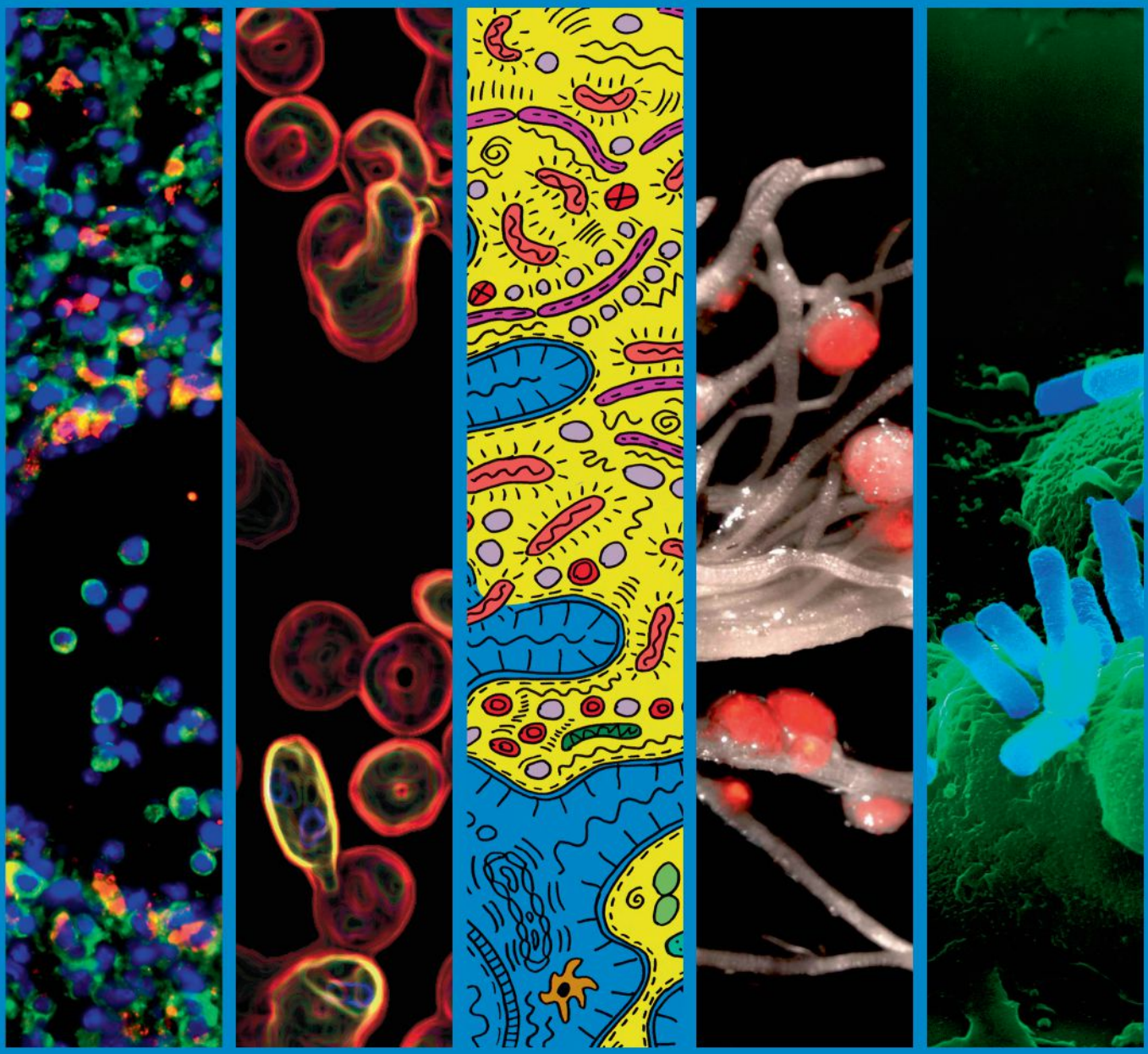
# Cell Host & Microbe

Best of 2014



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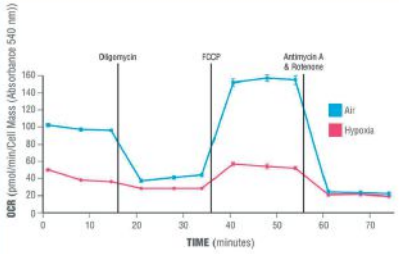
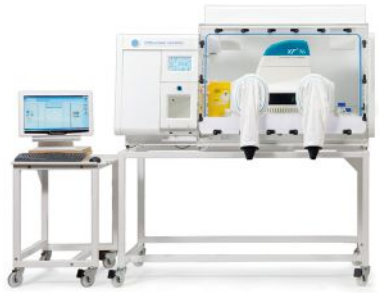



From: Ayse Latif, Experimental Oncology Group, University of Manchester, "Interrogating gynecological cancer cell metabolism at different oxygen tensions: Results using a novel atmosphere handling system"



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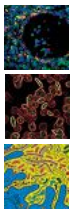
# Foreword



Three years ago, Cell Press launched the "Best of" reprint collections across a number of our journals, including *Cell Host & Microbe*. We proudly welcome you to the 2014 edition of *Best of Cell Host & Microbe*, published for presentation at the 2015 ASM meeting. In looking back at the papers published during 2014, we wanted to provide our readers with a sense of the various topics and findings in which they and their colleagues have shown significant interest. In order to take into consideration the amount of time since publication, we have selected seven of the most accessed articles from 2014, and we have given attention and weighting to the time since publication. We use the number of requests for PDF and full-text HTML versions of a given article up until the end of March 2015 to identify the article as one of our "most accessed." We acknowledge that no single measurement can truly be indicative of "the best" research papers over a given period of time. This is especially true when the community hasn't had sufficient time to fully appreciate the relative importance of a discovery. That said, we think it is still informative to look back at what the community found most interesting in *Cell Host & Microbe* over the course of 2014.

In this collection, we present for your consideration three reviews and seven research articles from 2014. You will see a range of exciting topics, including a review of microbes and microbiota in colon cancer and an examination of the treatment-naive microbiome in new-onset Crohn's disease.

We hope that you will enjoy reading this special collection (visit www.cell.com/bestof to access the entire "Best of" Cell Press collection). To check out the latest findings that we have had the privilege to publish, visit www.cell.com/cell-host-microbe/home, and follow us on twitter @cellhostmicrobe. Also, be sure to visit www.cell.com to find other high-quality papers published in the full collection of Cell Press journals.

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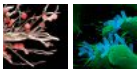
**Cell Host & Microbe**



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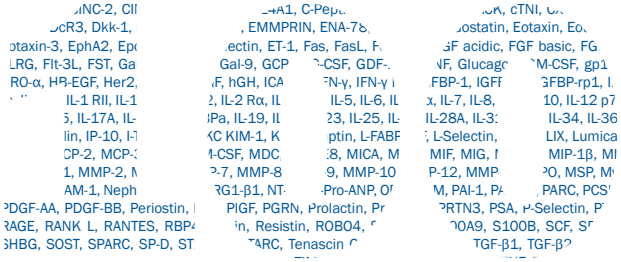


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# Cell Host & Microbe

Best of 2014

## Reviews



## Articles


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Cell Host & Microbe
Review

# Microbes, Microbiota, and Colon Cancer

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**Colorectal cancer (CRC) presents a considerable disease burden worldwide. The human colon is also an anatomical location with the largest number of microbes. It is natural, therefore, to anticipate a role for microbes, particularly bacteria, in colorectal carcinogenesis. The increasing accessibility of microbial meta'omics is fueling a surge in our understanding of the role that microbes and the microbiota play in CRC. In this review, we will discuss recent insights into contributions of the microbiota to CRC and explore conceptual frameworks for evaluating the role of microbes in cancer causation. We also highlight new findings on candidate CRC-potentiating species and current knowledge gaps. Finally, we explore the roles of microbial metabolism as it relates to bile acids, xenobiotics, and diet in the etiology and therapeutics of CRC.**

## Introduction

The human large bowel is a common site for adenocarcinomas and also one of the most densely populated microbial ecosystems on our planet. Colorectal cancers (CRCs) affect over a quarter of a million people each year. In industrialized nations, the lifetime risk of developing CRC is approximately 5%, and the lifetime risk of developing an adenoma, a noncancerous colon tumor that can develop into CRC, is 20%. When the disease is local or confined, cure rates range from 70%–90%; however, advanced CRC has a high mortality rate, consistently ranking in the top three causes of cancer-related death around the globe. There has been long-standing curiosity about the role of bacteria in colorectal carcinogenesis, because of the large disease burden of CRC and the microbial load of the colon; and recent heightened interest in the gut microbiome in CRC, because of the increasing accessibility of microbial meta'omics.

Sequencing technologies have vastly expanded our understanding of the human genetic landscape of CRC. Similarly, efforts at sequencing CRC microbiomes are providing leads into how a microbe's interactions with an individual's entire colonic microbial community, clades within that community, or the human holobiont (Gordon et al., 2013), the entirety of the assemblage of both human and microbe, may be associated with colorectal carcinogenesis. Studies of candidate species in model systems have been useful in evaluating cancer causality and are in keeping with reductionist scientific experimental paradigms. However, an equally plausible concept is that consortia of microbes contribute to CRC risk over time, which can be a far more challenging concept to observationally or experimentally interrogate. This concept is well-aligned with human genetic-based models of colorectal carcinogenesis, namely that molecular alterations in multiple genes underlie the development of a hyperplastic epithelium and propel progression onto adenoma and then toward adenocarcinoma. Mutations in human genes that influence adenoma and adenocarcinoma development may shape the growth rate of colonic epithelial cells (CECs), reduce their susceptibility to cell death, endow them with metabolic specializations, and confer on them abilities to commandeer immune cells to further promote growth and spread. Similarly, microbes can be viewed as collections of gene networks that affect cancer genomic stability, metabolism, and immune responsiveness. In turn, it is possible that the characteristics of transformed CECs render them more sensitive to microbially influenced carcinogenesis.

Herein, we will discuss recent insights into contributions of the microbiota to CRC. We explore conceptual frameworks for evaluating the role of microbes in cancer causation as we highlight new findings on candidate CRC-potentiating species and knowledge gaps. We will not summarize how microbially elicited inflammation or host microbial-sensing pathways affect carcinogenesis, as these topics have been the subject of several recent reviews (Dejea et al., 2013; Kostic et al., 2013a; Jobin, 2012; Schwabe and Jobin, 2013; and see review in this issue of *Cell Host & Microbe* by Goldszmid et al. [2014]). Instead, we concentrate on microbial metabolism in colorectal carcinogenesis with a focus on bile acids and also touch upon xenobiotics and food, all of which are areas where the microbiota has the potential to explicate observations about host gene-environmental interactions in carcinogenesis.

## Causality Theory

CRC is essentially a genetic disease (Figure 1). Gradual accumulation of oncogenic gene mutations leads to autonomous CEC proliferation that slowly progresses typically over 10–40 years,

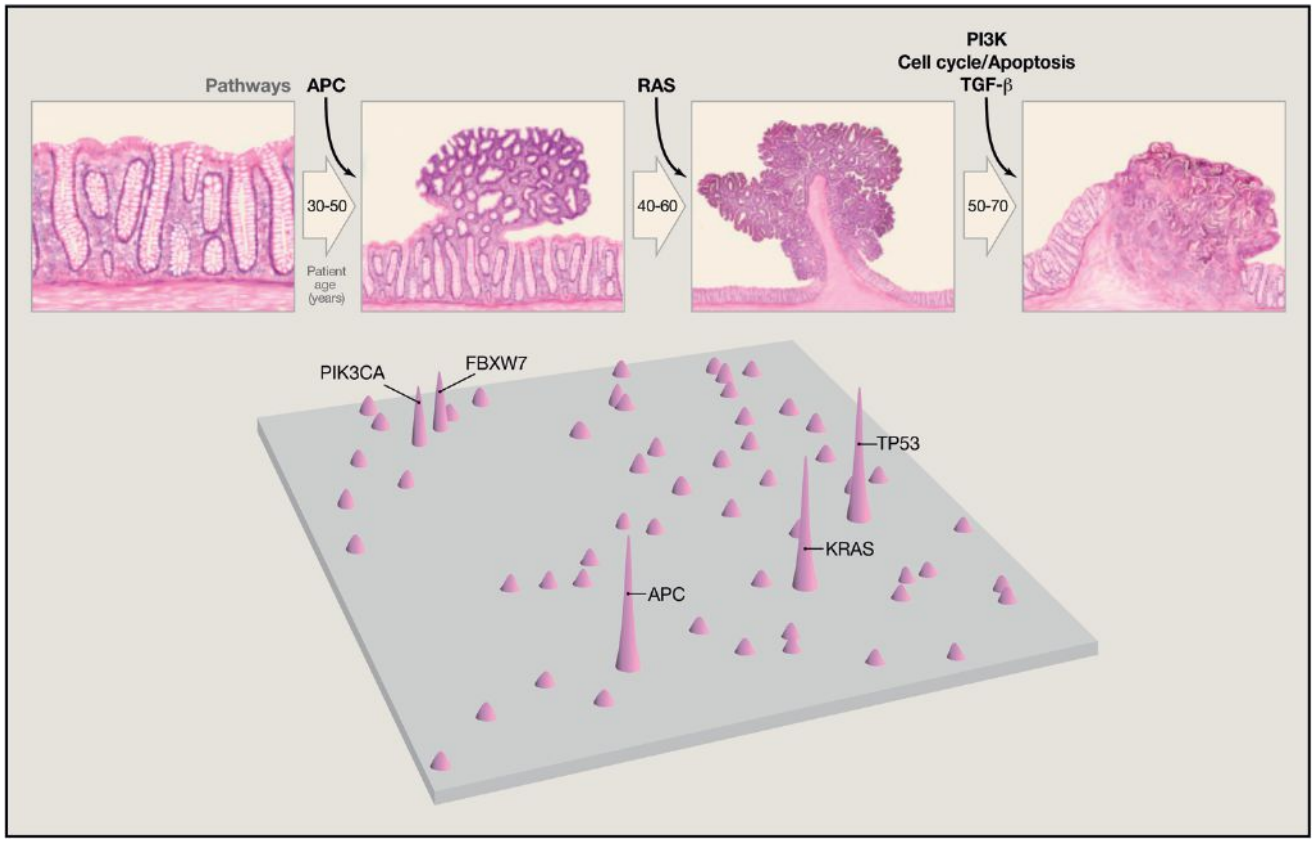


**Figure 1. Genetic Alterations and the Progression of CRC**
The major signaling pathways that drive the development of CRC are shown at the transitions between each tumor stage. One of several driver genes in each signaling pathway can be altered in an individual tumor. Patient age indicates the time interval during which the driver genes are usually mutated. The classic "Vogelgram" shown in the upper panel is adapted from Vogelstein et al. (2013). A map of genes mutated in CRC is shown in the lower panel, with peak height indicating that a large percentage of human colorectal tumors harbor such mutations (adapted from Wood et al., 2007).

resulting in first colon adenomas and then, in a minority of individuals, cancers. Initiation of colon tumors (adenomas, adenocarcinomas) refers to events yielding biologic changes fostering CEC proliferation; progression refers to the subsequent events that liberate growth of the incipient colon tumor and, ultimately, transformation to cancer. Yet, it remains unknown with any precision what events precipitate either the initial, disease-initiating mutation(s) or foster the subsequent disease progression. The microbiome, however, is a prime suspect for triggering the initiation and/or progression of colonic carcinogenesis. Certainly, in murine disease models, colon mucosal inflammation, often induced by mucosal irritants (dextran sulfate sodium, 2,4,6-trinitrobenzenesulfonic acid) combined with administration of a carcinogen (often azoxymethane, a compound in engine fuel), yields colon tumorigenesis, substantiating the intersection of inflammation and exposure to carcinogens in colon tumorigenesis. Certain engineered murine gene knockouts, with potential mucosal inflammation sequelae, also yield colon carcinogenesis that is ameliorated in germ-free animals or sometimes merely by a vivarium change, often considered a proxy for acquisition of a new microbiota. While these models support the hypothesis that the microbiota contributes to colon carcinogenesis, they poorly mimic human disease development. Until recently, the contribution of the human colon environment, home to the largest and most complex microbial mass of human ecology, was not integral to the analytical framework of translational CRC research.

What can guide us as we seek to determine if and how the colon microbiome contributes to the pathogenesis of sporadic human CRC? One clear limitation in seeking a microbe as the cause of a chronic disease is the possibility that the inciting microbe is no longer present at the time the disease is identified, perhaps gradually eliminated by changes in tumor microenvironment no longer hospitable to the microbe or, alternatively, because the microbe acts by a "hit and run" mechanism whereby limited microbial exposure is sufficient to incite disease. For example, in the case of *H. pylori* and gastric cancer, isolation of *H. pylori* declines with advancing gastric cancer, although detection of *H. pylori* exposure is usually still possible by serology (Ota et al., 1998). This reinforces the importance of utilizing multiple approaches in seeking to link a microbe to disease "causation."

Figure 2 provides a framework for considering the microbiota and specific members of the microbiota as either primary (initiators) or secondary (fostering progression) contributors to human CRC pathogenesis (Sears and Pardoll, 2011). We consider three models by which specific microbes (model 1), a microbial community (model 2), or the two acting sequentially

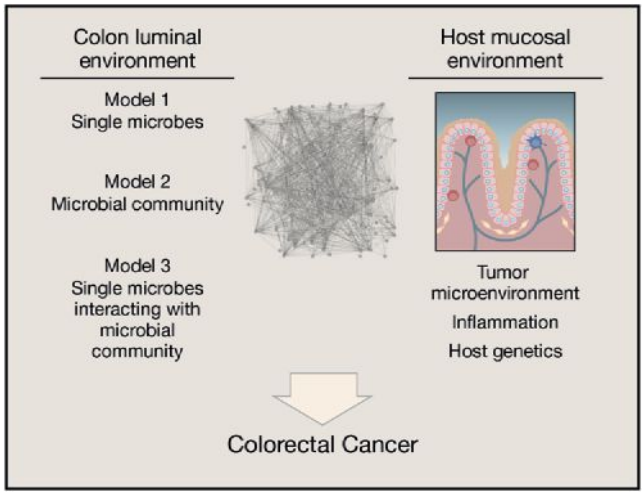


**Figure 2. Microbial Contributions to the Pathogenesis of Colorectal Cancer**

Complex host-microbiota interactions are considered probable primary or secondary contributors to the pathogenesis of CRC. From the microbiota perspective, several hypotheses are actively under investigation, including disease instigation or promotion through individual microbes (model 1), the collective microbiota (model 2), or an interactive model in which single microbes drive the emergence of a modified, disease-generating microbiota (model 3). From the host perspective, the microbiota may alter tumor-associated inflammation with consequences for tumor biology, or conversely, the tumor microenvironment or associated inflammation may induce microbiota shifts with the potential to further inhibit or promote tumor biology. Host genetic polymorphisms that modify immune and metabolic responses are predicted to play a key role in host-microbiota interactions during colonic carcinogenesis. See text for details.

and/or in synergy (model 3) influence colon carcinogenesis. With respect to the first model, we well understand that individual microbes such as the pneumococcus, the meningococcus, *Helicobacter pylori*, or hepatitis viruses are established etiologies of host pathology. We consider that these microbial pathogens possess sufficient virulence mechanisms enabling them to act alone in disease causation. In contrast, as a second model, inflammatory bowel disease stands as the best prototype for microbial community disease causation (Sears and Pardoll, 2011). In this disease, host genetics is the presumed initiator permissive to development of a dysbiotic (implying dysfunctional, disease-initiating or -amplifying) microbiome with an ensuing cycle of host gene-microbiota interactions causing intestinal and possibly extraintestinal disease. Experimental work definitively supports this pathogenetic sequence in that dysbiotic colonic microbiota develop in at least select mice with gene knockouts (e.g., *Tlr5*, *Il10*, *T-bet*, and *Rag2*) and that this emergent dysbiotic microbiome alone possesses the capacity to transmit to a healthy mouse (without any gene mutations) the disease of interest (Garrett et al., 2007, 2010; Vijay-Kumar et al., 2010). Importantly, these murine experiments strongly support the idea that the dysbiotic microbiota can, as a community, encode tissue-specific (e.g., colitis) as well as systemic disease (e.g., metabolic syndrome, obesity) (Ridaura et al., 2013; Vijay-Kumar et al., 2010).

Human CRC provides an opportunity to consider individually the above disease causation scenarios as well as a novel hypothesis in which these theories blend with individual or limited bacterial species acting in concert with a locally modified microbiome to cause CRC (Figure 2, model 3). Nuances of a limited microbial consortium inducing colon tumorigenesis include the possibilities of sequential microbial exposure or polymicrobial disease causation. For this third disease model, we lack clear, clinically relevant historical examples to guide us except for the requirement of the hepatitis B virus for replication and disease induction by the hepatitis D virus. For example, while we understand that the pneumococcus or the meningococcus invades the host from among a complex microbiota in the respiratory tree or nasopharynx, we have no clear data suggesting that the composition of these microbial communities is required for or contributes to the disease causation potential of these bacteria. Similarly, the intestinal microbiota has been shown to hinder or exacerbate viral infections in murine models, but the responsible bacteria are not known (Wilks et al., 2013).

It will not be easy to discern among these potential disease models that provide a framework for defining the microbe contributions to human CRC pathogenesis. Carefully designed studies that consider the Bradford Hill criteria (Bradford Hill, 1965) are needed to link the microbiota and/or select microbes with CRC initiation and progression. A view from the colon lumen may be insufficient. Rather, microbial data must be considered in the context of key host parameters such as the host immunologic response (including the tumor microenvironment) and host gene polymorphisms that influence the host immune response as well as host susceptibility to CEC gene mutations.

## The Microbiome Community as Protagonist

The seminal work of Eckburg et al. clarified the complexity of the fecal and mucosal colon microbiota, importantly illustrating two key points relevant to the colon microbiome as causal in colonic carcinogenesis (Eckburg et al., 2005). First, the majority of microbes, predominantly bacteria, within the colon microbial community are ''noncultivatible.'' While this concept has been challenged by subsequent work, it remains clear that a complete cultured or genome sequence catalog of the colon microbiome with strain-level resolution is still far from our reach (Lagier et al., 2012). Thus, at this time, associations between the microbiota and CRC rely on approaches to broadly define the composition or function of the colon microbiome using various ''omic'' approaches (16S rRNA gene sequencing, metagenomics, transcriptomics, proteomics, metabolomics). Second, while the colon mucosal community may not vary substantially along the axis of the colon, the mucosa-associated community differs from the intraluminal microbiome. These data raise the important and yet unanswered question of whether the fecal microbiome alone will sufficiently mirror mucosal events to allow ''causation'' to be established.

The data set to address microbial community associations in human CRC is limited and has focused on defining bacterial communities associated with colon tumorigenesis. The available studies yield several worthy, albeit preliminary, observations (Dejea et al., 2013). First, the bacterial community composition in colon adenoma or CRC patients, both in mucosal samples and feces, differs from the examined control samples, although consistent associations of bacterial groups with tumor samples or tumor hosts is not yet discernible. Second, ''on-tumor'' and ''off-tumor'' mucosa bacterial populations differ within the tumor