Journal of Gastrointestinal Cancer (2021) 52:1314–1319
https://doi.org/10.1007/s12029-021-00748-1

INVITED REVIEWS

The Gut Microbiome and Hepatocellular Carcinoma

Hikmet Akkız[1]

Accepted: 28 October 2021 / Published online: 6 December 2021

Abstract

The microbiome modulates key processes in metabolism, inflammation, and immunity and plays pivotal roles in many gastrointestinal and liver diseases. Recent experimental studies have demonstrated a key role of the microbiome in hepatocarcinogenesis. Dysfunctions of the gut bacterial flora have a significant effect on liver disease. Dysbiosis is found to be associated with chronic liver diseases. Hepatocellular carcinoma (HCC) is the sixth most common cancer and the third leading cause of cancer-related mortality. The majority of HCC develops in patients with chronic liver disease, caused by chronic viral hepatitis, nonalcoholic fatty liver disease (NAFLD), and alcohol-related fatty liver disease. This review discusses molecular mechanisms of gut microbiome–related hepatocarcinogenesis and the impact of dysbiosis on chronic liver disease progression.

Keywords Inflammation · Hepatocellular carcinoma (HCC) · Microbiome · Microbiota

Abbreviations

BAs	Bile acids
CCL_4	Carbon tetrachloride
CLD	Chronic liver disease
DCA	Deoxycholic acid
DEN	Diethylnitrosamine
DMBA	7,12-Dimethylbenz[a]anthracene
Ereg	Epiregulin
EtOH	Ethanol
HCC	Hepatocellular carcinoma
HSCs	Hepatic stellate cells
LPS	Lipopolysaccharide
LSECs	Liver sinusoidal cells
LTA	Lipoteichoic acid
MAMPs	Microbiota-associated molecular patterns
MDSCs	Myeloid-derived suppressor cells
nAFLD	Nonalcoholic fatty liver disease
NASH	Nonalcoholic steatohepatitis
NKT	Natural killer T cell
ROS	Reactive oxygen species
SASP	Senescence-associated secretory phenotype
SCFAs	Short-chain fatty acids
TLR	Toll-like receptor
TMA	Trimethylamine
TMAO	Trimethylamine N-oxide

Introduction

Hepatocellular carcinoma (HCC) is the sixth most common cancer worldwide and the third leading cause of cancer-related death [1, 2]. HCC is the most common primary liver cancer [1, 2]. HCC can be caused by chronic hepatitis B virus (HBV) infection, hepatitis C virus (HCV) infection, nonalcoholic fatty liver disease (NAFLD), and alcoholic liver disease [1–3]. Most HCCs develop in patients with long-term inflammatory liver damage and cirrhosis [1, 2]. HCCs are usually diagnosed at an advanced stage. Clinical outcomes remain poor, with only approximately one-third of patients eligible for curative treatments such as ablation, surgical resection, and liver transplantation [4, 5]. Currently, over 50% of patients with HCC are given systemic therapies that are barely effective and cause considerable toxic damage to the remaining normal liver [4, 5].

There is growing evidence that dysbiosis contributes to the development of obesity, metabolic disease, and chronic liver diseases [6, 7]. Dysbiosis also promotes HCC development [6, 7]. The liver is not in direct contact with microbiota. However, the liver has anatomic links to the gut [8]. The physiologic transport of nutrient-rich blood from the intestine to the liver accompanied by low-grade exposure to

This is part of Topical Collection: Clinical and basic research on hepatocellular carcinoma in Turkey.

✉ Hikmet Akkız
hakkiz@superonline.com

[1] Department of Gastroenterology and Hepatology, The University of Çukurova, Adana, Turkey

gut-derived metabolites and products, known as microbiota-associated molecular patterns (MAMPs) [6]. Kupffer cells in the liver act as a firewall mediating mutualism to protect from bacterial infections [9]. Chronic liver disease (CLD) is found to be associated with qualitative and quantitative alterations of the gut microbiota, known as dysbiosis [6, 10]. Additionally, changes in the intestinal barrier promote leakiness, causing hepatic exposure to MAMPs and bacterial metabolites in CLD [6, 10, 11]. All studies show that gut microbiota plays a key role in the progression of CLD and the development of HCC [10, 11]. Data accumulated during the last decade also suggest that the gut microbiome play a pivotal role in the regulation of immune responses, including antitumor responses following immunotherapy and chemotherapy [10, 11]. Given all data, the gut microbiome has dual roles which include promotion of cancer development in patients with CLD and the positive regulation of antitumor response. This review will focus on different roles of gut microbiome in the development and clinical outcomes of HCC.

The Gut Microbiota and Chronic Liver Diseases

CLD is characterized by a chronic inflammatory injury to the liver parenchyma that causes liver fibrosis and end-stage liver disease [7]. The cycle of hepatocyte injuries and regenerations causes persistent inflammation and disrupts the regulation of intricately balanced relationship between the gastrointestinal tract and the liver [7]. The gut microbiome mediates this bidirectional interaction through the unique hepatic portal system, the enterohepatic circulation of bile acids (BAs), and the systemic circulation [7]. Homeostasis within gut-liver axis is further preserved through extensive relationship between the gut microbiome and the immune, metabolic, and neuroendocrine system [6, 7]. In the gut-liver axis, the composition and function of the gut microbiota are critical [6, 7]. At the beginning, majority of gut microbiota are commensal organisms, such as Firmicutes and Verrucomicrobia phyla, that maintain this balance by preventing the overgrowth of pathogenic organisms, including Bacteroidetes and Protobacteria phyla [7]. Additionally, a healthy microbiome modifies BAs to be potent signals in the gut-liver axis and immune system, and metabolize dietary contents into short-chain fatty acids (SCTAs) as intermediates to the metabolic system [6, 7]. An intact gut epithelium exhibits an integral role in modulating the gut-liver axis. Disruption of this barrier which results in increased intestinal permeability may be due to overgrown pathogenic bacteria [7].

During the last two decades, microbiome-related discoveries have been made, using new gene sequencing technologies and bioinformatics [7, 12]. The human microbiota is found on all surfaces of the human body, including the skin, the aerodigestive tract, the vaginal cavity in females, and the gastrointestinal lumen [7, 12]. Each of the surfaces contains a specific niche of the microbiome forming its own unique characteristics. The gut microbiome plays an integral role in metabolism, immune tolerance, and immunocompetence [7, 12]. The gut microbiota which contains many microorganisms, including bacteria, viruses, fungi, and archaea, is considered to have over 100 trillion cells [7]. The gut microbiome genome is characterized by over 9 million genes, which outnumbers the human genome by 500-fold [13]. Developing the microbiome starts during birth and alters during life by environmental and behavioral factors [14, 15]. Additionally, in the setting of diseases, including inflammatory bowel disease, liver cirrhosis, and gastrointestinal tumors, the gut microbiota is found to be associated with significant alterations [16]. It is well-known that the liver plays an essential role in the metabolic processes and immune system via the gut-liver axis, which creates its unique connection between the gastrointestinal tract and portal circulation[17, 18]. Currently, sequencing of 16S ribosomal RNA (rRNA) has provided the detailed data on profiling of human gut microbiome. The 16S rRNA is a part of the small 30S subunit of bacterial ribosomes and it is a reliable phylogenic marker [19].

The majority of liver cancers develop in patients with liver cirrhosis. The leaky gut and dysbiosis are significant properties of the liver cirrhosis. The leaky gut and dysbiosis are thought to contribute to the hepatocarcinogenesis in cirrhotic patients [6, 7]. Increased bacterial translocation and dysbiosis are observed in earlier stage of CLD. These processes promote inflammation, fibrogenesis, and progression to cirrhosis. Therefore, dysbiosis and leaky gut are significant properties of all stages of CLD and promote progression to fibrosis, cirrhosis, and HCC [6, 9, 10]. While dysbiosis may contribute to a more permeable intestinal barrier, a leaky gut enables bacterial metabolites and MAMPs to more translocate and reach the liver [6]. Lipopolysaccharide (LPS) is a cell wall component of gram-negative bacteria that promotes inflammation through Toll-like receptor (TLR) 4 that is the most commonly used marker of MAMPs [6, 7]. Portal levels of LPS increase in consistent with the degree of CLD [6, 9, 14]. The highest levels of LPS have been observed in patients with Child–Pugh C cirrhosis [6, 7]. Bacterial DNA levels, an agonist of TLR9, are increased in CLD. In patients with CLD, TLR ligands and other bacterial products and metabolites affect the chronically injured liver [6, 20]. Recent studies have shown dysbiosis in different stage of CLD. However, dysbiosis is the most prominent in cirrhotic and in HCC patients. Dysbiosis alters some processes that affect CLD progression and the subsequent development of liver cancer, such as inflammation, injury, fibrogenesis,

regeneration, and immunity [21, 22]. Studies have shown that the most prominent alterations in the gut microbiota are identified between healthy individuals and cirrhotic patients. However, data also indicated that there are differences between patients with cirrhosis and HCC [21]. Intestinal overgrowth of *Escherichia coli* in HCC patients has first been observed in 2016 [6]. Recently, a study has investigated microbial diversity in patients with cirrhosis and HCC, by using the microbiome as a biomarker for HCC. Microbial diversity has been decreased from healthy individuals to cirrhosis. However, microbial diversity was increased from cirrhosis to early-stage HCC. In this study, Actinobacteria was found to be increased and Verrucomicrobia has been observed to be decreased [22]. The researchers have identified 30 microbial biomarkers in this study. Ponziani and colleagues published a study that compared the gut microbiome in patients with HBV-related HCC and non-HBV non-HCV-related HCC [23]. The researchers investigated whether specific variations of the gut microbiota and inflammatory cytokines are increased in patients with NAFLD and HCC. The study included 21 patients with NAFLD-related cirrhosis and HCC, 20 patients with NAFLD-related cirrhosis without HCC, and 20 healthy controls [23]. In addition to age, cohorts with cirrhosis have been matched on severity of liver disease and portal hypertension. The researchers observed similar dysfunctions of gut permeability in patients with cirrhosis. However, the authors suggested an increased intestine-derived permeability in patients with cirrhosis and HCC, revealed by increases in lipopolysaccharides and zonulin-1 plasma level compared with healthy individuals [23, 24]. The cytokine pattern supports an inflammatory microenvironment specific to patients with cirrhosis with or without HCC [23, 24]. Myeloid-derived suppressor cells (MDSCs) are bone morrow–derived cells, and their diagnostic and prognostic significance have been observed in HCC patients [23, 24]. MDSCs promote HCC progression by suppressing antitumor immunity and favoring tumor angiogenesis. Ponziani and colleagues found that proinflammatory molecules such as interleukin 8 (IL-8), IL-13, chemokine (C–C motif) ligand 3 (CCL3), CCL4, and CCL5 have been significantly increased in patients with HCC and positively correlated with the levels of circulating activated monocytes and MDSCs [23, 24].

The Gut Microbiota and Hepatocarcinogenesis

Most HCC develops in patients with CLD, particularly in cirrhotic patients, but can also develop in patients with chronic hepatitis B infection or NAFLD who have no liver cirrhosis [6]. In chronic inflammatory microenvironment, hepatocytes accumulate genetic and epigenetic reverse transcriptase (TERT) promoter, P53 gene, Wnt/B-catenin signaling pathway genes, playing pivotal roles in hepatocarcinogenesis [1]. Additionally, tumor microenvironment has been thought to play relevant role in the development of HCC. The HCC microenvironment contains many cells, including HCC cells, hepatocytes, cancer-associated fibroblasts (CAFs), MDSCs, regulatory T cells (Treg), tumor-associated macrophages (TAMs), tumor-associated neutrophils (TANs), dendritic cells (DCs), hepatic stellate cells (HSCs), $CD4^+$ T cells, $CD8^+$ T cells, natural killer cells (NK), and Kupffer cells (KCs) [25–27]. Tumor-associated cells have strong immunosuppressive effects which play critical roles both in the development and progression of HCC [28]. The HCC microenvironment also contains precancerous hepatocytes surrounded by fibrotic tissue and the innate and adaptive cells [7, 25]. These cells include KCs, HSCs, NKCs, NKTCs, and other innate lymphocytes [7, 25]. As the liver processes foreign products through portal circulation, a mildly immunosuppressive environment is maintained by these cells with a balanced release of pro-inflammatory and inflammatory cytokines to prevent over activation of the immune system [25]. It is suggested that with persistent chronic inflammation and fibrosis, this balance is disrupted and results in reduced immune surveillance and hepatocarcinogenesis. Recently, studies have shown that the gut microbiota has an important role in this process [7].

To date, the relationship between gut-microbiota-liver axis and hepatocarcinogenesis was investigated in mice models. It has been suggested that CLD-related inflammation causes dysbiosis and intestinal barrier dysfunction (Fig. 1). There are no clinical studies demonstrating a correlation of specific microbial profiles with HCC risk or modulation of HCC risk [6]. Trials in germ-free mice and mice treated with microbial metabolites or MAMPs showed that microbiota activates pathways to contribute to hepatocarcinogenesis [6]. MYC overexpression, the combination of 7,12-dimethylbenz[a] anthracene (DMBA) and high-fat diet, and the combination of major urinary protein-urokinase plasminogen activator (MUP-uPA) overexpression with high-fat diet have been found significantly reduced in gut-sterilized or germ-free mice [29–32]. The majority of tumor-promoting signals from the leaky gut occur in late stages of DEN + CCl_4-induced hepatocarcinogenesis. The leaky gut in CLD results in high circulating levels of MAMPs, including LPS in hepatocarcinogenesis through multiple mechanisms. Bacterial translocation causes a chronic inflammation that was attributed to LPS and its receptor TLR4 in DEN and DEN + CCl_4-induced hepatocarcinogenesis. TLR4 also promotes liver fibrosis and upregulated the expression of epiregulin in HSCs [6, 24, 25]. Genetic TLR4 inhibition suppresses DEN + CCl_4-induced hepatocarcinogenesis. Low-dose LPS infusion promotes hepatocarcinogenesis in mice. TLR4 promotes inflammatory gene expression; additionally, TLR4 also promotes

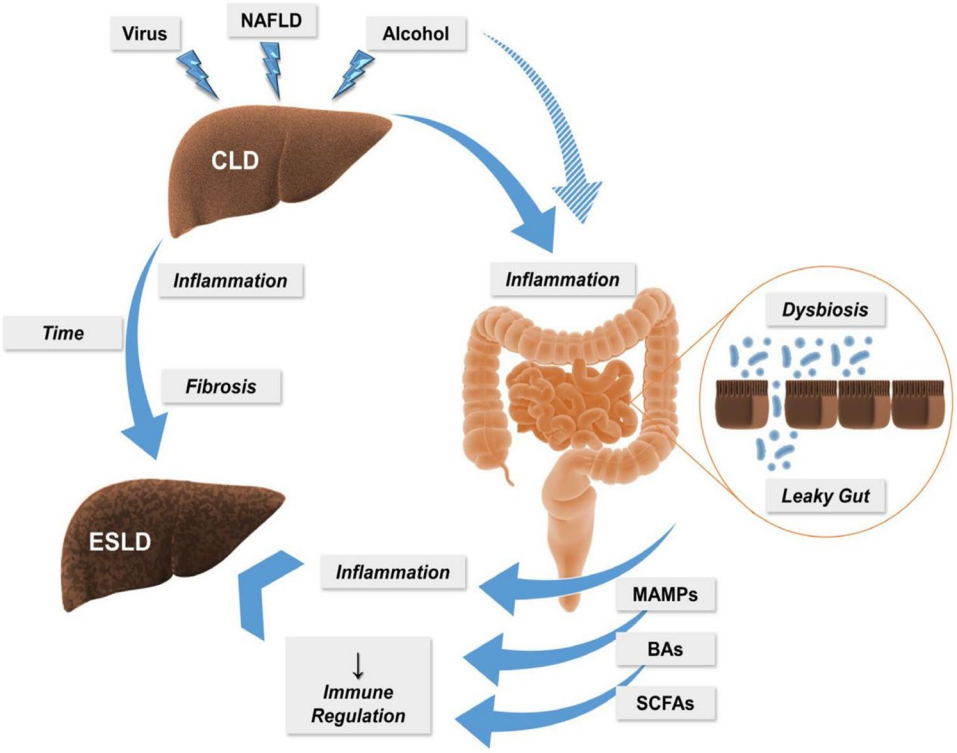

Fig. 1 Gut microbiome–related mechanisms in hepatocarcinogenesis

tumor proliferation, preventing tumor death [6]. The impacts of TLR2 in NAFLD-related HCC is mediated by HSCs. Yu and colleagues demonstrated that elevated levels of plasma LPS have been observed in rats that developed HCC after DEN exposure [33]. The researchers also showed that the impacts of LPS are mediated by TLR4 in knock models. Lower incidence of HCC has been observed in TLR knockout models [33]. This models also showed lower levels of inflammatory cytokines, including IL-6 and tumor necrosis factor alpha (TNF-a). Other studies have also shown this impact on dysbiosis in DEN-exposed models having an increased incidence of LPS-containing gram-negative bacteria, including *Escherichia coli*, *Atopobium*, *Collinsella*, *Eggerthella*, and *Cariobacterium* [7]. The use of high-dose probiotics in these animals improves dysbiosis and reduces the number and size HCC lesions. CLD-related dysbiosis can also cause suppression of the immune system, resulting in further progression of HCC [34]. The induction of HSCs in the stroma into a senescence-associated secretory phenotype (SASP) has been associated with the suppression of antitumor immunity [7, 35]. Yoshimato and colleagues demonstrated that the development of the SASP HSCs in obese animal models has not been observed to be mediated by the leaky gut pathway or TLRs, but it was associated with increased of deoxycholic acid (DCA) with increased prevalence in obesity-induced dysbiosis [35]. Other studies revealed that the gut microbiota in NAFLD-associated HCC animal models contains increased Bifidobacterium and Clostridiales, also known as Clostridium cluster XVIII. Both of these groups play roles in BA modification and implicated in the conversion of primary BAs to secondary BA [7, 32, 36].

Dysbiosis also changes metabolic pathways in the gut microbiota. One of the major alterations developing in mice fed a high-fat diet is the accumulation of gram-positive bacteria with an enhanced ability for the conversion of bile acids [6, 30]. Thus, a high-fat diet causes increased levels of secondary bile acids (DCA) that are generated by bacteria that promote hepatocarcinogenesis [6, 30]. DCA has been observed to increase TLR2 in HSCs that resulted in the tumor-promoting SASP [6, 30]. Gut microbiota–dependent alterations in the bile acid metabolism regulate HCC growth by driving hepatic expression of CXCL16- and CXCL16-mediated NKT cell recruitment [29]. Experiments in mice treated with antibiotic or cholestyramine have shown a key role for primary bile acids in upregulating CXC16 in liver sinusoidal endothelial cells that causes the NKT cell recruitment [29]. In another study, in NASH-related HCC, caused by a high cholesterol-high-fat NASH diet, antibiotics was observed to inhibit HCC development that was associated with a strong reduction of secondary bile acids (Yamada). Secondary bile acids, such as DCA, trigger the mTOR pathway in hepatocytes. However, mTOR activation was found to be reduced when NASH diet-fed mice have been treated with antibiotics [32]. Short-chain fatty acids, generated by bacterial fermentation of fiber and resistant starch, are thought to be health-promoting for the health of colonic

epithelial cells; recent studies have demonstrated that diets with high inulin, which is converted to butyrate, promote hepatocarcinogenesis in mice with dysbiosis [22]. In addition to the tumor-promoting effect of butyrate, the researchers also observed that depletion of bile acids by cholestyramine prevented the tumor-promoting effects of short-chain fatty acids (Shing). Other metabolites generated by gut bacteria, including trimethylamine (TMA) and its metabolite trimethylamine N-oxide (TMAO) as well as ethanol, may contribute to the development of HCC. TMA is generated by gut bacteria from dietary choline and carnitine and is converted to TMAO in the liver after intestinal absorption (Schwabe). TMAO has been found to be associated with cardiometabolic and colorectal carcinogenesis risk [37]. Decreased levels of choline may contribute to hepatotoxicity (Schwabe). Additionally, high TMAO levels are associated with insulin resistance; therefore, it can contribute to the progression of NAFLD and to NASH-related HCC [38]. However, the effects of TMAO in NASH and HCC development remain unclear. Ethanol is constantly generated by the intestinal microbiota and is increased in NASH patients [6].

HCC often develops in the setting of chronic inflammation. Although immune cells are key drivers in immunosurveillance, they may exhibit pro-tumor activity in hepatocarcinogenesis in patients with CLD through increasing inflammation [25]. Antitumor effects of B and T cells have been shown in mice injected with DEN (Schwabe). $CD4^+$ T and $CD8^+$ T cells and NK cells eradicate the tumor cells. However, recent studies suggested that $CD8^+$ T cells may promote inflammation in the liver and probably promote development of NASH [39]. In another study, researchers have reported similar findings in a fumarylacetoacetate hydrolase knockout mice that develop HCC in the setting of chronic inflammation [40]. $CD4^+$ T cells confer significant contribution to HCC immunosurveillance in the setting of NASH. A study suggested that NASH causes an accumulation of linoleic acid, which can induce reactive oxygen species–mediated $CD4^+$ T cell death promoting tumor growth [29]. B cells also contribute to HCC immunosurveillance. Immunosuppressive cells in stroma, such as MDSCs and Treg, have been demonstrated to impair effector T and NK cell function. How does gut microbiome affect hepatic immunosurveillance? Studies suggest that there has been a strong association between the gut microbiota and immune system. The gut microbiota drives both local immunity in the intestine and systemic immunity [6]. The gut microbiota stimulates myelopoiesis and different myeloid cells, including macrophages, dendritic cells, and neutrophils. Effects may be related to MAMPs and bacterial metabolites, such as SCFAs and BAs. A study showed that the gut microbiota drives immune responses to HBV in mice and causes the failure to clear HBV [41]. Given all data, microbial alterations can modulate immunosurveillance in HCC patients. Recent studies in mice and patients showed a key role of the gut microbiota in controlling antitumor immune responses in the setting of chemotherapy and immunotherapy [6].

Ponziani et al. performed a study regarding the gut microbiota composition by using the 16S RNA-based metagenomic analysis and found that patients with cirrhosis have a lower gut microbial diversity compared to healthy controls, independent of the presence HCC [23]. The researchers demonstrate cirrhosis affect the gut microbial composition. The dysbiotic fingerprint in patients with cirrhosis has alterations of specific bacterial families and genera. They detected the relative abundance in Enterobactericeae, Streptococcus, Bacteroides, and Ruminococcus and the reduction in Bifidobacterium and Akkermansia [23].

Conclusion

A better understanding of the gut microbiome will be useful in many ways, including for prevention, diagnosis, and treatment of HCC. The intestinal microbiota and related alterations are associated with CLD and HCC development. The alterations in intestinal microbiota generate an inflammatory hepatic microenvironment. Dysbiosis promotes to impair intestinal barrier that enhances TLR-mediated liver inflammation. Although there are clear relationship between dysbiosis-associated inflammation and HCC in animal models, we need further human microbiome studies to explain key role of the intestinal microbiome in hepatocarcinogenesis. The studies can generate new therapeutic approaches that improve overall survival.

Declarations

Conflict of Interest The author declares no competing interests.

References

1. Liovet JM, Kelley RK, Villonueva A, et al. Hepatocellular carcinoma Nat Rev Dis Primers. 2021;6:1–37.
2. Naulth J-C, Cheng A-L, Sangro B, et al. Milestones in the pathogenesis and management of primary liver cancer. J hepatol. 2020;72(2):209-14.
3. Rebouissou S, Naulth J-C. Advances in molecular classification and precision oncology in hepatocellular carcinoma. J Hepatol. 2020;72:215–29.
4. Calderaro J, Ziol M, Paradis V, et al. Molecular and histological correlations in liver cancers. J Hepatol. 2019;71:616–30.
5. Hou J, Zhang H, Sun B, et al. The immunobiology of hepatocellular carcinoma in humans and mice: Basic concepts and therapeutic implications. J Hepatol. 2020;72:167–82.
6. Schwabe RF, Greeten TF. Gut microbiome in HCC – mechanisms, diagnosis and therapy. J Hepatol. 2020;72:230–8.

7. Rattan P, Minacapelli CD, Rustgi V. The microbiome and hepatocellular carcinoma. Liver Transpl. 2020;26:1316–27.
8. Anstee QM, Reeves HL, Kotsiliti E, et al. From NASH to HCC: current concepts and future challenges. Nat Rev Gastroenterol Hepatol. 2019;16:411–28.
9. Balmer MI, Slack E, de Gottardi A, et al. the liver may act as a firewall mediating mutualism between the host and its gut commensal microbiota. Sci Transl Med. 2014;6:237–66.
10. Tripathi A, Debelius J, Brenner Da, et al. The gut-liver axis and the intersection with the microbiome. Nat Rev Gastroneterol Hepatol 2018;15:397–411.
11. Yu LX, Schwabe RF. The gut microbiome and liver cancer: mechanisms and clinical translation. Nat Rev Gastroenterol Hepatol. 2017;14:527–39.
12. Cho I, Blaser MJ. The human microbiome: at the interface of health and disease. Nat Rev Genet. 2012;13:260–70.
13. Nicholson JK, Holmes E, Kinros J, et al. Host-gut microbiota metabolic interactions. Science. 2012;336:1262–7.
14. Li J, Jia H, Cai X, et al. an integrated catalog of reference genes in the human gut microbiome. Nat Biotechnol. 2014;6:834–41.
15. Dominguez-Bello MG, Godoy-Vitorino F, Knight R, et al. Role of the microbiome in human development. Gut. 2019;68:1108–14.
16. Ni J, Wu GD, Albenberg I, et al. Gut microbiota and IBD: causation or correlation? Nat Rev Gastroenterol Hepatol. 2017;19:573–84.
17. Starkel P, Schnabl B. Bidirectional communication between liver and gut during alcoholic liver disease. Semin Liver Dis. 2016;20:331–9.
18. Szabo G. Gut-liver axis in alcoholic. Liver disease Gastroenterology. 2015;148:30–6.
19. Hamady M, Knight R. Microbial community profiling for human microbiome projects: tools, techniques, and challenges. Genome Res. 2009;1:1141–52.
20. Bellot P, Garcia-Pagan JC, Frances R, et al. Bacterial DNA translocation is associated with systemic circulatory abnormalities and intrahepatic endothelial dysfunction in patients with cirrhosis. Hepatology. 2010;52:2044–52.
21. Loomba R, Seguritan V, Li W, et al. Gut microbiome-based metagenomic signature for non-invasive detection of advanced fibrosis in human nonalcoholic fatty liver disease. Cell Metab. 2017;25:1054–62.
22. Singh V, Yeoh BS, Chassaing B, et al. Dysregulated microbial fermentation of soluble fiber induces cholestatic liver cancer. Cell. 2018;175:679–94.
23. Ponziani FR, Bhoori S, Castelli C, et al. Hepatocellular carcinoma. İs associated with gut microbiota profile and inflammation in nonalcoholic fatty liver disease. Hepatology 2019;69:107–120.
24. Cassano M, Dufour J-F. Inflammation and microbiota fingerprint: Delphi's oracle for nonalcoholic fatty liver disease – Related Hepatocellular Carcinoma. Hepatology. 2019;69:12–5.
25. Ringelhan M, Pfister D, O'connar T, et al. The immunology of hepatocellular carcinoma. Nat Immunol. 2018;19:222–32.
26. Lu C, Rong D, Zhang B, et al. current perspectives on the immunosuppressive tumor microenvironment in hepatocellular carcinoma: challenges and opportunities. BMC Molecular Cancer. 2019;18:1–12.
27. Song MJ, He JY, Pan QZ, et al. Cancer-associated fibroblast-mediated cellular crosstalk supports hepatocellular carcinoma progression. Hepatology 202173:1717–1734.
28. Lei MML, Lee TKW. Cancer-associated fibroblasts: orchestrating the crosstalk between liver cancer cells and neutrophils through the cardiotrophin-like cytokine factor 1-mediated chemokine (C-X-C motif) ligand 6/TGF-B axis. Hepatology. 2021;73:1631–3.
29. Ma C, Han M, Heinrich B, et al. Gut-microbiome-mediated bile acid metabolism regulates liver cancer via NKT cells. Science. 2018;8:360–9.
30. Shalapour S, Lin XJ, Bastian IN, et al. Inflammation-induced IgA^+ cells dismantle anti-liver cancer immunity. Nature. 2017;551:340–5.
31. Yoshimoto S, Loo Tm, Atarashi K, et al. Obesity-induced gut microbial metabolite promotes liver cancer through senescence secretome. Nature 2013;499:97–101.
32. Yamada S, Takashina Y, Watanabe M, et al. Bile acid metabolism regulated by the gut microbiota promotes non-alcoholic steatohepatitis-associated hepatocellular carcinoma in mice. Oncotarget. 2018;9:9925–39.
33. Yu LX, Yan HX, Liu Q, et al. Endotoxin accumulation prevents carcinogen-induced apoptosis and promotes liver tumorigenesis in rodents. Hepatology. 2010;52:1322–3.
34. Li J, Sung CYJ, Ni Y, et al. Probiotics modulated gut microbiota suppresses hepatocellular carcinoma growth in mice. Proc Natl Acad Sci USA. 2016;113:E1306–15.
35. Yoshimoto S, Loo TM, Atarashi K, et al. Obesity-induced gut microbial metabolite promotes liver cancer through senescence secretome. Nature. 2013;499:97–101.
36. Xie G, Wang X, Huang F, et al. Dysregulated hepatic bile acids collaboratively promote liver carcinogenesis. Int J Cancer. 2016;139:1764–75.
37. Ufnal M, Zadio A, Ostaszewski R. TMAO: a small molecule of great expectations. Nature. 2011;472:57–63.
38. Oeligaard J, winther SA, Hansen TS, et al. Trimethylamine N-oxide (TMAO) as a new potential therapeutic target for insulin resistance and cancer. Curr Pharm Des 2017;23:3699–3712.
39. Wolf MJ, Adili A, Piotrowitz K, et al. Metabolic activation of intrahepatic $CD8^+$ T cells and NKT cells causes nonalcoholic steatohepatitis and liver cancer via cross-talk hepatocytes. Cancer Cell. 2014;26:549–64.
40. Endig J, Buitrago-Molina LE, Marhenke S, et al. Dual role of the adaptive immune system in liver injury and hepatocellular carcinoma development. Cancer Cell. 2016;30:308–23.
41. Chou HH, Chien WH, Wu LL, et al. Age-related immune clearance of hepatitis B virus infection requires the establishment of gut microbiota. Proc Natl Acad Sci USA. 2015;112:2175–80.