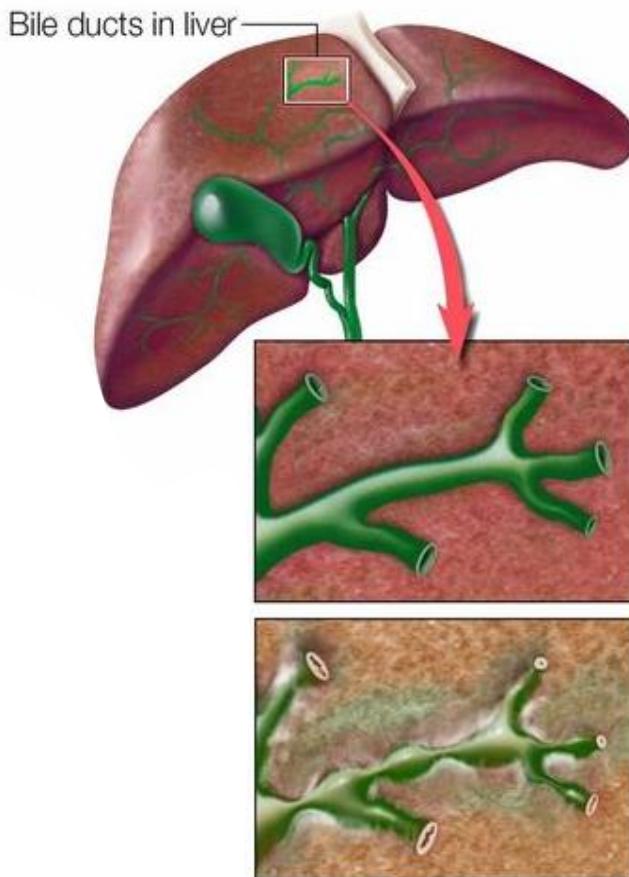


PRIMARY BILIARY CHOLANGITIS/CIRRHOSIS



Internal Medicine Grand Rounds
UT Southwestern Medical Center
August 11, 2017

Marlyn J Mayo, MD
Division of Digestive & Liver Diseases

This is to acknowledge that Marlyn Mayo, MD has disclosed related relationships (clinical trial agreements) with Intercept Pharmaceuticals, Glaxo Smith Klein, Shire Pharmaceuticals, Cymabay, and NGM Pharmaceuticals, who have compounds in development for PBC. Dr. Mayo will be discussing off label uses in this presentation

Marlyn J Mayo, MD
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Marlyn Mayo, MD is an Associate Professor of Medicine in the Division of Digestive and Liver Diseases. Dr. Mayo attended medical school at Baylor College of Medicine, from which she obtained her M.D. in 1990. She then completed internship and residency at University of California, Irvine. She came to UT Southwestern to complete Gastroenterology and Hepatology fellowships and joined the faculty in 1999. Dr Mayo has been the recipient of grants from NIH and AASLD to study the immunopathogenesis of PBC. She currently serves as principal investigator for multiple clinical trials investigating new treatment options for patients with PBC, and she maintains an active faculty practice in general Hepatology. She has a secondary interest in medical education and served as the GI Fellowship Director from 2012-2017.

Interests: cholestatic and autoimmune liver diseases

Purposes:

- To improve clinical management of patients with PBC
- To show physicians and scientists how recent advances in our understanding of bile acid metabolism and inflammation may be relevant to the pathogenesis of PBC and may lead to new therapies

Learning Objectives

At the end of this presentation, the learner should be able to:

- Recall the new name for primary biliary cirrhosis of “primary biliary cholangitis” and understand why the name was changed
- Recognize the full spectrum of clinical presentation of PBC
- Know the two FDA-approved therapies for PBC, and their mechanism of action

Clinical Presentation

Primary biliary cholangitis or primary biliary cirrhosis (abbreviated PBC) is a chronic cholestatic liver disease characterized by inflammatory destruction of the small to medium sized bile ducts in the liver. The healthy bile ducts become inflamed and the bile duct epithelial cells die of apoptosis. The bile duct lumens are progressively obliterated, impairing excretion of bile from the liver. Retained bile damages hepatocytes and biliary cells. Fibrosis accumulates over time and leads to cirrhosis.

PBC is a predominantly female disease, with a female to male ratio of approximately 12:1. Almost all patients have autoantibodies to mitochondrial antigens (AMA). The most common clinical symptoms are itching and fatigue. Cholestatic patients have an increased risk of osteoporosis and fat soluble vitamin deficiencies. The typical lab abnormalities are elevated alkaline phosphatase and GGT, which is how many patients are often identified in the early, asymptomatic phase. Bilirubin rises as the disease progresses and thus is an excellent prognostic marker. Transaminases may be normal, but may also reach into the several-hundred range, as there is a spectrum of the degree of associated hepatitis. Serum cholesterol is also elevated as a consequence of chronic cholestasis, and another characteristic feature is an elevation of polyclonal IgM.

Histology

PBC progresses through 4 histological stages. The first stage is defined by inflammation centered on the bile ducts. In the second stage, inflammation begins to spread beyond the limiting plate of the portal triad and fibrosis begins to develop. In both of these stages, the disease can be quite patchy and potentially missed on a small liver biopsy. Stage 3 is characterized by fibrosis that bridges between the adjacent branches of the biliary tree and loss of bile ducts in some areas. Stage 4 is synonymous with cirrhosis, where the fibrosis encircles nodules of liver trying to regenerate.

The inflammation centered around the bile duct is usually a cluster of CD4 and CD8 positive T cells admixed with plasma cells and a few eosinophils. There may be granulomatous destruction of the bile duct, which is considered so specific to this disease that it is called a "Florid Duct Lesion"

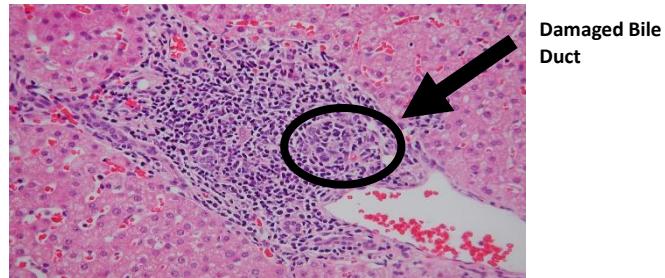


Figure 1. Florid Duct Lesion of PBC

Although the histological features are often diagnostic, liver biopsy is no longer required for the diagnosis if the patient has these clinical features. If the patient is a middle aged female, AMA positive, elevated alkaline phosphatase for > 6 months, transaminases below 500 and no clinical suspicion for another liver disease. This constellation of findings has a 95% positive predictive for the diagnosis of PBC, and thus liver biopsy is not needed.

Natural History

PBC begins with a long asymptomatic period. The very first abnormality that usually occurs is the development of anti-mitochondrial antibodies, before one can see much histological inflammation of the bile ducts on biopsy and before the alkaline phosphatase becomes elevated in the serum. Later, symptoms will develop: fatigue and pruritus. The fatigue may be associated with orthostatic hypotension or cognitive defects. The pruritus characteristically involves the palms and soles and has an interesting circadian rhythm, where it is worst in the evenings. As the disease progresses, fibrosis accumulates. Once cirrhosis develops, patients may develop portal hypertension.

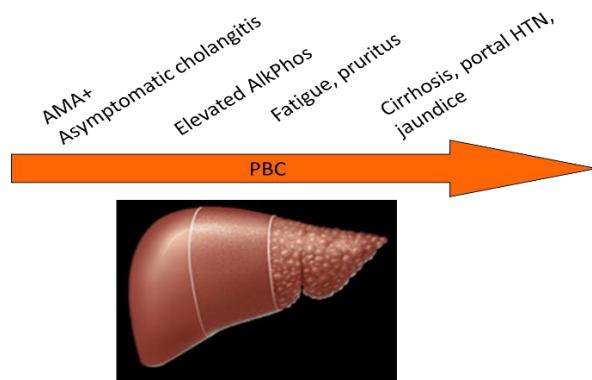


Figure 2. Natural History of PBC

Primary Biliary Cholangitis: A New Name

Primary biliary cholangitis is the same disease as primary biliary cirrhosis. Patient representatives wanted to remove the label of "cirrhosis," which is inaccurate for those with stage 1,2 or 3 disease because they felt the word "cirrhosis" carried a stigma. The new name was decided by vote on a conference call of experts and a single article was published in the 8 leading liver journals of the world, proclaiming the new name. The new name has been accepted by the scientific and regulatory communities.

PBC is Becoming Milder and More Common

The change in name is appropriate, given the changing clinical presentation of PBC, which is being diagnosed increasingly at earlier stages, due to increased awareness as well as widespread availability of chemistry panels including alkaline phosphatase and AMA testing. This table below illustrates how PBC patients who were diagnosed in the 1970s and early 1980s were usually already symptomatic with fatigue and pruritus. However, now it is much more common to diagnose patients in the asymptomatic phase. Evidence of more advanced disease, such as jaundice, splenomegaly, hyperpigmentation and xanthomas are actually now uncommon at the time of diagnosis. This is contrasted with the clinical presentation of PSC, which, despite the advent of magnetic resonance imaging for diagnosis, has not changed much.

Table 1. Changing Clinical Presentation of PBC

Symptoms	PSC 1970-2007	PBC1970-1985	PBC1985-2007
Fatigue	75%	90%	67%
Pruritus	70%	57%	17%
Weight Loss	40%	-	-
Fever	35%	-	-
Abdominal Pain	15%	20%	17%
Hepatomegaly	55%	3%	+
Jaundice	50%	28%	15%
Splenomegaly	30%	3%	+
Hyperpigmentation	25%	++	+
Xanthomata	4%	++	+

The Global PBC group, an international group of investigators, followed over 4,000 patients with PBC over a 40 year period and demonstrated that the proportion of patients being diagnosed at early stage disease has increased significantly with each decade since the 1980s. The mean age at diagnosis has also increased from 47 in the 1970s to 57 at present. We do not know if the age shift is due to increased awareness of the disease, or a change in disease biology.

The prevalence of PBC is increasing over time; an estimated 1/3000 people in the US have PBC. PBC affects all races, but good data on the worldwide racial and ethnic distribution are lacking. It does appear that minorities with PBC present with more advanced disease and have poorer outcomes, which could be due to differences in either health care access or disease biology.

Physical Exam Findings

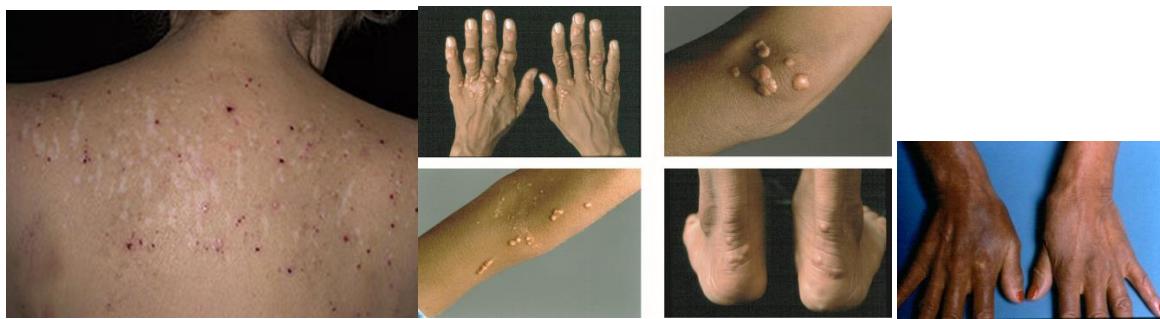


Figure 3. Left to Right: Excoriations from pruritus with “butterfly sparing,” xanthomas , and hyperpigmentation.

Extrahepatic Associations

Internists should be aware of the numerous extrahepatic associations with PBC, illustrated in the figure below:

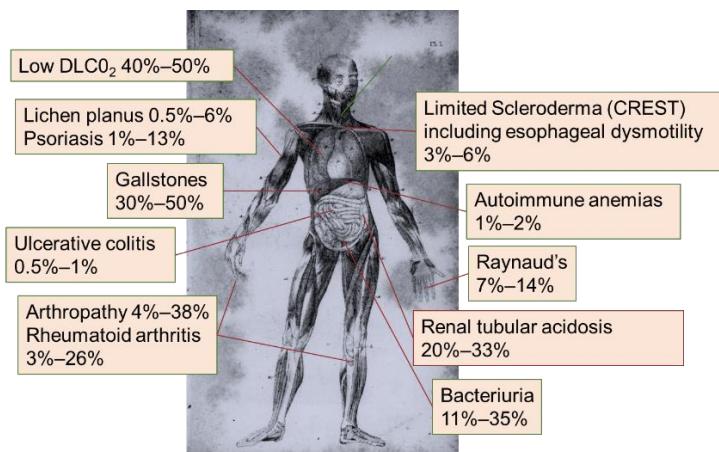


Figure 4. Extrahepatic associations with PBC

Pathophysiology

The exact etiology of PBC remains unknown. The disease is characterized by cholestasis and immune mediated destruction of bile ducts, but which comes first is not certain.

Loss of tolerance to the mitochondrial antigens may lead to the autoimmune destruction of bile ducts, with resultant cholestasis. However, it is also possible that the bile ducts are damaged by cholestasis first, and the damaged bile duct epithelial cells induce a secondary immune reaction.

Risk Factors

The strongest risk factor for developing PBC is a positive family history of PBC or to a lesser degree, other autoimmune diseases. Prior hormonal supplementation is also associated with increased risk of developing the disease, and never being pregnant appears to be protective. Weak (but statistically significant) risks of toxin exposures to cigarette smoke, nail polish, and hair dye have received considerable attention in the media.

Table 2 Risk Factors for PBC

	OR	95% CI	P
Medical/Family history			
Family history of PBC	10.736	4.227 - 27.268	<0.0001
Family history of SLE	2.234	1.261 – 3.957	0.0059
Family history of Sjögren's	5.814	1.279 - 26.435	0.0227
Lifestyle factors			
Ever smoked >100 cigarettes	1.569	1.292 – 1.905	<0.0001
Uses of nail polish / year	1.002	1.000-1.003	0.0136
Each smoker in household	0.5078	0.3167- 0.8143	0.0041
Reproductive history			
Ever used hormonal replacement	1.548	1.273-1.882	<0.0001
Never pregnant	0.6118	0.4489-0.8338	0.0012
Age of first pregnancy	0.9541	0.9331 – 0.9755	<0.0001

In genome wide association studies, the most important genetic risk factor was HLA and the specific association was HLADQ1, emphasizing the importance of both heredity and the immune system in developing PBC. GWAS also demonstrated a risk association with IL 12, the IL12 receptor, and STAT4 which mediates T cell responses to IL12. IL12 is a cytokine important in T cell induction of gamma interferon, and differentiation of TH1 cells.

Based on this finding as well as data showing upregulation of IL12 in humans with PBC, a clinical trial was undertaken using ustekinemab, a monoclonal antibody against IL12, but the drug did not have any measurable effect on clinically established PBC.

Anti-mitochondrial Antibodies

The role which antimitochondrial antibodies play in the pathogenesis of the disease continues to be debated. AMAs recognize the E2 subunit of the 2-oxo-acid dehydrogenase complexes, usually pyruvate dehydrogenase. PDC is highly conserved across species

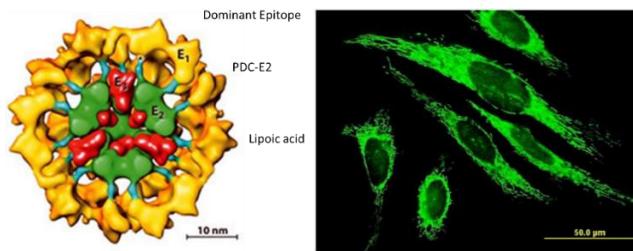


Figure 5. Pyruvate Dehydrogenase (left) is the target autoantigen of antimitochondrial antibodies, shown via immunofluorescence (right)

An important discovery was that biliary ductal epithelial cells in PBC aberrantly express PDC-E2 on their cell surface (it should be only in the mitochondria). It was hypothesized that this exposure to the immune system ignited the autoimmune destruction of the bile duct cells. Indeed, Autoreactive B and T cells in the liver do recognize this epitope. Apoptosis of bile duct cells causes them to express PDC-E2 on the surface, a feature which is unique to this cell type.

Molecular Mimicry

The factors that lead to loss of tolerance to PDCE-2 may include molecular mimicry, whereby one would be exposed to foreign mitochondrial antigens in the setting of an infection. The immune response mounted against the foreign mitochondrial antigens is then misdirected at our own native mitochondrial antigens. This theory is attractive because of the strong sequence homology of PDC-E2 across species.

Xenobiotics

Toxic chemical exposure may also play a role in the pathogenesis of PBC. There is an increased prevalence of PBC reported in survivors of the Nagasaki atomic bomb explosion and in persons who live near toxic waste sites in New York City. Most chemicals are metabolized in the liver, and during metabolism they may form reactive metabolites, which may modify cellular proteins to form neo-antigens. Xenobiotics may incite a reaction against native mitochondrial antigens by chemically modifying them to make them more immunogenic. This hypothesis has been proven in concept with animal models.

The Bicarbonate Umbrella Hypothesis

The bile duct epithelial cell normally secretes bicarbonate into the lumen. It has two mechanisms: cAMP dependent and Ca⁺⁺ dependent pathways to do this. This bicarbonate rich fluid serves as a buffer to protect the cell against injury from bile acids in the lumen and subsequent apoptosis. PBC could be a genetic or acquired predisposition to impaired formation

of a stable biliary HCO₃⁻ umbrella, rendering the biliary epithelial cell vulnerable to damage from the bile acids. Secretin drives this process by pumping chloride into the lumen, which is exchanged for the bicarbonate via the anion exchange protein (AE2). In patients with PBC, secretin induced excretion of bicarbonate is reduced. There is reduced expression of AE2 in the liver and in blood. AE2-deficient mice develop elevated liver enzymes, histological cholangitis, and positive AMA. Polymorphisms of AE2 are associated with risk of PBC. A specific microRNA-506 is increased in the liver of PBC patients, particularly overexpressed in the cholangiocytes. Mi-506 binds directly to the 3' untranslated region of AE2 mRNA, inhibiting protein translation, and resulting in decreased AE2 activity. There is some interest in pursuing secretin homologues or inhibitors of microRNA as a potential therapy for PBC.

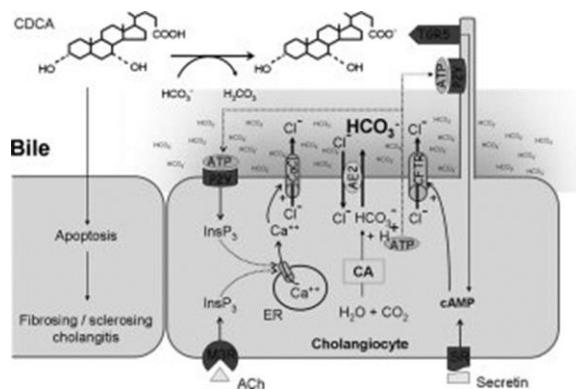


Figure 6. The bicarbonate umbrella (Hepatology 2010, 57:1989)

Treatment

Ursodeoxycholic Acid (Ursodiol, Urso)

There have been multiple randomized, double blind, placebo controlled clinical trials demonstrating that urso improves biochemical liver tests, delays histological progression, and prolongs expected survival without liver transplantation. None of the controlled trials were long enough to directly demonstrate the survival benefit, but compared to multiple prognostic models and historical controls, survival is improved with urso. Unfortunately, the transplant-free survival rate of UDCA-treated patients remains significantly lower than that of an age-matched and sex-matched control population, illustrating that better therapies are needed. Since the introduction of urso, the number of PBC patients listed liver transplantation has dropped, but has not gone away completely.

Overall survival is poorer due to a subset of patients called “ursodiol non-responders.”

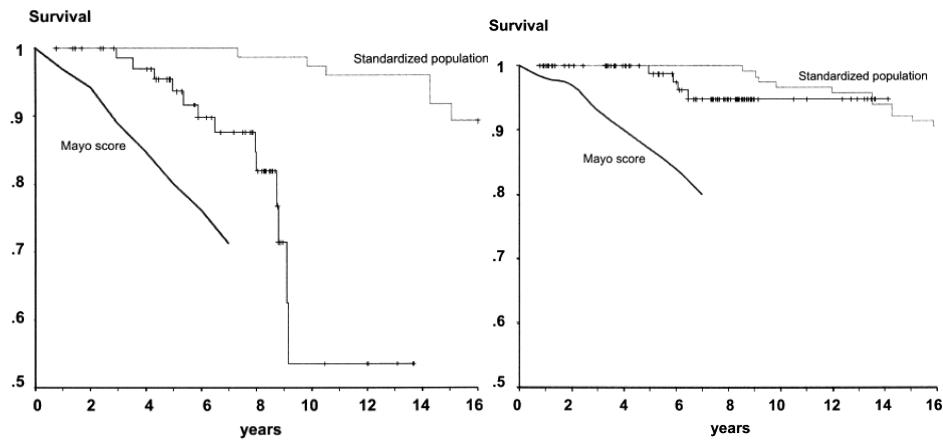


Figure 7. Patients with PBC who achieve a reduction of alkaline phosphatase to 40 % of baseline or normal after a year of ursodiol treatment have a transplant free survival that is no different from a control population (right plot). However, those that do not achieve this reduction in alkaline phosphatase have markedly reduced survival (left plot). Mayo Score shows the predicted survival of patients not treated with ursodiol.

In a French cohort treated with ursodiol, those who achieved a Bilirubin ≤ 1 , ALP ≤ 3 XULN, and AST ≤ 2 XULN after a year of ursodiol treatment had normal survival, whereas those who do not achieve those benchmarks, fared just as poorly as the natural history of untreated PBC. Approximately 20-40% of PBC patients fall into this non-responder or ineffectively-treated partial-responder category. It is this population for which additional treatments are needed.

Bilirubin is the single strongest marker in PBC, but jaundice does not develop until later in the disease process. Alkaline phosphatase response to ursodiol is an earlier marker of survival. The Globe Score is a prognostic model that combines bilirubin, alk phos, INR, age, to most accurately predict short and long term survival. There is an online calculator at this website:



www.globalpbc.com

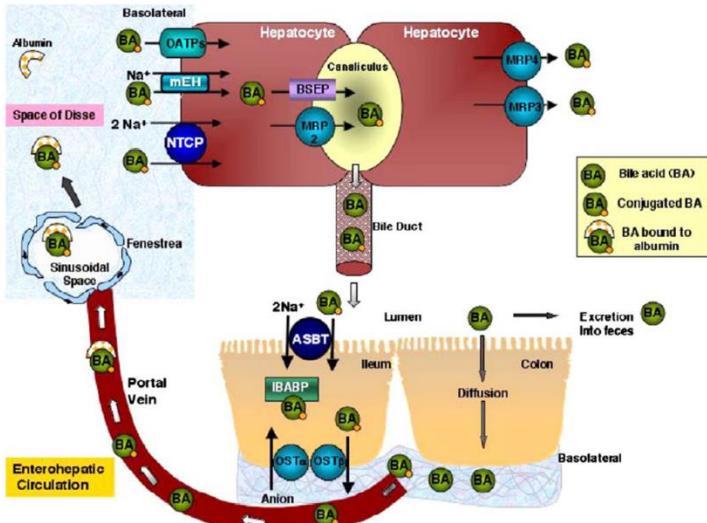


Figure 8. The Enterohepatic Circulation (Alfari, Pharm Research 2007)

Obeticholic Acid

In May 2016, the FDA approved a new therapy for PBC called obeticholic acid (OCA). OCA is an engineered bile acid in which an ethyl group is added to the 6 position of chenodeoxycholic acid. Adding the ethyl group results in 100 fold more potent stimulation of farnesoid X receptor (FXR). Ursodeoxycholic acid has negligible FXR activity, so even though both are bile acid therapies, OCA represents a new mechanism of action.

FXR plays an important role in bile acid metabolism and enterohepatic circulation. Bile acids are synthesized by the liver and excreted via bile salt export protein into the bile canaliculus where they travel through the biliary ductal system. They are re-absorbed from the gut lumen through the apical sodium-dependent bile acid transporter and then the organic solute transporters carry them into the bloodstream where they travel to the liver and are taken up by the organic anion transporting polypeptides to complete the enterohepatic cycle.

The above underlined transporters are under transcriptional control by FXR, which is why FXR agonists are useful in cholestatic liver diseases. They increase excretion of bile acids into the bile and decrease their uptake from the gut. FXR is also a key part of the negative feedback cycle that bile acids in the gut have to inhibit bile acid synthesis.

In clinical trials, OCA was tested as adjunctive therapy to ursodeoxycholic acid (ursodeo) in the subset of patients who did not respond biochemically to ursodeo alone, which is its primary FDA-approved indication. All three doses of the obeticholic acid resulted in a significant drop in ALP of about 24% of baseline.

Pruritus was a significant adverse event in clinical trials, and it was dose related. The doses that were FDA approved (5 and 10 mg) did not have significantly more itching than placebo. This

also makes a strong argument for bile acids being important in the etiology of cholestatic pruritus. The other adverse event that was more common in the OCA group was nausea, in about 10% of patients.

FGF-19 Analogues

The enterohepatic circulation has a negative feedback loop, such that when bile acid concentrations in the gut are high, de novo synthesis in the liver is downregulated.

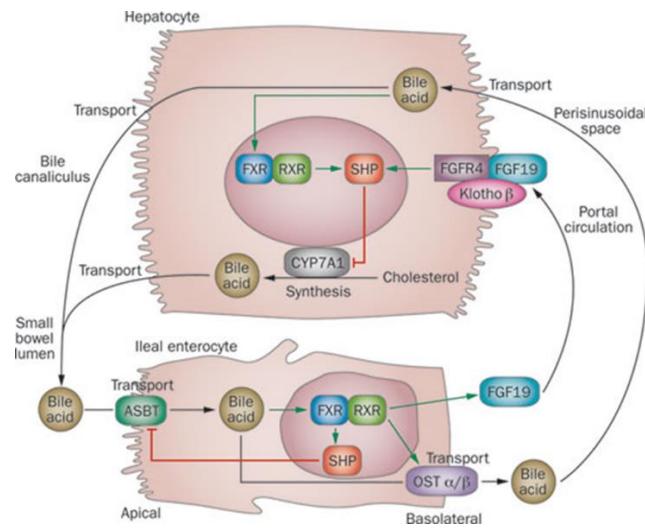


Figure 9. As bile acids are taken up by the apical sodium dependent bile acid transporter into the enterocyte, they bind to FXR and this leads to secretion of Fibroblast Growth Factor-19 (FGF-19). FGF-19 travels back to the hepatocytes and suppresses CYP 7A1, the rate limiting enzyme in de novo synthesis of bile acids from cholesterol.

Decreasing the size of the total bile acid pool using FGF-19 leads to reduction of liver damage in cholestatic animal models, which is why it is of potential interest as a therapeutic agent in PBC.

NGM-282, an engineered recombinant FGF-19, was given to PBC patients with elevated alkaline phosphatase despite treatment with ursodeoxycholic acid in a proof of concept study. NGM-282 led to a significant drop in alkaline phosphatase compared to placebo. Further testing is needed, but this is the first biologic for PBC with positive results.

PPAR Alpha, Delta Agonists

Another nuclear hormone receptor therapeutic target in PBC is PPARs, or Peroxisome Proliferator Activated Receptors, both the alpha and the delta. Fibrates such as fenofibrate and bezafibrate activate PPAR- α . This leads to activation several genes that decrease fatty acid metabolism and transport. PPAR alpha also downregulates transcription of COX-2, IL-1, IL-6,

and tumor necrosis factor (TNF), thus decreasing inflammation. Importantly, fibrates also increase expression of MDR-3 mRNA, which increases secretion of biliary phospholipids which protects the biliary epithelium from bile salt damage in cholestasis by forming micelles with the hydrophobic bile acids. Understanding these mechanisms led to the experimental use of fibrates in PBC- first in Japan, then Europe, and most recently in the US.

A pilot study of 20 patients in the US showed that the addition of fenofibrate to ursodiol non-responders resulted in a 50 % reduction in ALP. Fibrates also have a known risk of hepatotoxicity. 20% of patients will develop increased transaminases, with 3-5% reaching 3 time the upper limit of normal. There are also reports of acute liver failure due to fibrates, so the FDA has issued a warning not to use this drug off label in PBC until more data are gathered.

Bezafibrate is not available in the US, but the French just completed a phase 3 trial of bezafibrate in ursodeoxycholic acid (UDCA) non-responders. In that trial, 100 patients with inadequate biochemical response to UDCA were randomized 1:1 to a 2-year treatment period with either bezafibrate 400 mg/d or placebo in combination with UDCA 13-15 mg/kg/d. Alkaline phosphatase normalization occurred in 67% and 0% of patients, respectively ($p < 0.0001$). They also saw significant improvements in liver stiffness and (unlike OCA), significant improvements in pruritus. Enthusiasm for fibrates as adjunctive therapy to ursodeoxycholic acid has really taken off in Europe and Japan. So far, hepatotoxicity has not been an issue.

PPAR delta is a nuclear hormone receptor involved in cell differentiation and lipid accumulation. It was originally targeted for the treatment of hyperlipidemia, although agonists were found to also improve cholestasis. In a double blind randomized clinical trial, a selective PPAR delta agonist was compared to placebo in PBC ursodeoxycholic acid non-responders. Significant reductions in alkaline phosphatase were seen compared to placebo. Most patients completely normalized their alkaline phosphatase within 8 weeks. Further studies are needed to assess the benefit on clinical outcomes.

Liver Transplantation

Liver transplantation is the definitive salvage therapy for PBC. Only 3% of liver transplants are for patients with PBC, and that number is dropping by about 7 persons per year. Recurrent PBC does occur in 20-40%, but due to the slow progression, a second transplant is rarely needed.

Fatigue

The fatigue of PBC can be quite debilitating, but no therapy has been shown to be effective in clinical trials. At first modafinil, the drug used for narcolepsy, was considered an option based on small case series, but then a randomized, controlled trial showed that it had no significant effect on any of the validated measures of fatigue or quality of life in PBC patients.

Interestingly, there is epidemiological data that coffee consumption at a dose of 2 cups a day reduces progression of hepatic fibrosis in many chronic liver diseases; so without a good therapeutic option for the fatigue, we often recommend drinking coffee.

Pruritus

In contrast to fatigue, several medications have been tested and shown to be helpful at improving pruritus in patients with PBC. Bile acid binding resins, such as cholestyramine, are the first line treatment. Rifampin, and phenobarbital in children, are second line. There are a number of third line treatments: opioid blockers, sertraline, UV light, nasobiliary drainage, and plasmapheresis. Lack of understanding of the etiology of pruritus has hampered our therapeutic approach. Retained bile acids have often been suspected but not proved to be the cause. There are ongoing trial of inhibitors of bile salt resorption from the ileum. Fibrates have also shown early promise for improving itching. The mechanism is uncertain, although their benefit correlates with reduction in serum phospholipids.

Two recent trials of inhibitors of bile acid reabsorption for the treatment of pruritus in PBC have been completed. In one, pruritus was equally improved by both study drug and placebo. In the other, the study drug was significantly better than placebo.

In Summary,

- PBC is now called primary biliary cholangitis because the minority of modern-era patients actually have cirrhosis
- The etiology is still unclear, potentially molecular mimicry, xenobiotics, and/or loss of bicarbonate umbrella.
- Primary therapy is with ursodeoxycholic acid 13-15mg/kg.
- Consider adjuvant therapy if alkaline phosphatase does not drop to 1.5-2X ULN
- Obeticholic acid (5-10mg) is FDA approved for adjuvant therapy and many other possibilities are in development (FXR agonists, PPAR agonists)
- Liver transplant is a very effective salvage therapy, but rarely needed

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(1-17)

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