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**Colitis-associated colon cancer: Is it in your genes?**

Van Der Kraak L *et al.* Genetics of CA-CRC

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**Abstract**

Colitis-associated colorectal cancer (CA-CRC) is the cause of death in 10%-15% of inflammatory bowel disease (IBD) patients. CA-CRC results from the accumulation of mutations in intestinal epithelial cells and progresses through a well-characterized inflammation to dysplasia to carcinoma sequence. Quantitative estimates of overall CA-CRC risks are highly variable ranging from 2% to 40% depending on IBD severity, duration and location, with IBD duration being the most significant risk factor associated with CA-CRC development. Recently, studies have identified IBD patients with similar patterns of colonic inflammation, but that differ with respect to CA-CRC development, suggesting a role for additional non-inflammatory risk factors in CA-CRC development. One suggestion is that select IBD patients carry polymorphisms in various low penetrance disease susceptibility genes, which pre-dispose them to CA-CRC development, although these loci have proven difficult to identify in human genome wide association studies. Mouse models of CA-CRC have provided a viable alternative for the discovery, validation and study of individual genes in CA-CRC pathology. In this review, we summarize the current CA-CRC literature with a strong focus on genetic pre-disposition and highlight an emerging role for mouse models in the search for CA-CRC risk alleles.

**Key words:** Colitis-associated colorectal cancer; Inflammatory bowel disease; Forward genetics; Susceptibility genes; Azoxymethane; Dextran sulfate sodium; Mouse models

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**Core tip:** Colitis-associated colorectal cancer (CA-CRC) is the cause of death in 10%-15% of inflammatory bowel disease (IBD) patients. Quantitative estimates of overall CA- CRC risk are highly variable and depend of the severity, duration and location of active IBD. Recently, studies have identified IBD patients with similar patterns of colonic inflammation, but that differ with respect to CA-CRC development. Suggesting a role for additional non-inflammatory risk factors in CA-CRC development. In this review, we summarize the current CA-CRC literature with a strong focus on genetic pre-disposition and highlight an emerging role for mouse models in the search for CA-CRC risk alleles.

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**INTRODUCTION**

Inflammatory bowel disease (IBD) is an umbrella term used to describe chronic-relapsing inflammatory conditions of the intestinal tract[[1](#_ENREF_1)]. While there are several subtypes of IBD, the two most common are Crohn’s disease (CD) and ulcerative colitis (UC). CD is characterized by inflammation throughout the entire gastro-intestinal tract with lesions most commonly found in the small intestine and proximal colon. Approximately 60% of CD patients have colonic involvement, with only 20% having isolated colonic disease[[2](#_ENREF_2)]. In CD, the inflammation is transmural, traversing multiple layers of the intestine, and typically occurs in patches[[1](#_ENREF_1)]. In UC, inflammation arises in the rectum and spreads proximally in a continuous manner, rarely extending into the small intestine and is confined to the mucosal layer. Worldwide IBD incidence rates are highly variable (UC: < 1-24.3/100000, CD: 1-20.2/100000), with higher incidence recorded in Western and Northern Europe, Australia and North America and lower incidence in Africa (excluding South Africa), Asia and South America[[3-5](#_ENREF_3)]. A large-scale meta-analysis of 107 IBD studies (57 CD and 50 UC) recently determined that CD incidence increased in 75% of CD studies and 60% of UC studies over a period of at least 10 years[[4](#_ENREF_4)]. As IBD patients exhibit a low mortality rate, the global prevalence of IBD is expected to increase in the coming years.

Colitis-associated colorectal cancer (CA-CRC), which develops in areas of active colonic inflammation, is listed as cause of death in 10%-15% of all IBD patients[[6](#_ENREF_6),[7](#_ENREF_7)]. As rates of IBD increase, rates of subsequent CA-CRC are also predicted to increase. From a colon cancer perspective, inflammation is the third most common CRC risk factor, after the hereditary CRC syndromes familial adenomatous polyposis coli (FAP) and hereditary non-polyposis colon cancer (HNPCC). However, unlike FAP and HNPCC, whose etiologies are well characterized, the specific etiologies underlying increased CA-CRC are still being elucidated. In this review, we briefly highlight the current literature with respect to CA-CRC etiology and epidemiology and compare and contrast CA-CRCs relative to non-inflammatory CRC conditions and IBD. In addition, we speculate on a possible function for genetic pre-disposing risk factors in CA-CRC and a role for animal models in elucidating these genetic effects.

**EPIDEMIOLOGY, ETIOLOGY AND SURVEILLANCE**

CA-CRC is listed as cause of death in 10-15% of IBD patients[[7](#_ENREF_7)]. CA-CRC mortality is approximately 50% (CD: 46%, UC: 50%) and this suggests that between 20%-30% of IBD patients will develop CA-CRC within their lifetime[[6](#_ENREF_6)]. Both UC- and CD-CRC are early-onset conditions presenting with an average age of onset between 40-55 years of age[[6](#_ENREF_6),[8-10](#_ENREF_8)]. UC-CRC is primarily identified in the rectum and sigmoid colon, whereas CD-CRC is more evenly distributed between the right-colon (ascending), sigmoid colon and rectum, although only a small proportion of CD patients have colonic disease[[6](#_ENREF_6),[11](#_ENREF_11)]. The differences with respect to tumor location may reflect differences in location of active IBD as 76% of CD-CRCs and 100% of UC-CRCs arise in areas of macroscopic IBD. CA-CRC patients often present at diagnosis with multiple synchronous carcinomas (CD-CRC: 11%, UC-CRC: 12%) and with lesions showing a high proportion of mucinous and signet ring features (CD-CRC: 29%, UC-CRC: 21%)[[6](#_ENREF_6)].

According to the American Cancer Society, individuals at increased risk for CA-CRC should undergo routine colonoscopy at 1-2 year intervals starting 8-12 years post-disease diagnosis ([www.ccfa.com](http://www.ccfa.com)). It is also recommended that at least four random colonic biopsies be taken for every 10 cm of colon examined during these routine colonoscopies, as approximately 20%-50% of colon dysplasia cannot be detected by visual inspection alone[[12](#_ENREF_12),[13](#_ENREF_13)]. Intraepithelial neoplasms are highly variable with respect to appearance and may present as raised (pendunculated or sessile) or flat (plaque or bump) lesions[[14](#_ENREF_14)]. Flat lesions are a unique feature to CA-CRC, rarely being detected in familial or sporadic CRC, and are generally associated with high risk of transformation into CA-CRC[[15](#_ENREF_15)]. The identification of CA-CRC can also be further complicated by large benign inflammatory pseudopolyps, which form during mucosal regeneration and ulcer healing.

***Inflammation in CA-CRC pathogenesis***

Quantitative estimates of overall CA-CRC risks are highly variable ranging from 2% to 40% depending on IBD severity, duration and location[[7](#_ENREF_7)]. CD patients with disease isolated to the small intestine only are not at increased risk of CD-CRC supporting the strong link between inflammation and CA-CRC[[6](#_ENREF_6)]. CRC risk in UC has been estimated at 2% after 10 years, 8% after 20 years and 18% after 30 years of disease[[8](#_ENREF_8)]. Studies of UC-CRC have also noted a high concordance between CA-CRC risks with location/extent of disease. For example, Ekbom *et al*[[16](#_ENREF_16)] identified a standardized incidence ratio (SIR) of 1.7 for proctitis (rectal only), 2.8 for left-sided colitis and 14.8 for pancolitis (defined as extensive colitis, or colitis involving the entire colon).

Studies of CD-CRC are complicated by vast heterogeneity with respect to CD anatomical sites. However, as with UC, CA-CRC risk associations have been correlated with duration/severity of disease. The relative risk (RR) of CD-CRC based on duration of disease was calculated to be 2.9, 5.6 and 8.3 after 10, 20 and 30 years of disease, respectively[[17](#_ENREF_17)]. In 2007, a CRC meta-analysis by disease site estimated a RR of 0.85, 4.3 and 13.4 for small bowel only, ileocolic and colon CD, respectively[[18](#_ENREF_18)]. CD-CRC RR is increased to 18.2 in patients with extensive disease.

One of the oldest and most prevalent treatments in IBD is administration of the non-steroidal anti-inflammatory (NSAID) drug 5-aminosalicylic acid (5-ASA) or its derivatives. 5-ASA modulates mucosal inflammation through several mechanisms including: the down regulation cyclooxygenase 2; inhibition of tumor necrosis factor alpha (TNF-α) and interleukin 1 beta (IL-1β); decreased nuclear factor kappa beta (NF-κβ) activation and modulation of peroxisome-proliferator activated receptor gamma (PPAR-γ)[[19](#_ENREF_19)]. While the protective effects of 5-ASA in IBD are well established, the literature examining 5-ASA as a chemo preventative agent in CA-CRC is controversial. Some studies have demonstrated up to a 97% reduction in CA-CRC risk in patients receiving regular 5-ASA therapy[[20-22](#_ENREF_20)]. However, recent studies tend to support no protective effects of regular 5-ASA use on CA-CRC risk[[23-25](#_ENREF_23)]. These discrepancies highlight the complex nature of CA-CRC. It also leads to questions regarding whether there may be certain non-inflammatory factors, such as genetic predisposition that may influence the efficacy of 5-ASA therapeutics.

CA-CRC initiation and progression is dependent on the accumulation of mutations in various tumor suppressors and oncogenes in intestinal epithelial cells[[26](#_ENREF_26)]. Support for inflammation as a key mediator in CA-CRC pathogenesis comes from animal studies showing increased DNA damage and tumor formation following extended periods of colitis in mice in the absence of a known DNA mutagen[[27](#_ENREF_27)]. The specific mechanisms through which inflammation regulates CA-CRC initiation and progression are not well understood. It has been suggested that reactive oxygen species (ROS) produced by immune cells during colitis may play a crucial role in promoting DNA damage. Epigenetics, cytokines and the microflora are also thought to be important in mediating cross talk between increased inflammation and CA-CRC and are reviewed in[[28](#_ENREF_28)].

***Primary sclerosing cholangitis***

Primary sclerosing cholangitis (PSC) is a rare disease characterized by inflammation, fibrosis and subsequent narrowing of the common bile duct. This narrowing leads to the accumulation of bile in the liver resulting in cirrhosis and future liver failure thus reducing life expectancy[[29](#_ENREF_29)]. There is a strong correlation between IBD and PSC, with approximately 70% (CI: 46.5% to 98.7%) of PSC patients presenting with concomitant IBD, usually in the form of UC[[30](#_ENREF_30)]. This corresponds to 8% of IBD patients developing coexisting PSC[[31](#_ENREF_31),[32](#_ENREF_32)]. The specific etiology underlying PSC development is complex, but similar to IBD is thought to arise due to a combination of genetic, environmental and microbial risk factors[[33](#_ENREF_33),[34](#_ENREF_34)].

In 2002, a large-scale meta-analysis concluded that PSC patients were at increased risk of developing CA-CRC compared to both IBD patients without PSC and the general population[[35](#_ENREF_35)]. While there has since been conflicting data concerning CA-CRC in PSC-IBD patients[[32](#_ENREF_32)], it is generally accepted that PSC is a risk factor associated with CA-CRC development. The explanation behind increased CRC in PSC-IBD patients remains elusive, but may be associated with increased levels of bile acid. Co-diagnosis of IBD and PSC is important to clinicians, as there is some evidence to suggest that treatment with ursodeoxycholic acid (UDCA) may reduce risk of CA-CRC, although additional testing is still necessary[[36](#_ENREF_36)].

***Evidence for non-inflammatory factors in CA-CRC pathogenesis***

In addition to strong evidence linking extent and duration of colonic inflammation to CA-CRC risk in IBD patients, there have recently been several observations in humans and mice to suggest a role for non-inflammatory factors in CA-CRC initiation/progression. Family history of CRC development is an important parameter to assess in IBD patients as a positive family history of CRC is associated with a 2-fold greater risk of developing CA-CRC[[37](#_ENREF_37)]. Studies of human UC and CD-CRC have also shown increased risk of CA-CRC in patients diagnosed with IBD at a young age. For UC-CRC, the absolute CRC risk 35 years post-diagnosis was 40% *vs* 30% in early-onset (age 15 or less) and late-onset UC patients diagnosed with pancolitis, respectively[[16](#_ENREF_16)]. This was subsequently confirmed in a large scale meta-analysis whereby patients with UC diagnosed prior to 25 years of age were 13 and 70 times more likely to develop CA-CRC compared to older UC patients and the general population, respectively[[38](#_ENREF_38)]. A similar trend was seen in CD with an increased RR of 21.5 *vs* 1.6 in patients younger and older than 25, respectively and subsequently confirmed in second unrelated CD cohort[[18](#_ENREF_18),[39](#_ENREF_39)]. The specific etiology underlying increased CA-CRC risk in younger onset IBD is still being investigated.

In 2014, Connolly *et al*[[40](#_ENREF_40)] compiled a cohort of UC patients to study the role of select IBD genes in CA-CRC. In this study, two cohorts were identified; patients with CA-CRC and those without, despite having similar amounts of UC-inflammation in both cohorts. Similar observations in mice with high levels of colonic inflammation, but low levels of CA-CRC have also been reported and will be discussed further in the mouse model section of this review[[41](#_ENREF_41),[42](#_ENREF_42)]. CA-CRC divergence among individuals with similar IBD status suggests a role for other non-inflammatory factors in mediating CA-CRC initiation or progression and it has been suggested that similar to many other complex traits, select IBD patients are genetically pre-disposed to developing CA-CRC.

**GENETIC ASSOCIATIONS IN HUMAN CA-CRC**

The “common disease, common variant” hypothesis, stipulates that common complex diseases, such as cancer, diabetes and IBD, arise in part due to common genetic variants (single nucleotide polymorphisms, SNPs) within the genome[[43](#_ENREF_43)]. To understand the rationale for hypothesizing genetic predisposition in CA-CRC, we must reiterate the similarities with respect to cancer progression between CA-CRC and a type of non-inflammatory CRC, often referred to as familial CRC.

***Familial vs colitis-associated colorectal cancers***

Genetically, CRCs can be categorized on a sliding scale of pre-disposing risk, which describes the predicted effect size of a given CRC risk variant compared to the minor allele frequency [(MAF), the abundance of the minor allele within a reference population][[44](#_ENREF_44)]. At one extreme, there are the rare, but well-characterized Mendelian or Hereditary CRC syndromes, such as FAP or HNPCC, whose mutations are associated with a high penetrance of disease symptoms and are easily identified in large families with multiple affected individuals. At the other extreme are familial CRCs, which present with fewer affected individuals per family, and arise, in part, due to common genetic variants within a class of genes known as low penetrance tumor susceptibility genes[[45](#_ENREF_45)].

In familial CRCs, like most cancers, the balance between cell proliferation, differentiation and apoptosis becomes progressively disrupted through the accumulation of mutations in several signaling pathways encompassing *WNT*, *RAS*, *p53*, *DCC* and *TGF-β* genes.This is referred to as the adenoma-carcinoma sequence progression[[46](#_ENREF_46)]. Analysis of invasive familial and CA tumors show a similar pattern of acquired molecular alterations and hence CA-CRC was originally categorized as a subtype of familial CRC. This led to speculation that low penetrance tumor susceptibility genes, which are important in familial CRC, could also be important in CA-CRC initiation and progression.

However, the timing and frequency of these genetic events differ between familial and CA-CRC and therefore it has been hypothesized that variants in different genes may be associated with both cancers. For example, mutations/deletions of *p53* are early events in CA-CRC with 50% of ulcerative colitis (UC) patients having *p53* mutations compared to ~10% of non-inflammatory adenomas (Figure 1)[[28](#_ENREF_28),[47](#_ENREF_47)]. But *APC* mutations are rare events in CA-CRC (27.5% of high grade dysplasia) compared to 50% in non-CA-CRC adenomas[[28](#_ENREF_28),[48](#_ENREF_48)].

CA-CRCs progress through the colitis-dysplasia-carcinoma sequence associated with the development of inflammation, indefinite, low-grade and high-grade dysplasia, with eventual progression to carcinoma (Figure 1)[[26](#_ENREF_26)]. Dysplasia describes the abnormal growth and development of colon cells, with indefinite dysplasia describing early changes that cannot be categorized as either negative or positive for dysplasia. It is interesting to note that key inflammatory mediators such as reactive oxygen and nitrogen species (ROS and NOS), as well as chemokines and cytokines (IL-6, STAT3, TNF-α, IL-10, IL-12 and IL-23) all participate to orchestrate the conversion of a normal epithelium to indefinite dysplasia, which again highlights a variable role for inflammation in CA-CRC transformation[[28](#_ENREF_28)].

***Genome wide association studies***

The completion of the Human Genome Project, the International HapMap Project and increased technological power has led to the advent of genome-wide association studies (GWAS)[[49](#_ENREF_49)]. GWAS compare the prevalence of thousands of common genetic variants [single nucleotide polymorphisms (SNPs)] within healthy (control) and disease (case) cohorts looking for allelic imbalance indicative of disease association[[49](#_ENREF_49)].

Both IBD and familial CRC have been associated with polymorphisms in low penetrance disease susceptibility genes, with numerous positive associations detected in GWAS. For IBD, more than 200 loci have been identified, the largest number for any common complex disease (<http://genome.gov/gwastudies>). As IBD pathogenesis is driven by aberrant immune responses against the commensal bacteria of the lumen, it is not surprising that a large number of genes within IBD loci have been associated with epithelial barrier maintenance and permeability, cytokine signaling and pathogen recognition/clearance[[50](#_ENREF_50)]. Some of the most well characterized genetic associations are *NOD2*, *IL-23R* and *ATG16L1* involved in bacterial sensing, the IL-23 inflammatory response and autophagy, respectively[[51](#_ENREF_51)]. To date more than 40 loci have been associated with familial CRC (<http://genome.gov/gwastudies>), with many of the SNPs mapping to regions in strong linkage disequilibrium (LD) with members of the TGF-β signaling pathway, highlighting an important role for TGF-β signaling in CRC[[52](#_ENREF_52)]. There is little overlap between known IBD and CRC loci in humans suggesting different etiologies to both diseases.

***Lack of GWAS for colitis-associated colon cancer***

Unlike familial CRC and IBD, there have been very few GWAS performed to identify genetic loci regulating susceptibility to CA-CRC. In part, this maybe due to high numbers of IBD patients undergoing colectomy, making identification of IBD patients with and without CA-CRC difficult[[53](#_ENREF_53),[54](#_ENREF_54)].CA-CRC is influenced by numerous risk factors including age at IBD-onset, and duration/extent of IBD colonic involvement[[7](#_ENREF_7),[38](#_ENREF_38)]. While not essential for early CA-CRC GWAS it may also be important to segregate CA-CRC patients into categories associated with differences with respect to age of diagnosis, ethnicity and extent of inflammation, as different genes may underlie CA-CRC in different colonic microenvironments.

In 2009, the UK IBD Genetics Consortium identified and published a novel UC locus situated on chr. 16 (16q22)[[55](#_ENREF_55)]. Interestingly, this locus had previously been associated with increased CRC risk[[56](#_ENREF_56)]. Therefore, it has been speculated that this locus may also play an important role in CA-CRC. However, a recent study showed no association between any known UC loci and UC-CRC risk, disproving this hypothesis[[40](#_ENREF_40)]. Recently, the STAT3 locus has been associated with both IBD and CA-CRC, exerting its effects in a TP53-dependent manner. This is a promising step in the identification of CA-CRC loci in humans [[57](#_ENREF_57)].

**MOUSE MODELS OF COLITIS-ASSOCIATED COLON CANCER**

The complex and heterogeneous genetic component of complex diseases can be difficult to tease apart in human populations due to confounding environmental and lifestyle variables. However, these traits can be dissected in genetically well-defined inbred and recombinant congenic mouse strains[[58](#_ENREF_58)]. Mice are not particularly prone to the spontaneous development of IBD or CRC and therefore disease induction in mice can be performed using dietary modifications, infectious agents, genetic mutation or chemical reagents[[59](#_ENREF_59)]. To date more than 100 different mouse models of CRC, IBD, and CA-CRC have been published. For a comprehensive review of these, see[[58](#_ENREF_58),[60-62](#_ENREF_60)].

We have narrowed the focus of this review to three relevant areas; the *Il-10* knockout genetic model of colitis/CA-CRC, the AOM/DSS model of CA-CRC and the mapping of genetic loci regulating susceptibility to CA-CRC using forward genetic approaches.

***The Il-10 model of colitis and CA-CRC***

Many common colitis and CA-CRC models involve deleting the expression of a specific gene or multiple genes. These models are associated with an increase in IBD (either UC or CD) with or without subsequent CA-CRC[[60](#_ENREF_60)]. The most-well characterized of these genetic models involves the deletion of the *Il-10* gene encoding a pleiotropic anti-inflammatory cytokine produced by monocytes and lymphocytes that acts to dampen and terminate immune responses[[63](#_ENREF_63)]. In 1993, Kuhn *et al*[[64](#_ENREF_64)]generated the 129/B6 *Il-10-/-*knockout mouse line (*Il-10tm1Cgn*). These mice showed a high incidence of weight loss, anemia and enterocolitis 1-3 mo after birth. Enterocolitis was first detected in the proximal colon and then in the remaining colon, the duodenum and the proximal jejunum of the small intestine and mimics human CD, associated with discontinuous, transmural inflammation, ulceration and thickening of the bowel wall[[64-66](#_ENREF_64)].

Enterocolitis in *Il-10*-/- mice is strain-dependent, suggesting a strong role for genetic factors in disease pathogenesis. The most sensitive genetic backgrounds are C3H/HeJBir and 129/Sv, with 100% of the mice developing severe colitis before 3 mo of age[[66](#_ENREF_66),[67](#_ENREF_67)]. C3H/HeJBir mice, with a wild type *1l-10* gene are also susceptible to spontaneous colitis[[60](#_ENREF_60)]. However, CA-CRC susceptibility has not been assessed in the C3H/HeJBir *Il-10*-/- mice[[67](#_ENREF_67)]. On the 129/Sv background, 67% of the mice develop adenocarcinomas in the first 6 months of life[[66](#_ENREF_66)]. As evaluated by histopathology, BALB/cJ *1l-10*-/- mice have a higher incidence of spontaneous colitis (100%) compared to B6 *1l-10*-/- mice (57%) at 3 mo of age, but a lower incidence of colonic tumors (29%) at 6 mo of age compared to 129/Sv *1l-10*-/- mice. B6 *1l-10*-/- mice do not develop colonic adenocarcinomas within this timeframe. NOD/LtJ *1l-10*-/- mice also develop severe colitis, associated with 100% incidence of rectal prolapse, although the time frame for disease development was not specified[[68](#_ENREF_68)]. These NOD/LtJ *1l-10*-/- mice are not good models for CA-CRC as high incidence of rectal prolapse prevents long-term studies in these mice. Together, these studies highlight an important role for genetic backgrounds in colitis and CA-CRC susceptibility.

Generally, experiments of colitis and CA-CRC in *Il-10*-deficient mice support a strong role for inflammation as the driving factor underlying increased CA-CRC risk. However, an exception to this is a study from Arthur *et al*[[69](#_ENREF_69)] who demonstrated similar inflammatory profiles in *Il-10-/-* mice infected with *E. faecalis* and *E. coli*, with only the latter being associated with increased CA-CRC, supporting a role for non-inflammatory mediators of CA-CRC.

***The AOM/DSS model of CA-CRC***

Chemical models of colitis and CA-CRC are advantageous as treatments are relatively inexpensive and easy to administer producing highly reproducible results. These models offer a distinct advantage compared to genetic models as time of onset, duration and severity of colitis/CA-CRC can be adjusted by changing the dose and/or length of the treatment protocol. In addition, unlike genetic models of colitis and CA-CRC, the inflammatory agents can be removed and thus the healing/regeneration process can be studied in detail. In addition, these models highlight a probable role for genetic factors in CA-CRC, with some mice developing high levels of colonic inflammation, yet low levels of CA-CRC and *vice versa*.

In 2003, Tanaka *et al*[[70](#_ENREF_70)] published results showing that a single azoxymethane (AOM) injection in CD-1 mice, followed a week later by a 7-d dextran sulfate sodium (DSS) treatment, was sufficient to induce macroscopically visible tumors 20 wk post-initiation. Mice treated with only a single AOM or single DSS injection did not develop tumors within this 20-wk period, suggesting that combined administration of AOM and DSS is essential for tumorigenesis. This AOM/DSS protocol has since become one of the most popular models to study the influence of dietary, microbial and genetic factors of CA-CRC progression and initiation[[58](#_ENREF_58)]. Interestingly, permissive mice given multiple injections of AOM develop CRC, reminiscent of human familial CRC, while those given DSS-alone develop an UC-like phenotype. This allows for common and unique genetic signatures to be identified between the AOM/DSS CA-CRC protocol and the AOM-only CRC and DSS-only IBD protocols.

AOM is a colon specific carcinogen that, when activated, generates a methyl cation that can react with deoxyguanosine at either the N7 or O6 position; with the latter leading to the formation of deoxymethylguanosine, resulting in mismatched base pairing and subsequent G to A transitions. DSS is a long chain (5-140 kDa), negatively charged polysaccharide derived from the esterification of dextran and chlorosulfonic acid[[71](#_ENREF_71)]. When administered to rodents in drinking water, DSS is a highly potent inducer of colitis, mimicking human UC[[72](#_ENREF_72)]. The location of colitis is highly dependent on the DSS molecular weight, with low weight DSS (5 kDa) inducing lesions in the cecum and proximal colon, mid weight (40 kDa) DSS provoking lesions in the mid and distal colon and high weight DSS (500 kDa) failing to induce colitis in mice[[73](#_ENREF_73)]. All future mention of DSS refers to mid-weight (~36-54 kDa) DSS.

Inbred strains of mice differ with respect to AOM/DSS-induced CA-CRC susceptibility, with strains such as BALB/c, Swiss Webster, CBA/J, CD1, A/J and FVB/NJ behaving as susceptible and strains such as C3H/HeJ, C57Bl/6 (B6) and DBA/2J being resistant[[70](#_ENREF_70),[74-78](#_ENREF_74)]. Testing for DSS-induced colitis in some strains, such as BALB/c, CBA/J and DBA/2J, suggests that the extent of colonic inflammation is an important driver for CA-CRC[[41](#_ENREF_41),[72](#_ENREF_72)]. However, C3H/HeJ mice are highly susceptible to DSS-induced colitis, yet resistant to CA-CRC, suggesting that inflammation alone does not determine CA-CRC susceptibility[[41](#_ENREF_41)]. We have also shown that A/J mice, while more susceptible to CA-CRC than B6 mice, develop lower levels of overall colonic inflammation compared to B6 mice following AOM/DSS treatment[[78](#_ENREF_78)]. Studies of myeloid translocation gene, related 1 (*Mtgr1*) gene deficiency in mice have demonstrated reduced tumor burden following AOM/DSS treatment despite an increased colonic inflammation, again suggesting a role for non-inflammatory, possibly genetic factors in CA-CRC[[79](#_ENREF_79)].

Recently, Gao *et al*[[80](#_ENREF_80)] compared global gene expression patterns in untreated BALB/c inbred mice, to those treated with AOM/DSS, AOM-only and DSS-only. As expected, both the DSS- and AOM/DSS-treated mice showed evidence of increased colonic inflammation, which was notably absent in the AOM-only and untreated mice. However, despite the strong influence of inflammation in the AOM/DSS-treated mice, ~50% of the identified differentially expressed genes were unique to the AOM/DSS treatment group and were not observed in the AOM or DSS-only groups. Li *et al*[[81](#_ENREF_81)] also recorded unique genetic signatures association with AOM/DSS-induced CA-CRC, compared to chronic murine colitis, confirming this observation. Collectively, these studies suggest that CA-CRC susceptibility is associated with unique genetic signatures. Identification of genes specific to CA-CRC may aid in the identification of IBD patients with rapid onset CA-CRC or those who develop CA-CRC despite low levels of colonic inflammation.

***Mouse loci identified using forward genetic studies***

The identification of genes associated with the development of various complex traits, can be identified using either forward or reverse genetic approaches in mice. Forward genetics is a phenotype-driven approach in which mutations are identified underlying disease traits through the generation of informative mouse crosses followed by linkage analysis[[82](#_ENREF_82)]. This is the converse of reverse genetics in which a range of phenotypes is characterized for a given genetic mutation[[83](#_ENREF_83)]. Reverse studies are easier to conduct and are shorter in duration than forward genetic studies, but can be hampered by inefficient knockdown or genetic background effects[[83](#_ENREF_83),[84](#_ENREF_84)]. In addition, forward genetic screens are advantageous as they are conducted without bias as to the types of mutations detected, with mutations mapping to genes that are often unlikely to be tested using reverse genetic approaches and represent a spectrum of mutations more likely to be detected in human disease. Forward genetic studies typically use 4 distinct types of mouse populations; F2, N2, recombinant congenic mice (RCS) or recombinant inbred mice (RI), often using more than one informative population.

Numerous non-inflammatory CRC and IBD susceptibility loci have been mapped in mice using a forward genetics approach and therefore, it is not improbable to hypothesize that CA-CRC loci could be identified using the same approach. These forward genetic approaches are possible as inbred mice and differ with respect to susceptibility to all three of the above diseases. Figure 2 summarizes the known IBD, CRC and CA-CRC loci mapped using a forward genetics approach. With respect to IBD, these loci have been mapped using spontaneous (SAMP1/YitFC), chemical (DSS, TNBS), genetic (*Il-10-/-, Gnai-/-, Gpx1/2-/-*) and infectious (*Helicobacter, Trichuris muris*) models of colitis, while non-inflammatory CRC loci have been mapped using the *ApcMin+/-* mouse model of CRC (mimicking FAP) and AOM (or the AOM precursor dimethylhydrazine)-only models[[42](#_ENREF_42),[85-101](#_ENREF_85)]. Despite differences with respect to strains of mice tested and models of disease induction used, these studies share a common feature, *i.e.,* each cross identified multiple genetic loci regulating susceptibility and each locus controls a small proportion of the phenotypic variation (< 20%).

However, only 3 CA-CRC loci have been identified (Figure 2). The first locus referred to as *Hiccs*, regulates *Helicobacter* *hepaticus*-induced colitis and CA-CRC susceptibility[[88](#_ENREF_88)]. This *Helicobacter* model however, is a poor recapitulation of human disease, with mice developing lesions exclusively in the proximal colon.

Our laboratory has also mapped two additional loci that regulate CA-CRC susceptibility. To map these loci, we first defined that A/J mice, contrary to B6 mice, were susceptible to AOM/DSS-induced CA-CRC. Then, using forward genetics and (A/J X B6) F1 and F2 cohorts, we identified and mapped a novel A/J-derived CA-CRC susceptibility locus to mouse chromosome 9, centered around marker D9Mit67. This novel locus was named *Ccs4*[[78](#_ENREF_78)]. Further analyses of (A/J X B6) F2 mice homozygous for A/J alleles at *Ccs4* identified a second locus on the distal part of mouse chromosome 14 (peak marker rs13482311, 93.5 Mb) that acts to regulate tumor susceptibility in an additive fashion with the *Ccs4* locus. F2 mice homozygous for A/J alleles at both loci (*Ccs4* and chromosome 14) are as susceptible to CA-CRC as the A/J controls, while mice homozygous for B6 alleles are as resistant as the B6 controls, thus supporting the role of two loci in this CA-CRC model. Two locus systems are rarely identified in human GWAS studies, in part due to the low penetrance of the second locus. The ability to detect such interactions in mice provides a framework to search for such associations in humans. In our studies, we also detected higher levels of inflammation in the resistant B6 colons, suggesting that elevated inflammation is not the primary driver of this differential CA-CRC susceptibility. It is interesting to note that an unrelated locus on chromosome 3, namely *Ccs3*, is the primary driver of AOM-induced CRC susceptibility in these same strains, suggesting that these CRC loci may be specific to CA-CRC[[102](#_ENREF_102)]. The success of this initial genetic screen has led us to hypothesize that other novel genetic factors may also regulate susceptibility to CA-CRC in different inbred mouse strains, which we are currently assessing.

#

**CONCLUSION**

# CA-CRC is a complex disease arising from a combination of dietary, lifestyle, microbial and genetic factors. In addition, disease risk is tightly correlated with severity, location and duration of colonic inflammation (IBD). CA-CRC risk is increased in early-onset IBD patients and this specific subset of IBD patients is increasing in North America, suggesting that CA-CRC may be a growing concern for future generations[[103](#_ENREF_103)]. It has been well established that various reverse genetic approaches are ideal in identifying and mapping novel genes associated with increased inflammation and subsequent CA-CRC. However, we have recently shown that we can use the common AOM/DSS model of CA-CRC to identify and map novel loci regulating susceptibility to CA-CRC. By identifying parental strains for mapping, discordant with respect to their colitis and CA-CRC phenotype, we can increase the probability of identifying genetic factors specific to CA-CRC and not factors associated with increased colitis. Such loci can then be assessed in human cohorts, with the hope of identifying patients at high risk for colitis to CA-CRC transformation.

**REFERENCES**

1 Crohn's and Colitis Foundation of Canada. The Impact of Inflammatory Bowel Disease in Canada: 2012 Final Report and Recommendations, 2012

2 **Loftus EV**, Schoenfeld P, Sandborn WJ. The epidemiology and natural history of Crohn's disease in population-based patient cohorts from North America: a systematic review. *Aliment Pharmacol Ther* 2002; **16**: 51-60 [PMID: 11856078]

3 **O'Brien KD**, Corkill CM. The specialist orthodontic practitioner. The 1989 survey. *Br Dent J* 1990; **168**: 471-475 [PMID: 2369542 DOI: 10.1097/MOG.0b013e32836229fb]

4 **Cho J**, Pastorino S, Zeng Q, Xu X, Johnson W, Vandenberg S, Verhaak R, Cherniack AD, Watanabe H, Dutt A, Kwon J, Chao YS, Onofrio RC, Chiang D, Yuza Y, Kesari S, Meyerson M. Glioblastoma-derived epidermal growth factor receptor carboxyl-terminal deletion mutants are transforming and are sensitive to EGFR-directed therapies. *Cancer Res* 2011; **71**: 7587-7596 [PMID: 22001862 DOI: 10.1053/j.gastro.2011.10.001]

5 **Cosnes J**, Gower-Rousseau C, Seksik P, Cortot A. Epidemiology and natural history of inflammatory bowel diseases. *Gastroenterology* 2011; **140**: 1785-1794 [PMID: 21530745 DOI: 10.1053/j.gastro.2011.01.055]

6 **Choi PM**, Zelig MP. Similarity of colorectal cancer in Crohn's disease and ulcerative colitis: implications for carcinogenesis and prevention. *Gut* 1994; **35**: 950-954 [PMID: 8063223]

7 **Mattar MC**, Lough D, Pishvaian MJ, Charabaty A. Current management of inflammatory bowel disease and colorectal cancer. *Gastrointest Cancer Res* 2011; **4**: 53-61 [PMID: 21673876]

8 **Eaden JA**, Abrams KR, Mayberry JF. The risk of colorectal cancer in ulcerative colitis: a meta-analysis. *Gut* 2001; **48**: 526-535 [PMID: 11247898]

9 **Jess T**, Rungoe C, Peyrin-Biroulet L. Risk of colorectal cancer in patients with ulcerative colitis: a meta-analysis of population-based cohort studies. *Clin Gastroenterol Hepatol* 2012; **10**: 639-645 [PMID: 22289873 DOI: 10.1016/j.cgh.2012.01.010]

10 **Lakatos L**, Mester G, Erdelyi Z, David G, Pandur T, Balogh M, Fischer S, Vargha P, Lakatos PL. Risk factors for ulcerative colitis-associated colorectal cancer in a Hungarian cohort of patients with ulcerative colitis: results of a population-based study. *Inflamm Bowel Dis* 2006; **12**: 205-211 [PMID: 16534422]

11 **Rutter MD**, Saunders BP, Wilkinson KH, Kamm MA, Williams CB, Forbes A. Most dysplasia in ulcerative colitis is visible at colonoscopy. *Gastrointest Endosc* 2004; **60**: 334-339 [PMID: 15332019]

12 **Bernstein CN**, Shanahan F, Weinstein WM. Are we telling patients the truth about surveillance colonoscopy in ulcerative colitis? *Lancet* 1994; **343**: 71-74 [PMID: 7903776]

13 **Mescoli C**, Frego M, Rugge M. Pathology of dysplasia and cancer in inflammatory bowel disease. *Ann Ital Chir* 2010; **82**: 11-18 [PMID: 21657150]

14 **Bressenot A**, Cahn V, Danese S, Peyrin-Biroulet L. Microscopic features of colorectal neoplasia in inflammatory bowel diseases. *World J Gastroenterol* 2014; **20**: 3164-3172 [PMID: 24696602 DOI: 10.3748/wjg.v20.i12.3164]

15 **Geboes K**. Review article: what are the important endoscopic lesions for detection of dysplasia in inflammatory bowel disease? *Aliment Pharmacol Ther* 2006; **24 Suppl 3**: 50-55 [PMID: 16961746]

16 **Ekbom A**, Helmick C, Zack M, Adami HO. Ulcerative colitis and colorectal cancer. A population-based study. *N Engl J Med* 1990; **323**: 1228-1233 [PMID: 2215606]

17 **Canavan C**, Abrams KR, Mayberry J. Meta-analysis: colorectal and small bowel cancer risk in patients with Crohn's disease. *Aliment Pharmacol Ther* 2006; **23**: 1097-1104 [PMID: 16611269]

18 **von Roon AC**, Reese G, Teare J, Constantinides V, Darzi AW, Tekkis PP. The risk of cancer in patients with Crohn's disease. *Dis Colon Rectum* 2007; **50**: 839-855 [PMID: 17308939]

19 **Perrotta C**, Pellegrino P, Moroni E, De Palma C, Cervia D, Danelli P, Clementi E. Five-aminosalicylic Acid: an update for the reappraisal of an old drug. Gastroenterol Res Pract 2015; 2015: 456895 [PMID: PMC4320793 DOI: 10.1155/2015/456895]

20 **Eaden J**, Abrams K, Ekbom A, Jackson E, Mayberry J. Colorectal cancer prevention in ulcerative colitis: a case-control study. *Aliment Pharmacol Ther* 2000; **14**: 145-153 [PMID: 10651654]

21 **Velayos FS**, Terdiman JP, Walsh JM. Effect of 5-aminosalicylate use on colorectal cancer and dysplasia risk: a systematic review and metaanalysis of observational studies. *Am J Gastroenterol* 2005; **100**: 1345-1353 [PMID: 15929768]

22 **van Staa TP**, Card T, Logan RF, Leufkens HG. 5-Aminosalicylate use and colorectal cancer risk in inflammatory bowel disease: a large epidemiological study. *Gut* 2005; **54**: 1573-1578 [PMID: 15994215]

23 **Terdiman JP**, Steinbuch M, Blumentals WA, Ullman TA, Rubin DT. 5-Aminosalicylic acid therapy and the risk of colorectal cancer among patients with inflammatory bowel disease. *Inflamm Bowel Dis* 2007; **13**: 367-371 [PMID: 17206695]

24 **Bernstein CN**, Blanchard JF, Metge C, Yogendran M. Does the use of 5-aminosalicylates in inflammatory bowel disease prevent the development of colorectal cancer? *Am J Gastroenterol* 2003; **98**: 2784-2788 [PMID: 14687833]

25 **Nguyen GC**, Wu H, Gulamhusein A, Rosenberg M, Thanabalan R, Yeo EL, Bernstein CN, Steinhart AH, Margolis M. The utility of screening for asymptomatic lower extremity deep venous thrombosis during inflammatory bowel disease flares: a pilot study. *Inflamm Bowel Dis* 2013; **19**: 1053-1058 [PMID: 23429463 DOI: 10.1097/MIB.0b013e3182802a65]

26 **Itzkowitz SH**, Yio X. Inflammation and cancer IV. Colorectal cancer in inflammatory bowel disease: the role of inflammation. *Am J Physiol Gastrointest Liver Physiol* 2004; **287**: G7-17 [PMID: 15194558]

27 **Grivennikov SI**. Inflammation and colorectal cancer: colitis-associated neoplasia. *Semin Immunopathol* 2013; **35**: 229-244 [PMID: 23161445 DOI: 10.1007/s00281-012-0352-6]

28 **Foersch S**, Neurath MF. Colitis-associated neoplasia: molecular basis and clinical translation. *Cell Mol Life Sci* 2014; **71**: 3523-3535 [PMID: 24830703 DOI: 10.1007/s00018-014-1636-x]

29 **Navaneethan U**, Shen B. Hepatopancreatobiliary manifestations and complications associated with inflammatory bowel disease. *Inflamm Bowel Dis* 2010; **16**: 1598-1619 [PMID: 20198712 DOI: 10.1002/ibd.21219]

30 **de Vries AB**, Janse M, Blokzijl H, Weersma RK. Distinctive inflammatory bowel disease phenotype in primary sclerosing cholangitis. *World J Gastroenterol* 2015; **21**: 1956-1971 [PMID: 25684965 DOI: 10.3748/wjg.v21.i6.1956]

31 **Loftus EV**, Harewood GC, Loftus CG, Tremaine WJ, Harmsen WS, Zinsmeister AR, Jewell DA, Sandborn WJ. PSC-IBD: a unique form of inflammatory bowel disease associated with primary sclerosing cholangitis. *Gut* 2005; **54**: 91-96 [PMID: 15591511]

32 **Wang R**, Leong RW. Primary sclerosing cholangitis as an independent risk factor for colorectal cancer in the context of inflammatory bowel disease: a review of the literature. *World J Gastroenterol* 2014; **20**: 8783-8789 [PMID: 25083052 DOI: 10.3748/wjg.v20.i27.8783]

33 **Eaton JE**, Talwalkar JA, Lazaridis KN, Gores GJ, Lindor KD. Pathogenesis of primary sclerosing cholangitis and advances in diagnosis and management. *Gastroenterology* 2013; **145**: 521-536 [PMID: 23827861 DOI: 10.1053/j.gastro.2013.06.052]

34 **Henriksen EK**, Melum E, Karlsen TH. Update on primary sclerosing cholangitis genetics. *Curr Opin Gastroenterol* 2014; **30**: 310-319 [PMID: 24565892 DOI: 10.1097/MOG.0000000000000052]

35 **Soetikno RM**, Lin OS, Heidenreich PA, Young HS, Blackstone MO. Increased risk of colorectal neoplasia in patients with primary sclerosing cholangitis and ulcerative colitis: a meta-analysis. *Gastrointest Endosc* 2002; **56**: 48-54 [PMID: 12085034]

36 **Broomé U**, Bergquist A. Primary sclerosing cholangitis, inflammatory bowel disease, and colon cancer. *Semin Liver Dis* 2006; **26**: 31-41 [PMID: 16496231]

37 **Askling J**, Dickman PW, Karlén P, Broström O, Lapidus A, Löfberg R, Ekbom A. Family history as a risk factor for colorectal cancer in inflammatory bowel disease. *Gastroenterology* 2001; **120**: 1356-1362 [PMID: 11313305]

38 **Lutgens MW**, van Oijen MG, van der Heijden GJ, Vleggaar FP, Siersema PD, Oldenburg B. Declining risk of colorectal cancer in inflammatory bowel disease: an updated meta-analysis of population-based cohort studies. *Inflamm Bowel Dis* 2013; **19**: 789-799 [PMID: 23448792 DOI: 10.1097/MIB.0b013e31828029c0]

39 **Ekbom A**, Helmick C, Zack M, Adami HO. Increased risk of large-bowel cancer in Crohn's disease with colonic involvement. *Lancet* 1990; **336**: 357-359 [PMID: 1975343]

40 **Connelly TM**, Berg AS, Harris LR, Brinton DL, Hegarty JP, Deiling SM, Stewart DB, Koltun WA. Ulcerative colitis neoplasia is not associated with common inflammatory bowel disease single-nucleotide polymorphisms. *Surgery* 2014; **156**: 253-262 [PMID: 24947639 DOI: 10.1016/j.surg.2014.03.017]

41 **Mähler M**, Bristol IJ, Leiter EH, Workman AE, Birkenmeier EH, Elson CO, Sundberg JP. Differential susceptibility of inbred mouse strains to dextran sulfate sodium-induced colitis. *Am J Physiol* 1998; **274**: G544-G551 [PMID: 9530156]

42 **Mähler M**, Bristol IJ, Sundberg JP, Churchill GA, Birkenmeier EH, Elson CO, Leiter EH. Genetic analysis of susceptibility to dextran sulfate sodium-induced colitis in mice. *Genomics* 1999; **55**: 147-156 [PMID: 9933561]

43 **Iyengar SK**, Elston RC. The genetic basis of complex traits: rare variants or "common gene, common disease"? *Methods Mol Biol* 2007; **376**: 71-84 [PMID: 17984539]

44 **Chung CC**, Magalhaes WC, Gonzalez-Bosquet J, Chanock SJ. Genome-wide association studies in cancer--current and future directions. *Carcinogenesis* 2010; **31**: 111-120 [PMID: 19906782 DOI: 10.1093/carcin/bgp273]

45 **Houlston RS**, Peto J. The search for low-penetrance cancer susceptibility alleles. *Oncogene* 2004; **23**: 6471-6476 [PMID: 15322517]

46 . Kinzler, K., Vogelstein B., Lessons from hereditary colorectal cancers. Cell, 1996. 87: p. 159-70.

47 **Baker SJ**, Preisinger AC, Jessup JM, Paraskeva C, Markowitz S, Willson JK, Hamilton S, Vogelstein B. p53 gene mutations occur in combination with 17p allelic deletions as late events in colorectal tumorigenesis. *Cancer Res* 1990; **50**: 7717-7722 [PMID: 2253215]

48 **Goss KH**, Groden J. Biology of the adenomatous polyposis coli tumor suppressor. *J Clin Oncol* 2000; **18**: 1967-1979 [PMID: 10784639]

49 **Melum E**, Franke A, Karlsen TH. Genome-wide association studies--a summary for the clinical gastroenterologist. *World J Gastroenterol* 2009; **15**: 5377-5396 [PMID: 19916168]

50 **Khor B**, Gardet A, Xavier RJ. Genetics and pathogenesis of inflammatory bowel disease. *Nature* 2011; **474**: 307-317 [PMID: 21677747 DOI: 10.1038/nature]

51 **Naser SA**, Arce M, Khaja A, Fernandez M, Naser N, Elwasila S, Thanigachalam S. Role of ATG16L, NOD2 and IL23R in Crohn's disease pathogenesis. *World J Gastroenterol* 2012; **18**: 412-424 [PMID: 22346247 DOI: 10.3748/wjg.v18.i5.412]

52 **Tomlinson IP**, Carvajal-Carmona LG, Dobbins SE, Tenesa A, Jones AM, Howarth K, Palles C, Broderick P, Jaeger EE, Farrington S, Lewis A, Prendergast JG, Pittman AM, Theodoratou E, Olver B, Walker M, Penegar S, Barclay E, Whiffin N, Martin L, Ballereau S, Lloyd A, Gorman M, Lubbe S, Howie B, Marchini J, Ruiz-Ponte C, Fernandez-Rozadilla C, Castells A, Carracedo A, Castellvi-Bel S, Duggan D, Conti D, Cazier JB, Campbell H, Sieber O, Lipton L, Gibbs P, Martin NG, Montgomery GW, Young J, Baird PN, Gallinger S, Newcomb P, Hopper J, Jenkins MA, Aaltonen LA, Kerr DJ, Cheadle J, Pharoah P, Casey G, Houlston RS, Dunlop MG. Multiple common susceptibility variants near BMP pathway loci GREM1, BMP4, and BMP2 explain part of the missing heritability of colorectal cancer. *PLoS Genet* 2011; **7**: e1002105 [PMID: 21655089 DOI: 10.1371/journal.pgen.1002105]

53 **Gillen CD**, Walmsley RS, Prior P, Andrews HA, Allan RN. Ulcerative colitis and Crohn's disease: a comparison of the colorectal cancer risk in extensive colitis. *Gut* 1994; **35**: 1590-1592 [PMID: 7828978]

54 **Winther KV**, Jess T, Langholz E, Munkholm P, Binder V. Long-term risk of cancer in ulcerative colitis: a population-based cohort study from Copenhagen County. *Clin Gastroenterol Hepatol* 2004; **2**: 1088-1095 [PMID: 15625654]

55 **UK IBD Genetics Consortium**, Barrett JC, Lee JC, Lees CW, Prescott NJ, Anderson CA, Phillips A, Wesley E, Parnell K, Zhang H, Drummond H, Nimmo ER, Massey D, Blaszczyk K, Elliott T, Cotterill L, Dallal H, Lobo AJ, Mowat C, Sanderson JD, Jewell DP, Newman WG, Edwards C, Ahmad T, Mansfield JC, Satsangi J, Parkes M, Mathew CG; Wellcome Trust Case Control Consortium 2, Donnelly P, Peltonen L, Blackwell JM, Bramon E, Brown MA, Casas JP, Corvin A, Craddock N, Deloukas P, Duncanson A, Jankowski J, Markus HS, Mathew CG, McCarthy MI, Palmer CN, Plomin R, Rautanen A, Sawcer SJ, Samani N, Trembath RC, Viswanathan AC, Wood N, Spencer CC, Barrett JC, Bellenguez C, Davison D, Freeman C, Strange A, Donnelly P, Langford C, Hunt SE, Edkins S, Gwilliam R, Blackburn H, Bumpstead SJ, Dronov S, Gillman M, Gray E, Hammond N, Jayakumar A, McCann OT, Liddle J, Perez ML, Potter SC, Ravindrarajah R, Ricketts M, Waller M, Weston P, Widaa S, Whittaker P, Deloukas P, Peltonen L, Mathew CG, Blackwell JM, Brown MA, Corvin A, McCarthy MI, Spencer CC, Attwood AP, Stephens J, Sambrook J, Ouwehand WH, McArdle WL, Ring SM, Strachan DP. Genome-wide association study of ulcerative colitis identifies three new susceptibility loci, including the HNF4A region. *Nat Genet* 2009; **41**: 1330-1334 [PMID: 19915572 DOI: 10.1038/ng.483]

56 **Houlston RS**, Cheadle J, Dobbins SE, Tenesa A, Jones AM, Howarth K, Spain SL, Broderick P, Domingo E, Farrington S, Prendergast JG, Pittman AM, Theodoratou E, Smith CG, Olver B, Walther A, Barnetson RA, Churchman M, Jaeger EE, Penegar S, Barclay E, Martin L, Gorman M, Mager R, Johnstone E, Midgley R, Niittymäki I, Tuupanen S, Colley J, Idziaszczyk S, Thomas HJ, Lucassen AM, Evans DG, Maher ER, Maughan T, Dimas A, Dermitzakis E, Cazier JB, Aaltonen LA, Pharoah P, Kerr DJ, Carvajal-Carmona LG, Campbell H, Dunlop MG, Tomlinson IP. Meta-analysis of three genome-wide association studies identifies susceptibility loci for colorectal cancer at 1q41, 3q26.2, 12q13.13 and 20q13.33. *Nat Genet* 2010; **42**: 973-977 [PMID: 20972440 DOI: 10.1038/ng.670]

57 **Ryan BM**, Wolff RK, Valeri N, Khan M, Robinson D, Paone A, Bowman ED, Lundgreen A, Caan B, Potter J, Brown D, Croce C, Slattery ML, Harris CC. An analysis of genetic factors related to risk of inflammatory bowel disease and colon cancer. *Cancer Epidemiol* 2014; **38**: 583-590 [PMID: 25132422 DOI: 10.1016/j.canep.2014.07.003]

58 **Rosenberg DW**, Giardina C, Tanaka T. Mouse models for the study of colon carcinogenesis. *Carcinogenesis* 2009; **30**: 183-196 [PMID: 19037092 DOI: 10.1093/carcin/bgn267]

59 Jackson Laboratory. Available from: URL: http: //www.jax.org/. 2010

60 **Mizoguchi A**. Animal models of inflammatory bowel disease. *Prog Mol Biol Transl Sci* 2012; **105**: 263-320 [PMID: 22137435 DOI: 10.1016/B978-0-12-394596-9.00009-3]

61 **Kanneganti M**, Mino-Kenudson M, Mizoguchi E. Animal models of colitis-associated carcinogenesis. *J Biomed Biotechnol* 2011; **2011**: 342637 [PMID: 21274454 DOI: 10.1155/2011/342637]

62 **Westbrook AM**, Szakmary A, Schiestl RH. Mechanisms of intestinal inflammation and development of associated cancers: lessons learned from mouse models. *Mutat Res* 2010; **705**: 40-59 [PMID: 20298806 DOI: 10.1016/j.mrrev.2010.03.001]

63 **Paul G**, Khare V, Gasche C. Inflamed gut mucosa: downstream of interleukin-10. *Eur J Clin Invest* 2012; **42**: 95-109 [PMID: 21631466 DOI: 10.1111/j.1365-2362.2011.02552.x]

64 **Kühn R**, Löhler J, Rennick D, Rajewsky K, Müller W. Interleukin-10-deficient mice develop chronic enterocolitis. *Cell* 1993; **75**: 263-274 [PMID: 8402911]

65 **Davidson NJ**, Leach MW, Fort MM, Thompson-Snipes L, Kühn R, Müller W, Berg DJ, Rennick DM. T helper cell 1-type CD4+ T cells, but not B cells, mediate colitis in interleukin 10-deficient mice. *J Exp Med* 1996; **184**: 241-251 [PMID: 8691138]

66 **Berg DJ**, Davidson N, Kühn R, Müller W, Menon S, Holland G, Thompson-Snipes L, Leach MW, Rennick D. 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 [PMID: 8770874]

67 **Bristol IJ**, Farmer MA, Cong Y, Zheng XX, Strom TB, Elson CO, Sundberg JP, Leiter EH. Heritable susceptibility for colitis in mice induced by IL-10 deficiency. *Inflamm Bowel Dis* 2000; **6**: 290-302 [PMID: 11149562]

68 **Rajagopalan G**, Kudva YC, Sen MM, Marietta EV, Murali N, Nath K, Moore J, David CS. IL-10-deficiency unmasks unique immune system defects and reveals differential regulation of organ-specific autoimmunity in non-obese diabetic mice. *Cytokine* 2006; **34**: 85-95 [PMID: 16740391]

69 **Arthur JC**, Perez-Chanona E, Mühlbauer M, Tomkovich S, Uronis JM, Fan TJ, Campbell BJ, Abujamel T, Dogan B, Rogers AB, Rhodes JM, Stintzi A, Simpson KW, Hansen JJ, Keku TO, Fodor AA, Jobin C. Intestinal inflammation targets cancer-inducing activity of the microbiota. *Science* 2012; **338**: 120-123 [PMID: 22903521 DOI: 10.1126/science.1224820]

70 **Tanaka T**, Kohno H, Suzuki R, Yamada Y, Sugie S, Mori H. A novel inflammation-related mouse colon carcinogenesis model induced by azoxymethane and dextran sodium sulfate. *Cancer Sci* 2003; **94**: 965-973 [PMID: 14611673]

71 **Tanaka T**. Development of an inflammation-associated colorectal cancer model and its application for research on carcinogenesis and chemoprevention. *Int J Inflam* 2012; **2012**: 658786 [PMID: 22518340 DOI: 10.1155/2012/658786]

72 **Okayasu I**, Hatakeyama S, Yamada M, Ohkusa T, Inagaki Y, Nakaya R. A novel method in the induction of reliable experimental acute and chronic ulcerative colitis in mice. *Gastroenterology* 1990; **98**: 694-702 [PMID: 1688816]

73 **Kitajima S**, Takuma S, Morimoto M. Histological analysis of murine colitis induced by dextran sulfate sodium of different molecular weights. *Exp Anim* 2000; **49**: 9-15 [PMID: 10803356]

74 **Clapper ML**, Cooper HS, Chang WC. Dextran sulfate sodium-induced colitis-associated neoplasia: a promising model for the development of chemopreventive interventions. *Acta Pharmacol Sin* 2007; **28**: 1450-1459 [PMID: 17723178]

75 **Okayasu I**, Ohkusa T, Kajiura K, Kanno J, Sakamoto S. Promotion of colorectal neoplasia in experimental murine ulcerative colitis. *Gut* 1996; **39**: 87-92 [PMID: 8881816]

76 **Saud SM**, Young MR, Jones-Hall YL, Ileva L, Evbuomwan MO, Wise J, Colburn NH, Kim YS, Bobe G. Chemopreventive activity of plant flavonoid isorhamnetin in colorectal cancer is mediated by oncogenic Src and β-catenin. *Cancer Res* 2013; **73**: 5473-5484 [PMID: 23824743 DOI: 10.1158/0008-5472.CAN-13-0525]

77 **Suzuki R**, Kohno H, Sugie S, Nakagama H, Tanaka T. Strain differences in the susceptibility to azoxymethane and dextran sodium sulfate-induced colon carcinogenesis in mice. *Carcinogenesis* 2006; **27**: 162-169 [PMID: 16081511]

78 **Van Der Kraak L**, Meunier C, Turbide C, Jothy S, Gaboury L, Marcus V, Chang SY, Beauchemin N, Gros P. A two-locus system controls susceptibility to colitis-associated colon cancer in mice. *Oncotarget* 2010; **1**: 436-446 [PMID: 21311099]

79 **Barrett CW**, Fingleton B, Williams A, Ning W, Fischer MA, Washington MK, Chaturvedi R, Wilson KT, Hiebert SW, Williams CS. MTGR1 is required for tumorigenesis in the murine AOM/DSS colitis-associated carcinoma model. *Cancer Res* 2011; **71**: 1302-1312 [PMID: 21303973 DOI: 10.1158/0008-5472.CAN-10-3317]

80 **Gao Y**, Li X, Yang M, Zhao Q, Liu X, Wang G, Lu X, Wu Q, Wu J, Yang Y, Yang Y, Zhang Y. Colitis-accelerated colorectal cancer and metabolic dysregulation in a mouse model. *Carcinogenesis* 2013; **34**: 1861-1869 [PMID: 23615396 DOI: 10.1093/carcin/bgt135]

81 Li, X., et al., Identification of gene expression changes from colitis to CRC in the mouse CAC model. PLoS One, 2014. 9(4): p. e95347.

82 **Beutler B**, Du X, Xia Y. Precis on forward genetics in mice. *Nat Immunol* 2007; **8**: 659-664 [PMID: 17579639]

83 **Justice MJ**, Siracusa LD, Stewart AF. Technical approaches for mouse models of human disease. *Dis Model Mech* 2011; **4**: 305-310 [PMID: 21558063 DOI: 10.1242/dmm.000901]

84 **Eisener-Dorman AF**, Lawrence DA, Bolivar VJ. Cautionary insights on knockout mouse studies: the gene or not the gene? *Brain Behav Immun* 2009; **23**: 318-324 [PMID: 18822367 DOI: 10.1016/j.bbi.2008.09.001]

85 **Bouma G**, Kaushiva A, Strober W. Experimental murine colitis is regulated by two genetic loci, including one on chromosome 11 that regulates IL-12 responses. *Gastroenterology* 2002; **123**: 554-565 [PMID: 12145808]

86 **Pizarro TT**, Pastorelli L, Bamias G, Garg RR, Reuter BK, Mercado JR, Chieppa M, Arseneau KO, Ley K, Cominelli F. SAMP1/YitFc mouse strain: a spontaneous model of Crohn's disease-like ileitis. *Inflamm Bowel Dis* 2011; **17**: 2566-2584 [PMID: 21557393 DOI: 10.1002ibd.21638]

87 **Borm ME**, He J, Kelsall B, Peña AS, Strober W, Bouma G. A major quantitative trait locus on mouse chromosome 3 is involved in disease susceptibility in different colitis models. *Gastroenterology* 2005; **128**: 74-85 [PMID: 15633125]

88 **Boulard O**, Kirchberger S, Royston DJ, Maloy KJ, Powrie FM. Identification of a genetic locus controlling bacteria-driven colitis and associated cancer through effects on innate inflammation. *J Exp Med* 2012; **209**: 1309-1324 [PMID: 22734048 DOI: 10.1084/jem.20120239]

89 **Hillhouse AE**, Myles MH, Taylor JF, Bryda EC, Franklin CL. Quantitative trait loci in a bacterially induced model of inflammatory bowel disease. *Mamm Genome* 2011; **22**: 544-555 [PMID: 21717222 DOI: 10.1007/s00335-011-9343-5]

90 **Esworthy RS**, Aranda R, Martín MG, Doroshow JH, Binder SW, Chu FF. Mice with combined disruption of Gpx1 and Gpx2 genes have colitis. *Am J Physiol Gastrointest Liver Physiol* 2001; **281**: G848-G855 [PMID: 11518697]

91 **Esworthy RS**, Kim BW, Larson GP, Yip ML, Smith DD, Li M, Chu FF. Colitis locus on chromosome 2 impacting the severity of early-onset disease in mice deficient in GPX1 and GPX2. *Inflamm Bowel Dis* 2011; **17**: 1373-1386 [PMID: 20872835 DOI: 10.1002/ibd.21479]

92 **Mähler M**, Most C, Schmidtke S, Sundberg JP, Li R, Hedrich HJ, Churchill GA. Genetics of colitis susceptibility in IL-10-deficient mice: backcross versus F2 results contrasted by principal component analysis. *Genomics* 2002; **80**: 274-282 [PMID: 12213197]

93 **Baran AA**, Silverman KA, Zeskand J, Koratkar R, Palmer A, McCullen K, Curran WJ, Edmonston TB, Siracusa LD, Buchberg AM. The modifier of Min 2 (Mom2) locus: embryonic lethality of a mutation in the Atp5a1 gene suggests a novel mechanism of polyp suppression. *Genome Res* 2007; **17**: 566-576 [PMID: 17387143]

94 **Cormier RT**, Bilger A, Lillich AJ, Halberg RB, Hong KH, Gould KA, Borenstein N, Lander ES, Dove WF. The Mom1AKR intestinal tumor resistance region consists of Pla2g2a and a locus distal to D4Mit64. *Oncogene* 2000; **19**: 3182-3192 [PMID: 10918573]

95 **Crist RC**, Roth JJ, Lisanti MP, Siracusa LD, Buchberg AM. Identification of Mom12 and Mom13, two novel modifier loci of Apc (Min) -mediated intestinal tumorigenesis. *Cell Cycle* 2011; **10**: 1092-1099 [PMID: 21386660]

96 **Nnadi SC**, Watson R, Innocent J, Gonye GE, Buchberg AM, Siracusa LD. Identification of five novel modifier loci of Apc(Min) harbored in the BXH14 recombinant inbred strain. *Carcinogenesis* 2012; **33**: 1589-1597 [PMID: 22637734 DOI: 10.1093/carcin/bgs185]

97 **Haines J**, Johnson V, Pack K, Suraweera N, Slijepcevic P, Cabuy E, Coster M, Ilyas M, Wilding J, Sieber O, Bodmer W, Tomlinson I, Silver A. Genetic basis of variation in adenoma multiplicity in ApcMin/+ Mom1S mice. *Proc Natl Acad Sci U S A* 2005; **102**: 2868-2873 [PMID: 15710876]

98 **Moen CJ**, Groot PC, Hart AA, Snoek M, Demant P. Fine mapping of colon tumor susceptibility (Scc) genes in the mouse, different from the genes known to be somatically mutated in colon cancer. *Proc Natl Acad Sci U S A* 1996; **93**: 1082-1086 [PMID: 8577718]

99 **van Wezel T**, Ruivenkamp CA, Stassen AP, Moen CJ, Demant P. Four new colon cancer susceptibility loci, Scc6 to Scc9 in the mouse. *Cancer Res* 1999; **59**: 4216-4218 [PMID: 10485458]

100 **van Wezel T**, Stassen AP, Moen CJ, Hart AA, van der Valk MA, Demant P. Gene interaction and single gene effects in colon tumour susceptibility in mice. *Nat Genet* 1996; **14**: 468-470 [PMID: 8944029]

101 **Ruivenkamp CA**, Csikós T, Klous AM, van Wezel T, Demant P. Five new mouse susceptibility to colon cancer loci, Scc11-Scc15. *Oncogene* 2003; **22**: 7258-7260 [PMID: 14562056]

102 **Meunier C**, Kwan T, Turbide C, Beauchemin N, Gros P. Genetic control of susceptibility to carcinogen-induced colorectal cancer in mice: the Ccs3 and Ccs5 loci regulate different aspects of tumorigenesis. *Cell Cycle* 2011; **10**: 1739-1749 [PMID: 21543896]

103 **Benchimol EI**, Fortinsky KJ, Gozdyra P, Van den Heuvel M, Van Limbergen J, Griffiths AM. Epidemiology of pediatric inflammatory bowel disease: a systematic review of international trends. *Inflamm Bowel Dis* 2011; **17**: 423-439 [PMID: 20564651 DOI: 10.1002/ibd.21349]

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**Figure 1 Progression of colitis-associated colorectal cancer.** Colitis-associated colorectal cancer progresses through a colitis-dysplasia-carcinoma sequence associated with the development of inflammation, low-grade, high-grade dysplasia and eventually carcinoma due to molecular alterations.



**Figure 2 Mouse inflammatory bowel disease and colorectal cancer susceptibility loci.** Summary of the current inflammatory bowel disease and colorectal cancer (CRC) loci mapped in inbred mice using forward genetic studies. Arranged by chromosome, each locus has been drawn to scale based on the current mapping data for each. Putative loci or loci that lack mapping data have been excluded. Loci whose precise map location is unknown (indicated with a \*) have been drawn centered over the peak marker of association. Loci mapped by our lab are shown with names in red font. *Ccs*: Colon cancer susceptibility; *Cdcs*: Cytokine deficiency in colitis (*Il-10*-/- mouse model of colitis); *Dssc*: Dextran sulfate sodium-induced colitis; *Gpdc*: *G protein deficient colitis; Hiccs*: *Helicobacter hepaticus*-induced colitis and associated cancer susceptibility; *Ibdq*: Inflammatory bowel disease quantitative trait loci (Spontaneous *SAMP1/YitFC* model of colitis); *Mom*: Modifier of min (*ApcMin+/-* model of CRC); *Scc*: Susceptibility to colon cancer; *Tm*: *Trishuris muris*-induced colitis; *Tnbs*: Trinitrobenzene sulfonic acid susceptibility.