

Update on primary sclerosing cholangitis

Tom H. Karlsen^{1,2}, Kirsten Muri Boberg^{1,3,4,*}

¹Norwegian PSC Research Center, Department of Transplantation Medicine, Division of Cancer Medicine, Surgery and Transplantation, Oslo University Hospital, Rikshospitalet, Oslo, Norway; ²Division of Gastroenterology, Institute of Medicine, University of Bergen, Bergen, Norway; ³Institute of Clinical Medicine, University of Oslo, Oslo, Norway; ⁴Section of Gastroenterology, Department of Transplantation Medicine, Division of Cancer Medicine, Surgery and Transplantation, Oslo University Hospital, Rikshospitalet, Oslo, Norway

Summary

Primary sclerosing cholangitis (PSC) remains one of the most challenging conditions of clinical hepatology. There has been a steady growth in research to overcome this fact and the present review aims at summarizing the most recently published literature. The main emphasis will be put on the link of recent pathogenetic insights to clinical characteristics and patient management. With regard to pathogenesis, there is no consensus yet as to whether immune mediated injury or factors related to bile acid physiology are the most important. It also remains to be clarified whether PSC is a mixed bag of various secondary etiologies yet to be defined, or a disease entity predominantly represented by sclerosing cholangitis in the context of inflammatory bowel disease. Most important, there is no available medical therapy with proven influence on clinical end points, and timing of liver transplantation and patient follow-up are challenging due to the unpredictable and high risk of cholangiocarcinoma.

© 2013 European Association for the Study of the Liver. Published by Elsevier B.V. All rights reserved.

Introduction

To our knowledge, sclerosing cholangitis was introduced as a medical term in 1867 by Hoffman [1]. In the mid 1960s several case series were reviewed, establishing the link to inflammatory bowel disease (IBD) and describing several other clinical characteristics of a primary form of sclerosing cholangitis (PSC). The introduction of endoscopic retrograde cholangiography (ERC) throughout the 1970s greatly facilitated diagnosis, and the clinical, radiological and histopathological criteria for PSC were stated by three publications in 1980 from the US (Rochester), UK (London), and Norway

(Oslo). Later, magnetic resonance cholangiography (MRC) has been recommended as the primary diagnostic modality in suspected cases of PSC (Clinical Points 1). There has been a steady growth in research activity around the many clinical challenges associated with PSC (Fig. 1), including the founding of an international PSC study group (www.ipscsg.org) in 2010. The aim of the present article is to summarize insights provided by the most recent research (published since our previous update [2]).

Clinical Points 1. Diagnosis of primary sclerosing cholangitis (PSC) [114, 117]

- In patients with a cholestatic biochemical profile not otherwise explained and where causes of secondary sclerosing cholangitis have been excluded, a diagnosis of PSC is made when magnetic resonance cholangiography (MRC) shows typical findings
- Endoscopic retrograde cholangiography (ERC) can be considered if high-quality MRC is uncertain and in patients with inflammatory bowel disease with normal high-quality MRC but high suspicion of PSC
- A liver biopsy is not necessary for the diagnosis of PSC in patients with typical cholangiographic findings
- A liver biopsy is recommended to diagnose small duct PSC if high-quality MRC (or ERC) is normal and in patients with disproportionately elevated aminotransferases to identify additional or alternative disease processes

The principle challenges in PSC all derive from the fact that etiology and pathogenesis are still largely unknown. Since the development of sclerosing cholangitis represents a “final common pathway” for multiple underlying mechanisms of bile duct injury, *in vivo* data in patients with established PSC do not necessarily reflect etiology. The first part of this review elaborates on recent insights into the pathogenesis of PSC, with a particular emphasis on research of relevance to novel treatment strategies currently in the testing phase. An update on aspects relevant to diagnosis of PSC will also be given, with a particular emphasis

Keywords: Primary sclerosing cholangitis; Inflammatory bowel disease; Cholangiocarcinoma.

Received 4 February 2013; received in revised form 12 March 2013; accepted 14 March 2013

* Corresponding author. Address: Department of Transplantation Medicine, Division of Cancer Medicine, Surgery and Transplantation, Oslo University Hospital, Rikshospitalet, Postboks 4950 Nydalen, N-0424 Oslo, Norway. Tel.: +47 2307 2468; fax: +47 2307 3928.

E-mail address: kboberg@ous-hf.no (K.M. Boberg).



ELSEVIER

Review

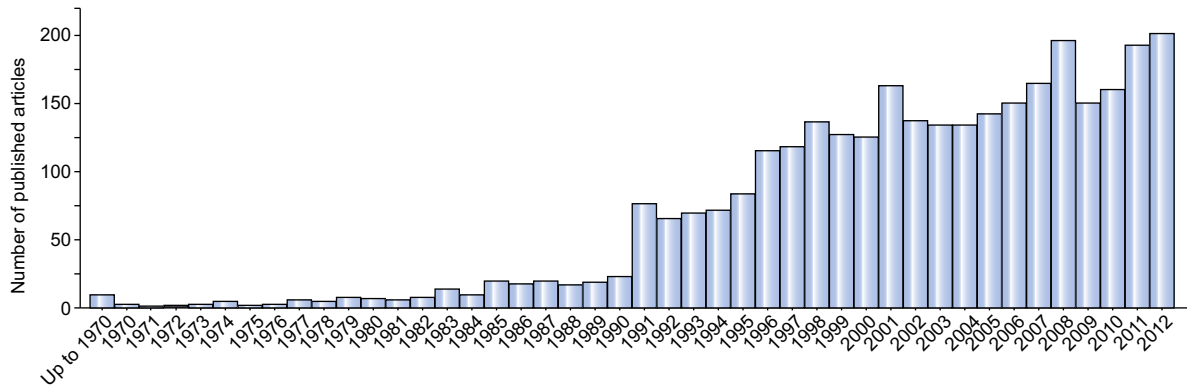


Fig. 1. Published articles reporting the search term “primary sclerosing cholangitis” (<http://www.ncbi.nlm.nih.gov/pubmed/>). The present review will focus on articles published from 2010 to 2012.

on the recent insights into PSC associated clinical aspects, including elevated levels of serum IgG4, IBD, cholangiocarcinoma, pruritus, and concurrent autoimmune hepatitis. Finally, we will summarize the updates regarding treatment and cancer surveillance of PSC patients.

Pathogenesis of PSC – toxicity or immunology?

There is no universally accepted explanation for the bile duct injury in PSC. Mechanistic aspects of the development of similar bile duct lesions are under intense study both in human conditions and rodent models, nurturing an ongoing discussion as to whether the primary injury is caused by immune mediated mechanisms or biochemical aspects related to bile physiology and how these two aspects can potentially be integrated in one model. The definition of a subgroup of PSC patients characterized by high serum levels of IgG4 in 2006 [3] was in support of the possibility that the PSC patient population may be heterogeneous, and that one pathogenetic mechanism cannot be expected to account for all cases. Nevertheless, from a clinical perspective in Northern Europe and the US, a relatively demarcated “syndrome” of concurrent bile duct fibrosis, predominant right-sided colitis and a neoplastic propensity at both these sites, seems to comprise 70–80% of the PSC patient population. For this group of patients, it is not unreasonable to expect a relatively uniform pathogenesis.

Genetic association studies

The genetic susceptibility to PSC aligns with prototypical autoimmune diseases as much as with IBD (Fig. 2). Indeed, the hallmark of an “autoimmune” susceptibility at a genetic level; i.e., a predominant HLA association, also accounts for the overall genetic architecture of PSC (Table 1). As for most HLA associated diseases (celiac disease being a notable exception), the immunological implications of PSC associated HLA variants are unknown. Several review articles have assessed the theoretical knowledge associated with each non-HLA susceptibility locus (Table 1) [4–7]. Only speculations are possible for the potential disease mechanisms represented by these loci, since most of the functional studies that serve as the basis of these review articles were performed prior to, and independently from the knowledge of genetic asso-

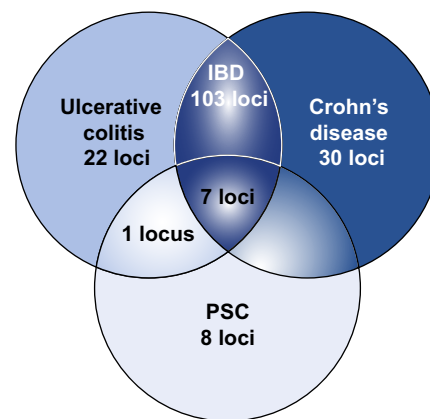


Fig. 2. Venn diagram illustrating the genetic overlap between primary sclerosing cholangitis (PSC) and inflammatory bowel disease (IBD). A total of 163 IBD susceptibility loci and 16 PSC susceptibility loci were included in the plot [9,158]. The major histocompatibility complex (MHC) associations in PSC include several independent association signals and are counted as one PSC specific locus.

ciations in PSC. Furthermore, the “pool” of risk loci is incompletely defined (for reasons discussed elsewhere [8]). Importantly, in our opinion, there is also a risk of oversimplifying implications from genetic association study outcomes if too wide assumptions are formed on the basis of each individual locus (as would only be appropriate in a monogenous trait).

Despite these limitations, the largest genetic study in PSC warrants specific mentioning [9]. The study represents a major accomplishment by multiple centers within the international PSC study group. Genetic risk for PSC was assessed by means of case-control association analysis of a total of 3789 PSC cases to 25,079 controls across 130,422 single-nucleotide polymorphisms (SNPs) genotyped using the ImmunoChip [10]. The ImmunoChip is a targeted genotyping array covering 186 known disease loci from various immune-mediated diseases. Outside of these 186 loci, ImmunoChip also assays thousands of SNPs of intermediate significance from previous studies in these other diseases. A total of 9 novel risk loci for PSC were detected in the analysis. Furthermore, by taking *a priori* knowledge on genetic associations in other diseases into account (Crohn's disease, celiac disease, psoriasis, rheumatoid arthritis, sarcoidosis, type 1 diabetes, and ulcerative colitis), there was *a posteriori* evidence for another 33

Table 1. Genetic susceptibility loci in primary sclerosing cholangitis. Loci are defined according to genome-wide significance threshold conservatively set at $p < 5 \times 10^{-8}$. For each locus, overlapping associations reported in other diseases are observed. Additional risk loci may be defined by taking *a priori* knowledge on genetic associations reported in these other phenotypes into account (see [9] for further details).

Locus	Notable gene(s) nearby	Shared associations
1p36	<i>MMEL1, TNFRSF14</i>	CeD, PBC, RA, UC
2q13	<i>BCL2L11</i>	CLL
2q33	<i>CD28</i>	AA, CeD, CHD, GD, Ht, MI, RA, T1D
2q37	<i>GPR35</i>	UC
3p21	<i>MST1</i>	CD, UC
4q27	<i>IL2, IL21</i>	AA, CD, CeD, RA, T1D, UC
6p21	<i>HLA class I and II, other genes</i>	Multiple diseases
6q15	<i>BACH2</i>	CD, CeD, MS, T1D, Vi
10p15	<i>IL2RA</i>	AA, MS, RA, T1D, Vi
11q23	<i>SIK2</i>	Colorectal cancer
12q13	<i>HDAC7</i>	CD, UC
12q24	<i>SH2B3, ATXN2</i>	BP, CeD, Ch, CKD, EC, He, Hg, Ht, PBC, RVC, T1D
18q21	<i>TCF4</i>	Sch, FCD
18q22	<i>CD226</i>	T1D
19q13	<i>PRKD2, STRN4</i>	CLL, T1D
21q22	<i>PSMG1</i>	AS, CD, UC

AA, alopecia areata; AS, ankylosing spondylitis; BP, blood pressure; CD, Crohn's disease; CeD, celiac disease; Ch, cholesterol; CHD, coronary heart disease; CKD, chronic kidney disease; CLL, chronic lymphocytic leukaemia; EC, eosinophil counts; FCD, Fuchs's corneal dystrophy; GD, Grave's disease; He, haematocrit; Hg, haemoglobin; Hr, hypothyroidism; MI, myocardial infarction; MS, multiple sclerosis; PBC, primary biliary cirrhosis; RA, rheumatoid arthritis; RVC, retinal vascular calibre; Sch, schizophrenia; T1D, type 1 diabetes; UC, ulcerative colitis; Vi, vitiligo.

For gene name abbreviations, see <http://www.ncbi.nlm.nih.gov/gene>.

risk loci (including previously reported loci at 2q35 [encompassing several genes including *TGR5* and interleukin 8 receptor *IL8RA* and *IL8RB*] and 19q13 [fucosyltransferase 2; *FUT2*]). The most notable novel pathway detected relates to the functionally connected *PRKD2*, *HDAC7*, and *SIK2*, but whether these associations represent aberrations of T-cell activation or bile acid homeostasis [11,12], or other biological aspects, can only be speculated.

Bile acid toxicity

The concept of sclerosing cholangitis in the context of "bile toxicity" has evolved during the years following the description of the *Abcb4*^{-/-} mouse and the characterization of nuclear receptor regulation of bile acid homeostasis. Extensive research on the potentially beneficial effects of ursodeoxycholic acid (UDCA) in cholestatic liver diseases has also contributed to the concept [13]. In 2010, previous notions could be unified by the launching of the hypothesis that deficiencies of a biliary bicarbonate "umbrella" (i.e., loss of alkalization of cholangiocyte apical membrane proximity) may increase membrane permeation of protonated (and thus hydrophobic) bile acids leading to bile duct injury [14]. The integrity of the cholangiocyte apical glycocalyx also

appears critical for the maintenance of cholangiocyte protection [15]. The "umbrella" hypothesis further involves the interplay between the Na⁺-independent Cl⁻/HCO₃⁻ anion exchanger (AE2) and active Cl⁻-transporters, most notably the ATP-driven cystic fibrosis transmembrane conductance regulator (CFTR), but also the more recently defined Ca⁺⁺-driven anoctamin 1 channel [16]. The bile acid receptor TGR5 is most likely expressed at the cilia on the biliary epithelium [17], and may be involved in the regulation of these systems [18].

Sclerosing cholangitis in the context of deficient Cl⁻-secretion in cystic fibrosis occurs in up to 1/3 of the patients. Cystic fibrosis-related cholangiopathy has been linked to innate immune regulation, partly based on the fact that *Cftr*^{-/-} mice only develop biliary lesions in the context of induced colitis [19]. The mechanism in this model seems to involve altered regulation of toll-like receptor 4 (TLR4) signaling in these mice [20]. Intriguingly, the cholangiopathy in *Cftr*^{-/-} mice was resistant to treatment with *nor*-ursodeoxycholic acid (*nor*-UDCA), but responded to treatment with oral neomycin and polymyxin B, suggesting a potential role of the gut microbiota in driving the increased NF-κB signaling [20]. These aspects add to the prevailing hypothesis of CFTR dysfunction leading to dehydrated mucosal surfaces and impaired mucociliary clearance. In PSC, a possible role of alterations in mucus secretion from peribiliary glands has not been studied [21,22], but warrants incorporation in future CFTR-centered assessments.

Regulatory aspects of the enterohepatic circulation of bile acids and bile acid detoxification systems may be relevant to human PSC. Several recent review articles summarize the principles of these mechanisms [23,24]. Key updates include the important role of farnesoid X receptor (FXR) activation in ameliorating cholangitis development in the *Abcb4*^{-/-} model [25,26], as well as the regulatory function of fibroblast growth factor 19 (FGF19) in bile homeostasis [27–29]. Genetic associations in ulcerative colitis and to a lesser extent PSC at 2q35 encompass several genes [30,31], including *TGR5* and *IL8RA* and *IL8RB* genes [30,31]. Mechanistically, direct evidence so far available on TGR5 on PSC related aspects seems to concern immunosuppressive effects of receptor activation [32–36], more than aspects of bile homeostasis [17,18,37,38]. Interpretation of the genetic findings at 2q35 in PSC is difficult, since IL8 has also been implicated in cholestasis [39,40]. Likely there are interactions and synergism between several of the regulatory pathways in the context of cholestasis (e.g., FXR and TGR5 [41]) and other receptors may also be involved, e.g., pregnane X receptor (PXR) and G-protein coupled receptor 35 (GPR35) [42,43]. Importantly, bile homeostasis closely integrates with regulation of lipid metabolism, as also reflected by studies in the *Abcb4*^{-/-} model [44].

The gut-liver axis

The earliest theories of PSC development derive from the potential relationship between IBD and inflammatory affection in the portal tracts ("the leaky gut"). At present, there is renewed enthusiasm and great expectations concerning the opportunities associated with characterization of gut microbiota with genomic technologies [45–47]. The gut microbiota is shaped by host gender [48], genetics [49], immune function [50], as well as environmental factors (e.g., diet and xenobiotics [51]), and is likely to represent one component of the pathogenesis of several metabolic and inflammatory conditions. One example illustrating

Review

how the specific genetic constitution of an individual influences the gut microbial environment is given by the *FUT2* association in PSC (and Crohn's disease) [52,53]. Even in healthy individuals, homozygous state for the disease associated variant is associated with substantial alterations in gut microbial community composition [53,54]. The impact of dietary fat on IBD susceptibility represents another example of gene-environment interactions. The interleukin 10 gene (*IL10*) is an established susceptibility factor for human ulcerative colitis [55]. Saturated fat enriched diet in *IL10*^{-/-} mice induces specific changes in the bile acid pool (an increased fraction of taurocholic acid) that lead to alterations in the gut microbial community composition ultimately increasing susceptibility to inflammatory bowel disease in these mice from 25–30% to over 60% [56]. Ongoing research at many sites presently explores the role of similar effects in human PSC patients and established mouse models of sclerosing cholangitis.

An important aspect of immune-mediated liver diseases and liver fibrosis is the recruitment process by which immune-cells and monocytes are recruited from circulation into sites of injury and inflammation. As reviewed elsewhere [57], gut activated T-lymphocytes in the context of IBD may contribute to portal inflammation in PSC due to overlapping adhesion molecule profiles of gut and liver endothelium (i.e., mucosal vascular addressin cell adhesion molecule 1 [MadCAM-1] and vascular cell adhesion molecule 1 [VCAM-1] expression along with chemokine C-C motif ligand 25 [CCL25] secretion). Generation of the corresponding T-lymphocyte phenotype (i.e., $\alpha 4\beta 7$, $\alpha 4\beta 1$, and chemokine C-C motif receptor 9 [CCR9], respectively) is retinoic acid dependent [58], predominantly occurs in the gut associated lymphoid tissues [59], and only under special circumstances in the liver [60]. This “enterohepatic circulation of lymphocytes” is clearly of potential relevance to PSC pathogenesis, and the MadCAM-1 expression observed in PSC affected livers seems to be dependent on amine oxidase activity of vascular adhesion protein 1 (VAP-1) in the presence of tumor necrosis factor alpha (TNF α) [61]. Inhibitors of CCR9 (GSK1605786) and $\alpha 4\beta 7$ (vedolizumab) are already in clinical trials for IBD (www.clinicaltrials.gov). In addition to these, there may be a role in PSC for liver-specific therapeutic manipulation of lymphocyte recruitment via VAP-1 [62]. Mechanisms of the recruitment of other inflammatory cells to the liver (monocytes, Th17 cells, T regulatory cells, and B cells) have also been explored and may offer similar opportunities for manipulating inflammatory processes in PSC [63–67].

Pathogenesis or therapeutic opportunities?

To what extent these axes of most active research are representative of primary disease mechanisms in PSC can only be speculated (Fig. 3). Largely, the key mechanism of IBD associated PSC phenotype remains obscure. However, regardless of the driving forces of the disease process in this “core” predominant PSC phenotype, efforts now delineating mechanisms for disease propagation and bile duct fibrosis are likely to be useful in the definition of novel treatment modalities.

Clinical characteristics of PSC – what is primary and what is secondary?

The list of causes of secondary sclerosing cholangitis strictly speaking only pertains to defined etiologies with histopatholo-

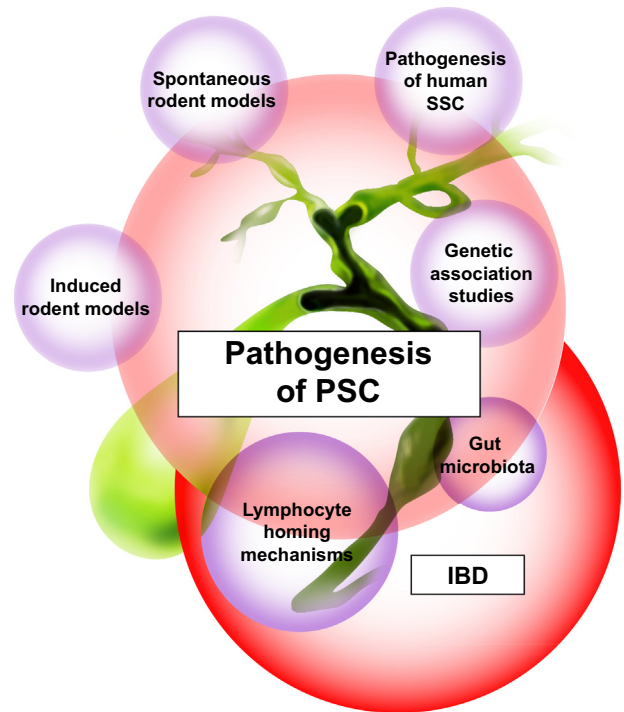


Fig. 3. Relationship between pathogenesis of primary sclerosing cholangitis (PSC) and ongoing studies aimed at elucidating disease mechanisms and potential therapeutic targets. Importantly, the core processes of PSC development remain obscure. However, efforts now delineating mechanisms for bile duct injury and hepatobiliary inflammation in general, as well as in secondary sclerosing cholangitis (SSC), are likely to be useful in the definition of novel treatment strategies even though they may only partly relate to PSC pathogenesis. The only approach fully relevant to human PSC is the human genetic association studies, but they only account for a fraction of the liability in PSC [9], underscoring the role of environmental factors, including the gut microbiota and factors interacting with the gut microbiota [51]. IBD; inflammatory bowel disease.

gical appearance similar to PSC (Table 2). Examples include genetic causes on the one hand, and environmental and iatrogenic causes on the other (Fig. 4). In clinical practice, it is however important to recognize that also infiltrative diseases and various types of malformations may mimic PSC on cholangiography (Table 2). As evident from the discussion on pathogenesis, it remains to be clarified whether the remaining “bag” of PSC after these causes have been excluded is a “mixed bag” or a relatively defined disease entity with a predominant representation of the IBD associated cholangitis. Given the relatively higher frequency of IBD among PSC patients in Northern Europe and the US (62–83%) than in Southern Europe (approximately 50%) and Asia (20–37%), there may be geographic differences (i.e., the IBD-associated PSC is likely represented by a smaller fraction outside of populations of Northern European descent). The term “IBD-related sclerosing cholangitis” has been proposed for this group of patients [68], but is not established.

IgG4 and sclerosing cholangitis

IgG4 associated cholangitis is a sclerosing cholangitis that has been acknowledged during the recent years. Due to the corticosteroid responsiveness, this condition is an important differential diagnosis to PSC. Etiology is not defined, but may represent an

Table 2. Secondary sclerosing cholangitis. Conditions listed include both sclerosing cholangitis according to defined etiologies and liver affections that may mimic primary sclerosing cholangitis (PSC) on cholangiography. According to clinical practice guidelines, IgG4 related disease is now considered in the context of secondary sclerosing cholangitis [114,117].

Infection	Bacterial/parasitic cholangitis Recurrent pyogenic cholangitis
Immunodeficiency related (infections)	Congenital immunodeficiency Acquired immunodeficiency (e.g. HIV) Combined immunodeficiencies Angioimmunoblastic lymphadenopathy
Mechanic/toxic	Cholelithiasis/choledocholithiasis Surgical bile duct trauma Intra-arterial chemotherapy
Ischaemic	Vascular trauma Hepatic allograft arterial insufficiency Paroxysmal nocturnal haemoglobinuria
Pancreatic disease	Chronic pancreatitis IgG4 related systemic disease
Others	Cystic fibrosis cholangiopathy ABCB4 associated cholangiopathy Sclerosing cholangitis of critical illness Hypereosinophilic syndrome Sarcoidosis Graft-versus-host disease Amyloidosis Systemic mastocytosis Caroli's disease Congenital hepatic fibrosis Other types of ductal plate abnormalities Hodgkin's disease Cholangitis glandularis proliferans Neoplastic/metastatic disease Langerhans cell histiocytosis Hepatic allograft rejection

antigen directed specific immune response to environmental triggers [69]. Diagnostic criteria for IgG4 associated cholangitis are elaborated upon elsewhere [70] and nearly align with the HISORT criteria for diagnosing autoimmune pancreatitis [71]. Patients are thus diagnosed on the basis of two or more main manifestations (elevated serum IgG4, suggestive pancreatic imaging findings, other organ involvement and bile duct or papilla Vateri biopsy with >10 IgG4 positive cells/hpf) in combination with a significant corticosteroid treatment response defined as markedly improved biliary strictures allowing stent removal, liver enzymes <2x ULN and significant decreases in serum IgG4 and CA19-9 levels. Biopsies of the duodenal papilla are easily accessible [72], yet only moderately sensitive, and precaution must be taken to avoid papilla injury with an increased risk of complicating pancreatitis. IgG4 is also detectable in bile [73]. Differential diagnostic assessments versus malignancy (e.g., cholangiocarcinoma) may sometimes represent a challenge [74]. Importantly, slight elevations (up to 5 g/L) of serum IgG4 of uncertain relevance occur also among PSC patients not fulfilling criteria for IgG4 associated cholangitis. Among these patients, a more aggressive disease course is often seen [75], but the pathogenetic significance of the detected serum IgG4 elevation and management of these patients have not been defined.

IBD in PSC

Concurrent IBD represents the most common inflammatory comorbidity in PSC [76]. There is some evidence to suggest that comorbidities in PSC (e.g., IBD and other autoimmune diseases) are of prognostic importance [77–80], but there is no consensus definition of these subgroups. Although IBD is most often classified as ulcerative colitis, there are several distinct features of IBD in PSC (summarized in Table 3). Most characteristically, affection does not follow a “distal-to-proximal” distribution. Rather, there is a right sided predominance of inflammation, with also inflammatory affections observed in the ileum, i.e., distribution seems to maximize in the vicinity of the valvula Bauhinii, also consistent with predominance of right-sided colonic carcinomas in PSC patients with IBD [81,82]. There is at present no molecular basis known to explain these features. There is an increased risk of ulcerative colitis even without PSC in siblings of PSC patients, suggesting the presence of a shared genetic predisposition between PSC and IBD [83]. However, in the most recent large-scale assessment of this genetic overlap [9], only 8 out of 163 *bona fide* IBD risk loci associate with PSC (Fig. 2), and for half of the PSC loci, no consistent associations could be detected in IBD. Out of the 8 loci associating with both PSC and IBD in this analysis, one preferentially associates with ulcerative colitis (Fig. 2) while the others associate also with Crohn's disease. Clinical classification of IBD in PSC as Crohn's disease that in some cases is in line with this overlap. In our view, the overall evidence and practical implications (e.g., the increased risk of dysplasia [84]) support the notion of a distinct sub-entity of IBD (“PSC-IBD”) and it may soon be worth considering to implement this term in clinical practice [85].

Cholangiocarcinoma in PSC

Recently reported patient series support the notion that up to 50% of cholangiocarcinomas in PSC are diagnosed within the first year after the diagnosis of PSC [86–88] and the challenge of making the diagnosis remains unresolved [89]. In up to 1/3 of the

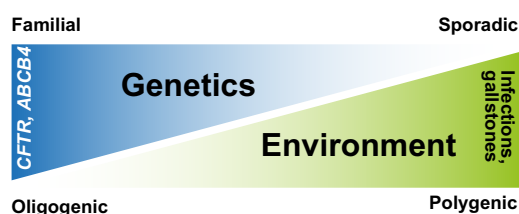


Fig. 4. Concept of the relationship between environmental and genetic risk factors for sclerosing cholangitis. Secondary sclerosing cholangitis (SSC) entities cluster at both ends of this spectrum (e.g., genetic causes versus infectious causes, respectively), whereas primary sclerosing cholangitis (PSC) is a complex phenotype involving multiple, interacting genetic and environmental factors. Factors involved in the development of SSC may modify or complicate disease course in PSC (e.g., ABCB4 variants and cholangitis, respectively).

Review

Table 3. Selected studies from the US, Europe and Asia summarizing key features of patients with primary sclerosing cholangitis (PSC) and inflammatory bowel disease (IBD) [154–156]. (See below-mentioned references for further information)

Study, [Ref.]	N _{PSC-IBD}	Pancolitis (%)	Backwash ileitis (%)	Rectal sparing (%)	Neoplasia (%)
Faubion <i>et al.</i> [154]	39	80	n.a.	27	10
Loftus <i>et al.</i> [85]	71	87	51	52	15
Joo <i>et al.</i> [155]	40	85	36	18	15
Jørgensen <i>et al.</i> [156]	155	55	20	65	n.a.
Ye <i>et al.</i> [157]	21	95	43	38	14

n.a., not available.

cases referred for liver transplantation [90], a pre-transplant diagnosis is impossible to confirm. The importance of considering also the gallbladder epithelium at increased risk of malignancies in PSC is now established [91,92], but there is not yet consensus as to what size of lesions should trigger cholecystectomy [93,94]. The difficulties associated with the detection of increased levels of CA19-9 outside the context of malignancy emphasize that all assessments for cholangiocarcinoma in PSC need to be multimodal [95,96]. Furthermore, it should be kept in mind that for genetic reasons some individuals do not produce CA19-9 [97].

Recent studies confirm an added value of fluorescent *in situ* hybridization (FISH) to conventional brush cytology [98–100], and at referral centers serving large volumes of PSC patients, the method should be considered implemented. As is the case for conventional brush cytology, specificity of FISH is generally good, whereas the reported sensitivity varies between investigators and study populations in the range of 50% and 87% [101,102]. The technique of brushing is likely also of importance, but has not been studied systematically. High quality samples have been obtained at our center by sampling non-dilated strictures, using an over-the-wire brush [103]. Brushing with the whole catheter, as opposed to moving the brush in and out of the catheter, may increase the yield, avoiding losing material against the edge of the catheter. The value of detecting cholangiocellular dysplasia in the absence of suspected cholangiocarcinoma (e.g., dominant strictures) is controversial since up to 1/3 of PSC patients may show such features [98,104,105]. However, considering biliary dysplasia a precursor of cholangiocarcinoma and a reported specificity of 95% for cytological findings classified as high-grade dysplasia, it can be argued that PSC patients with cytological dysplasia should be referred for liver transplantation [103]. As for conventional cytology, serial findings of FISH abnormalities associate with an increased risk of cholangiocarcinoma [99].

Cholestatic pruritus

Cholestatic pruritus may be debilitating for patients with PSC. Treatment is difficult and reviewed elsewhere [106]. In refractory cases, plasmapheresis and albumin dialysis may be effective [107], but sometimes pruritus may represent an indication for liver transplantation. There seems to be underlying differences in the patient population biology since some patients may be highly cholestatic without pruritus, whereas other patients are severely affected even when cholestasis is modest. Genetic reasons (e.g., modifiers reflecting underlying cholestatic predisposition [108]) or differences in immune reaction profiles between the patients [109] may be involved in causing the pruritic predisposition. At some level, the mechanisms causing cholestatic pruritus seem to involve the conversion of lysophosphatidylcholine into lysophosphatidic acid (LPA) by autotaxin [110]. Autotaxin activity in cholestatic patients correlates with pruritus, which is not the case for other candidate pruritogens like bile salts, histamine, tryptase, substance P or endogenous opioids. The mechanisms behind the increased autotaxin activity remain to be established. At another level, accumulated bile acids may be directly involved in cholestatic pruritus via activation of TGR5 on sensory neurons transmitting pruritus and pain [111].

PSC with features of autoimmune hepatitis

In a position paper published by the International Autoimmune Hepatitis Group (IAIHG), the demarcations between PSC, autoimmune hepatitis (AIH) and primary biliary cirrhosis (PBC) are elaborated upon [112]. Importantly, it is argued for abandoning the term “overlap syndromes” and opting for a primary diagnosis of either one of the three conditions. In both PSC and PBC, there is a propensity for non-biliary autoimmunity, and it is not surprising that in some patients this may also include hepatocyte affection and features of AIH (in 7–14% and 2–19%, respectively). In cases where a primary diagnosis of PSC and PBC can be made, the IAIHG scoring systems for AIH should not be applied in the diagnosing of such features [113]. Probably, sound clinical judgment of biochemical (ALT at least 5x ULN and IgG at least 2x ULN) and histological (suspected features of AIH is the main indication for liver biopsy in PSC) parameters should form the basis of diagnosing such features. Although not evidence based, immunosuppressive therapy along standard guidelines for treatment of AIH is recommended for PSC patients with overlapping features with AIH [112,114]. Treatment response in terms of delaying cirrhosis development is likely less pronounced than in AIH without a primary diagnosis of PSC [115,116]. It is thus important to be aware of the risk of side effects and to assess treatment response. From a mechanistic perspective, it is interesting that co-occurrence of PSC and PBC features rarely occurs. There is no evident explanation, yet it is interesting to note that for a genetic risk locus where a shared effect between PSC and PBC has been reported (1p36, see Table 1), the effect (odds ratio) of the risk variant goes in the opposite direction in PSC compared with PBC.

Management – treatment or surveillance?

The almost parallel publication of the European Association for the Study of the Liver (EASL) and American Association for the study of Liver Diseases (AASLD) practice guidelines for PSC in 2009 and 2010 [114,117] provided comprehensive directions

for the diagnosis and management of PSC patients (Clinical Points 1 and 2). In most aspects, the guidelines agree (reviewed elsewhere [2]), yet the discrepancy on principle directions for prescription of UDCA caused a still ongoing discussion on which one of the conclusions was most appropriate. The EASL guidelines concluded that limitations in available data did not yet allow a specific recommendation for the general use of UDCA in PSC. The same guidelines opened up to the use of moderate dose UDCA (15–20 mg/day) in individuals with strong family history of colorectal cancer, previous colorectal neoplasia or longstanding extensive colitis, not for the improvement of liver disease, but to prevent neoplasia. The AASLD guidelines unambiguously recommend against the use of UDCA in adult PSC patients.

Clinical Points 2. Treatment of primary sclerosing cholangitis [114, 117]

- The role for ursodeoxycholic acid (UDCA) in PSC is currently debated. There is no definite evidence that UDCA improves survival or is efficacious in the prevention of colorectal or biliary neoplasia. Practice varies, however, between centers. It has been the tradition of the current authors to be reluctant to start PSC patients on UDCA
- Although not evidence-based, PSC patients with features overlapping with those of autoimmune hepatitis should be considered for therapy with corticosteroids and/or other immunosuppressive agents. Therapy should be individualized and adjusted according to response and the risk of side effects
- In PSC patients with dominant bile duct strictures causing biochemical and/or clinical signs of cholestasis, endoscopic dilatation with or without stenting is recommended. Short-term (2-3 weeks) stenting is favored by many, including the current authors. A multicenter, prospective, randomized intervention trial to compare the efficacy of single session balloon dilatation and short-term stenting is currently ongoing. Prophylactic antimicrobial therapy is recommended during the procedure
- Liver transplantation is a curative treatment modality in PSC patients with end-stage liver disease and in selected cases with severe symptoms of cholestasis. Transplantation may also be considered in patients with evidence of biliary epithelium dysplasia

Bile acid therapy

We will not re-review the literature that led to the slightly differing conclusions on UDCA therapy in PSC [2], but rather mention some subsequent reports that have elaborated on the complexity of the problem. Serum alkaline phosphatase (ALP) level is a long established risk factor for disease progression in PSC [118]. Genetic risk factors for PSC (i.e., *FUT2*) overlap with genetic factors influencing serum levels of ALP in healthy individuals [119,120], but how these factors are involved in the prognostic importance of ALP has not been studied. In a retrospective analysis of 139 PSC patients [121], those receiving UDCA and achieving an improvement of ALP to <1.5x ULN had significantly longer survival without

end points compared with patients without ALP normalization. In series involving patients from previous UDCA treatment trials, the association between ALP normalization and PSC behavior seemed to be independent of UDCA administration [122,123]. The interpretation of all these data requires some caution; in principle both UDCA administration and PSC behavior seem to independently influence ALP levels. At present, it cannot be claimed that ALP normalization in the context of UDCA administration is representative of drug efficacy. Accounting for *FUT2* related biology, there is a strong basis for further studies to clarify these mechanisms that seem to converge on a robust representative of disease intensity in PSC, potentially also amendable by means of bile acid therapy.

The effects of high-dose UDCA (28–30 mg/kg/day) treatment on the bile acid pool warrant reflections [124]. In PSC patients receiving such treatment, there was both an expansion and compositional changes of the total bile acid pool. While only the expansion seemed to associate significantly with adverse outcomes (progression to cirrhosis, development of varices, cholangiocarcinoma, liver transplantation, and death), the increased proportion of lithocholic acid found in this trial is theoretically harmful. It needs to be noted that in another study of bile acid pool compositional effects from UDCA treatment, only UDCA itself was significantly enriched [125]. Nevertheless, given the disease distribution in PSC, predominantly affecting the “proximities of the enterohepatic circulation”, the influence of bile acid treatment, or treatment aiming at modifying the regulation of bile acid homeostasis (e.g., FXR agonists), needs further consideration. Importantly, the bile acid pool serves not only as a potential “toxic medium” to epithelial surfaces, but profoundly interacts with the metabolism and immune function via effects on the gut microbiota [56,126,127]. There is growing interest in the influence of this interaction on IBD and liver diseases [128–130], and it may be wise to advise for future clinical trials in PSC to include assessments of bile acid composition and metagenomic parameters in the basic study readout [51].

Antibiotics and beyond

Of the many approaches currently explored for treatment of PSC, antibiotics most clearly link up with the intersections between inflammation, bile acid homeostasis, and gut microbiota. Distinguishing between non-absorbable (i.e., local effects on the gut microbiota only) and absorbable (i.e., including a portal and systemic anti-microbial effect) is reasonable. Non-absorbable antibiotics like vancomycin exert effects on systemic immune function, e.g., by influencing regulatory T-cell subsets. In a trial of 14 pediatric patients with PSC [131], significant improvement in hepatic biochemistries was observed, paralleling increased levels of regulatory T-cells. Regarding absorbable antibiotics, improvements in hepatic biochemistries have also been observed for metronidazole [132], azithromycin [133] and minocycline [134], altogether suggesting that liver affection in PSC may be influenced by antimicrobial therapies. We anticipate that upon the further characterization of metagenomic components in PSC pathogenesis (Fig. 3), specific attempts to manipulate these (by antibiotics, probiotics, prebiotics or dietary means) will be made.

The paradox of immunosuppression

The strong HLA association along with multiple shared risk loci between PSC and prototypical autoimmune diseases (Table 1),

Review

more pronounced than for IBD, strongly suggests that PSC pathogenesis has an “autoimmune” component [2]. Antibodies against biliary as well as colonic epithelial cells have been reported, and the preferential usage of particular T-cell receptor gene segments of hepatic T-cells also suggests the existence of tissue-specific antigens in PSC pathogenesis. In spite of this, no immunosuppressive drug tested to date has shown to significantly improve clinical outcomes. Of particular interest are cyclosporine A (CsA) and tacrolimus that exert immunosuppressive actions via inhibition of calcineurin, which again is indispensable for cytokine induction, interleukin 2 (IL2) in particular, upon engagement of the T-cell receptor. Genetic associations in PSC are reported both at the *IL2* and IL2 receptor alpha (*IL2RA*) loci (Table 1), directly suggesting an involvement of the IL2-signaling pathway in PSC pathogenesis. Treatment trials show some influence from CsA on IBD activity in PSC [135], and from tacrolimus on hepatic biochemistries [136], but no effects on clinical end points are evident and side effects preclude a broad application. Effects of CsA and tacrolimus may also differ, at least concerning IBD in the context of PSC. This was recently underscored by the finding of enhanced IBD activity after liver transplantation for PSC in patients receiving tacrolimus and mycophenolate compared with patients on CsA and azathioprine [137]. Mechanistic elucidation of the immunological consequences of PSC susceptibility loci *in vivo* needs to be performed before rational application of immunosuppression can be drafted on the basis of genetics.

Antifibrotic therapy

To some extent, it may be argued that present clinical trials in PSC have concerned patients in whom fibrotic lesions are already manifested and efficacy of immunosuppressive and antibiotic treatment cannot be expected. Antifibrotic treatment strategies in preparation largely build on studies on the *Abcb4*^{-/-} mouse model. Some effects converge on nuclear receptor signaling pathways, including TGR5, FXR, and peroxisome proliferator activated receptor gamma (PPAR γ) [26,138]. So far, no nuclear receptor has been identified as a target of *nor*-UDCA [139]. Other components of the *Abcb4*^{-/-} fibrogenesis are currently being targeted in treatment trials, e.g., lysyl oxidase-like 2 [140] by monoclonal antibody GS6624 (<http://clinicaltrials.gov/ct2/show/NCT01672853>). Up to the point where a diagnosis of PSC may be made at a pre-clinical, pre-fibrotic stage, therapies derived from this model may hold promise.

Cholangiocarcinoma surveillance

The most challenging aspect of PSC follow-up remains the unpredictable occurrence of cholangiocarcinoma. Management and surveillance strategies are reviewed elsewhere [89], and novel aspects predominantly derive from proteomic and epigenetic approaches. In serum, a proteomic study report the potential diagnostic improvement achieved by including serum leucine-rich α -2-glycoprotein (LRG1) and IL6 in prediction models for cholangiocarcinoma [141]. In bile, a panel of 22 peptides detected in a screening panel was shown to discriminate between cholangiocarcinoma and PSC with an area under the receiver operating characteristics curve of 87% [142], correctly detecting 8 out of 10 cases of cholangiocarcinoma complicating PSC. In urine, a similar approach led to comparable results [143], with correct classification of 10 out of 10 cases of cholangiocarcinoma arising in the context of PSC. Based on experience in other cancers and preli-

minary data [144,145], epigenetic markers are also potential biomarkers for early cholangiocarcinoma development. This accounts for genes affected by CpG hypermethylation [146], as well as cholangiocarcinoma-associated serum micro RNA profiles. The prospective clinical utility of all these approaches is presently under study at several sites and it is likely that some approaches will be incorporated in patient management in the near future. This will ease timing for liver transplantation and thus ultimately assist in reducing cholangiocarcinoma-related deaths in the PSC population.

Liver transplantation

Due to the lack of effective medical treatment, liver transplantation remains the principle therapeutic option in PSC. The present literature on the high risk of acute cellular rejections in PSC following liver transplantation and PSC recurrence in the allograft was recently reviewed elsewhere [147]. One key ongoing discussion relates to whether the presence of an intact colon in PSC patients with IBD influences disease recurrence, but due to the lack of reproduction so far [148], these data are only useful for pathophysiological considerations and should not prompt pre-transplantation colectomy on a general basis. In our center, biliary epithelium dysplasia by brush cytology is considered an indication for liver transplantation in PSC, but this practice has not been prospectively evaluated. Also, in highly selected cases of hilar cholangiocarcinoma, liver transplantation in conjunction with neoadjuvant chemotherapy and radiation should be considered [149,150], in particular since patients with PSC may present with favorable tumor characteristics compared with cholangiocarcinoma outside of the context of PSC [151]. Further studies are needed to clarify the utility of liver transplantation for intrahepatic cholangiocarcinoma [152], which is presently advised against. In areas where prioritizations for liver transplantations are made on the basis of model of end-stage liver disease (MELD) assessments, issues related to malignancies in PSC add to an already ongoing discussion on exception criteria for patients with PSC [153].

Conclusions

There is a growing interest in the hepatological community to resolve the main challenges associated with PSC. Ongoing efforts to delineate pathogenesis are of key importance, since they ultimately may guide the rational management of remaining clinical challenges, i.e., early diagnosis of PSC in patients with IBD, early diagnosis of cholangiocarcinoma in PSC, determining surrogate markers for disease progression and disease activity; and finally, treatment modalities that significantly influence clinical outcome.

Conflict of interest

The authors declared that they do not have anything to disclose regarding funding or conflict of interest with respect to this manuscript.

References

- [1] Hoffman CEE. Verschluss der gallenwege durch verdickung der wandungen. Arch Pathol Anat Physiol 1867;39:206–215.

- [2] Karlsen TH, Schrumpf E, Boberg KM. Update on primary sclerosing cholangitis. *Dig Liver Dis* 2010;42:390–400.
- [3] Mendes FD, Jorgensen R, Keach J, Katzmann JA, Smyrk T, Donlinger J, et al. Elevated serum IgG4 concentration in patients with primary sclerosing cholangitis. *Am J Gastroenterol* 2006;101:2070–2075.
- [4] Karlsen TH. A lecture on the genetics of primary sclerosing cholangitis. *Dig Dis* 2012;30:32–38.
- [5] Karlsen TH, Kaser A. Deciphering the genetic predisposition to primary sclerosing cholangitis. *Semin Liver Dis* 2011;31:188–207.
- [6] Folseraas T, Melum E, Franke A, Karlsen TH. Genetics in primary sclerosing cholangitis. *Best Pract Res Clin Gastroenterol* 2011;25:713–726.
- [7] Naess S, Shiryayev A, Hov JR, Franke A, Karlsen TH. Genetics in primary sclerosing cholangitis. *Clin Res Hepatol Gastroenterol* 2012;36:325–333.
- [8] Manolio TA, Collins FS, Cox NJ, Goldstein DB, Hindorf LA, Hunter DJ, et al. Finding the missing heritability of complex diseases. *Nature* 2009;461:747–753.
- [9] Liu JZ, Hov JE, Folseraas T, Ellinghaus E, Rushbrook SM, Doncheva NT, et al. Dense genotyping of immune-related disease regions identifies nine new risk loci for primary sclerosing cholangitis. *Nature Genetics* 2013;45:670–675.
- [10] Cortes A, Brown MA. Promise and pitfalls of the Immunochip. *Arthritis Res Ther* 2011;13:101.
- [11] Mitro N, Godio C, De Fabiani E, Scotti E, Galmozzi A, Gilardi F, et al. Insights in the regulation of cholesterol 7 alpha-hydroxylase gene reveal a target for modulating bile acid synthesis. *Hepatology* 2007;46:885–897.
- [12] Clark K, MacKenzie KF, Petkevicius K, Kristariyanto Y, Zhang J, Choi HG, et al. Phosphorylation of CRT3 by the salt-inducible kinases controls the interconversion of classically activated and regulatory macrophages. *Proc Natl Acad Sci U S A* 2012;109:16986–16991.
- [13] Beuers U. Drug insight: mechanisms and sites of action of ursodeoxycholic acid in cholestasis. *Nat Clin Pract Gastroenterol Hepatol* 2006;3:318–328.
- [14] Beuers U, Hohenester S, De Buy Wenniger LJ, Kremer AE, Jansen PL, Elferink RP. The biliary HCO(3)(-) umbrella: a unifying hypothesis on pathogenetic and therapeutic aspects of fibrosing cholangiopathies. *Hepatology* 2010;52:1489–1496.
- [15] Hohenester S, Wenniger LM, Paulusma CC, van Vliet SJ, Jefferson DM, Elferink RP, et al. A biliary HCO3- umbrella constitutes a protective mechanism against bile acid-induced injury in human cholangiocytes. *Hepatology* 2012;55:173–183.
- [16] Dutta AK, Khimji AK, Kresge C, Bugde A, Dougherty M, Esser V, et al. Identification and functional characterization of TMEM16A, a Ca2+-activated Cl- channel activated by extracellular nucleotides, in biliary epithelium. *J Biol Chem* 2011;286:766–776.
- [17] Keitel V, Ullmer C, Haussinger D. The membrane-bound bile acid receptor TGR5 (Gpbar-1) is localized in the primary cilium of cholangiocytes. *Biol Chem* 2010;391:785–789.
- [18] Keitel V, Cupisti K, Ullmer C, Knoefel WT, Kubitz R, Haussinger D. The membrane-bound bile acid receptor TGR5 is localized in the epithelium of human gallbladders. *Hepatology* 2009;50:861–870.
- [19] Blanco PG, Zaman MM, Junaidi O, Sheth S, Yantiss RK, Nasser IA, et al. Induction of colitis in Cfr-/- mice results in bile duct injury. *Am J Physiol Gastrointest Liver Physiol* 2004;287:G491–G496.
- [20] Fiorotto R, Scirpo R, Trauner M, Fabris L, Hoque R, Spirli C, et al. Loss of CFTR affects biliary epithelium innate immunity and causes TLR4-NF-kappaB-mediated inflammatory response in mice. *Gastroenterology* 2011;141:1498–1508, e1491–1495.
- [21] McMinn RM, Kugler JH. The glands of the bile and pancreatic ducts: autoradiographic and histochemical studies. *J Anat* 1961;95:1–11.
- [22] Spitz L, Petropoulos A. The development of the glands of the common bile duct. *J Pathol* 1979;128:213–220.
- [23] Halilbasic E, Claudel T, Trauner M. Bile acid transporters and regulatory nuclear receptors in the liver and beyond. *J Hepatol* 2013;58:155–168.
- [24] Trauner M, Halilbasic E. Nuclear receptors as new perspective for the management of liver diseases. *Gastroenterology* 2011;140:1120–1125, e1121–1112.
- [25] Modica S, Petruzzelli M, Bellafante E, Murzilli S, Salvatore L, Celli N, et al. Selective activation of nuclear bile acid receptor FXR in the intestine protects mice against cholestasis. *Gastroenterology* 2012;142:355–365, e351–354.
- [26] Baghdasaryan A, Claudel T, Gumhold J, Silbert D, Adorini L, Roda A, et al. Dual farnesoid X receptor/TGR5 agonist INT-767 reduces liver injury in the Mdr2-/- (Abcb4-/-) mouse cholangiopathy model by promoting biliary HCO output. *Hepatology* 2011;54:1303–1312.
- [27] Schaap FG, van der Gaag NA, Gouma DJ, Jansen PL. High expression of the bile salt-homeostatic hormone fibroblast growth factor 19 in the liver of patients with extrahepatic cholestasis. *Hepatology* 2009;49:1228–1235.
- [28] Triantis V, Saeland E, Bijl N, Oude-Elferink RP, Jansen PL. Glycosylation of fibroblast growth factor receptor 4 is a key regulator of fibroblast growth factor 19-mediated down-regulation of cytochrome P450 7A1. *Hepatology* 2010;52:656–666.
- [29] Zweers SJ, Booij KA, Komuta M, Roskams T, Gouma DJ, Jansen PL, et al. The human gallbladder secretes fibroblast growth factor 19 into bile: towards defining the role of fibroblast growth factor 19 in the enterobiliary tract. *Hepatology* 2012;55:575–583.
- [30] Anderson CA, Boucher G, Lees CW, Franke A, D'Amato M, Taylor KD, et al. Meta-analysis identifies 29 additional ulcerative colitis risk loci, increasing the number of confirmed associations to 47. *Nat Genet* 2011;43:246–252.
- [31] Hov JR, Keitel V, Laerdahl JK, Spomer L, Ellinghaus E, Elsharawy A, et al. Mutational characterization of the bile acid receptor TGR5 in primary sclerosing cholangitis. *PLoS One* 2010;5:e12403.
- [32] Wang YD, Chen WD, Yu D, Forman BM, Huang W. The G-protein-coupled bile acid receptor, Gpbar1 (TGR5), negatively regulates hepatic inflammatory response through antagonizing nuclear factor kappa light-chain enhancer of activated B cells (NF-kappaB) in mice. *Hepatology* 2011;54:1421–1432.
- [33] Cipriani S, Mencarelli A, Chini MG, Distrutti E, Renga B, Bifulco G, et al. The bile acid receptor GPBAR-1 (TGR5) modulates integrity of intestinal barrier and immune response to experimental colitis. *PLoS One* 2011;6:e25637.
- [34] Keitel V, Donner M, Winandy S, Kubitz R, Haussinger D. Expression and function of the bile acid receptor TGR5 in Kupffer cells. *Biochem Biophys Res Commun* 2008;372:78–84.
- [35] Kawamata Y, Fujii R, Hosoya M, Harada M, Yoshida H, Miwa M, et al. A G protein-coupled receptor responsive to bile acids. *J Biol Chem* 2003;278:9435–9440.
- [36] Chen WD, Yu D, Forman BM, Huang W, Wang YD. Deficiency of G-protein-coupled bile acid receptor Gpbar1 (TGR5) enhances chemically induced liver carcinogenesis. *Hepatology* 2013;57:656–666.
- [37] Li T, Holmstrom SR, Kir S, Umetani M, Schmidt DR, Kiewer SA, et al. The G protein-coupled bile acid receptor, TGR5, stimulates gallbladder filling. *Mol Endocrinol* 2011;25:1066–1071.
- [38] Keitel V, Haussinger D. TGR5 in the biliary tree. *Dig Dis* 2011;29:45–47.
- [39] Isse K, Harada K, Nakanuma Y. IL-8 expression by biliary epithelial cells is associated with neutrophilic infiltration and reactive bile ductules. *Liver Int* 2007;27:672–680.
- [40] Bansal AS, Thomson A, Steadman C, Le Gros G, Hogan PG, Kerlin P, et al. Serum levels of interleukins 8 and 10, interferon gamma, granulocyte-macrophage colony stimulating factor and soluble CD23 in patients with primary sclerosing cholangitis. *Autoimmunity* 1997;26:223–229.
- [41] Baghdasaryan A, Claudel T, Gumhold J, Silbert D, Adorini L, Roda A, et al. Dual farnesoid X receptor/TGR5 agonist INT-767 reduces liver injury in the Mdr2-/- (Abcb4-/-) mouse cholangiopathy model by promoting biliary HCO(-)3 output. *Hepatology* 2011;54:1303–1312.
- [42] Karlsen TH, Lie BA, Frey Froslee K, Thorsby E, Broome U, Schrumpf E, et al. Polymorphisms in the steroid and xenobiotic receptor gene influence survival in primary sclerosing cholangitis. *Gastroenterology* 2006;131:781–787.
- [43] Ellinghaus D, Folseraas T, Holm K, Ellinghaus E, Melum E, Balschun T, et al. Genome-wide association analysis in primary sclerosing cholangitis and ulcerative colitis identifies risk loci at GPR35 and TCF4. *Hepatology* 2013. <http://dx.doi.org/10.1002/hep.25977>, in press.
- [44] Moustafa T, Fickert P, Magnes C, Guelly C, Thueringer A, Frank S, et al. Alterations in lipid metabolism mediate inflammation, fibrosis, and proliferation in a mouse model of chronic cholestatic liver injury. *Gastroenterology* 2012;142:e112.
- [45] Weinstock GM. Genomic approaches to studying the human microbiota. *Nature* 2012;489:250–256.
- [46] The Human Microbiome Project Consortium. Structure, function and diversity of the healthy human microbiome. *Nature* 2012;486:207–214.
- [47] Kuczynski J, Lauber CL, Walters WA, Parfrey LW, Clemente JC, Gevers D, et al. Experimental and analytical tools for studying the human microbiome. *Nat Rev Genet* 2012;13:47–58.
- [48] Markle JG, Frank DN, Mortin-Toth S, Robertson CE, Feazel LM, Rolle-Kampczyk U, et al. Sex differences in the gut microbiome drive hormone-dependent regulation of autoimmunity. *Science* 2013;339:1084–1088.
- [49] Spor A, Koren O, Ley R. Unravelling the effects of the environment and host genotype on the gut microbiome. *Nat Rev Microbiol* 2011;9:279–290.

Review

- [50] Garrett WS, Lord GM, Punit S, Lugo-Villarino G, Mazmanian SK, Ito S, et al. Communicable ulcerative colitis induced by T-bet deficiency in the innate immune system. *Cell* 2007;131:33–45.
- [51] Maurice CF, Haïser HJ, Turnbaugh PJ. Xenobiotics shape the physiology and gene expression of the active human gut microbiome. *Cell* 2013;15:39–50.
- [52] McGovern DP, Jones MR, Taylor KD, Marcianti K, Yan X, Dubinsky M, et al. Fucosyltransferase 2 (FUT2) non-secretor status is associated with Crohn's disease. *Hum Mol Genet* 2010;19:3468–3476.
- [53] Rausch P, Rehman A, Kunzel S, Hasler R, Ott SJ, Schreiber S, et al. Colonic mucosa-associated microbiota is influenced by an interaction of Crohn disease and FUT2 (secretor) genotype. *Proc Natl Acad Sci U S A* 2011;108:19030–19035.
- [54] Wacklin P, Makivuokko H, Alakulppi N, Nikkila J, Tenkanen H, Rabina J, et al. Secretor genotype (FUT2 gene) is strongly associated with the composition of Bifidobacteria in the human intestine. *PLoS One* 2011;6:e20113.
- [55] Franke A, Balschun T, Karlsen TH, Sventoraityte J, Nikolaus S, Mayr G, et al. Sequence variants in IL10, ARPC2 and multiple other loci contribute to ulcerative colitis susceptibility. *Nat Genet* 2008;40:1319–1323.
- [56] Devkota S, Wang Y, Musch MW, Leone V, Fehlner-Peach H, Nadimpalli A, et al. Dietary-fat-induced taurocholic acid promotes pathobiont expansion and colitis in IL10^{-/-} mice. *Nature* 2012;487:104–108.
- [57] Adams DH, Eksteen B. Aberrant homing of mucosal T cells and extra-intestinal manifestations of inflammatory bowel disease. *Nat Rev Immunol* 2006;6:244–251.
- [58] Iwata M, Hirakiyama A, Eshima Y, Kagechika H, Kato C, Song SY. Retinoic acid imprints gut-homing specificity on T cells. *Immunity* 2004;21:527–538.
- [59] Eksteen B, Mora JR, Haughton EL, Henderson NC, Lee-Turner L, Villablanca EJ, et al. Gut homing receptors on CD8 T cells are retinoic acid dependent and not maintained by liver dendritic or stellate cells. *Gastroenterology* 2009;137:320–329.
- [60] Neumann K, Kruse N, Szilagy B, Erben U, Rudolph C, Flach A, et al. Connecting liver and gut: murine liver sinusoidal endothelium induces gut tropism of CD4⁺ T cells via retinoic acid. *Hepatology* 2012;55:1976–1984.
- [61] Liaskou E, Karikoski M, Reynolds GM, Lalor PF, Weston CJ, Pullen N, et al. Regulation of mucosal addressin cell adhesion molecule 1 expression in human and mice by vascular adhesion protein 1 amine oxidase activity. *Hepatology* 2011;53:661–672.
- [62] Lalor PF, Tuncer C, Weston C, Martin-Santos A, Smith DJ, Adams DH. Vascular adhesion protein-1 as a potential therapeutic target in liver disease. *Ann N Y Acad Sci* 2007;1110:485–496.
- [63] Shetty S, Bruns T, Weston CJ, Stamataki Z, Oo YH, Long HM, et al. Recruitment mechanisms of primary and malignant B cells to the human liver. *Hepatology* 2012;56:1521–1531.
- [64] Oo YH, Weston CJ, Lalor PF, Curbishley SM, Withers DR, Reynolds GM, et al. Distinct roles for CCR4 and CXCR3 in the recruitment and positioning of regulatory T cells in the inflamed human liver. *J Immunol* 2010;184:2886–2898.
- [65] Spinall AI, Curbishley SM, Lalor PF, Weston CJ, Liaskou E, Adams RM, et al. CX(3)CR1 and vascular adhesion protein-1-dependent recruitment of CD16(+) monocytes across human liver sinusoidal endothelium. *Hepatology* 2010;51:2030–2039.
- [66] Oo YH, Banz V, Kavanagh D, Liaskou E, Withers DR, Humphreys E, et al. CXCR3-dependent recruitment and CCR6-mediated positioning of Th-17 cells in the inflamed liver. *J Hepatol* 2012;57:1044–1051.
- [67] Barth MC, Ahluwalia N, Anderson TJ, Hardy GJ, Sinha S, Alvarez-Cardona JA, et al. Kynurenic acid triggers firm arrest of leukocytes to vascular endothelium under flow conditions. *J Biol Chem* 2009;284:19189–19195.
- [68] Ponsioen CY. Novel developments in IBD-related sclerosing cholangitis. *Best Pract Res Clin Gastroenterol* 2011;25:S15–S18.
- [69] de Buy Wenniger LJ, Doorenspleet ME, Klarenbeek PL, Verheij J, Baas F, Elferink RP, et al. IgG4⁺ clones identified by next-generation sequencing dominate the b-cell receptor repertoire in IgG4-associated cholangitis. *Hepatology* 2013. <http://dx.doi.org/10.1002/hep.26232>, in press.
- [70] Maillette de Buy Wenniger L, Rauws EA, Beuers U. What an endoscopist should know about immunoglobulin-G4-associated disease of the pancreas and biliary tree. *Endoscopy* 2012;44:66–73.
- [71] Dite P, Novotny I, Trna J, Svecikova A. Autoimmune pancreatitis. *Best Pract Res Clin Gastroenterol* 2008;22:131–143.
- [72] Moon SH, Kim MH, Park do H, Song TJ, Eum J, Lee SS, et al. IgG4 immunostaining of duodenal papillary biopsy specimens may be useful for supporting a diagnosis of autoimmune pancreatitis. *Gastrointest Endosc* 2010;71:960–966.
- [73] Vosskuhl K, Negm AA, Framke T, Weismuller T, Manns MP, Wedemeyer H, et al. Measurement of IgG4 in bile: a new approach for the diagnosis of IgG4-associated cholangiopathy. *Endoscopy* 2012;44:48–52.
- [74] Oseini AM, Chaiteerakij R, Shire AM, Ghazale A, Kaiya J, Moser CD, et al. Utility of serum immunoglobulin G4 in distinguishing immunoglobulin G4-associated cholangitis from cholangiocarcinoma. *Hepatology* 2011;54:940–948.
- [75] Bjornsson E, Chari S, Silveira M, Gossard A, Takahashi N, Smyrk T, et al. Primary sclerosing cholangitis associated with elevated immunoglobulinG4: clinical characteristics and response to therapy. *Am J Ther* 2010;18:198–205.
- [76] Lamberts LE, Janse M, Haagsma EB, van den Berg AP, Weersma RK. Immune-mediated diseases in primary sclerosing cholangitis. *Dig Liver Dis* 2011;43:802–806.
- [77] Halliday JS, Djordjevic J, Lust M, Culver EL, Braden B, Travis SP, et al. A unique clinical phenotype of primary sclerosing cholangitis associated with Crohn's disease. *J Crohn's Colitis* 2012;6:174–181.
- [78] Rupp C, Mummelthei A, Sauer P, Weiss KH, Schirmacher P, Stiehl A, et al. Non-IBD immunological diseases are a risk factor for reduced survival in PSC. *Liver Int* 2013;33:86–93.
- [79] Ngu JH, Geary RB, Wright AJ, Stedman CA. Inflammatory bowel disease is associated with poor outcomes of patients with primary sclerosing cholangitis. *Clin Gastroenterol Hepatol* 2011;9:1092–1097.
- [80] Rudolph G, Gotthardt D, Kloeters-Plachky P, Rost D, Kulaksiz H, Stiehl A. In PSC with dominant bile duct stenosis, IBD is associated with an increase of carcinomas and reduced survival. *J Hepatol* 2010;53:313–317.
- [81] Broome U, Bergquist A. Primary sclerosing cholangitis, inflammatory bowel disease, and colon cancer. *Semin Liver Dis* 2006;26:31–41.
- [82] Claessen MM, Lutgens MW, van Buuren HR, Oldenburg B, Stokkers PC, van der Woude CJ, et al. More right-sided IBD-associated colorectal cancer in patients with primary sclerosing cholangitis. *Inflamm Bowel Dis* 2009;15:1331–1336.
- [83] Bergquist A, Montgomery SM, Bahmanyar S, Olsson R, Danielsson A, Lindgren S, et al. Increased risk of primary sclerosing cholangitis and ulcerative colitis in first-degree relatives of patients with primary sclerosing cholangitis. *Clin Gastroenterol Hepatol* 2008;6:939–943.
- [84] Lindstrom L, Lapidus A, Ost A, Bergquist A. Increased risk of colorectal cancer and dysplasia in patients with Crohn's colitis and primary sclerosing cholangitis. *Dis Colon Rectum* 2011;54:1392–1397.
- [85] Loftus Jr EV, Harewood GC, Loftus CG, Tremaine WJ, Harmsen WS, Zinsmeister AR, et al. PSC-IBD: a unique form of inflammatory bowel disease associated with primary sclerosing cholangitis. *Gut* 2005;54:91–96.
- [86] Chapman MH, Webster GJ, Bannoo S, Johnson GJ, Wittmann J, Pereira SP. Cholangiocarcinoma and dominant strictures in patients with primary sclerosing cholangitis: a 25-year single-centre experience. *Eur J Gastroenterol Hepatol* 2012;24:1051–1058.
- [87] Fevery J, Henckaerts L, Van Oirbeek R, Vermeire S, Rutgeerts P, Nevens F, et al. Malignancies and mortality in 200 patients with primary sclerosing cholangitis: a long-term single-centre study. *Liver Int* 2012;32:214–222.
- [88] Boberg KM, Bergquist A, Mitchell S, Pares A, Rosina F, Broome U, et al. Cholangiocarcinoma in primary sclerosing cholangitis: risk factors and clinical presentation. *Scand J Gastroenterol* 2002;37:1205–1211.
- [89] Boberg KM, Lind GE. Primary sclerosing cholangitis and malignancy. *Best Pract Res Clin Gastroenterol* 2011;25:753–764.
- [90] Chalasani N, Baluyut A, Ismail A, Zaman A, Sood G, Ghalib R, et al. Cholangiocarcinoma in patients with primary sclerosing cholangitis: a multicenter case-control study. *Hepatology* 2000;31:7–11.
- [91] Said K, Glaumann H, Bergquist A. Gallbladder disease in patients with primary sclerosing cholangitis. *J Hepatol* 2008;48:598–605.
- [92] Karlsen TH, Schruppf E, Boberg KM. Gallbladder polyps in primary sclerosing cholangitis: not so benign. *Curr Opin Gastroenterol* 2008;24:395–399.
- [93] Schramm C, Lohse AW. Gallbladder polyps in primary sclerosing cholangitis: indication for early intervention. *Hepatology* 2012;56:396.
- [94] Eaton JE, Thackeray EW, Lindor KD. Likelihood of malignancy in gallbladder polyps and outcomes following cholecystectomy in primary sclerosing cholangitis. *Am J Gastroenterol* 2012;107:431–439.
- [95] Sinakos E, Saenger AK, Keach J, Kim WR, Lindor KD. Many patients with primary sclerosing cholangitis and increased serum levels of carbohydrate antigen 19-9 do not have cholangiocarcinoma. *Clin Gastroenterol Hepatol* 2011;9:e431.
- [96] Razumilava N, Gores GJ. Classification, diagnosis, and management of cholangiocarcinoma. *Clin Gastroenterol Hepatol* 2013;11:e11.
- [97] Narimatsu H, Iwasaki H, Nakayama F, Ikehara Y, Kudo T, Nishihara S, et al. Lewis and secretor gene dosages affect CA19-9 and DU-PAN-2 serum levels

- in normal individuals and colorectal cancer patients. *Cancer Res* 1998;58:512–518.
- [98] Halme L, Arola J, Numminen K, Krogerus L, Makisalo H, Farkkila M. Biliary dysplasia in patients with primary sclerosing cholangitis: additional value of DNA ploidy. *Liver Int* 2012;32:783–789.
- [99] Barr Fritcher EG, Kipp BR, Voss JS, Clayton AC, Lindor KD, Halling KC, et al. Primary sclerosing cholangitis patients with serial polysomy fluorescence in situ hybridization results are at increased risk of cholangiocarcinoma. *Am J Gastroenterol* 2011;106:2023–2028.
- [100] Barr Fritcher EG, Caudill JL, Blue JE, Djuric K, Feipel L, Maritim BK, et al. Identification of malignant cytologic criteria in pancreatobiliary brushings with corresponding positive fluorescence in situ hybridization results. *Am J Clin Pathol* 2011;136:442–449.
- [101] Kipp BR, Stadheim LM, Halling SA, Pochron NL, Harmsen S, Nagorney DM, et al. A comparison of routine cytology and fluorescence in situ hybridization for the detection of malignant bile duct strictures. *Am J Gastroenterol* 2004;99:1675–1681.
- [102] Charatcharoenwitthaya P, Enders FB, Halling KC, Lindor KD. Utility of serum tumor markers, imaging, and biliary cytology for detecting cholangiocarcinoma in primary sclerosing cholangitis. *Hepatology* 2008;48:1106–1117.
- [103] Boberg KM, Jepsen P, Clausen OP, Foss A, Aabakken L, Schrupf E. Diagnostic benefit of biliary brush cytology in cholangiocarcinoma in primary sclerosing cholangitis. *J Hepatol* 2006;45:568–574.
- [104] Bangarulingam SY, Bjornsson E, Enders F, Barr Fritcher EG, Gores G, Halling KC, et al. Long-term outcomes of positive fluorescence in situ hybridization tests in primary sclerosing cholangitis. *Hepatology* 2010;51:174–180.
- [105] Lewis JT, Talwalkar JA, Rosen CB, Smyrk TC, Abraham SC. Precancerous bile duct pathology in end-stage primary sclerosing cholangitis, with and without cholangiocarcinoma. *Am J Surg Pathol* 2010;34:27–34.
- [106] Imam MH, Gossard AA, Sinakos E, Lindor KD. Pathogenesis and management of pruritus in cholestatic liver disease. *J Gastroenterol Hepatol* 2012;27:1150–1158.
- [107] Pares A, Herrera M, Aviles J, Sanz M, Mas A. Treatment of resistant pruritus from cholestasis with albumin dialysis: combined analysis of patients from three centers. *J Hepatol* 2010;53:307–312.
- [108] Pauli-Magnus C, Meier PJ, Stieger B. Genetic determinants of drug-induced cholestasis and intrahepatic cholestasis of pregnancy. *Semin Liver Dis* 2010;30:147–159.
- [109] Gay M, Pares A, Carrascal M, Bosch-i-Crespo P, Gorga M, Mas A, et al. Proteomic analysis of polypeptides captured from blood during extracorporeal albumin dialysis in patients with cholestasis and resistant pruritus. *PLoS One* 2011;6:e21850.
- [110] Kremer AE, Martens JJ, Kulk W, Rueff F, Kuiper EM, van Buuren HR, et al. Lysophosphatidic acid is a potential mediator of cholestatic pruritus. *Gastroenterology* 2010;139:1008–1018, e1001.
- [111] Alemi F, Kwon E, Poole DP, Lieu T, Lyo V, Cattaruzza F, et al. The TGR5 receptor mediates bile acid-induced itch and analgesia. *J Clin Invest* 2013;123:1513–1530.
- [112] Boberg KM, Chapman RW, Hirschfeld GM, Lohse AW, Manns MP, Schrupf E. Overlap syndromes: The International Autoimmune Hepatitis Group (IAIHG) position statement on a controversial issue. *J Hepatol* 2010;54:374–385.
- [113] Hennes EM, Zeniya M, Czaja AJ, Pares A, Dalekos GN, Krawitt EL, et al. Simplified criteria for the diagnosis of autoimmune hepatitis. *Hepatology* 2008;48:169–176.
- [114] European Association for the Study of the Liver. EASL clinical practice guidelines: management of cholestatic liver diseases. *J Hepatol* 2009;51:237–267.
- [115] Luth S, Kanzler S, Frenzel C, Kasper HU, Dienes HP, Schramm C, et al. Characteristics and long-term prognosis of the autoimmune hepatitis/primary sclerosing cholangitis overlap syndrome. *J Clin Gastroenterol* 2008;43:75–80.
- [116] Al-Chalabi T, Portmann BC, Bernal W, McFarlane IG, Heneghan MA. Autoimmune hepatitis overlap syndromes: an evaluation of treatment response, long-term outcome and survival. *Aliment Pharmacol Ther* 2008;28:209–220.
- [117] Chapman R, Fevery J, Kallou A, Nagorney DM, Boberg KM, Shneider B, et al. Diagnosis and management of primary sclerosing cholangitis. *Hepatology* 2010;51:660–678.
- [118] Farrant JM, Hayllar KM, Wilkinson ML, Karani J, Portmann BC, Westaby D, et al. Natural history and prognostic variables in primary sclerosing cholangitis. *Gastroenterology* 1991;100:1710–1717.
- [119] Chambers JC, Zhang W, Sehmi J, Li X, Wass MN, Van der Harst P, et al. Genome-wide association study identifies loci influencing concentrations of liver enzymes in plasma. *Nat Genet* 2011;43:1131–1138.
- [120] Folseraas T, Melum E, Rausch P, Juran BD, Ellinghaus E, Shiryayev A, et al. Extended analysis of a genome-wide association study in primary sclerosing cholangitis detects multiple novel risk loci. *J Hepatol* 2012;57:366–375.
- [121] Al Mamari S, Djordjevic J, Halliday JS, Chapman RW. Improvement of serum alkaline phosphatase to <1.5 upper limit of normal predicts better outcome and reduced risk of cholangiocarcinoma in primary sclerosing cholangitis. *J Hepatol* 2013;58:329–334.
- [122] Lindstrom L, Hultcrantz R, Boberg KM, Friis-Liby I, Bergquist A. Association between reduced levels of alkaline phosphatase and survival times of patients with primary sclerosing cholangitis. *Clin Gastroenterol Hepatol* 2013. <http://dx.doi.org/10.1016/j.cgh.2012.12.032>, in press.
- [123] Stanich PP, Bjornsson E, Gossard AA, Enders F, Jorgensen R, Lindor KD. Alkaline phosphatase normalization is associated with better prognosis in primary sclerosing cholangitis. *Dig Liver Dis* 2011;43:309–313.
- [124] Sinakos E, Marschall HU, Kowdley KV, Befeler A, Keach J, Lindor K. Bile acid changes after high-dose ursodeoxycholic acid treatment in primary sclerosing cholangitis: Relation to disease progression. *Hepatology* 2010;52:197–203.
- [125] Rost D, Rudolph G, Kloeters-Plachky P, Stiehl A. Effect of high-dose ursodeoxycholic acid on its biliary enrichment in primary sclerosing cholangitis. *Hepatology* 2004;40:693–698.
- [126] Turnbaugh PJ. Microbiology: fat, bile and gut microbes. *Nature* 2012;487:47–48.
- [127] Tremaroli V, Backhed F. Functional interactions between the gut microbiota and host metabolism. *Nature* 2012;489:242–249.
- [128] Duboc H, Rajca S, Rainteau D, Benarous D, Maubert MA, Quervain E, et al. Connecting dysbiosis, bile-acid dysmetabolism and gut inflammation in inflammatory bowel diseases. *Gut* 2013;62:531–539.
- [129] Kakiyama G, Pandak WM, Gillevet PM, Hylemon PB, Heuman DM, Daita K, et al. Modulation of the Fecal Bile Acid Profile by Gut Microbiota in Cirrhosis. *J Hepatol* 2013;58:949–955.
- [130] Henao-Mejia J, Elinav E, Jin C, Hao L, Mehal WZ, Strowig T, et al. Inflammation-mediated dysbiosis regulates progression of NAFLD and obesity. *Nature* 2012;482:179–185.
- [131] Abarbanel DN, Seki SM, Davies Y, Marlen N, Benavides JA, Cox K, et al. Immunomodulatory effect of vancomycin on Treg in pediatric inflammatory bowel disease and primary sclerosing cholangitis. *J Clin Immunol* 2013;33:397–406.
- [132] Farkkila M, Karvonen AL, Nurmi H, Nuutinen H, Taavitsainen M, Pikkarainen P, et al. Metronidazole and ursodeoxycholic acid for primary sclerosing cholangitis: a randomized placebo-controlled trial. *Hepatology* 2004;40:1379–1386.
- [133] 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 Pharm* 2007;20:847–849.
- [134] Silveira MG, Torok NJ, Gossard AA, Keach JC, Jorgensen RA, Petz JL, et al. Minocycline in the treatment of patients with primary sclerosing cholangitis: results of a pilot study. *Am J Gastroenterol* 2009;104:83–88.
- [135] Sandborn WJ, Wiesner RH, Tremaine WJ, Larusso NF. Ulcerative colitis disease activity following treatment of associated primary sclerosing cholangitis with cyclosporin. *Gut* 1993;34:242–246.
- [136] Talwalkar JA, Gossard AA, Keach JC, Jorgensen RA, Petz JL, Lindor RN. Tacrolimus for the treatment of primary sclerosing cholangitis. *Liver Int* 2007;27:451–453.
- [137] Jorgensen KK, Lindstrom L, Cvancarova M, Karlsen TH, Castedal M, Friman S, et al. Immunosuppression after liver transplantation for primary sclerosing cholangitis influences activity of inflammatory bowel disease. *Clin Gastroenterol Hepatol* 2013;11:517–523.
- [138] Baghdasaryan A, Claudel T, Kosters A, Gumhold J, Silbert D, Thuringer A, et al. Curcumin improves sclerosing cholangitis in Mdr2^{-/-} mice by inhibition of cholangiocyte inflammatory response and portal myofibroblast proliferation. *Gut* 2010;59:521–530.
- [139] Trauner M, Baghdasaryan A, Claudel T, Fickert P, Halilbasic E, Moustafa T, et al. Targeting nuclear bile acid receptors for liver disease. *Dig Dis* 2011;29:98–102.
- [140] Nakken KE, Nygard S, Haaland T, Berge KE, Arnkvaern K, Odegaard A, et al. Multiple inflammatory-, tissue remodelling- and fibrosis genes are differentially transcribed in the livers of Abcb4^(-/-) mice harbouring chronic cholangitis. *Scand J Gastroenterol* 2007;42:1245–1255.
- [141] Sandanayake NS, Sinclair J, Andreola F, Chapman MH, Xue A, Webster GJ, et al. A combination of serum leucine-rich alpha-2-glycoprotein 1, CA19-9

Review

- and interleukin-6 differentiate biliary tract cancer from benign biliary strictures. *Br J Cancer* 2011;105:1370–1378.
- [142] Lankisch TO, Metzger J, Negm AA, Vosskuhl K, Schiffer E, Siwy J, et al. Bile proteomic profiles differentiate cholangiocarcinoma from primary sclerosing cholangitis and choledocholithiasis. *Hepatology* 2011;53:875–884.
- [143] Metzger J, Negm AA, Plentz RR, Weismuller TJ, Wedemeyer J, Karlsen TH, et al. Urine proteomic analysis differentiates cholangiocarcinoma from primary sclerosing cholangitis and other benign biliary disorders. *Gut* 2013;62:122–130.
- [144] Kim BH, Cho NY, Choi M, Lee S, Jang JJ, Kang GH. Methylation profiles of multiple CpG island loci in extrahepatic cholangiocarcinoma versus those of intrahepatic cholangiocarcinomas. *Arch Pathol Lab Med* 2007;131:923–930.
- [145] Rawson JB, Bapat B. Epigenetic biomarkers in colorectal cancer diagnostics. *Expert Rev Mol Diagn* 2012;12:499–509.
- [146] Andresen K, Boberg KM, Vedeld HM, Honne H, Hektoen M, Wadsworth CA, et al. Novel target genes and a valid biomarker panel identified for cholangiocarcinoma. *Epigenetics* 2012;7:1249–1257.
- [147] Fosby B, Karlsen TH, Melum E. Recurrence and rejection in liver transplantation for primary sclerosing cholangitis. *World J Gastroenterol* 2012;18:1–15.
- [148] Alabraba E, Nightingale P, Gunson B, Hubscher S, Olliff S, Mirza D, et al. A re-evaluation of the risk factors for the recurrence of primary sclerosing cholangitis in liver allografts. *Liver Transpl* 2009;15:330–340.
- [149] Darwish Murad S, Kim WR, Harnois DM, Douglas DD, Burton J, Kulik LM, et al. Efficacy of neoadjuvant chemoradiation, followed by liver transplantation, for perihilar cholangiocarcinoma at 12 US centers. *Gastroenterology* 2012;143:88–98, e83; quiz e14.
- [150] Rosen CB, Darwish Murad S, Heimbach JK, Nyberg SL, Nagorney DM, Gores GJ. Neoadjuvant therapy and liver transplantation for hilar cholangiocarcinoma: is pretreatment pathological confirmation of diagnosis necessary? *J Am Coll Surg* 2012;215:31–38.
- [151] Darwish Murad S, Kim WR, Therneau T, Gores GJ, Rosen CB, Martenson JA, et al. Predictors of pretransplant dropout and posttransplant recurrence in patients with perihilar cholangiocarcinoma. *Hepatology* 2012;56:972–981.
- [152] Friman S, Foss A, Isoniemi H, Olausson M, Hockerstedt K, Yamamoto S, et al. Liver transplantation for cholangiocarcinoma: selection is essential for acceptable results. *Scand J Gastroenterol* 2011;46:370–375.
- [153] Goldberg DS, Camp A, Martinez-Camacho A, Forman L, Fortune B, Reddy KR. Risk of waitlist mortality in patients with primary sclerosing cholangitis and bacterial cholangitis. Liver transplantation: official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society 2013;19:250–258.
- [154] Faubion Jr WA, Loftus EV, Sandborn WJ, Freese DK, Perrault J. Pediatric “PSC-IBD”: a descriptive report of associated inflammatory bowel disease among pediatric patients with PSC. *J Pediatr Gastroenterol Nutr* 2001;33:296–300.
- [155] Joo M, Abreu-e-Lima P, Farraye F, Smith T, Swaroop P, Gardner L, et al. Pathologic features of ulcerative colitis in patients with primary sclerosing cholangitis: a case-control study. *Am J Surg Pathol* 2009;33:854–862.
- [156] Jorgensen KK, Grzyb K, Lundin KE, Clausen OP, Aamodt G, Schrumpf E, et al. Inflammatory bowel disease in patients with primary sclerosing cholangitis: Clinical characterization in liver transplanted and nontransplanted patients. *Inflamm Bowel Dis* 2012;18:536–545.
- [157] Ye BD, Yang SK, Boo SJ, Cho YK, Yang DH, Yoon SM, et al. Clinical characteristics of ulcerative colitis associated with primary sclerosing cholangitis in Korea. *Inflamm Bowel Dis* 2011;17:1901–1906.
- [158] Jostins L, Ripke S, Weersma RK, Duerr RH, McGovern DP, Hui KY, et al. Host-microbe interactions have shaped the genetic architecture of inflammatory bowel disease. *Nature* 2012;491:119–124.