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**Role of intestinal flora in primary sclerosing cholangitis and its potential therapeutic value**

Li ZJ *et al*. Intestinal flora in primary sclerosing cholangitis

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**Abstract**

Primary sclerosing cholangitis (PSC) is an autoimmune disease characterized by chronic cholestasis, a persistent inflammation of the bile ducts that leads to sclerotic occlusion and cholestasis. Gut microbes, consisting of microorganisms colonized in the human gut, play an important role in nutrient intake, metabolic homeostasis, immune regulation, and immune regulation; however, their presence might aid PSC development. Studies have found that gut-liver axis interactions also play an important role in the pathogenesis of PSC. Patients with PSC have considerably reduced intestinal flora diversity and increased abundance of potentially pathogenic bacteria. Dysbiosis of the intestinal flora leads to increased intestinal permeability, homing of intestinal lymphocytes, entry of bacteria and their associated metabolites, such as bile acids, into the liver, stimulation of hepatic immune activation, and promotion of PSC. Currently, PSC effective treatment is lacking. However, a number of studies have recently investigated the targeted modulation of gut microbes for the treatment of various liver diseases (alcoholic liver disease, metabolic fatty liver, cirrhosis, and autoimmune liver disease). In addition, antibiotics, fecal microbiota transplantation, and probiotics have been reported as successful PSC therapies as well as for the treatment of gut dysbiosis, suggesting their effectiveness for PSC treatment. Therefore, this review briefly summarizes the role of intestinal flora in PSC with the aim of providing new insights into PSC treatment.

**Key Words:** Primary sclerosing cholangitis; Intestinal flora; Antibiotics; Fecal microbiota transplantation; Probiotics; Bile acids

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**Core Tip:** Primary sclerosing cholangitis (PSC) is an autoimmune disease that currently lacks treatment. The intestinal flora comprises microorganisms that colonize the human gut and play essential roles in nutrient intake, metabolic homeostasis, immune regulation, and PSC development. Thus, the intestinal flora may be a potential therapeutic target for PSC, and many recent studies have attempted to regulate it. In this review, we have reviewed the role of the intestinal flora in PSC. We believe that our study makes a significant contribution to the literature because our paper demonstrated the great potential of the gut flora as a therapeutic target for PSC treatment.

**INTRODUCTION**

Primary sclerosing cholangitis (PSC) is an autoimmune-mediated chronic cholestatic liver disease characterized by progressive bile duct inflammation, leading to intra- and extrahepatic bile duct stenosis and occlusion and cholestatic cirrhosis[1]. Patients with PSC have a greatly increased risk of developing cancers (cholangiocarcinoma, gallbladder cancer, hepatocellular carcinoma, and colorectal cancer); approximately half of patients with PSC develop cancer, ultimately leading to death[2]. Although the etiology of PSC remains unclear, it is generally accepted that interactions between genetics and the environment determine PSC development[3]. Owing to the close anatomical and physiological connection between the intestine and the liver, the intestinal flora is closely related to liver disease[4]. Several studies have suggested that the intestinal flora is involved in PSC development through the gut-liver axis[5,6]. Moreover, patients with PSC have significantly reduced intestinal flora diversity and an increased abundance of potentially pathogenic bacteria[7,8]. Intestinal flora dysbiosis leads to increased intestinal permeability, intestinal lymphocyte homing, entry of bacteria and their associated metabolites [*e.g.* bile acids (BAs)] into the liver, activation of the hepatic immune response, and bile duct inflammation and fibrosis[9].

The incidence of PSC is increasing, but an effective treatment does not exist currently. PSC can eventually progress to cirrhosis or liver failure, but these conditions are still symptomatically treated[10,11]. For patients with end-stage PSC, liver transplantation is the sole option; however, transplantations are not universally available owing to high costs and potential transplant rejection. Furthermore, approximately 30%-50% of patients experience PSC recurrence within 10 years of transplantation[12].

Many studies have reported that the gut flora is a promising therapeutic target for PSC, and that antimicrobial therapy based on gut flora modulation, fecal microbiota transplantation (FMT), and probiotics is an emerging therapeutic options[13,14]. Thus, in this review, we discuss the latest research findings on the role of intestinal flora in PSC and provide important insights into potential microbe-altering interventions.

**PSC PATHOPHYSIOLOGY AND THE GUT-LIVER AXIS**

PSC is a rare disease with an incidence of 0.91-1.30/100000. The incidence of small bile duct PSC is approximately 0.15/100000, with the highest prevalence in the Nordic countries, reaching an incidence of 16.2/100000[15]. PSC mostly occurs in men aged 40-50 years, with age of diagnosis at 30-40 years and a male to female ratio of approximately 2:1[1]. The pathogenesis of PSC is complex, but it is currently believed that interactions between genetic susceptibility factors and environmental promoters plays a role in the occurrence and development of PSC[16]. Human leukocyte antigen is strongly associated with PSC pathogenesis[17]. However, the risk ratio for first-degree relatives is approximately 11, suggesting that environmental factors play a more critical role in the pathogenesis of PSC[18]. In addition, the geographic distribution of PSC pathogenesis may indicate that the disease is influenced by the environment[19]. Interactions of the gut-liver axis, such as damage to the intestinal mucosal barrier, dysbiosis, and immune interactions, also play a role in the pathogenesis of PSC[1].

The gut-liver axis refers to the bidirectional relationship between the intestine, its microbiota, and the liver, resulting from the interaction of dietary, genetic, and environmental factors[20]. The close association between the intestine and the liver begins during embryonic development, with both organs originating together in the ventral foregut endoderm. From a physiological point of view, the gut-liver axis is one of the most important links between the intestinal flora and the liver[21]. The liver, which receives approximately 70% of its blood from the portal vein, is a receiver and filter of nutrients and bacterially produced toxins and their metabolites. The secretion of substances such as BAs and antibodies into the intestine acts as a feedback mechanism that affects intestinal homeostasis[22].

Approximately 70% of patients with PSC have concomitant inflammatory bowel disease (IBD) and more commonly ulcerative colitis (UC)[23-25]. Patients with PSC have a reduced risk of death after a colectomy, and after receiving liver transplantation, colectomy significantly reduces the risk of PSC recurrence. This close association between PSC and IBD suggests that intestinal flora may play a key role in the pathogenesis of PSC[26,27] through the gut-liver axis[28].

**PATIENTS WITH PSC AND THEIR DYSBIOSIS OF INTESTINAL FIORA**

***Intestinal flora dysbiosis***

Under normal physiological conditions, the human body contains a large and diverse community of intestinal microorganisms, reaching up to 1014 organisms that comprise more than 1000 species; this is collectively known as the intestinal flora[29]. A normal intestinal flora is primarily composed of *Firmicutes, Bacteroidetes, Proteobacteria*, and *Actinobacteria*, which promote nutrient digestion and absorption, defend against foreign pathogens, regulate immunity, and participate in metabolic processes[30].

In healthy populations, the intestinal microenvironment is in homeostasis due to the mutual regulation of various flora. Intestinal flora dysbiosis is a disruption of the dynamic balance between intestinal flora when various factors cause disturbances in the human body environment, and changes in the number, species, and ratio of favorable, conditionally pathogenic, and harmful bacteria[31,32]. The manifestations of intestinal flora dysbiosis include intestinal flora translocation (transfer of the original colonizing bacteria from the intestine to the deep mucosa or from the intestine to other sites) and intestinal flora imbalance (decrease in the abundance of the original beneficial intestinal flora and increase in the abundance of pathogenic flora). Dysbiosis of the intestinal flora leads to impairment of the intestinal barrier, increased endotoxemia, and a breakdown of the liver's immune tolerance to the intestinal flora and its metabolites, which in turn causes a strong immune response in the liver[33].

***The intestinal flora of patients with PSC***

It was found that patients with PSC have a marked dysbiosis of the intestinal flora compared with the healthy population. Rossen *et al*[34] performed the first 16S rRNA analysis of the microbiota of the intestinal mucosa in the ileocecal region of patients with PSC and found that, at the genus level, the relative abundance of uncultured *Clostridium II* was notably lower in patients with PSC compared with patients with UC and non-inflammatory controls. In addition, the mucosal adherent microbiota at the level of the ileocecal region in patients with PSC showed considerably reduced diversity and abundance. Torres *et al*[8], using bacterial 16S rRNA next-generation sequencing, reported that patients with PCS had similar overall microbiome characteristics at different locations in the gut, exhibiting enrichment in *Blautia* and *Barnesiellaceae spp*. A more in-depth taxon analysis at the operational taxonomic unit (OTU) level revealed the most significant changes in *Clostridiales*, with 3 decreases and 66 OTU enrichments. Sabino’s[35] study found that species richness (defined as the number of different OTUs observed in the samples) was reduced in patients with PSC compared with healthy controls, that *Enterococcus, Clostridium, Lactobacillus,* and *Streptococcus* were enriched, and that an operational taxonomic unit belonging to the *Enterococcus* genus is positively correlated with the levels of alkaline phosphatase (ALP) levels (a marker of disease severity). In addition, PSC has its own unique gut microbial profile that is not associated with IBD co-morbidity. Subsequently, a study by Kummen *et al*[36] also confirmed the unique gut microbiota of PSC independent of the receipt of antibiotics and ursodeoxycholic acid (UDCA) treatment, with a marked reduction in bacterial diversity in patients with PSC. Furthermore, Quraishi *et al*[37] explored the intestinal mucosal flora of PSC-IBD patients, further complementing the study by Kummen *et al*[36] *Escherichia, Lachnospiraceae,* and *Megasphera* were markedly increased, whereas *Prevotella* and *Roseburia* (butyrate producers) were decreased in abundance in PSC-IBD patients compared with controls. They hypothesized that intestinal microecological dysregulation in patients with PSC may prompt dysregulation associated with mucosal immunity by modulating abnormal homing of intestinal-specific lymphocytes and intestinal permeability. This is consistent with the hypothesis of Kummen *et al*[36] Rühlemann *et al*[38,39] also showed that PSC itself drives the observed changes in fecal microbiota and that the findings of Kummen *et al*[36] regarding microbiota as a diagnostic marker are promising.

In the last two years, studies on the PSC gut flora have gained more interest. Quraishi *et al*[40] tried to unravel the PSC disease mechanism by integrating mucosal transcriptomics, immunophenotyping, and mucosal microbial analysis; their study reported that PSC-IBD patients had considerably higher abundance of *Bacteroides fragilis, Roseburia spp., Shewanella sp.,* and *Clostridium ramosum* species, which was associated with changes in the BA metabolic pathway. In addition, the amine oxidase-expressing bacterium *Sphingomonas sp*. is upregulated in PSC-IBD. Amine oxidase is associated with abnormal homing of intestinal lymphocytes to the liver[41]. The upper gastrointestinal tract and bile ducts of patients with PSC are equally affected by microbial ecological dysbiosis. Liwinski *et al*[42] showed that the biliary microbiome of patients with PSC exhibited the most extensive changes, including reduced biodiversity and expansion of pathogenic bacteria, with the marked increase of *Enterococcus spp.* directly causing epithelial barrier damage and mucosal inflammation. Lapidot *et al*[43] found that microbial alterations in PSC were consistent in saliva and gut, with 27 bacterial species present in both the salivary and gut microbiomes, including *Clostridium perfringens XlVa, Veillonella, Lachnospiraceae, Streptococcus,* and *Blautia*. Of these, *Lactobacillus, Ruminococcus gnavus,* and *Streptococcus salivarius* were extensively enriched. The study by Lemoinne *et al*[44] confirmed previous findings of altered bacterial microbiota composition in patients with PSC, such as *Faecalibacterium* and *Ruminococcus* in reduced proportions, and reported for the first time the occurrence of fungal ecological dysbiosis in patients with PSC. PSC-associated fungal ecological dysbiosis is characterized by increased biodiversity (alpha diversity) and altered composition compared with in healthy subjects or patients with IBD, including a marked increase in the abundance of *Exophiala spp*. In some patients. Kummen *et al*[45] provided a detailed functional analysis of microbial genes encoding enzymes and metabolic pathways by using metagenomic shotgun sequencing. *Clostridium spp.* increased in the intestinal flora of patients with PSC, while *Eubacterium spp.* and *Ruminococcus obeum* decreased. Targeted metabolomics revealed reduced concentrations of vitamin B6 and branched-chain amino acids in PSC. Microbial metabolism of essential nutrients and circulating metabolites associated with the disease process were considerably altered in patients with PSC compared with in healthy individuals, suggesting that microbial function may be related to the disease process in PSC.

Most of these studies used 16S gene sequencing to examine the microbiota in the intestinal mucosa and feces of patients with PSC. Although these studies came from all over the world, some of them had relatively small sample sizes, and dietary and lifestyle habits varied between the samples of each study, possibly affecting the final gut floral composition of patients with PSC and limiting the generalization of the results. However, these studies also yielded some common findings that reveal to some extent the gut microbiota characteristics of patients with PSC[46]. Patients with PSC suffer intestinal dysbiosis, which has its own unique biological characteristics, as evidenced by a decrease in gut bacterial α-diversity (average species diversity of the ecosystem) and marked changes in β-diversity (spatial variation in species composition), a decrease in specialized anaerobic bacteria, and an increase in the abundance of potentially pathogenic bacteria (Table 1)[7,35-39,42-44,47]. Among which, *Veillonella, Enterococcus, Streptococcus, Clostridium,* and *Lactobacillus spp.* were markedly elevated[36,38,42-44]. An increase in *Veillonella* species, a potential pathogen in humans, can serve as a biomarker of the severity of certain diseases, such as autoimmune liver disease and cirrhosis[48,49].

**INTESTINAL FLORA AND PSC-MECHANISTIC INSIGHTS**

The leaky gut hypothesis was first proposed by Bjarnason *et al*[50] in 1984, providing theoretical support for the involvement of the intestinal flora in the development of PSC. Normal intestinal flora plays the role of maintaining the balance of the intestinal environment and preventing pathogenic bacteria and toxins from entering the blood circulation[51]. The germ-free (GF) multidrug resistance 2 knockout (Mdr2-/-) mice is a well-studied PSC model that shows a lack of microbial regulation, which is direct evidence that intestinal flora has a key role in PSC development[52]. Intestinal flora dysbiosis damages the intestinal barrier in patients with PSC, allowing bacteria and enteric-derived endotoxins to enter the liver *via* the portal vein, triggering an immune response[53]. Simultaneously, when liver function is impaired, Kupffer cells cannot inactivate endotoxins as efficiently, impairing bile excretion. Furthermore, this increases intestinal permeability, intestinal lymphocyte nesting, and the entry of bacteria and their metabolites [*i.e.* pathogen-associated molecular patterns (PAMPs)] into the liver, impairs normal BA metabolism, and promotes bile duct inflammation and fibrosis (Figure 1)[54,55].

***Intestinal flora dysbiosis activates liver immunity***

**Intestinal flora dysbiosis damages the intestinal barrier:** Lapidot *et al*[43] found that in patients with PSC, a decrease in the relative abundance of commensal bacteria in the intestinal flora, including *Bacteroides thetaiotaomicron* and *Faecalibacterium prausnitzii*, and bacterial diversity led to decreased short-chain fatty acids (SCFAs) with anti-inflammatory effects, such as acetate and butyric acid. This decrease caused intestinal barrier dysfunction and lack of antimicrobial peptides, exacerbating the leaky gut syndrome. When intestinal flora dysregulation occurs in patients with PSC, PAMPs in the gut bind to Toll-like receptors (TLRs) and NOD-like receptors (NLRPs) on the surface of dendritic cells. This event activates the cytoplasmic downstream nuclear transcription factor κB (NF-κB), causing the production and secretion of inflammatory cytokines and chemokines. Disruption of intestinal epithelium tight junctions and the normal intestinal barrier leads to increased intestinal permeability[31,56]. Furthermore, *Enterococcus faecalis* (*E. faecalis*), which increased the most in the intestinal flora of patients with PSC, produces gelatinase, which damages the intestinal epithelium and causes impaired intestinal barrier function[35]. Nakamoto *et al*[57] also found that increased *Klebsiella pneumoniae* during PSC forms pores by disrupting the intestinal epithelium, leading to increased intestinal permeability, thus prompting other bacteria (*e.g.* *Proteus mirabilis* and *Enterococcus gallinarum*) to cross the intestinal barrier. In turn, a Th17 cell-mediated inflammatory response initiates in the liver. Finally, Manfredo *et al*[58] demonstrated that *Enterococcus gallinarum* could reach several organs, such as the mesentery, mesenteric lymph nodes, liver, and spleen, after crossing the damaged intestinal epithelium, causing autoimmune diseases such as PSC.

In addition, PSC recurrence in patients who had undergone liver transplantation was associated with specific intestinal flora changes before transplantation. The rate of PSC recurrence was decreased in patients with a higher abundance of *Shigella spp*. in the intestinal flora before transplantation, suggesting that *Shigella spp*. may reduce bacterial translocation and endotoxemia by improving the intestinal mucus layer function and repairing the intestinal barrier[59].

**Intestinal bacterial translocation induces liver inflammation:** Secondary bacterial overgrowth in the small intestine of rats, achieved by using a blind jejunal loop, led to the translocation of intestinal flora and its metabolite. Consequently, the intestines exhibited characteristic pathological changes of PSC, such as irregular dilatation and bead-like changes in the intra- and extrahepatic bile ducts[60]. Furthermore, Tedesco *et al*[61] found elevated serum interleukin (IL)-17 levels in PSC mice; enriched *Lactobacillus gasseri*, peribiliary collagen deposition, and periportal fibrosis; and increased numbers of IL-17A+ and γδTCR+ cells in mouse liver tissues, which are characteristic inflammatory responses. Additionally, Liao *et al*[62] used Mdr2-/- mice to investigate the role of intestinal flora in PSC, reporting that Mdr2-/- mice had intestinal flora dysbiosis. This caused the NLRP3-mediated innate immune response in the liver, amplified by intestinal barrier failure and enhanced bacterial translocation. Finally, Dhillon *et al*[63] compared the serum soluble cluster of differentiation 14 (sCD14) and lipopolysaccharide-binding protein (LBP) levels of patients with PSC and healthy controls, finding that patients with PSC had elevated levels of sCD14 and LBP. The sCD14 and LBP bind to lipopolysaccharides (typical bacterial translocation markers in humans) in response to significant intestinal flora translocation in patients with PSC[64].

The liver contains many immune cells, including Kupffer cells, natural killer (NK) cells, NK T cells, T cells, and B cells, and is a vital immune organ. In healthy individuals, only a few translocated bacterial products make it to the liver. The liver immune system tolerates these bacterial products to avoid harmful reactions, known as hepatic immune tolerance[65]. The intestinal flora dysbiosis in PSC impairs the intestinal barrier function, allowing bacteria and their products to enter the liver continuously. Thus, the hepatic immune tolerance breaks, inducing local inflammation and immune responses by activating TLR-based pattern recognition receptors on hepatic immune cells. Gram-positive bacteria mainly mediate TLR2 activation, endotoxins mediate TLR4 activation, bacterial flagella mediate TLR5 activation, and unmethylated CpG DNA mediates TLR9 activation[66]. TLR activation promotes a downstream inflammatory cascade that activates the MyD88-mediated NF-κB pathway to induce liver fibrosis[67]. Simultaneously, inflammatory cytokines and chemokines [*e.g.* IL-6 and tumor necrosis factor-α (TNF-α)] are overexpressed, inflammatory cells infiltrate, and oxidative stress and endoplasmic reticulum stress occur in the bile duct epithelium. Eventually, bile duct sclerosis and occlusion, cholestasis, and bile duct fibrosis develop[54,68].

***Intestinal lymphocyte homing exacerbates liver inflammation***

Up to 70% of patients with PSC also develop IBD, suggesting a correlation between the intestine and the liver in patients with PSC and IBD. The discovery of reciprocal transport pathways of lymphocytes to target tissues, as well as the expression of gut-specific adhesion molecules and chemokines in the liver, suggest the homing of intestinal lymphocytes as a contributing factor to PSC pathogenesis[69,70]. Endothelial cells in the hepatic sinusoids of patients with PSC overexpress mucosal vascular addressin cell adhesion molecule 1 (an endothelial adhesion molecule) and C-C motif chemokine ligand 25 (a chemokine), which bind to α4β7 integrin and C-C motif chemokine receptor expressed by intestinal mucosal lymphocytes. This event prompts the recruitment of lymphocytes of an intestinal origin into the liver, which then recognizes the corresponding antigen and triggers an autoimmune response, causing liver injury[71,72]. Trivedi *et al*[41] suggested that this mechanism is associated with hepatic vascular adhesion protein-1 (VAP-1) overexpression. Increased *Veillonella* in the gut of patients with PSC results in primary amine metabolism, which participates in VAP-1 synthesis (as a VAP-1 substrate). Furthermore, hepatic interstitial cells express VAP-1, which recruits intestine-derived T cells to the liver, promoting liver inflammation and fibrosis[73]. Moro-Sibilot *et al*[74] found that elevated levels of sVAP-1 were associated with poor disease outcomes in PSC. High sVAP-1 Levels correlate with the expression of mucosal addressin cell adhesion molecule 1 in the liver, which contributes to the homing of intestinally activated T cells to the hepatobiliary tract[75]. Meanwhile, sVAP-1 triggers oxidative stress in hepatocytes and aggravates liver injury[76]. B cells in the liver are also derived from intestine-associated lymphoid tissue. B cells are activated by intestinal bacteria and enter the liver, producing antibacterial molecules, such as immunoglobin A, that aggravate liver damage.

***Intestinal flora affects PSC through BAs metabolism***

It has been established that several intestinal bacterial genera produce BA hydrolases, such as *Lactobacillus, Clostridium, Enterococcus,* and *Bifidobacterium*. Normal microbial metabolism increases BA diversity as well as hydrophobicity, which facilitates BA excretion[77,78]. Intestinal flora plays a key role in the pathogenesis of PSC by mediating BA biosynthesis and farnesol X receptor (FXR) signaling. FXR regulates BA synthesis through a negative feedback loop thereby affecting the intestinal flora[79]. BAs can directly damage intestinal bacterial cell membranes and indirectly affect the intestinal flora composition by binding to FXR and enhancing the action of antimicrobial peptides. Intestinal flora can also alter BA metabolism by affecting the ab initio synthesis of BAs and enterohepatic circulation[80]. Liwinski *et al*[42] found that patients with PSC had increased taurolithocholic acid concentrations in their bile, which causes inflammation; the levels were closely related to the abundance of *Enterococcus.* BA hydrolase expression, which catalyzes the conversion of primary BAs to secondary BAs, is highest when the human intestinal flora contains *E. faecalis*. Thus, a significant increase in *E. faecalis* in the bile of patients with PSC may affect BA metabolism and cause excessive accumulation of secondary BAs in the body, exacerbating PSC[7,42,81]. Tabibian *et al*[82] found that Mdr2-/- mice produced similar biochemical and histological features of PSC (confirmed by liver pathology and hydroxyproline assays) compared to conventionally reared Mdr2-/- mice; these mice were deficient in secondary BAs due to lack of intestinal flora. Further studies showed that GF-Mdr2-/- mice and antibiotic-induced specific pathogen-free Mdr2-/- mice showed imbalance in BA homeostasis, increased BA reuptake, and accelerated accumulation of harmful BAs in the liver due to dysregulation of intestinal microecology[83].

A recent study showed that *Prevotella copri* in the human gut regulates BA metabolism and transport pathways through gut microbiota interactions, especially the FXR signaling pathway, significantly improving chlorosis and liver fibrosis in 3,5-diethoxy-carbonyl-1,4-dihydropyridine-induced PSC mice[84]. Another study showed that intestinal flora attenuates liver damage by promoting UDCA production. The mechanism of UDCA, which has antioxidant, immunomodulatory, hepatocyte-protective, and membrane-maintaining functions, includes re-establishing the intestinal flora, and is widely used to treat PSC[85]. Lee *et al*[86] found that *Ruminococcus gnavus N53* and *Collinsella aerofaciens* in normal human intestinal flora catalyze the conversion of goose deoxycholic acid to UDCA by expressing the 7β-hydroxysteroid dehydrogenase gene, which increases UDCA acid, thereby reducing liver damage in pathological conditions.

**TARGETED INTESTINAL FLORA MODULATION FOR PSC TREATMENT**

There are no clear and effective options for treating PSC. Pharmacological and endoscopic treatments exist; however, these treatments primarily target the symptoms, and the only effective treatment for end-stage PSC is liver transplantation[16]. In recent years, the incidence of PSC has increased, but intestinal flora research has also expanded, resulting in antimicrobial therapy based on intestinal flora modulation and FMT as potential PSC treatment options[87]. Studies have found that antibiotics, probiotics, and FMT improve intestinal flora disorders, thereby treating PSC (Table 2)[88,89].

***Antibiotics***

Studies have shown that patients with PSC treated with vancomycin had significant reductions in their serum ALP and bilirubin levels and Mayo PSC risk scores (MRSs) and significant improvements in clinical symptoms, such as fatigue and pruritus[90,91]. An open-label prospective therapeutic clinical trial study showed that oral vancomycin was well tolerated in patients with PSC, with peripheral blood γ-gamma-glutamyl transpeptidase (GGT), alanine aminotransferase (ALT) concentrations, white blood cell counts, and neutrophil counts returning to normal from pre-treatment elevated levels within 3 mo of oral administration. Cholangiography, histological, and liver stiffness assessment at the end of follow-up showed improved results, and the trial also showed that that peripheral blood levels of CD4 + CD25hiCD127 Lo and CD4 + FoxP3 + regulatory T cells were also elevated in PSC-IBD patients treated with oral vancomycin[92,93]. Furthermore, Britto *et al*[94] found fewer potentially pathogenic bacteria, such as *Fusobacterium, Haemophilus,* and *Neisseria*, in the intestinal flora of patients with PSC after oral vancomycin treatment. A significant recovery in flora diversity was also observed, suggesting that vancomycin treatment indirectly leads to a secondary increase in bacterial diversity by prompting the intestinal flora to suppress mucosal inflammation. The efficacy of vancomycin for PSC may be related to its selectivity for *Clostridium* *perfringens*[95]. Shah *et al*[96] reported that vancomycin has a relatively narrow antibiotic spectrum and specifically targets *Clostridiales.* Consequently, vancomycin affects the abundance of *Clostridiales* in the intestinal flora of the distal small intestine and colon by reducing primary BA dehydroxylation and preventing excessive secondary BA accumulation, thereby reducing PSC activity. In addition, Davies *et al*[97] demonstrated that vancomycin directly attenuates the inflammatory response to periportal inflammation and liver injury during PSC.

Studies in animal models have demonstrated that metronidazole also has a therapeutic effect on liver injury in PSC[60]. For example, Karvonen *et al*[98] found that treating patients with PSC with both UDCA and metronidazole significantly reduces the serum glutamyl transpeptidase and ALP levels, and significantly improves the MRS and pathological staging compared with those treated with only UDCA. Furthermore, Krehmeier *et al*[99] reported that metronidazole reduced intestinal permeability, decreased bacterial endotoxin entry into the blood, inhibited endotoxin-induced TNF-α production, inhibited hepatic Kupffer cells and macrophage activation, reduced chemokine and cytokine secretion by biliary epithelial cells, attenuated liver inflammation, and prevented PSC-like bead-like liver injury. Finally, Silveira *et al*[100] showed that minocycline is a safe and effective PSC treatment, significantly reducing the ALP level and MRS after one year of oral minocycline administration.

***FMT***

FMT is the transplantation of fecal flora from healthy individuals into a patient’s intestine to replenish or restore normal intestinal flora. This procedure aims to reverse intestinal dysbiosis, regulate product metabolism, and improve clinical symptoms to treat the disease (*Clostridium* difficile infection, IBD, diabetes mellitus, cancer, liver cirrhosis, gut-brain disease and others)[101,102]. FMT restores the health of the intestinal flora, further reducing the transport of harmful metabolites, such as endotoxins to the liver, and reducing the damage caused by metabolites to the liver[103]. FMT uses the principle of bacterial therapy to restore the health of the intestinal flora. The transplanted beneficial bacteria (*Bifidobacteria*, *etc.*) are involved in the conversion of polysaccharides to monosaccharides, producing SCFAs such as acetate, propionate, and butyrate[104]. These metabolites regulate normalization of the intestinal flora and reduce intestinal permeability in patients with liver disease, to further reduce the transport of metabolites such as endogenous ethanol and endotoxins to the liver, thus, reducing the damage to the liver[103,105,106]. Studies have shown intestinal flora normalization, a significant improvement in intestinal flora diversity, reduced cholestasis, and decreased ALP levels in PSC patients after FMT. Allegretti *et al*[107] performed the first human FMT trial in ten patients with PSC who had ALP levels more than three times the normal upper limit. After FMT, 30% of the patients had decreased ALP levels by 50%, and 70% had a 30% reduction in the levels of serum liver transaminases (ALT and aspartate aminotransferase). One week after FMT, the recipients’ intestinal flora diversities were higher than the baseline level of all patients and continued increasing for 24 wk. Furthermore, Philips *et al*[108] found that fecal flora diversity improved in patients with PSC after FMT, with a decrease in the relative abundance of *Proteobacteria* and an increase in the abundances of *Bacteroidetes* and *Firmicutes*; this intestinal flora composition was more similar to that of healthy individuals. The blood biochemistry and total BA indicators also significantly improved.

***Probiotics***

Probiotic is a general term for a group of active microorganisms that have beneficial roles by regulating intestinal flora growth and improving the host’s intestinal microecology. They regulate the intestinal microenvironment metabolism, increase SCFAs production, and reduce the permeability of the intestinal barrier[109,110]. Additionally, probiotics upregulate intestinal epithelial tight junction protein expression, improve intestinal motility[110,111], increase adhesion and colonization of intestinal flora, reduce TNF-α production, and maintain tissue homeostasis[112]. One study demonstrated that oral administration of probiotic preparations (consisting of six strains of viable and freeze-dried bacteria: *Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus salivarius, Lactococcus lactis, Bifidobacterium bifidum,* and *Bifidobacterium lactis*) decreased the serum alkaline phosphatase level by 15 % in patients with PSC compared to healthy individuals[113]. Furthermore, Shimizu *et al*[114] treated a patient with PSC with a combination of prednisolone, salazosulfapyridine, and probiotics, and reported that the patient’s symptoms and tests improved after two weeks. Additionally, repeat pathological biopsies at 30 mo showed significant improvements in liver inflammatory cell infiltration and peribiliary fibrosis. *Lactobacillus plantarum Lp2* has the potential to ameliorate liver injury by inhibiting the activation of LPS-induced inflammatory pathways in the liver, reducing inflammation, and decreasing oxidative damage and apoptosis[115]. Therefore, probiotics have a therapeutic effect on PSC by suppressing intestinal inflammation and maintaining intestinal flora homeostasis.

***BAs and other metabolites***

Compared to conventional mice, germ-free mice show higher concentrations of BA in the plasma and significantly reduced concentrations in the feces. Additionally, FXR signaling is significantly inhibited, resulting in reduced BA synthesis in germ-free mice[116,117]. Colonization of germ-free mice with human feces activates the expression of FXR target genes and increases the levels of BAs in the liver and ileal tissue[118]. FXR agonists inhibit cholesterol 7α-hydroxylase activity and, thus, intracellular BA synthesis. These agonists can activate transcription of the bile salt export pump on the hepatocyte membrane, enhancing the transport of BAs from hepatocytes to bile ducts and promoting BA excretion. Simultaneously, These agonists can inhibit the expression of extracellular matrix proteins in hepatic astrocytes and, thus, prevent the transformation of liver fibrosis in patients with PSC[119]. Obeticholic acid (OCA) is one of FXR agonists representative drugs that alleviates the cholestatic symptoms of PSC by reducing the BA pool[120]. OCA is also approved for the treatment of PSC[121,122]. In fact, there are phase II clinical trials demonstrating the efficacy and safety of OCA in patients with PSC[123].

***Relevant clinical trials***

In addition to the above-mentioned studies, there are currently several relevant clinical trials demonstrating the efficacy of treatments targeting intestinal flora and its metabolites in PSC (Table 3). From these clinical studies, we found that oral vancomycin is the most established for the treatment of PSC, and all phase IV clinical trials using vancomycin have been completed. Vancomycin can significantly reduce biochemical indexes such as ALP and ALT and reduce MRS in patients with PSC[92,124]. One case study also described a decrease in serum γ-GGT, which reached normal levels at 195 d, in pediatric patients with PSC-UC who were administered vancomycin[94]. *Fusobacterium, Haemophilus,* and *Neisseria*, which generally have a significantly high abundance in PSC, showed decreased abundance in the saliva and feces of these patients[40,42,43,47]. Results of meta-analyses have also shown vancomycin to be beneficial in patients with PSC[96]. Currently, there are clinical guidelines recommending the use of antimicrobial agents and FXR agonists for the treatment of PSC[125,126]. Clinical trials of UDCA for PSC are also well established[127]. UDCA is a hydrophilic dihydroxy BA, and pharmacological studies have confirmed that UDCA has a strong affinity in bile, promoting bile secretion, protecting bile duct cells from the cytotoxicity of hydrophobic BAs, and protecting hepatocytes from BA-induced apoptosis[128]. It promotes the formation of liquid cholesterol crystal complexes, accelerates cholesterol excretion and clearance to the intestine, acts as a cholagogue, and competitively inhibits endogenous hepatic BA absorption in the small intestine, reducing serum BA levels[129]. 24-norUDCA is a side chain shortened congener of C23UDCA, which makes a bile hepatic shunt possible. Based on its pharmacological properties of relative amidation resistance and reduced secondary BA production, norUDCA is a promising drug for a range of cholestatic liver and bile duct diseases[130]. Some clinical trials have shown that norUDCA improved cholestasis and significantly reduced serum alkaline phosphatase levels in patients after 12 wk in a dose-dependent manner. Importantly, norUDCA treatment has shown a good safety profile[131]. OCA is a potent FXR agonist that affects the hepatic transport of conjugated BAs in humans and reduces duration of hepatocyte exposure to potentially cytotoxic BAs[132,133]. Clinical trials have demonstrated the efficacy and safety of OCA in patients with PSC; Treatment with OCA 5-10 mg resulted in a significant reduction in ALP in patients with PSC after 24 wk[123]. In addition, clinical studies of probiotics, FMT, and other approaches targeting intestinal flora for the treatment of PSC are ongoing to highlight their efficacy and safety in PSC and demonstrate their therapeutic potential[108,134].

**CONCLUSION**

PSC is a chronic progressive autoimmune disease that can develop into cirrhosis or liver failure, thereby severely affecting the patient’s quality of life if not actively and effectively treated. Intestinal flora dysbiosis is crucial in the occurrence and development of PSC, as it destroys the intestinal barrier and prompts intestinal lymphocyte homing and translocation of bacteria and their metabolites, thus aggravating the immune damage to the liver. The intestinal flora also interacts with BAs and participates in PSC development.

Our understanding of the gut flora has expanded with the development of genomics, metabolomics, and high-throughput sequencing technologies. These research approaches help elucidate the complex role of the gut flora in diseases, such as PSC. Technological advances have also provided individualized treatment options for patients with PSC that target the intestinal flora with good clinical results. Treatments, including antibiotics, FMT, and probiotics, have offered new ideas for managing PSC. More precise therapies, such as probiotics, synbiotics, and phages, have shown promising results in PSC patients. However, there remain some challenges in the use of intestinal flora for PSC treatment. The intestinal flora regulation mechanisms for PSC are not fully understood, and the optimal method and timing have not been standardized. Future prospective studies with a large sample size or multi-center studies are warranted to provide direct evidence of the role of the intestinal flora in PSC and establish a therapeutic protocol for the use of the intestinal flora. If these issues are resolved, targeted regulation of the intestinal flora will become a new option for PSC treatment.

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**REFERENCES**

1 **Dyson JK**, Beuers U, Jones DEJ, Lohse AW, Hudson M. Primary sclerosing cholangitis. *Lancet* 2018; **391**: 2547-2559 [PMID: 29452711 DOI: 10.1016/S0140-6736(18)30300-3]

2 **Fung BM**, Lindor KD, Tabibian JH. Cancer risk in primary sclerosing cholangitis: Epidemiology, prevention, and surveillance strategies. *World J Gastroenterol* 2019; **25**: 659-671 [PMID: 30783370 DOI: 10.3748/wjg.v25.i6.659]

3 **Rabiee A**, Silveira MG. Primary sclerosing cholangitis. *Transl Gastroenterol Hepatol* 2021; **6**: 29 [PMID: 33824933 DOI: 10.21037/tgh-20-266]

4 **Bruneau A**, Hundertmark J, Guillot A, Tacke F. Molecular and Cellular Mediators of the Gut-Liver Axis in the Progression of Liver Diseases. *Front Med (Lausanne)* 2021; **8**: 725390 [PMID: 34650994 DOI: 10.3389/fmed.2021.725390]

5 **Mack CL**, Adams D, Assis DN, Kerkar N, Manns MP, Mayo MJ, Vierling JM, Alsawas M, Murad MH, Czaja AJ. Diagnosis and Management of Autoimmune Hepatitis in Adults and Children: 2019 Practice Guidance and Guidelines From the American Association for the Study of Liver Diseases. *Hepatology* 2020; **72**: 671-722 [PMID: 31863477 DOI: 10.1002/hep.31065]

6 **Cao RR**, He P, Lei SF. Novel microbiota-related gene set enrichment analysis identified osteoporosis associated gut microbiota from autoimmune diseases. *J Bone Miner Metab* 2021; **39**: 984-996 [PMID: 34338852 DOI: 10.1007/s00774-021-01247-w]

7 **Iwasawa K**, Suda W, Tsunoda T, Oikawa-Kawamoto M, Umetsu S, Inui A, Fujisawa T, Morita H, Sogo T, Hattori M. Characterisation of the faecal microbiota in Japanese patients with paediatric-onset primary sclerosing cholangitis. *Gut* 2017; **66**: 1344-1346 [PMID: 27670376 DOI: 10.1136/gutjnl-2016-312533]

8 **Torres J**, Bao X, Goel A, Colombel JF, Pekow J, Jabri B, Williams KM, Castillo A, Odin JA, Meckel K, Fasihuddin F, Peter I, Itzkowitz S, Hu J. The features of mucosa-associated microbiota in primary sclerosing cholangitis. *Aliment Pharmacol Ther* 2016; **43**: 790-801 [PMID: 26857969 DOI: 10.1111/apt.13552]

9 **Tornai T**, Palyu E, Vitalis Z, Tornai I, Tornai D, Antal-Szalmas P, Norman GL, Shums Z, Veres G, Dezsofi A, Par G, Par A, Orosz P, Szalay F, Lakatos PL, Papp M. Gut barrier failure biomarkers are associated with poor disease outcome in patients with primary sclerosing cholangitis. *World J Gastroenterol* 2017; **23**: 5412-5421 [PMID: 28839442 DOI: 10.3748/wjg.v23.i29.5412]

10 **Mehta TI**, Weissman S, Fung BM, Sotiriadis J, Lindor KD, Tabibian JH. Global incidence, prevalence and features of primary sclerosing cholangitis: A systematic review and meta-analysis. *Liver Int* 2021; **41**: 2418-2426 [PMID: 34224208 DOI: 10.1111/liv.15007]

11 **Floreani A**, De Martin S. Treatment of primary sclerosing cholangitis. *Dig Liver Dis* 2021; **53**: 1531-1538 [PMID: 34011480 DOI: 10.1016/j.dld.2021.04.028]

12 **Karlsen TH**, Folseraas T, Thorburn D, Vesterhus M. Primary sclerosing cholangitis - a comprehensive review. *J Hepatol* 2017; **67**: 1298-1323 [PMID: 28802875 DOI: 10.1016/j.jhep.2017.07.022]

13 **Liu C**, Wang YL, Yang YY, Zhang NP, Niu C, Shen XZ, Wu J. Novel approaches to intervene gut microbiota in the treatment of chronic liver diseases. *FASEB J* 2021; **35**: e21871 [PMID: 34473374 DOI: 10.1096/fj.202100939R]

14 **Gallo C**, Howardson BO, Cristoferi L, Carbone M, Gershwin ME, Invernizzi P. An update on novel pharmacological agents for primary sclerosing cholangitis. *Expert Opin Ther Targets* 2022; **26**: 69-77 [PMID: 35040733 DOI: 10.1080/14728222.2022.2030707]

15 **Nicoletti A**, Maurice JB, Thorburn D. Guideline review: British Society of Gastroenterology/UK-PSC guidelines for the diagnosis and management of primary sclerosing cholangitis. *Frontline Gastroenterol* 2021; **12**: 62-66 [PMID: 33456743 DOI: 10.1136/flgastro-2019-101343]

16 **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]

17 **Liu JZ**, Hov JR, Folseraas T, Ellinghaus E, Rushbrook SM, Doncheva NT, Andreassen OA, Weersma RK, Weismüller TJ, Eksteen B, Invernizzi P, Hirschfield GM, Gotthardt DN, Pares A, Ellinghaus D, Shah T, Juran BD, Milkiewicz P, Rust C, Schramm C, Müller T, Srivastava B, Dalekos G, Nöthen MM, Herms S, Winkelmann J, Mitrovic M, Braun F, Ponsioen CY, Croucher PJ, Sterneck M, Teufel A, Mason AL, Saarela J, Leppa V, Dorfman R, Alvaro D, Floreani A, Onengut-Gumuscu S, Rich SS, Thompson WK, Schork AJ, Næss S, Thomsen I, Mayr G, König IR, Hveem K, Cleynen I, Gutierrez-Achury J, Ricaño-Ponce I, van Heel D, Björnsson E, Sandford RN, Durie PR, Melum E, Vatn MH, Silverberg MS, Duerr RH, Padyukov L, Brand S, Sans M, Annese V, Achkar JP, Boberg KM, Marschall HU, Chazouillères O, Bowlus CL, Wijmenga C, Schrumpf E, Vermeire S, Albrecht M; UK-PSCSC Consortium, Rioux JD, Alexander G, Bergquist A, Cho J, Schreiber S, Manns MP, Färkkilä M, Dale AM, Chapman RW, Lazaridis KN; International PSC Study Group, Franke A, Anderson CA, Karlsen TH; International IBD Genetics Consortium. Dense genotyping of immune-related disease regions identifies nine new risk loci for primary sclerosing cholangitis. *Nat Genet* 2013; **45**: 670-675 [PMID: 23603763 DOI: 10.1038/ng.2616]

18 **Park JW**, Kim JH, Kim SE, Jung JH, Jang MK, Park SH, Lee MS, Kim HS, Suk KT, Kim DJ. Primary Biliary Cholangitis and Primary Sclerosing Cholangitis: Current Knowledge of Pathogenesis and Therapeutics. *Biomedicines* 2022; **10** [PMID: 35740310 DOI: 10.3390/biomedicines10061288]

19 **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]

20 **Albillos A**, de Gottardi A, Rescigno M. The gut-liver axis in liver disease: Pathophysiological basis for therapy. *J Hepatol* 2020; **72**: 558-577 [PMID: 31622696 DOI: 10.1016/j.jhep.2019.10.003]

21 **Wang Y**, Liu Y. Gut-liver-axis: Barrier function of liver sinusoidal endothelial cell. *J Gastroenterol Hepatol* 2021; **36**: 2706-2714 [PMID: 33811372 DOI: 10.1111/jgh.15512]

22 **Wang R**, Tang R, Li B, Ma X, Schnabl B, Tilg H. Gut microbiome, liver immunology, and liver diseases. *Cell Mol Immunol* 2021; **18**: 4-17 [PMID: 33318628 DOI: 10.1038/s41423-020-00592-6]

23 **Bambha K**, Kim WR, Talwalkar J, Torgerson H, Benson JT, Therneau TM, Loftus EV Jr, Yawn BP, Dickson ER, Melton LJ 3rd. Incidence, clinical spectrum, and outcomes of primary sclerosing cholangitis in a United States community. *Gastroenterology* 2003; **125**: 1364-1369 [PMID: 14598252 DOI: 10.1016/j.gastro.2003.07.011]

24 **Weismüller TJ**, Trivedi PJ, Bergquist A, Imam M, Lenzen H, Ponsioen CY, Holm K, Gotthardt D, Färkkilä MA, Marschall HU, Thorburn D, Weersma RK, Fevery J, Mueller T, Chazouillères O, Schulze K, Lazaridis KN, Almer S, Pereira SP, Levy C, Mason A, Naess S, Bowlus CL, Floreani A, Halilbasic E, Yimam KK, Milkiewicz P, Beuers U, Huynh DK, Pares A, Manser CN, Dalekos GN, Eksteen B, Invernizzi P, Berg CP, Kirchner GI, Sarrazin C, Zimmer V, Fabris L, Braun F, Marzioni M, Juran BD, Said K, Rupp C, Jokelainen K, Benito de Valle M, Saffioti F, Cheung A, Trauner M, Schramm C, Chapman RW, Karlsen TH, Schrumpf E, Strassburg CP, Manns MP, Lindor KD, Hirschfield GM, Hansen BE, Boberg KM; International PSC Study Group. Patient Age, Sex, and Inflammatory Bowel Disease Phenotype Associate With Course of Primary Sclerosing Cholangitis. *Gastroenterology* 2017; **152**: 1975-1984.e8 [PMID: 28274849 DOI: 10.1053/j.gastro.2017.02.038]

25 **Liang H**, Manne S, Shick J, Lissoos T, Dolin P. Incidence, prevalence, and natural history of primary sclerosing cholangitis in the United Kingdom. *Medicine (Baltimore)* 2017; **96**: e7116 [PMID: 28614231 DOI: 10.1097/MD.0000000000007116]

26 **Palmela C**, Peerani F, Castaneda D, Torres J, Itzkowitz SH. Inflammatory Bowel Disease and Primary Sclerosing Cholangitis: A Review of the Phenotype and Associated Specific Features. *Gut Liver* 2018; **12**: 17-29 [PMID: 28376583 DOI: 10.5009/gnl16510]

27 **Da Cunha T**, Vaziri H, Wu GY. Primary Sclerosing Cholangitis and Inflammatory Bowel Disease: A Review. *J Clin Transl Hepatol* 2022; **10**: 531-542 [PMID: 35836773 DOI: 10.14218/JCTH.2021.00344]

28 **Lazaridis KN**, LaRusso NF. The Cholangiopathies. *Mayo Clin Proc* 2015; **90**: 791-800 [PMID: 25957621 DOI: 10.1016/j.mayocp.2015.03.017]

29 **Yu D**, Meng X, de Vos WM, Wu H, Fang X, Maiti AK. Implications of Gut Microbiota in Complex Human Diseases. *Int J Mol Sci* 2021; **22** [PMID: 34884466 DOI: 10.3390/ijms222312661]

30 **Weinstock GM**. Genomic approaches to studying the human microbiota. *Nature* 2012; **489**: 250-256 [PMID: 22972298 DOI: 10.1038/nature11553]

31 **Kho ZY**, Lal SK. The Human Gut Microbiome - A Potential Controller of Wellness and Disease. *Front Microbiol* 2018; **9**: 1835 [PMID: 30154767 DOI: 10.3389/fmicb.2018.01835]

32 **Hou K**, Wu ZX, Chen XY, Wang JQ, Zhang D, Xiao C, Zhu D, Koya JB, Wei L, Li J, Chen ZS. Microbiota in health and diseases. *Signal Transduct Target Ther* 2022; **7**: 135 [PMID: 35461318 DOI: 10.1038/s41392-022-00974-4]

33 **Tranah TH**, Edwards LA, Schnabl B, Shawcross DL. Targeting the gut-liver-immune axis to treat cirrhosis. *Gut* 2021; **70**: 982-994 [PMID: 33060124 DOI: 10.1136/gutjnl-2020-320786]

34 **Rossen NG**, Fuentes S, Boonstra K, D'Haens GR, Heilig HG, Zoetendal EG, de Vos WM, Ponsioen CY. The mucosa-associated microbiota of PSC patients is characterized by low diversity and low abundance of uncultured Clostridiales II. *J Crohns Colitis* 2015; **9**: 342-348 [PMID: 25547975 DOI: 10.1093/ecco-jcc/jju023]

35 **Sabino J**, Vieira-Silva S, Machiels K, Joossens M, Falony G, Ballet V, Ferrante M, Van Assche G, Van der Merwe S, Vermeire S, Raes J. Primary sclerosing cholangitis is characterised by intestinal dysbiosis independent from IBD. *Gut* 2016; **65**: 1681-1689 [PMID: 27207975 DOI: 10.1136/gutjnl-2015-311004]

36 **Kummen M**, Holm K, Anmarkrud JA, Nygård S, Vesterhus M, Høivik ML, Trøseid M, Marschall HU, Schrumpf E, Moum B, Røsjø H, Aukrust P, Karlsen TH, Hov JR. The gut microbial profile in patients with primary sclerosing cholangitis is distinct from patients with ulcerative colitis without biliary disease and healthy controls. *Gut* 2017; **66**: 611-619 [PMID: 26887816 DOI: 10.1136/gutjnl-2015-310500]

37 **Quraishi MN**, Sergeant M, Kay G, Iqbal T, Chan J, Constantinidou C, Trivedi P, Ferguson J, Adams DH, Pallen M, Hirschfield GM. The gut-adherent microbiota of PSC-IBD is distinct to that of IBD. *Gut* 2017; **66**: 386-388 [PMID: 27196590 DOI: 10.1136/gutjnl-2016-311915]

38 **Rühlemann M**, Liwinski T, Heinsen FA, Bang C, Zenouzi R, Kummen M, Thingholm L, Tempel M, Lieb W, Karlsen T, Lohse A, Hov J, Denk G, Lammert F, Krawczyk M, Schramm C, Franke A. Consistent alterations in faecal microbiomes of patients with primary sclerosing cholangitis independent of associated colitis. *Aliment Pharmacol Ther* 2019; **50**: 580-589 [PMID: 31250469 DOI: 10.1111/apt.15375]

39 **Rühlemann MC**, Heinsen FA, Zenouzi R, Lieb W, Franke A, Schramm C. Faecal microbiota profiles as diagnostic biomarkers in primary sclerosing cholangitis. *Gut* 2017; **66**: 753-754 [PMID: 27216937 DOI: 10.1136/gutjnl-2016-312180]

40 **Quraishi MN**, Acharjee A, Beggs AD, Horniblow R, Tselepis C, Gkoutos G, Ghosh S, Rossiter AE, Loman N, van Schaik W, Withers D, Walters JRF, Hirschfield GM, Iqbal TH. A Pilot Integrative Analysis of Colonic Gene Expression, Gut Microbiota, and Immune Infiltration in Primary Sclerosing Cholangitis-Inflammatory Bowel Disease: Association of Disease With Bile Acid Pathways. *J Crohns Colitis* 2020; **14**: 935-947 [PMID: 32016358 DOI: 10.1093/ecco-jcc/jjaa021]

41 **Trivedi PJ**, Tickle J, Vesterhus MN, Eddowes PJ, Bruns T, Vainio J, Parker R, Smith D, Liaskou E, Thorbjørnsen LW, Hirschfield GM, Auvinen K, Hubscher SG, Salmi M, Adams DH, Weston CJ. Vascular adhesion protein-1 is elevated in primary sclerosing cholangitis, is predictive of clinical outcome and facilitates recruitment of gut-tropic lymphocytes to liver in a substrate-dependent manner. *Gut* 2018; **67**: 1135-1145 [PMID: 28428344 DOI: 10.1136/gutjnl-2016-312354]

42 **Liwinski T**, Zenouzi R, John C, Ehlken H, Rühlemann MC, Bang C, Groth S, Lieb W, Kantowski M, Andersen N, Schachschal G, Karlsen TH, Hov JR, Rösch T, Lohse AW, Heeren J, Franke A, Schramm C. Alterations of the bile microbiome in primary sclerosing cholangitis. *Gut* 2020; **69**: 665-672 [PMID: 31243055 DOI: 10.1136/gutjnl-2019-318416]

43 **Lapidot Y**, Amir A, Ben-Simon S, Veitsman E, Cohen-Ezra O, Davidov Y, Weiss P, Bradichevski T, Segev S, Koren O, Ben-Ari Z, Safran M. Alterations of the salivary and fecal microbiome in patients with primary sclerosing cholangitis. *Hepatol Int* 2021; **15**: 191-201 [PMID: 32949377 DOI: 10.1007/s12072-020-10089-z]

44 **Lemoinne S**, Kemgang A, Ben Belkacem K, Straube M, Jegou S, Corpechot C; Saint-Antoine IBD Network, Chazouillères O, Housset C, Sokol H. Fungi participate in the dysbiosis of gut microbiota in patients with primary sclerosing cholangitis. *Gut* 2020; **69**: 92-102 [PMID: 31003979 DOI: 10.1136/gutjnl-2018-317791]

45 **Kummen M**, Thingholm LB, Rühlemann MC, Holm K, Hansen SH, Moitinho-Silva L, Liwinski T, Zenouzi R, Storm-Larsen C, Midttun Ø, McCann A, Ueland PM, Høivik ML, Vesterhus M, Trøseid M, Laudes M, Lieb W, Karlsen TH, Bang C, Schramm C, Franke A, Hov JR. Altered Gut Microbial Metabolism of Essential Nutrients in Primary Sclerosing Cholangitis. *Gastroenterology* 2021; **160**: 1784-1798.e0 [PMID: 33387530 DOI: 10.1053/j.gastro.2020.12.058]

46 **Kummen M**, Hov JR. The gut microbial influence on cholestatic liver disease. *Liver Int* 2019; **39**: 1186-1196 [PMID: 31125502 DOI: 10.1111/liv.14153]

47 **Bajer L**, Kverka M, Kostovcik M, Macinga P, Dvorak J, Stehlikova Z, Brezina J, Wohl P, Spicak J, Drastich P. Distinct gut microbiota profiles in patients with primary sclerosing cholangitis and ulcerative colitis. *World J Gastroenterol* 2017; **23**: 4548-4558 [PMID: 28740343 DOI: 10.3748/wjg.v23.i25.4548]

48 **Wei Y**, Li Y, Yan L, Sun C, Miao Q, Wang Q, Xiao X, Lian M, Li B, Chen Y, Zhang J, Li Y, Huang B, Li Y, Cao Q, Fan Z, Chen X, Fang JY, Gershwin ME, Tang R, Ma X. Alterations of gut microbiome in autoimmune hepatitis. *Gut* 2020; **69**: 569-577 [PMID: 31201284 DOI: 10.1136/gutjnl-2018-317836]

49 **Lang S**, Fairfied B, Gao B, Duan Y, Zhang X, Fouts DE, Schnabl B. Changes in the fecal bacterial microbiota associated with disease severity in alcoholic hepatitis patients. *Gut Microbes* 2020; **12**: 1785251 [PMID: 32684075 DOI: 10.1080/19490976.2020.1785251]

50 **Bjarnason I**, Peters TJ, Wise RJ. The leaky gut of alcoholism: possible route of entry for toxic compounds. *Lancet* 1984; **1**: 179-182 [PMID: 6141332 DOI: 10.1016/s0140-6736(84)92109-3]

51 **Milosevic I**, Vujovic A, Barac A, Djelic M, Korac M, Radovanovic Spurnic A, Gmizic I, Stevanovic O, Djordjevic V, Lekic N, Russo E, Amedei A. Gut-Liver Axis, Gut Microbiota, and Its Modulation in the Management of Liver Diseases: A Review of the Literature. *Int J Mol Sci* 2019; **20** [PMID: 30658519 DOI: 10.3390/ijms20020395]

52 **Li M**, Ping J, Xu LM. [Application of Mdr2 gene knockout mice in liver disease research]. *Zhonghua Gan Zang Bing Za Zhi* 2021; **29**: 585-590 [PMID: 34225436 DOI: 10.3760/cma.j.cn501113-20191007-00364]

53 **Zheng Y**, Ran Y, Zhang H, Wang B, Zhou L. The Microbiome in Autoimmune Liver Diseases: Metagenomic and Metabolomic Changes. *Front Physiol* 2021; **12**: 715852 [PMID: 34690796 DOI: 10.3389/fphys.2021.715852]

54 **Tripathi A**, Debelius J, Brenner DA, Karin M, Loomba R, Schnabl B, Knight R. The gut-liver axis and the intersection with the microbiome. *Nat Rev Gastroenterol Hepatol* 2018; **15**: 397-411 [PMID: 29748586 DOI: 10.1038/s41575-018-0011-z]

55 **Bozward AG**, Ronca V, Osei-Bordom D, Oo YH. Gut-Liver Immune Traffic: Deciphering Immune-Pathogenesis to Underpin Translational Therapy. *Front Immunol* 2021; **12**: 711217 [PMID: 34512631 DOI: 10.3389/fimmu.2021.711217]

56 **Maroni L**, Ninfole E, Pinto C, Benedetti A, Marzioni M. Gut-Liver Axis and Inflammasome Activation in Cholangiocyte Pathophysiology. *Cells* 2020; **9** [PMID: 32192118 DOI: 10.3390/cells9030736]

57 **Nakamoto N**, Sasaki N, Aoki R, Miyamoto K, Suda W, Teratani T, Suzuki T, Koda Y, Chu PS, Taniki N, Yamaguchi A, Kanamori M, Kamada N, Hattori M, Ashida H, Sakamoto M, Atarashi K, Narushima S, Yoshimura A, Honda K, Sato T, Kanai T. Gut pathobionts underlie intestinal barrier dysfunction and liver T helper 17 cell immune response in primary sclerosing cholangitis. *Nat Microbiol* 2019; **4**: 492-503 [PMID: 30643240 DOI: 10.1038/s41564-018-0333-1]

58 **Manfredo Vieira S**, Hiltensperger M, Kumar V, Zegarra-Ruiz D, Dehner C, Khan N, Costa FRC, Tiniakou E, Greiling T, Ruff W, Barbieri A, Kriegel C, Mehta SS, Knight JR, Jain D, Goodman AL, Kriegel MA. Translocation of a gut pathobiont drives autoimmunity in mice and humans. *Science* 2018; **359**: 1156-1161 [PMID: 29590047 DOI: 10.1126/science.aar7201]

59 **Visseren T**, Fuhler GM, Erler NS, Nossent YRA, Metselaar HJ, IJzermans JNM, Darwish Murad S, Peppelenbosch MP. Recurrence of primary sclerosing cholangitis after liver transplantation is associated with specific changes in the gut microbiome pretransplant - a pilot study. *Transpl Int* 2020; **33**: 1424-1436 [PMID: 33617049 DOI: 10.1111/tri.13692]

60 **Lichtman SN**, Keku J, Schwab JH, Sartor RB. Hepatic injury associated with small bowel bacterial overgrowth in rats is prevented by metronidazole and tetracycline. *Gastroenterology* 1991; **100**: 513-519 [PMID: 1985047 DOI: 10.1016/0016-5085(91)90224-9]

61 **Tedesco D**, Thapa M, Chin CY, Ge Y, Gong M, Li J, Gumber S, Speck P, Elrod EJ, Burd EM, Kitchens WH, Magliocca JF, Adams AB, Weiss DS, Mohamadzadeh M, Grakoui A. Alterations in Intestinal Microbiota Lead to Production of Interleukin 17 by Intrahepatic γδ T-Cell Receptor-Positive Cells and Pathogenesis of Cholestatic Liver Disease. *Gastroenterology* 2018; **154**: 2178-2193 [PMID: 29454797 DOI: 10.1053/j.gastro.2018.02.019]

62 **Liao L**, Schneider KM, Galvez EJC, Frissen M, Marschall HU, Su H, Hatting M, Wahlström A, Haybaeck J, Puchas P, Mohs A, Peng J, Bergheim I, Nier A, Hennings J, Reißing J, Zimmermann HW, Longerich T, Strowig T, Liedtke C, Cubero FJ, Trautwein C. Intestinal dysbiosis augments liver disease progression via NLRP3 in a murine model of primary sclerosing cholangitis. *Gut* 2019; **68**: 1477-1492 [PMID: 30872395 DOI: 10.1136/gutjnl-2018-316670]

63 **Dhillon AK**, Kummen M, Trøseid M, Åkra S, Liaskou E, Moum B, Vesterhus M, Karlsen TH, Seljeflot I, Hov JR. Circulating markers of gut barrier function associated with disease severity in primary sclerosing cholangitis. *Liver Int* 2019; **39**: 371-381 [PMID: 30269440 DOI: 10.1111/liv.13979]

64 **Wright SD**, Ramos RA, Tobias PS, Ulevitch RJ, Mathison JC. CD14, a receptor for complexes of lipopolysaccharide (LPS) and LPS binding protein. *Science* 1990; **249**: 1431-1433 [PMID: 1698311 DOI: 10.1126/science.1698311]

65 **Doherty DG**. Immunity, tolerance and autoimmunity in the liver: A comprehensive review. *J Autoimmun* 2016; **66**: 60-75 [PMID: 26358406 DOI: 10.1016/j.jaut.2015.08.020]

66 **Yamamoto M**, Takeda K. Current views of toll-like receptor signaling pathways. *Gastroenterol Res Pract* 2010; **2010**: 240365 [PMID: 21197425 DOI: 10.1155/2010/240365]

67 **Seki E**, Schnabl B. Role of innate immunity and the microbiota in liver fibrosis: crosstalk between the liver and gut. *J Physiol* 2012; **590**: 447-458 [PMID: 22124143 DOI: 10.1113/jphysiol.2011.219691]

68 **Anand G**, Zarrinpar A, Loomba R. Targeting Dysbiosis for the Treatment of Liver Disease. *Semin Liver Dis* 2016; **36**: 37-47 [PMID: 26870931 DOI: 10.1055/s-0035-1571276]

69 **de Krijger M**, Visseren T, Wildenberg ME, Hooijer GKJ, Verstegen MMA, van der Laan LJW, de Jonge WJ, Verheij J, Ponsioen CY. Characterization of gut-homing molecules in non-endstage livers of patients with primary sclerosing cholangitis and inflammatory bowel disease. *J Transl Autoimmun* 2020; **3**: 100054 [PMID: 32743534 DOI: 10.1016/j.jtauto.2020.100054]

70 **de Krijger M**, Wildenberg ME, de Jonge WJ, Ponsioen CY. Return to sender: Lymphocyte trafficking mechanisms as contributors to primary sclerosing cholangitis. *J Hepatol* 2019; **71**: 603-615 [PMID: 31108158 DOI: 10.1016/j.jhep.2019.05.006]

71 **Trivedi PJ**, Bruns T, Ward S, Mai M, Schmidt C, Hirschfield GM, Weston CJ, Adams DH. Intestinal CCL25 expression is increased in colitis and correlates with inflammatory activity. *J Autoimmun* 2016; **68**: 98-104 [PMID: 26873648 DOI: 10.1016/j.jaut.2016.01.001]

72 **Schippers A**, Hübel J, Heymann F, Clahsen T, Eswaran S, Schlepütz S, Püllen R, Gaßler N, Tenbrock K, Tacke F, Wagner N. MAdCAM-1/α4β7 Integrin-Mediated Lymphocyte/Endothelium Interactions Exacerbate Acute Immune-Mediated Hepatitis in Mice. *Cell Mol Gastroenterol Hepatol* 2021; **11**: 1227-1250.e1 [PMID: 33316453 DOI: 10.1016/j.jcmgh.2020.12.003]

73 **UniProt Consortium T**. UniProt: the universal protein knowledgebase. *Nucleic Acids Res* 2018; **46**: 2699 [PMID: 29425356 DOI: 10.1093/nar/gky092]

74 **Moro-Sibilot L**, Blanc P, Taillardet M, Bardel E, Couillault C, Boschetti G, Traverse-Glehen A, Defrance T, Kaiserlian D, Dubois B. Mouse and Human Liver Contain Immunoglobulin A-Secreting Cells Originating From Peyer's Patches and Directed Against Intestinal Antigens. *Gastroenterology* 2016; **151**: 311-323 [PMID: 27132185 DOI: 10.1053/j.gastro.2016.04.014]

75 **Tornai D**, Ven PL, Lakatos PL, Papp M. Serological biomarkers for management of primary sclerosing cholangitis. *World J Gastroenterol* 2022; **28**: 2291-2301 [PMID: 35800183 DOI: 10.3748/wjg.v28.i21.2291]

76 **Weston CJ**, Shepherd EL, Claridge LC, Rantakari P, Curbishley SM, Tomlinson JW, Hubscher SG, Reynolds GM, Aalto K, Anstee QM, Jalkanen S, Salmi M, Smith DJ, Day CP, Adams DH. Vascular adhesion protein-1 promotes liver inflammation and drives hepatic fibrosis. *J Clin Invest* 2015; **125**: 501-520 [PMID: 25562318 DOI: 10.1172/JCI73722]

77 **Horáčková Š**, Plocková M, Demnerová K. Importance of microbial defence systems to bile salts and mechanisms of serum cholesterol reduction. *Biotechnol Adv* 2018; **36**: 682-690 [PMID: 29248683 DOI: 10.1016/j.biotechadv.2017.12.005]

78 **Wahlström A**, Sayin SI, Marschall HU, Bäckhed F. Intestinal Crosstalk between Bile Acids and Microbiota and Its Impact on Host Metabolism. *Cell Metab* 2016; **24**: 41-50 [PMID: 27320064 DOI: 10.1016/j.cmet.2016.05.005]

79 **Fickert P**, Wagner M. Biliary bile acids in hepatobiliary injury - What is the link? *J Hepatol* 2017; **67**: 619-631 [PMID: 28712691 DOI: 10.1016/j.jhep.2017.04.026]

80 **Schneider KM**, Candels LS, Hov JR, Myllys M, Hassan R, Schneider CV, Wahlström A, Mohs A, Zühlke S, Liao L, Elfers C, Kilic K, Henricsson M, Molinaro A, Hatting M, Zaza A, Drasdo D, Frissen M, Devlin AS, Gálvez EJC, Strowig T, Karlsen TH, Hengstler JG, Marschall HU, Ghallab A, Trautwein C. Gut microbiota depletion exacerbates cholestatic liver injury via loss of FXR signalling. *Nat Metab* 2021; **3**: 1228-1241 [PMID: 34552267 DOI: 10.1038/s42255-021-00452-1]

81 **Chand D**, Panigrahi P, Varshney N, Ramasamy S, Suresh CG. Structure and function of a highly active Bile Salt Hydrolase (BSH) from Enterococcus faecalis and post-translational processing of BSH enzymes. *Biochim Biophys Acta Proteins Proteom* 2018; **1866**: 507-518 [PMID: 29325872 DOI: 10.1016/j.bbapap.2018.01.003]

82 **Tabibian JH**, O'Hara SP, Trussoni CE, Tietz PS, Splinter PL, Mounajjed T, Hagey LR, LaRusso NF. Absence of the intestinal microbiota exacerbates hepatobiliary disease in a murine model of primary sclerosing cholangitis. *Hepatology* 2016; **63**: 185-196 [PMID: 26044703 DOI: 10.1002/hep.27927]

83 **Awoniyi M**, Wang J, Ngo B, Meadows V, Tam J, Viswanathan A, Lai Y, Montgomery S, Farmer M, Kummen M, Thingholm L, Schramm C, Bang C, Franke A, Lu K, Zhou H, Bajaj JS, Hylemon PB, Ting J, Popov YV, Hov JR, Francis HL, Sartor RB. Protective and aggressive bacterial subsets and metabolites modify hepatobiliary inflammation and fibrosis in a murine model of PSC. *Gut* 2022 [PMID: 35705368 DOI: 10.1136/gutjnl-2021-326500]

84 **Jiang B**, Yuan G, Wu J, Wu Q, Li L, Jiang P. Prevotella copri ameliorates cholestasis and liver fibrosis in primary sclerosing cholangitis by enhancing the FXR signalling pathway. *Biochim Biophys Acta Mol Basis Dis* 2022; **1868**: 166320 [PMID: 34896545 DOI: 10.1016/j.bbadis.2021.166320]

85 **Poupon R**, Poupon RE. Ursodeoxycholic acid therapy of chronic cholestatic conditions in adults and children. *Pharmacol Ther* 1995; **66**: 1-15 [PMID: 7630925 DOI: 10.1016/0163-7258(94)00073-c]

86 **Lee JY**, Arai H, Nakamura Y, Fukiya S, Wada M, Yokota A. Contribution of the 7β-hydroxysteroid dehydrogenase from Ruminococcus gnavus N53 to ursodeoxycholic acid formation in the human colon. *J Lipid Res* 2013; **54**: 3062-3069 [PMID: 23729502 DOI: 10.1194/jlr.M039834]

87 **Shah A**, Macdonald GA, Morrison M, Holtmann G. Targeting the Gut Microbiome as a Treatment for Primary Sclerosing Cholangitis: A Conceptional Framework. *Am J Gastroenterol* 2020; **115**: 814-822 [PMID: 32250997 DOI: 10.14309/ajg.0000000000000604]

88 **Kanmani P**, Suganya K, Kim H. The Gut Microbiota: How Does It Influence the Development and Progression of Liver Diseases. *Biomedicines* 2020; **8** [PMID: 33207562 DOI: 10.3390/biomedicines8110501]

89 **Assis DN**, Levy C. Oral Vancomycin or Ursodeoxycholic Acid for Pediatric Primary Sclerosing Cholangitis? The Uncontroversial Need for Randomized Controlled Trials. *Hepatology* 2021; **73**: 887-889 [PMID: 33403699 DOI: 10.1002/hep.31702]

90 **Tabibian JH**, Weeding E, Jorgensen RA, Petz JL, Keach JC, Talwalkar JA, Lindor KD. Randomised clinical trial: vancomycin or metronidazole in patients with primary sclerosing cholangitis - a pilot study. *Aliment Pharmacol Ther* 2013; **37**: 604-612 [PMID: 23384404 DOI: 10.1111/apt.12232]

91 **Rahimpour S**, Nasiri-Toosi M, Khalili H, Ebrahimi-Daryani N, Nouri-Taromlou MK, Azizi Z. A Triple Blinded, Randomized, Placebo-Controlled Clinical Trial to Evaluate the Efficacy and Safety of Oral Vancomycin in Primary Sclerosing Cholangitis: a Pilot Study. *J Gastrointestin Liver Dis* 2016; **25**: 457-464 [PMID: 27981301 DOI: 10.15403/jgld.2014.1121.254.rah]

92 **Ali AH**, Damman J, Shah SB, Davies Y, Hurwitz M, Stephen M, Lemos LM, Carey EJ, Lindor KD, Buness CW, Alrabadi L, Berquist WE, Cox KL. Open-label prospective therapeutic clinical trials: oral vancomycin in children and adults with primary sclerosing cholangitis. *Scand J Gastroenterol* 2020; **55**: 941-950 [PMID: 32633158 DOI: 10.1080/00365521.2020.1787501]

93 **Abarbanel DN**, Seki SM, Davies Y, Marlen N, Benavides JA, Cox K, Nadeau KC, Cox KL. Immunomodulatory effect of vancomycin on Treg in pediatric inflammatory bowel disease and primary sclerosing cholangitis. *J Clin Immunol* 2013; **33**: 397-406 [PMID: 23054338 DOI: 10.1007/s10875-012-9801-1]

94 **Britto SL**, Hoffman KL, Tessier ME, Petrosino J, Miloh T, Kellermayer R. Microbiome Responses to Vancomycin Treatment in a Child With Primary Sclerosing Cholangitis and Ulcerative Colitis. *ACG Case Rep J* 2021; **8**: e00577 [PMID: 33997090 DOI: 10.14309/crj.0000000000000577]

95 **Tabibian JH**, Gossard A, El-Youssef M, Eaton JE, Petz J, Jorgensen R, Enders FB, Tabibian A, Lindor KD. Prospective Clinical Trial of Rifaximin Therapy for Patients With Primary Sclerosing Cholangitis. *Am J Ther* 2017; **24**: e56-e63 [PMID: 24914504 DOI: 10.1097/MJT.0000000000000102]

96 **Shah A**, Crawford D, Burger D, Martin N, Walker M, Talley NJ, Tallis C, Jones M, Stuart K, Keely S, Lewindon P, Macdonald GA, Morrison M, Holtmann GJ. Effects of Antibiotic Therapy in Primary Sclerosing Cholangitis with and without Inflammatory Bowel Disease: A Systematic Review and Meta-Analysis. *Semin Liver Dis* 2019; **39**: 432-441 [PMID: 31315136 DOI: 10.1055/s-0039-1688501]

97 **Davies YK**, Tsay CJ, Caccamo DV, Cox KM, Castillo RO, Cox KL. Successful treatment of recurrent primary sclerosing cholangitis after orthotopic liver transplantation with oral vancomycin. *Case Rep Transplant* 2013; **2013**: 314292 [PMID: 23509657 DOI: 10.1155/2013/314292]

98 **Färkkilä M**, Karvonen AL, Nurmi H, Nuutinen H, Taavitsainen M, Pikkarainen P, Kärkkäinen P. Metronidazole and ursodeoxycholic acid for primary sclerosing cholangitis: a randomized placebo-controlled trial. *Hepatology* 2004; **40**: 1379-1386 [PMID: 15565569 DOI: 10.1002/hep.20457]

99 **Krehmeier U**, Bardenheuer M, Voggenreiter G, Obertacke U, Schade FU, Majetschak M. Effects of antimicrobial agents on spontaneous and endotoxin-induced cytokine release of human peripheral blood mononuclear cells. *J Infect Chemother* 2002; **8**: 194-197 [PMID: 12111578 DOI: 10.1007/s101560200036]

100 **Silveira MG**, Torok NJ, Gossard AA, Keach JC, Jorgensen RA, Petz JL, Lindor KD. Minocycline in the treatment of patients with primary sclerosing cholangitis: results of a pilot study. *Am J Gastroenterol* 2009; **104**: 83-88 [PMID: 19098854 DOI: 10.1038/ajg.2008.14]

101 **Bakker GJ**, Nieuwdorp M. Fecal Microbiota Transplantation: Therapeutic Potential for a Multitude of Diseases beyond *Clostridium difficile*. *Microbiol Spectr* 2017; **5** [PMID: 28840809 DOI: 10.1128/microbiolspec.BAD-0008-2017]

102 **Zhang F**, Cui B, He X, Nie Y, Wu K, Fan D; FMT-standardization Study Group. Microbiota transplantation: concept, methodology and strategy for its modernization. *Protein Cell* 2018; **9**: 462-473 [PMID: 29691757 DOI: 10.1007/s13238-018-0541-8]

103 **Xu HM**, Huang HL, Zhou YL, Zhao HL, Xu J, Shou DW, Liu YD, Zhou YJ, Nie YQ. Fecal Microbiota Transplantation: A New Therapeutic Attempt from the Gut to the Brain. *Gastroenterol Res Pract* 2021; **2021**: 6699268 [PMID: 33510784 DOI: 10.1155/2021/6699268]

104 **Gupta M**, Krishan P, Kaur A, Arora S, Trehanpati N, Singh TG, Bedi O. Mechanistic and physiological approaches of fecal microbiota transplantation in the management of NAFLD. *Inflamm Res* 2021; **70**: 765-776 [PMID: 34212214 DOI: 10.1007/s00011-021-01480-z]

105 **Craven L**, Rahman A, Nair Parvathy S, Beaton M, Silverman J, Qumosani K, Hramiak I, Hegele R, Joy T, Meddings J, Urquhart B, Harvie R, McKenzie C, Summers K, Reid G, Burton JP, Silverman M. Allogenic Fecal Microbiota Transplantation in Patients With Nonalcoholic Fatty Liver Disease Improves Abnormal Small Intestinal Permeability: A Randomized Control Trial. *Am J Gastroenterol* 2020; **115**: 1055-1065 [PMID: 32618656 DOI: 10.14309/ajg.0000000000000661]

106 **Gu X**, Lu Q, Zhang C, Tang Z, Chu L. Clinical Application and Progress of Fecal Microbiota Transplantation in Liver Diseases: A Review. *Semin Liver Dis* 2021; **41**: 495-506 [PMID: 34261137 DOI: 10.1055/s-0041-1732319]

107 **Allegretti JR**, Kassam Z, Carrellas M, Mullish BH, Marchesi JR, Pechlivanis A, Smith M, Gerardin Y, Timberlake S, Pratt DS, Korzenik JR. Fecal Microbiota Transplantation in Patients With Primary Sclerosing Cholangitis: A Pilot Clinical Trial. *Am J Gastroenterol* 2019; **114**: 1071-1079 [PMID: 30730351 DOI: 10.14309/ajg.0000000000000115]

108 **Philips CA**, Augustine P, Phadke N. Healthy Donor Fecal Microbiota Transplantation for Recurrent Bacterial Cholangitis in Primary Sclerosing Cholangitis - A Single Case Report. *J Clin Transl Hepatol* 2018; **6**: 438-441 [PMID: 30637223 DOI: 10.14218/JCTH.2018.00033]

109 **Wieërs G**, Belkhir L, Enaud R, Leclercq S, Philippart de Foy JM, Dequenne I, de Timary P, Cani PD. How Probiotics Affect the Microbiota. *Front Cell Infect Microbiol* 2019; **9**: 454 [PMID: 32010640 DOI: 10.3389/fcimb.2019.00454]

110 **Maslennikov R**, Ivashkin V, Efremova I, Poluektova E, Shirokova E. Probiotics in hepatology: An update. *World J Hepatol* 2021; **13**: 1154-1166 [PMID: 34630882 DOI: 10.4254/wjh.v13.i9.1154]

111 **Gou HZ**, Zhang YL, Ren LF, Li ZJ, Zhang L. How do intestinal probiotics restore the intestinal barrier? *Front Microbiol* 2022; **13**: 929346 [PMID: 35910620 DOI: 10.3389/fmicb.2022.929346]

112 **Rodríguez-Pastén A**, Pérez-Hernández N, Añorve-Morga J, Jiménez-Alvarado R, Cariño-Cortés R, Sosa-Lozada T, Fernández-Martínez E. The Activity of Prebiotics and Probiotics in Hepatogastrointestinal Disorders and Diseases Associated with Metabolic Syndrome. *Int J Mol Sci* 2022; **23** [PMID: 35806234 DOI: 10.3390/ijms23137229]

113 **Vleggaar FP**, Monkelbaan JF, van Erpecum KJ. Probiotics in primary sclerosing cholangitis: a randomized placebo-controlled crossover pilot study. *Eur J Gastroenterol Hepatol* 2008; **20**: 688-692 [PMID: 18679073 DOI: 10.1097/MEG.0b013e3282f5197e]

114 **Shimizu M**, Iwasaki H, Mase S, Yachie A. Successful treatment of primary sclerosing cholangitis with a steroid and a probiotic. *Case Rep Gastroenterol* 2012; **6**: 249-253 [PMID: 22679413 DOI: 10.1159/000338834]

115 **Chen Y**, Guan W, Zhang N, Wang Y, Tian Y, Sun H, Li X, Wang Y, Liu J. *Lactobacillus plantarum* Lp2 improved LPS-induced liver injury through the TLR-4/MAPK/NFκB and Nrf2-HO-1/CYP2E1 pathways in mice. *Food Nutr Res* 2022; **66** [PMID: 35903291 DOI: 10.29219/fnr.v66.5459]

116 **Out C**, Patankar JV, Doktorova M, Boesjes M, Bos T, de Boer S, Havinga R, Wolters H, Boverhof R, van Dijk TH, Smoczek A, Bleich A, Sachdev V, Kratky D, Kuipers F, Verkade HJ, Groen AK. Gut microbiota inhibit Asbt-dependent intestinal bile acid reabsorption via Gata4. *J Hepatol* 2015; **63**: 697-704 [PMID: 26022694 DOI: 10.1016/j.jhep.2015.04.030]

117 **Jiang C**, Xie C, Li F, Zhang L, Nichols RG, Krausz KW, Cai J, Qi Y, Fang ZZ, Takahashi S, Tanaka N, Desai D, Amin SG, Albert I, Patterson AD, Gonzalez FJ. Intestinal farnesoid X receptor signaling promotes nonalcoholic fatty liver disease. *J Clin Invest* 2015; **125**: 386-402 [PMID: 25500885 DOI: 10.1172/JCI76738]

118 **Wahlström A**, Kovatcheva-Datchary P, Ståhlman M, Khan MT, Bäckhed F, Marschall HU. Induction of farnesoid X receptor signaling in germ-free mice colonized with a human microbiota. *J Lipid Res* 2017; **58**: 412-419 [PMID: 27956475 DOI: 10.1194/jlr.M072819]

119 **Tanaka N**, Aoyama T, Kimura S, Gonzalez FJ. Targeting nuclear receptors for the treatment of fatty liver disease. *Pharmacol Ther* 2017; **179**: 142-157 [PMID: 28546081 DOI: 10.1016/j.pharmthera.2017.05.011]

120 **Adorini L**, Pruzanski M, Shapiro D. Farnesoid X receptor targeting to treat nonalcoholic steatohepatitis. *Drug Discov Today* 2012; **17**: 988-997 [PMID: 22652341 DOI: 10.1016/j.drudis.2012.05.012]

121 **Hirschfield GM**, Mason A, Luketic V, Lindor K, Gordon SC, Mayo M, Kowdley KV, Vincent C, Bodhenheimer HC Jr, Parés A, Trauner M, Marschall HU, Adorini L, Sciacca C, Beecher-Jones T, Castelloe E, Böhm O, Shapiro D. Efficacy of obeticholic acid in patients with primary biliary cirrhosis and inadequate response to ursodeoxycholic acid. *Gastroenterology* 2015; **148**: 751-61.e8 [PMID: 25500425 DOI: 10.1053/j.gastro.2014.12.005]

122 **Nevens F**, Andreone P, Mazzella G, Strasser SI, Bowlus C, Invernizzi P, Drenth JP, Pockros PJ, Regula J, Beuers U, Trauner M, Jones DE, Floreani A, Hohenester S, Luketic V, Shiffman M, van Erpecum KJ, Vargas V, Vincent C, Hirschfield GM, Shah H, Hansen B, Lindor KD, Marschall HU, Kowdley KV, Hooshmand-Rad R, Marmon T, Sheeron S, Pencek R, MacConell L, Pruzanski M, Shapiro D; POISE Study Group. A Placebo-Controlled Trial of Obeticholic Acid in Primary Biliary Cholangitis. *N Engl J Med* 2016; **375**: 631-643 [PMID: 27532829 DOI: 10.1056/NEJMoa1509840]

123 **Kowdley KV**, Vuppalanchi R, Levy C, Floreani A, Andreone P, LaRusso NF, Shrestha R, Trotter J, Goldberg D, Rushbrook S, Hirschfield GM, Schiano T, Jin Y, Pencek R, MacConell L, Shapiro D, Bowlus CL; AESOP Study Investigators. A randomized, placebo-controlled, phase II study of obeticholic acid for primary sclerosing cholangitis. *J Hepatol* 2020; **73**: 94-101 [PMID: 32165251 DOI: 10.1016/j.jhep.2020.02.033]

124 **de Chambrun GP**, Nachury M, Funakoshi N, Gerard R, Bismuth M, Valats JC, Panaro F, Navarro F, Desreumaux P, Pariente B, Blanc P. Oral vancomycin induces sustained deep remission in adult patients with ulcerative colitis and primary sclerosing cholangitis. *Eur J Gastroenterol Hepatol* 2018; **30**: 1247-1252 [PMID: 30052539 DOI: 10.1097/MEG.0000000000001223]

125 **Chinese Society of Hepatology**, Chinese Medical Association. [Guidelines on the diagnosis and management of primary sclerosing cholangitis (2021)]. *Zhonghua Gan Zang Bing Za Zhi* 2022; **30**: 169-189 [PMID: 35359068 DOI: 10.3760/cma.j.cn112138-20211109-00786]

126 **Lindor KD**, Kowdley KV, Harrison ME; American College of Gastroenterology. ACG Clinical Guideline: Primary Sclerosing Cholangitis. *Am J Gastroenterol* 2015; **110**: 646-59; quiz 660 [PMID: 25869391 DOI: 10.1038/ajg.2015.112]

127 **Lindor KD**, Kowdley KV, Luketic VA, Harrison ME, McCashland T, Befeler AS, Harnois D, Jorgensen R, Petz J, Keach J, Mooney J, Sargeant C, Braaten J, Bernard T, King D, Miceli E, Schmoll J, Hoskin T, Thapa P, Enders F. High-dose ursodeoxycholic acid for the treatment of primary sclerosing cholangitis. *Hepatology* 2009; **50**: 808-814 [PMID: 19585548 DOI: 10.1002/hep.23082]

128 **Paumgartner G**, Beuers U. Ursodeoxycholic acid in cholestatic liver disease: mechanisms of action and therapeutic use revisited. *Hepatology* 2002; **36**: 525-531 [PMID: 12198643 DOI: 10.1053/jhep.2002.36088]

129 **Shah RA**, Kowdley KV. Current and potential treatments for primary biliary cholangitis. *Lancet Gastroenterol Hepatol* 2020; **5**: 306-315 [PMID: 31806572 DOI: 10.1016/S2468-1253(19)30343-7]

130 **Halilbasic E**, Steinacher D, Trauner M. Nor-Ursodeoxycholic Acid as a Novel Therapeutic Approach for Cholestatic and Metabolic Liver Diseases. *Dig Dis* 2017; **35**: 288-292 [PMID: 28249255 DOI: 10.1159/000454904]

131 **Fickert P**, Hirschfield GM, Denk G, Marschall HU, Altorjay I, Färkkilä M, Schramm C, Spengler U, Chapman R, Bergquist A, Schrumpf E, Nevens F, Trivedi P, Reiter FP, Tornai I, Halilbasic E, Greinwald R, Pröls M, Manns MP, Trauner M; European PSC norUDCA Study Group. norUrsodeoxycholic acid improves cholestasis in primary sclerosing cholangitis. *J Hepatol* 2017; **67**: 549-558 [PMID: 28529147 DOI: 10.1016/j.jhep.2017.05.009]

132 **Kjærgaard K**, Frisch K, Sørensen M, Munk OL, Hofmann AF, Horsager J, Schacht AC, Erickson M, Shapiro D, Keiding S. Obeticholic acid improves hepatic bile acid excretion in patients with primary biliary cholangitis. *J Hepatol* 2021; **74**: 58-65 [PMID: 32717289 DOI: 10.1016/j.jhep.2020.07.028]

133 **Gulamhusein AF**, Hirschfield GM. Primary biliary cholangitis: pathogenesis and therapeutic opportunities. *Nat Rev Gastroenterol Hepatol* 2020; **17**: 93-110 [PMID: 31819247 DOI: 10.1038/s41575-019-0226-7]

134 **Liu Q**, Duan ZP, Ha DK, Bengmark S, Kurtovic J, Riordan SM. Synbiotic modulation of gut flora: effect on minimal hepatic encephalopathy in patients with cirrhosis. *Hepatology* 2004; **39**: 1441-1449 [PMID: 15122774 DOI: 10.1002/hep.20194]

135 **Denoth L**, Juillerat P, Kremer AE, Rogler G, Scharl M, Yilmaz B, Bluemel S, On Behalf Of The Swiss Ibd Cohort Study. Modulation of the Mucosa-Associated Microbiome Linked to the PTPN2 Risk Gene in Patients with Primary Sclerosing Cholangitis and Ulcerative Colitis. *Microorganisms* 2021; **9** [PMID: 34442830 DOI: 10.3390/microorganisms9081752]

136 **Ostadmohammadi S**, Azimirad M, Houri H, Naseri K, Javanmard E, Mirjalali H, Yadegar A, Sadeghi A, Asadzadeh Aghdaei H, Zali MR. Characterization of the gut microbiota in patients with primary sclerosing cholangitis compared to inflammatory bowel disease and healthy controls. *Mol Biol Rep* 2021; **48**: 5519-5529 [PMID: 34304365 DOI: 10.1007/s11033-021-06567-8]

137 **Liu Q**, Li B, Li Y, Wei Y, Huang B, Liang J, You Z, Li Y, Qian Q, Wang R, Zhang J, Chen R, Lyu Z, Chen Y, Shi M, Xiao X, Wang Q, Miao Q, Fang JY, Gershwin ME, Lian M, Ma X, Tang R. Altered faecal microbiome and metabolome in IgG4-related sclerosing cholangitis and primary sclerosing cholangitis. *Gut* 2022; **71**: 899-909 [PMID: 34035120 DOI: 10.1136/gutjnl-2020-323565]

138 **Davies YK**, Cox KM, Abdullah BA, Safta A, Terry AB, Cox KL. Long-term treatment of primary sclerosing cholangitis in children with oral vancomycin: an immunomodulating antibiotic. *J Pediatr Gastroenterol Nutr* 2008; **47**: 61-67 [PMID: 18607270 DOI: 10.1097/MPG.0b013e31816fee95]

139 **Boner AL**, Peroni D, Bodini A, Delaini G, Piacentini G. Azithromycin may reduce cholestasis in primary sclerosing cholangitis: a case report and serendipitous observation. *Int J Immunopathol Pharmacol* 2007; **20**: 847-849 [PMID: 18179759 DOI: 10.1177/039463200702000423]

**Footnotes**

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**Provenance and peer review:** Unsolicited article; Externally peer reviewed.

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**Peer-review report’s scientific quality classification**

Grade A (Excellent): 0

Grade B (Very good): B

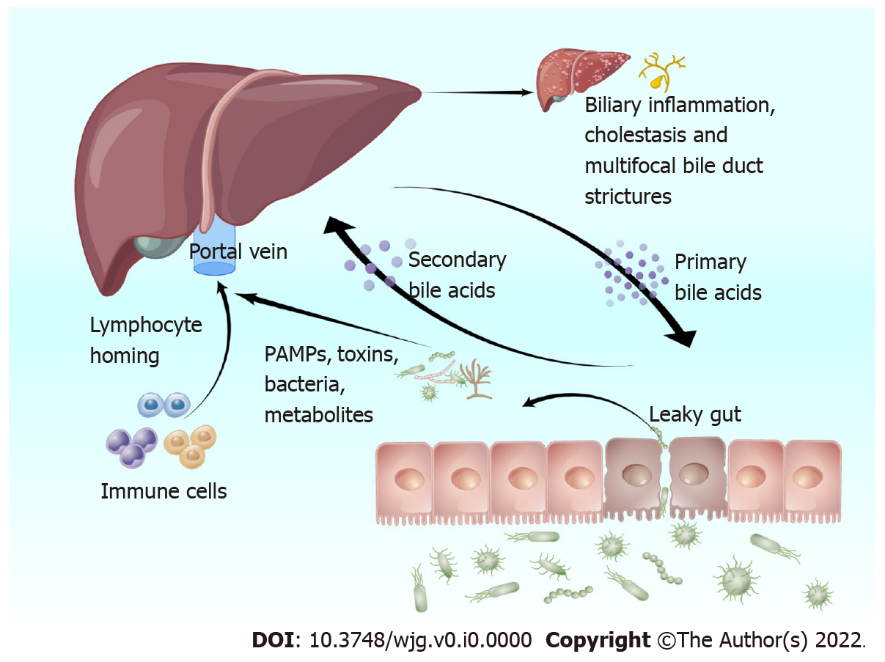
Grade C (Good): C

Grade D (Fair): 0

Grade E (Poor): 0

**P-Reviewer:** Cossiga V, Italy; Ker CG, Taiwan **S-Editor:** Zhang H **L-Editor:** A **P-Editor:** Zhang H

**Figure Legends**

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**Figure 1 The effect of intestinal flora dysbiosis in patients with primary sclerosing cholangitis.** Intestinal flora dysbiosis causes increased intestinal permeability, intestinal lymphocyte homing, and entry of bacteria and their metabolites (*i.e.* pathogen-associated molecular patterns) into the liver. It also impairs normal bile acid metabolism and promotes bile duct inflammation and fibrosis. PAMPs: Pathogen-associated molecular patterns. By Figdraw, www.figdraw.com.

# **Table 1 Changes in the intestinal flora of patients with primary sclerosing cholangitis**

|  |  |  |  |
| --- | --- | --- | --- |
| **Methods** | **Increased** | **Decreased** | **Ref.** |
| 16S RNA gene sequencing: ileum, colon, and rectal samples | *Actinobacillus*, *Bifidobacterium*, *Fusobacterium*, *Haemophilus*, *Roseburia* | *Bacteroides* | [135] |
| qPCR: fecal samples | *Enterobacteriaceae* | *Bifidobacterium*, *Lactobacillus* | [136] |
| 16S RNA gene sequencing: fecal samples | *Clostridium*, *Lactobacillus*, *Ruminococcus gnavus*, *Veillonella* | *Coprococcus*, *Faecalibacterium*, *Phascolarctobacterium*, *Ruminococcus* | [137] |
| 16S RNA gene sequencing: duodenal fluid, saliva, duodenal mucosa, and bile samples | Duodenal mucosa biopsy: *Escherichia coli*, *Veillonella dispar*; Bile fluid: *Enterococcus*, *Neisseria*, *Proteobacteria*, *Staphylococcus*, *Veillonella dispar* |  | [42] |
| Metagenomic shotgun sequencing | *Clostridiales* | *Eubacterium*, *Ruminococcus obeum* | [45] |
| 16S RNA gene sequencing: fecal and saliva samples | *Bacteroides fragilis*, *Blautia*, *Clostridium spp. Enterococcus*, *Enterobacteriaceae*, *Lactobacillus*, *Ruminococcus gnavus*, *Streptococcus salivarius Veillonella dispar* | *Bacteroides thetaiotaomicron*, *Faecalibacterium prausnitzii* | [43] |
| Sigmoid mucosal biopsies and 16S RNA gene sequencing | *Haemophilus parainfluenzae*, *Pseudomonas*, *Streptococcus* | *Lachnospiraceae* | [40] |
| 16S RNA gene sequencing: fecal samples | *Veillonella* | *Blautia*, *Faecalibacterium*, *Lachnoclostridium*, *Ruminococcus* | [44] |
| 16S RNA gene sequencing: fecal samples | *Enterococcus*, *Lactobacillus*, *Streptococcus*, *Veillonella* | *Clostridium cluster IV*, *Coprococcus*, *Desulfovibrio*, *Faecalibacterium*, *Holdemanella* | [38] |
| 16S RNA gene sequencing: fecal samples | *Veillonella* | *Coprococcus*, *Desulfovibrio*, *Phascolarctobacterium*, *Succinivibrio* | [36] |
| 16S RNA gene sequencing: fecal samples | *Clostridium*, *Enterococcus*, *Haemophilus*, *Rothia*, *Streptococcus*, *Veillonella* | *Adlercreutzia equolifaciens*, *Coprococcus catus*, *Faecalibacterium prausnitzii*, *Prevotella copri*, *Ruminococcus gnavus* | [47] |
| Colonic mucosal biopsies and 16S RNA gene sequencing | *Escherichia*, *Lachnospiraceae*, *Megasphaera* | *Bacteroides*, *Prevotella*, *Roseburia* | [37] |
| 16S RNA gene sequencing: fecal samples | *Veillonella* |  | [39] |
| 16S RNA gene sequencing: fecal samples | *Enterococcus*, *Streptococcus*, *Veillonella* |  | [7] |
| 16S RNA gene sequencing: fecal samples | *Bacteroidetes*, *Enterococcus*, *Fusobacterium*, *Lactobacillus*, *Morganella*, *Streptococcus*, *Veillonella* | *Anaerostipes* | [35] |
| Colonic mucosal biopsies and 16S RNA gene sequencing: ileal samples | *Barnesiellaceae*, *Blautia* |  | [8] |
| Mucosal biopsy of the ileocecal region and 16S RNA gene sequencing | *Clostridium clusters IV*  *and XIVa*, *Akkermansia sp.* (*Verrucomicrobia*), *Uncultured Clostridiales II* |  | [34] |

**Table 2 Intestinal flora regulation in primary sclerosing cholangitis treatment**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatment** | | | **Results** | **Ref.** |
| Oral Antibiotics | Vancomycin | 125 mg four times daily for 90 d | Fecal calprotectin and serum GGT levels returned to normal. | [94] |
| 125 mg four times daily for 12 wk | Significantly decreased ALP, MRS, ESR, and GGT levels. Fatigue, pruritus, diarrhea, and anorexia significantly improved. | [91] |
| 50 mg/kg/d for 30 to 118 mo | Decreased ALT, AST, GGT, and ESR levels. Jaundice improved. The overall rate of positive serum autoantibodies decreased after 3.5 mo. | [138] |
| Vancomycin and metronidazole | Vancomycin: 125 mg or 250 mg four times daily for 12 wk. Metronidazole: 250 mg or 500 mg three times daily for 12 wk. | Decreased ALP and MRS levels. The decrease in ALP level was more pronounced following vancomycin administration. | [90] |
| Metronidazole with ursodeoxycholic acid | Taken together for 36 mo | Significantly decreased ALP and MRS levels. | [98] |
| Azithromycin | 500 mg three days per week for 6 wk | Decreased ALP and TBIL levels and cholestasis-related symptoms. The urine was turned dark in color again | [139] |
| Rifaximin | 550 mg twice daily for 12 wk | Decreased GGT and CRP levels. Improved pruritus symptoms. | [95] |
| Minocycline | 100 mg twice daily for one year | Significantly decreased ALP and MRS levels. | [100] |
| Fecal microbiota transplantation at 24 wk | | | Significantly decreased ALP levels. Reduced AST levels (by at least 30%). | [107] |
| *Probiotics* | *Lactobacilli* and *Bifidobacteria* | Three months of: *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus salivarius*, and *Lactococcus lactis*, *Bifidobacterium bifidum*, *Bifidobacterium* | Reduced ALP levels (by 15%). | [113] |
| Combined | Prednisolone (30 mg/d), salazosulfapyridine (3000 mg/d), and *Lactobacillus casei* Shirota (3 g/d) for 2 wk | Decreased ALP, ALT, AST, and GGT levels. | [114] |

ALP: Alkaline phosphatase; AST: Aspartate aminotransferase; ALT: Alanine aminotransferase; MRS: Mayo primary sclerosing cholangitis risk score; ESR: Erythrocyte sedimentation rate; GGT: Gamma-glutamyl transpeptidase; CRP: C-reactive protein; TBIL: Total bilirubin.

**Table 3 Clinical trials related to primary sclerosing cholangitis treatment**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Study Phase** | **Study title** | **ClinicalTrials.gov identifier** | **Actual enrollment** | **Status** | **Interventions** |
| Phase 1 | A Pilot Study of Vancomycin or Metronidazole in Patients with Primary Sclerosing Cholangitis (PSC) | [NCT01085760](https://clinicaltrials.gov/show/NCT01085760) | 35 participants | Completed | Drug: vancomycin; Drug: metronidazole |
| Minocycline in Primary Sclerosing Cholangitis (PSC) | [NCT00630942](https://clinicaltrials.gov/show/NCT00630942) | 16 participants | Completed | Drug: minocycline |
| Treating Primary Sclerosing Cholangitis and Biliary Atresia with Vancomycin | [NCT02137668](https://clinicaltrials.gov/show/NCT02137668) | 200 participants | Recruiting | Drug: oral vancomycin |
| A Pilot Study to Characterize Bile Acid Metabolism and Dysbiosis in Primary Sclerosing Cholangitis | [NCT02464020](https://clinicaltrials.gov/show/NCT02464020) | 8 participants | Completed | Drug: vancomycin |
| Gastrointestinal Microbiota in Primary Sclerosing Cholangitis and Biliary Atresia with Vancomycin (PSC) | [NCT01322386](https://clinicaltrials.gov/show/NCT01322386) | 32 participants | Completed | Drug: vancomycin |
| Fecal Microbiota Transplantation for the Treatment of Primary Sclerosing Cholangitis. | [NCT02424175](https://clinicaltrials.gov/show/NCT02424175) | 10 participants | Completed | Biological: Fecal microbiota transplantation |
| Phase 2 | Norursodeoxycholic Acid in the Treatment of Primary Sclerosing Cholangitis (NUC-3) | [NCT01755507](https://clinicaltrials.gov/show/NCT01755507) | 159 participants | Completed | Drug: norursodeoxycholic acid; Drug: placebo |
| Obeticholic Acid (OCA) in Primary Sclerosing Cholangitis (PSC) (AESOP) | [NCT02177136](https://clinicaltrials.gov/show/NCT02177136) | 77 participants | Completed | Drug: OCA; Drug: placebo |
| Vancomycin for Primary Sclerosing Cholangitis | [NCT03710122](https://clinicaltrials.gov/show/NCT03710122) | 102 participants | Recruiting | Drug: vancomycin; Other: placebo |
| Trial of High-dose Urso in Primary Sclerosing Cholangitis | [NCT00059202](https://clinicaltrials.gov/show/NCT00059202) | 150 participants | Completed | Drug: ursodeoxycholic acid; Drug: placebo |
| Fecal Microbiota Transplantation for the Treatment of Primary Sclerosing Cholangitis. | [NCT02424175](https://clinicaltrials.gov/show/NCT02424175) | 10 participants | Completed | Biological: fecal microbiota transplantation |
| Phase 3 | Primary Sclerosing Cholangitis with Oral Vancomycin by the Study of Its Antimicrobial and Immunomodulating Effects (PSC) | [NCT01802073](https://clinicaltrials.gov/show/NCT01802073) | 34 participants | Completed | Drug: oral vancomycin |
| Probiotics in Patients with Primary Sclerosing Cholangitis | [NCT00161148](https://clinicaltrials.gov/show/NCT00161148) | 12 participants | Unknown1 | Drug: probiotics |
| Norursodeoxycholic Acid *vs* Placebo in PSC | [NCT03872921](https://clinicaltrials.gov/show/NCT03872921) | 300 participants | Recruiting | Drug: norursodeoxycholic acid |
| Vancomycin for Primary Sclerosing Cholangitis | [NCT03710122](https://clinicaltrials.gov/show/NCT03710122) | 102 participants | Recruiting | Drug: vancomycin; Other: placebo |
| Trial of High-dose Urso in Primary Sclerosing Cholangitis | [NCT00059202](https://clinicaltrials.gov/show/NCT00059202) | 150 participants | Completed | Drug: ursodeoxycholic acid; Drug: placebo |
| Phase 4 | Effect and Safety of Oral Vancomycin in Primary Sclerosing Cholangitis Patients | [NCT02605213](https://clinicaltrials.gov/show/NCT02605213) | 30 participants | Unknown1 | Drug: vancomycin; Drug: placebo |

1Study has passed its completion date and status has not been verified in more than two years.

PSC: Primary sclerosing cholangitis; OCA: Obeticholic Acid; AESOP: Aetiology and Ethnicity in Schizophrenia and Other Psychoses.