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Geoepidemiology of Primary Biliary Cholangitis: Lessons from Switzerland

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Abstract No data on primary biliary cholangitis (PBC) are available in Switzerland. We established a national patient cohort to obtain information on PBC phenotypes and disease course in Switzerland. Local databases in all university hospitals and in two large secondary centers were searched for case finding. In addition, all primary care physicians, gastroenterologists, rheumatologists, and dermatologists were invited to contribute patients from their own medical records. PBC diagnosis was centrally reviewed. Five hundred one PBC patients were identified, 474 were included in data analysis, and

Key Points
- This is a cross-sectional study on PBC in Switzerland, recruiting about one third of the Swiss PBC population by searching the database in tertiary and secondary centers and by a survey among physicians outside centers.
- Four hundred forty-seven patients were included in data analysis, median age at diagnosis was 53 years, 84% were women, and 86% were anti-mitochondrial antibody positive. Median follow up was 5.4 years. Ten-year transplant-free median survival was 85%.
- One quarter of patients and one half of male patients had splenomegaly at diagnosis, suggesting a diagnostic delay.
- We were able to externally validate of the UK-PBC risk score and Globe score.
- Patients included in this study are currently being enrolled in a prospective nationwide registry with biobank, taking advantage of the collaboration network generated by this study.

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449 of them were enrolled by tertiary centers. The catchment area accounts for approximately one third of the Swiss population or approximately 2.8 million inhabitants. The median age at diagnosis was 53 years, 84% were women, and 86% were anti-mitochondrial antibody positive. The median follow-up was 5.4 years, 12.6% experienced a liver-related endpoint. Splenomegaly was present at diagnosis in one quarter of patients and in half of male patients. Approximately one third were non-responders to ursodeoxycholic acid (UDCA). The median transplant-free survival at 10 years was 85%. The following variables were independently associated with poor outcome: low platelet count at baseline (HR = 0.99, $p < 0.0001$), elevated alkaline phosphatase at baseline (HR = 1.36, $p < 0.0001$), elevated bilirubin at baseline (HR = 1.11, $p = 0.001$), and elevated alanine aminotransaminase (HR = 1.35, $p = 0.04$) after 12 months of UDCA therapy. The AUROC for the *UK-PBC risk score* at 5, 10, and 15 years was 0.82. The AUROC for the *Globe score* at 5, 10, and 15 years was 0.77. Patients included in this study are currently being enrolled in a prospective nationwide registry with biobank, taking advantage of the collaboration network generated by this study. Our study provides the first snapshot of PBC in Switzerland, describing a diagnostic delay with one quarter of patients diagnosed when already in the cirrhotic stage. We were also able to externally validate the UK-PBC risk score and the Globe score. The ongoing nationwide prospective registry will be fundamental to improve disease awareness and interdisciplinary collaborations and will serve as a platform for clinical and translational research.

Trial registration number: clinicaltrials.gov: NCT02846896; SNCTP000001870

Keywords Primary biliary cholangitis · Switzerland · Cohort study · UK-PBC risk score · Globe score · Prospective registry

Abbreviations

PBC	Primary biliary cholangitis
UDCA	Ursodeoxycholic acid
AMA	Anti-mitochondrial antibodies
ANA	Anti-nuclear-antibodies
LT	Liver transplant
IgG	Immunoglobulin G
AIH	Autoimmune hepatitis
CRF	Case record form
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
AUROC	Area under the receiver operating characteristic curve
ULN	Upper limit of normal
CI	Confidence interval
HR	Hazard ratio

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Introduction

Primary biliary cholangitis (PBC) is a cholestatic liver disease, characterized by chronic inflammation and destruction of small bile ducts [1]. It is widely accepted that the disease is immune-mediated, as suggested by the presence of highly specific autoantibodies and autoreactive T cells [2]. PBC affects mainly women, with a female to male ratio of 9:1 [3]. First-line treatment is ursodeoxycholic acid (UDCA), and therapy end point is reduction of the biochemical cholestasis, defined according to international criteria [1, 3]. Second-line therapy for patients non-responding or intolerant to UDCA is obeticolic acid [1]. The course of the disease is highly variable: patients with a biochemical response to UDCA, in the absence of advanced disease, have a normal life expectancy, while patients not responding to UDCA face an increased risk of death or liver transplant [1]. Recently, continuous models to predict risk of death or liver transplant (LT) have been developed from large cohorts of PBC patients [4–6].

PBC diagnosis is based on the presence of two or more of the following criteria: cholestatic liver biochemistry, compatible or diagnostic liver histology, and positivity for anti-mitochondrial antibodies (AMA) at a titer > 1:40 [1, 7]. Sensitivity and specificity of the combination of biochemistry and serology is 95% [8]. AMA are found in 90–95% of PBC patients [1, 7], and are considered as the most disease-specific autoantibodies in human autoimmune diseases [9]. Identification of the target antigens of the M2 components, mainly the E2 subunit of the pyruvate dehydrogenase complex, has allowed the development of enzymatic tests for AMA detection, which are highly sensitive and specific [10]. A subgroup of anti-nuclear antibodies (ANA) represent additional disease-specific autoantibodies: ANA with a rim-like indirect immunofluorescence staining pattern on HEp-2 cells are mainly directed against the gp210 protein and are found in 10–40% of PBC patients. ANA showing a multiple-dots staining pattern on HEp-2 cells are mainly directed against the sp100 protein and are found in 20–40% of PBC patients. Both are associated with a more severe disease

course and are helpful diagnostic markers in AMA-negative PBC patients [11–13].

Due to simple, accurate, and widely available diagnostic tests, the epidemiology of this complex autoimmune disease, in theory, is rather easy to investigate. Better understanding of the epidemiology is undoubtedly a powerful tool for improving our understanding of the disease, its population-impact, and risk factors [14]. In particular, epidemiological studies have provided evidence of local clustering of PBC cases, suggesting the presence of environmental risk factors [15–17]. In addition, large cohorts of PBC patients allowed the identification of risk factors associated with a more severe disease course [18].

Epidemiological data from different European, American, and Asian-Pacific countries report a wide difference in disease frequency [19], ranging from an incidence of 0.9 to 5.8 per 100′000 inhabitants and a prevalence of 1.9 to 40.2 [20]. It was previously assumed that PBC frequency has a north-south gradient, but a recent well-designed study from Greece [21] reported a high prevalence, highlighting that the case-finding strategy is crucial to obtain reliable epidemiological data. Differences in reported frequency in epidemiological studies may be explained by the case-finding and ascertainment methods, the most reliable being probably based on AMA positivity [22]. However, even this method is limited by the need of laboratory expertise, presence of AMA-negative PBC cases, and the fact that AMA may be found in absence of liver disease [23, 24].

Data on PBC in Switzerland are lacking. The aim of our study is to describe the health burden, the clinical phenotype, and the disease course of PBC in Switzerland and put these data in the context of current epidemiological studies.

Patients and Methods

Study Area, Population, and Period We conducted a cross-sectional, observational study of patients with PBC living in Switzerland. Between January 1, 2005 and December 31, 2015, we searched for PBC cases throughout Switzerland. The study cohort included patients who had undergone LT and non-transplanted patients. Detailed clinical information was collected.

Case Finding Case-finding was performed by searching local databases in all five Swiss University hospitals, and in two large secondary centers. Furthermore, in an attempt to include patients not seen in centers, a survey among all Swiss gastroenterologists, primary care physicians, rheumatologists, and dermatologists was carried out. The latter categories were involved since up to 60% of PBC patients have extra-hepatic autoimmune diseases [25–27]. A total of 6120 physicians were sent a questionnaire by e-mail (Supplementary Table 1). A small monetary incentive was offered per included patient.

Data Collection

Detailed clinical information was collected as follows: date of diagnosis; sex; age at diagnosis; linguistic region of residency at diagnosis; ethnicity; height and weight at diagnosis; first and last visits; LT status; laboratory investigations at diagnosis including status of AMA, ANA, PBC-specific ANA, liver biochemistry (2 sets of values at least 12 months apart), platelet count, albumin, and immunoglobulin G (IgG) levels; spleen size at diagnosis; hepatic transient elastography at diagnosis; UDCA therapy status and dose; histologically proven and treated autoimmune hepatitis (AIH) overlap; concomitant autoimmune muco-cutaneous disease; concomitant hepatic disease; last alive date; and date and cause of death.

The case report form (CRF) was completed by the local investigators, or suitably trained students and research nurses.

Case-Ascertainment All completed CRFs were centrally evaluated for ascertainment of PBC diagnosis. The diagnosis of PBC was based on established diagnostic criteria. PBC-AIH overlap syndrome was defined as a combination of PBC, a liver biopsy showing moderate or severe periportal or periseptal lymphocytic piecemeal necrosis, and treatment for AIH.

Patients who were AMA-negative by immunofluorescence, but with antibodies against the major M2 components by enzymatic testing, and/or patients positive for PBC-specific ANA, in absence of a cholestatic biochemical pattern and of a histological evaluation, were included in the study, since these are considered specific markers of disease [10, 12, 13, 28].

The project was endorsed by the Swiss Association for the Study of the Liver. The study protocol conforms to the ethical guidelines of the Declaration of Helsinki of 1975 and was approved by all the Swiss Ethics Committees (Comitato etico cantonale del Canton Ticino, Ethikkommission Nordwest- und Zentralschweiz EKNZ, Kantonale Ethikkommission für die Forschung Bern, Commission cantonale d'éthique de la recherche CCER Genève, Ethikkommission St. Gallen EKSG, Kantonale Ethikkommission Zürich, Commission cantonale d'éthique de la recherche sur l'être humain CER-VD Vaud). Informed consent was obtained from each patient included in the study.

Statistical Analysis Analyses were performed using the statistical analysis software Stata statistical software 13.0 (StataCorp, College Station, TX, USA). Data were presented as median and range. Where data were distributed normally, comparisons were made between groups using unpaired *t* tests. Where data were distributed non-normally,

comparisons were made by the Mann–Whitney *U* test. Differences in proportions were determined using the chi-square test. Rates of survival were estimated by the Kaplan–Meier method. We performed univariate analysis of the following variables using Cox's proportional hazards regression model: age at diagnosis, gender, body mass index, date of LT, alkaline phosphatase (ALP), alanine aminotransferase (ALT), bilirubin levels at diagnosis and after 12 months of UDCA, albumin and platelet count at diagnosis, splenomegaly, AMA and ANA status, and IgG level. The variables transient elastography, anti-sp100, and anti-gp210 were not included in the model for the high rate of missing values. Variables that were statistically significant at $p < 0.05$ in univariate analysis were included in a multivariable Cox model. Liver-related end points were defined as LT for PBC and liver-related death. Those dying of non-liver-related causes were censored at the time of death. Patient data at baseline and after 12 months were used to calculate the UK-PBC risk score [6] and the Globe score [5]. To assess discrimination, we calculated the area under the receiver operating characteristic curve (AUROC) for each risk score at specific time-frames (5, 10 and 15 years).

Literature Search A systematic search of literature was performed in Medline (search last conducted October 1, 2017). The search strategy was as follows: (primary biliary cirrhosis) OR (primary biliary cholangitis) OR (PBC) AND (epidemiology) OR (incidence) OR (prevalence). References published after 2007 were included for further screening. Two authors (JM and BTP) screened the title and abstract of the articles returned by the search. As in the publication by Boonstra et al. we included population-based epidemiological studies which report the incidence of PBC in a geographical area of at least 100,000 inhabitants and performed an analogous data extraction recording the study area and period, the number of patients and number of inhabitants, incidence and prevalence per 100,000 inhabitants, method of case-finding, and case-ascertainment as well as the male to female ratio. Articles in all languages were considered.

Results

Data of 501 patients were collected, 27 were excluded from data analysis because they did not fulfill the inclusion criteria, mostly due to lack of data allowing a review of the diagnosis (Fig. 1).

Patients Enrolled by Secondary and Tertiary Centers

Four hundred seventy-six patients were enrolled by the secondary and tertiary care centers, accounting for the vast majority of recruited patients. Twenty-six of them were excluded from data analysis because available data were insufficient to review the diagnosis of PBC.

Survey Conducted Among Physicians Outside Tertiary Centers

The results of the survey among physicians outside centers are shown in Table 1. Only 8% of the primary care physicians, 16% of the gastroenterologists, 7% of the rheumatologists, and 6% of the dermatologists answered the survey; no patient was enrolled by the latter two categories.

A total number of 88 subjects diagnosed with PBC were identified across Switzerland using this case-finding system. Twenty-five of them were included in the study, 14 by primary care physicians, and 11 by gastroenterologists.

Patient Characteristics

The demographics, clinical, biochemical, and serological characteristics of the study population are outlined in Table 2. Eighty-five percent were female; median age at diagnosis was 53 years. Twenty percent of the patients were diagnosed before 2000, 40% were diagnosed in the first decade of this century, and 40% were diagnosed in the following 5 years, pointing to a more frequent PBC diagnosis in more recent years in Switzerland. This observation is consistent with data from other countries [20, 29]. Only 11% of patients were recruited from the French-speaking part of the country, which accounts for 22.6% of the Swiss population: in this part of Switzerland, PBC patients are mostly followed up by physicians outside centers, and therefore were missed by our study. According to the local clinical practice, only severe cases are referred to tertiary centers. Indeed, 15% of patients recruited from the French-speaking part of Switzerland underwent LT, compared to 6 and 10% of patients recruited from the Italian and German parts, respectively.

Four hundred forty-six patients (94%) were treated with UDCA. Nineteen (6%) were not treated and considered by the clinicians as pre-clinical PBC: indeed, the majority had ALP < 2 × ULN, normal albumin at diagnosis, median transient elastography at diagnosis < 8.1 kPa, and normal platelet count at diagnosis. Approximately 29% of the patients were non-responders to UDCA therapy, according to the Paris I criteria [30], 35% according to the Paris II criteria [31], and 27% according to the Toronto criterion [32].

Data on PBC-specific ANAs were available only in a small proportion of patients (< 10%), indicating that this test is not routinely performed in PBC patients in Switzerland.

Seventy patients (17%) were diagnosed with a biopsy-proven PBC/AIH overlap syndrome, and were on immunosuppressive treatment. We found that 33 patients (7%) had a concomitant autoimmune muco-cutaneous disease

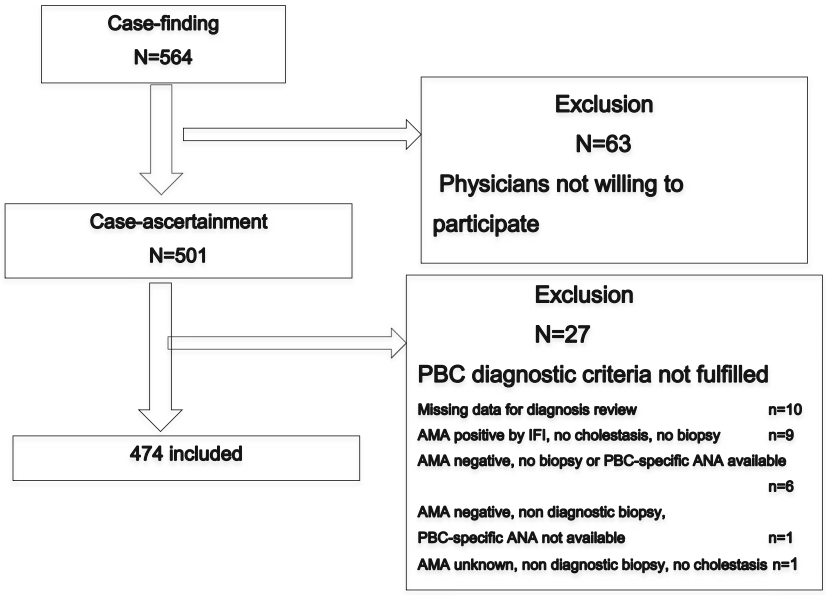

Fig. 1 Flowchart of patient inclusion. *PBC* primary biliary cholangitis, *AMA* anti-mitochondrial antibodies, *IFI* indirect immunofluorescence, *ANA* anti-nuclear antibodies

with Sjögren's syndrome having the highest prevalence (2.4%) (Table 3).

Survival

The median follow-up was 5.5 years (IQR, 2–12). During follow-up, 60 patients (12.6%) suffered an event, 52 patients (10.9%) underwent LT, and 8 patients (1.7%) died from liver-related causes. The overall event-free survival rate was 96% at 5 years, 85% at 10 years, and 75% at 15 years.

Univariate and multivariate Cox proportional hazard regression analyses were performed to identify variables associated with long-term outcome. After non-automatic backward selection, the best-fitting Cox model included five variables: platelet at baseline (hazard ratio (HR) = 0.99, CI = 0.98–0.99, $p < 0.0001$), and bilirubin (HR = 1.11, CI = 1.04–1.18, $p = 0.001$), ALT (HR = 1.35, CI = 1.00–1.80, $p = 0.04$) and ALP after 12 months of therapy with UDCA (HR = 1.36, CI = 1.15–1.62, $p < 0.0001$). We then assessed the performance of the existing, continuous predicting models of survival: the AUROC for the UK-PBC risk score at 5, 10 and 15 years was 0.82. The AUROC for the Globe score at 5, 10, and 15 years was 0.77.

Current PBC Geoepidemiology

The search returned 2084 references. Of these, 11 were included based on the abovementioned criteria (Table 4). Of the 11 studies, two report the incidence rates in Asia (Hong Kong and Korea), four in Europe (Denmark/Lombardia, Greece, England, Netherlands), and one each in the USA, Iceland, Israel, and New Zealand. Seven studies relied primarily on ICD-9/10 medical record databases for case finding; seven studies included AMA for case ascertainment. The highest incidence for PBC was described in England with 5.3 per 100,000 while the lowest incidences were found in Hong

Table 1 Number of identified and recruited patients by nationwide survey among physicians outside tertiary centers

	Primary care physicians	Gastroenterologists	Rheumatologists	Dermatologists	Total
Physicians involved	5004	295	372	449	6120
Replies (%)	406 (8)	48 (16)	25 (7)	29 (6)	508 (8)
Physicians having PBC patients	77	36	12	1	126
Still alive patients	116	157	12	1	286
Deceased patients	25	7	0	0	32
Total patients	141	164	12	1	318
Total number of enrolled patients	14	11	0	0	25

Table 2 Demographics (2a), clinical and laboratory characteristics (2b) of the study population

Parameter		Data available (%)
a		
Date of diagnosis–no. (%)		*N* = 473 (99)
Before 2000	93 (20)	
1.1.2001–31.12.2009	188 (40)	
1.1.2010–31.12.2015	190 (40)	
Female (%)	401 (85)	*N* = 474 (100)
Age at diagnosis–years		
Median (IQR)	53 (45–63)	*N* = 470 (99)
Male (IQR)	57 (48–64)	*N* = 73 (100)
Female (IQR)	52 (44–62)	*N* = 397 (99)
Region of residency–no. (%)		
German speaking	344 (73)	*N* = 463 (97)
French speaking	53 (11)	
Italian speaking	66 (14)	
Ethnicity–no. (%)		
White	443 (93.5)	*N* = 467 (98)
Asian	10 (2.1)	
Hispanic	8 (1.7)	
Black	1 (0.2)	
Body mass index–kg/m^2		
Median (IQR)	25.0 (22.2–29.08)	*N* = 352 (74)
Liver transplant–no. (%)	52 (10.9)	*N* = 473 (99)
Male	10 (13.5)	*N* = 73 (100)
Female	42 (10.5)	*N* = 400 (99)
b		
Alkaline phosphatase level at diagnosis, U/l		
Median (IQR)	1.6 × ULN (1.0–2.9)	*N* = 454 (96)
> 1.67 × ULN (%)	218 (48)	
> ULN (%)	339 (75)	
Alkaline phosphatase level at 1 year UDCA, U/l		
Median (IQR)	1.1 × ULN (0.8–1.7)	*N* = 416 (88)
> 1.67 × ULN (%)	114 (27)	
> ULN (%)	234 (56)	
Toronto non response overall (male-female)	27% (32–27%)	(*N* = 63–353)
Total bilirubin level at diagnosis, μmol/l		
Median (IQR)	0.5 × ULN (0.4–0.9)	
> ULN (%)	82 (19)	*N* = 429 (90)
Total bilirubin level after 1 year UDCA μmol/l		
Median (IQR)	0.5 × ULN (0.4–0.89)	
> ULN (%)	58 (15)	*N* = 391 (82)
ALT level at diagnosis, U/l		
Median (IQR)	1.5 (1.0–2.7)	*N* = 450 (95)
ALT level after 1 year UDCA		
Median (IQR)	0.8 (0.5–1.3)	*N* = 404 (85)
Platelet count at diagnosis, G/l		
Median (IQR)	242 (190–300)	*N* = 424 (89)
Male	220 (154–266)	*N* = 62 (85)
Female	246 (194–305)	*N* = 363 (90)

Table 2 (continued)

Parameter		Data available (%)
Albumin at diagnosis, g/l		
Median (IQR)	38 (35–42)	*N* = 393 (83)
Male	38 (34–42)	*N* = 55 (75)
Female	38 (36–42)	*N* = 329 (82)
Total IgG at diagnosis, g/l		
Median (IQR)	14 (11–17)	*N* = 312 (66)
Spleen size at diagnosis by ultrasound, cm > 12 (%)	99 (26)	*N* = 382 (81)
Male	27 (45)	*N* = 60 (82)
Female	72 (22)	*N* = 322 (80)
Transient elastography at diagnosis, kPa		
Median (IQR)	6.3 (4.6–9-9)	*N* = 296 (62)
Male	7.6 (4.6–11.7)	*N* = 44 (60)
Female	6.1 (4.5–9.35)	*N* = 252 (63)
Number of UDCA treated subjects (%)	446 (94)	*N* = 465 (98)
UDCA dose, mg/kg/day	13.6 (11.4–15.0)	*N* = 375 (79)
AMA positive (%)	415 (88)	*N* = 458 (97)
ANA ≥ 1:80 (%)	261 (63)	*N* = 412 (87)
Anti-sp100 positive (%)	38 (56)	*N* = 67 (14)
Anti-gp210 positive (%)	19 (39)	*N* = 49 (10)

Table 3 Concomitant autoimmune and hepatic diseases conditions

Concomitant conditions	*N* (%)	Data available
AIH-overlap (biopsy-proven and treated)	70 (17)	*N* = 407 (86)
Concomitant autoimmune muco-cutaneous disease (%)	33* (7)	*N* = 467 (98)
Sjögren's syndrome	11 (2.4)	
Systemic sclerosis	5 (1.1)	
Systemic lupus erythematosus	5 (1.1)	
Vitiligo	3 (0.6)	
Pemphigus	2 (0.4)	
Cutaneous lupus erythematosus	2 (0.4)	
Dermatitis herpetiformis Duhring	1 (0.2)	
Psoriasis	1 (0.2)	
Limited scleroderma	1 (0.2)	
Raynaud's syndrome	1 (0.2)	
Urticaria vasculitis	1 (0.2)	
Other	1 (0.2)	
Concomitant hepatic disease (%)	43 (9)	*N* = 472 (99)
Non alcoholic fatty liver disease	15 (3)	
HCV	9 (2)	
HBV	3 (0.6)	
Alcoholic liver disease	2 (0.4)	
Overlap PSC	2 (0.4)	
Hepatocellular carcinoma	2 (0.4)	
other	7 (1.5)	

* One patient had two concomitant skin diseases

Kong and New Zealand with 0.8 per 100,000. The highest prevalence of 38.3 was reported for Iceland; this study took advantage of the unique features of the country, being an island with AMA tested in one single laboratory.

Discussion

The present study provides the first assessment of the health burden due to PBC in Switzerland. We estimate that we were able to identify approximately one third of the Swiss PBC population, assuming a prevalence of 15–20 cases per 100,000 inhabitants. This estimate is corroborated by the high number of patients recruited from the Italian-speaking part of the country, were the coordinator center is located, and where the case-finding strategy is likely to be more effective due to the limited extension of the region, and close contact between primary care physicians and hepatologists, leading to the high referral rate of patients with suspected liver disease. In this region, accounting for 1/25 of the Swiss population (8.4 million inhabitants), 66 patients were recruited: assuming a similar prevalence of PBC across Switzerland, the estimated total number of PBC patients in the country is 1650, corresponding approximately to a prevalence of 20:100,000 inhabitants.

In order to reduce the selection bias due to recruitment from secondary and tertiary centers, we tried to identify patients followed in primary care, and in the private healthcare system.

Table 4 High-quality epidemiological papers on primary biliary cholangitis published after 2007

Study, country	Period	Number of patients	Population	Case-finding	Case ascertainment	Incidence per 100,000 inhabitants (95% CI)	Prevalence per 100,000 inhabitants (95% CI)	Male (%)
Cheung et al. [33], Hong Kong	2000–2015	1016	7,300,000	ICD-9 CDARS	I, IIb, III	0.8	8.2	21
Kanth et al. [34], USA	1992–2011	79	409,670	ICD-9 MESA	I, IIb, III	4.9		5
Lleo et.al [35], Lombardia and Denmark	2000–2009	2970 + 722	9,700,000 + 5,500,000	ICD-9/ICD-8 and 10		1.7 + 1.1	16	30 + 23
Kim et al. [36], South Korea	2009–2013	2824	38,424,000	ICD-10, rare intractable diseases registry	I, IIb, III	0.85	4.75	13.9
Koulentaki et al. [21], Crete Greece	1990–2010	245	600,000	Hospital database	I, IIb, III	2.08	36.5	12
McNally et al. [37], England	1987–2003	982	2,050,000	Personal registry physicians, hospital admission data, ICD-9, ICD-10, AMA, death certificates		5.35		9.7
Boonstra et al. [29], The Netherlands	2000–2008	992	5,855,000	Hospital database	I, IIa, IIb, III	1.1	13.2	10
Delgado et al. [38], Israel	1990–2010	138	1,076,600	Hospital database, ICD-9, physician records	I, IIa, IIb, III	1	25.5	5.1
Baldusdottir et al. [39], Iceland	1991–2010	168	317,630	AMA, ICD-9	I, IIb, III	2.5	38.3	18
Ngu et al. [40], New Zealand	2008	71	494,170	Hospital database, ICD-9, physician records		0.8	9.9	8

CDARS Clinical Data Analysis and Reporting System, *MESA* Marshfield Epidemiological Study Area, *AMA* anti-mitochondrial antibodies, *I* AMA, *IIa* elevated serum alkaline phosphatase ≥ 6 months, *IIb* cholestatic liver parameters, *III* liver biopsy, *IV* elevated IgM

To do so, we carried out a web-based survey research method. Unfortunately, primary care physicians proved to be a group with low survey response rates. Our survey results provide an example for the challenges faced when conducting healthcare research where response rates to surveys are often low. Also, findings from this study suggest that small monetary incentives such as the ones adopted in our study for each enrolled individual may not be the most appropriate way to increase response rates.

The epidemiology and geoepidemiology of PBC have been examined in numerous studies over the last 15 years. Truly population-based studies are lacking, and thus data come from analyses of probability samples of reference populations. Boonstra et al. did compile a comprehensive review of large epidemiological studies on PBC in 2012 [20]. In their review, studies that reported incidence and/or prevalence for at least 100,000 adult inhabitants in a defined geographical area were considered. For the years between 1972 and 2007, there were 23 papers describing the epidemiology of PBC that were reviewed. The majority of the studies were conducted in Western countries. Only after 2005 epidemiological data from outside the USA and Europe were published with papers from Israel [38], Brunei [41], Singapore, and China [42–44].

We have now extended the review of the epidemiological literature to 2017 and have included 11 new epidemiological studies (Table 4).

The cohort of PBC patients described in the present study is consistent with the cohorts reported in the literature with a female to male ratio of 9:1, a median age at diagnosis of 53 years, and a rate of AMA positive subjects of 88%, the majority treated with UDCA (94%) [5, 18, 29]. Some epidemiological studies reported a lower female to male ratio [35, 45]: they are based on administrative data-bases, suggesting that this case-finding method may lead to inclusion of pre-clinical cases, which are more common in males compared to clinical cases [24].

Approximately 20% of the female patients and 50% of the male patients had splenomegaly at diagnosis. This indicates that the diagnosis is often made after a longstanding disease course, particularly in men. This is true for those diagnosed before and after 2000 (data not shown), indicating that even in recent years the diagnosis has been made at a late disease stage in a high proportion of the patients. The proportion of patients who underwent LT is similar to recently described national cohorts [18] (e.g., 10% in our cohort, 8% in the UK cohort).

When we explored the gender impact on PBC, important differences were seen between men and women. Men with PBC presented at an older age, and more frequently have splenomegaly at diagnosis. The rate of transplanted patients is higher in men than in women. However, the UDCA response rate according to different criteria was not different between females and males. The lack of difference between men and women in these parameters is probably due to the small number (73 subjects) of male patients included in our cohort.

PBC-specific ANAs were positive in a high proportion of patients (Table 2). This might in part be explained by the characteristics of our cohort with a high rate of patients with a more severe disease course; indeed, these autoantibodies are associated with a higher risk of disease progression to jaundice and hepatic failure [11, 46].

We found 33 patients (7%) that presented with a concomitant autoimmune muco-cutaneous disease. The frequency of Sjögren's syndrome was low (2.4%), compared to that found by accurate assessment, being up to 34% [25], probably due to underdiagnosis/underreporting. Vitiligo and psoriasis, which are both frequent conditions, affecting 1 and 2% of the population in central Europe, respectively, were found only in 0.6 and 0.2% in our cohort, again most probably due to underreporting by non-dermatologists. We identified a few cases of unexpected concomitant autoimmune diseases, including pemphigus and dermatitis herpetiformis Duhring, which are both rare conditions and this finding could be due to chance.

Seventy patients (17%) were diagnosed with a biopsy-proven PBC/AIH overlap syndrome, and were on immunosuppressive treatment, the proportion being higher than the commonly reported frequency of 10% in PBC patients [47, 48]. This is possibly related to a referral bias.

We looked at baseline variables associated with UDCA response but we could not identify neither sex nor age at diagnosis as a negative predictor of response [18].

In the current study, we confirmed that the recently developed long-term prognostic models of PBC, i.e., UK-PBC risk score and Globe score, are accurate in predicting liver-related (UK-PBC risk score) and overall survival (Globe score), with AUROCs of 0.82 and 0.77, respectively. Efforts should be made in the future to explore whether there are additional biomarkers, e.g., PBC-specific ANAs, non-invasive markers, or pathological features, among others, that can improve the accuracy of such scoring systems.

Our case-finding strategy is limited by the characteristics of the Swiss Healthcare System. Switzerland has many unconnected (private and public) laboratories: this fact prevents the use of the case-finding strategy proposed by Metcalf and James [22], based on identification of AMA-positive blood samples. The same is true for a case-finding strategy including liver pathology reports.

Still the incidence and prevalence found in Switzerland is in concordance with the data from other recent epidemiological studies. These studies from 2007 until 2017 further confirm the previously reported wide range of prevalence and incidence for PBC. The studies from Asia and New Zealand [33, 49] report the lowest incidences while England, USA, Greece, and Iceland report the highest incidence rates [21, 34, 37, 39]. Of note, studies reporting the highest disease frequency include AMA in their case-finding strategy, which, as mentioned above, is not possible in our country. Like in Switzerland, several studies report an increase in incidence

over the observed period, which may in part be due to improved diagnostic tools and awareness as well as registration of cases. It is interesting to notice that the publication by Boonstra et al. identified 23 articles describing the incidence and prevalence of PBC between 1972 and 2007 but there are an additional 11 articles for the years 2007 to 2017. This seems to indicate that the interest in and awareness of PBC as an important health issue has increased drastically.

In order to assess the true incidence and prevalence of PBC, repeated screening of a well-defined population would be the best methodology, allowing inclusion of undiagnosed cases. It could be argued that such a screening in PBC is facilitated by the presence of serum hallmarks, AMA and PBC-specific ANA. However, it is well known that these markers can predate liver disease by decades [22, 23], thus hampering their use as a screening tool. In addition, use of correct laboratory methodologies is key to achieve high sensitivity and specificity [24]. Given these limitations, we believe that probably the best way to study PBC epidemiology is based on medical awareness of the condition, and thus education of primary care doctors. Diagnosed patients, including those positive for AMA and/or PBC-specific ANA without liver disease, should be included in prospective registries with biobank, in order to generate high-quality, standardized data. We took advantage of the collaboration networks generated by the present study to set-up such a registry in our country (NCT03146910). As mentioned before, our case-finding strategy was very effective in the region where the coordinator center is located, supporting our hypothesis that disease awareness and collaborations are key to improve diagnosis and treatment of PBC patients.

In conclusion, this study provides a snapshot of PBC in Switzerland between 2005 and 2015. We described a diagnostic delay in PBC, particularly in male patients. This should be reflected in the planning and delivery of clinical care. Moreover, we validated the UK-PBC risk score and the Globe score as accurate tools in predicting liver-related and overall survival, respectively, in Swiss patients with PBC. The running prospective registry will be of fundamental importance to both improve patient care and serve as a platform for clinical and translational research.

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Compliance with Ethical Standards The study protocol conforms to the ethical guidelines of the Declaration of Helsinki of 1975 and was approved by all the Swiss Ethics Committees (Comitato etico cantonale del Canton Ticino, Ethikkommission Nordwest- und Zentralschweiz EKNZ, Kantonale Ethikkommission für die Forschung Bern, Commission cantonale d'éthique de la recherche CCER Genève, Ethikkommission St. Gallen EKSG, Kantonale Ethikkommission Zürich, Commission cantonale d'éthique de la recherche sur l'être humain CER-VD Vaud).

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GST received consulting fees and support for an educational event from Intercept Switzerland.

MF received support for organization and attendance of educational events from Intercept Switzerland.

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References

1. European Association for the Study of the Liver (2017) EASL Clinical Practice Guidelines: The diagnosis and management of patients with primary biliary cholangitis. J Hepatol 67:145–172. https://doi.org/10.1016/j.jhep.2017.03.022
2. Selmi C, Gershwin ME (2010) The etiology mystery in primary biliary cirrhosis. Dig Dis Basel Switz 28(1):105–115. https://doi.org/10.1159/000282073
3. Carey EJ, Ali AH, Lindor KD (2015) Primary biliary cirrhosis. Lancet Lond Engl 386(10003):1565–1575. https://doi.org/10.1016/S0140-6736(15)00154-3
4. Lammers WJ, van Buuren HR, Hirschfield GM et al (2014) Levels of alkaline phosphatase and bilirubin are surrogate end points of outcomes of patients with primary biliary cirrhosis: an international follow-up study. Gastroenterology 147:1338–1349.e5; quiz e15. https://doi.org/10.1053/j.gastro.2014.08.029
5. Lammers WJ, Hirschfield GM, Corpechot C, Nevens F, Lindor KD, Janssen HL, Floreani A, Ponsioen CY, Mayo MJ, Invernizzi P, Battezzati PM, Parés A, Burroughs AK, Mason AL, Kowdley KV, Kumagi T, Harms MH, Trivedi PJ, Poupon R, Cheung A, Lleo A, Caballeria L, Hansen BE, van Buuren HR, Global PBC Study Group (2015) Development and validation of a scoring system to predict outcomes of patients with primary biliary cirrhosis receiving ursodeoxycholic acid therapy. Gastroenterology 149(7):1804–1812.e4. https://doi.org/10.1053/j.gastro.2015.07.061
6. Carbone M, Sharp SJ, Flack S, Paximadas D, Spiess K, Adgey C, Griffiths L, Lim R, Trembling P, Williamson K, Wareham NJ, Aldersley M, Bathgate A, Burroughs AK, Heneghan MA, Neuberger JM, Thorburn D, Hirschfield GM, Cordell HJ, Alexander GJ, Jones DEJ, Sandford RN, Mells GF, and the members of the UK-PBC Consortium (2016) The UK-PBC risk scores: derivation and validation of a scoring system for long-term prediction of end-stage liver disease in primary biliary cholangitis. Hepatol Baltim Md 63(3):930–950. https://doi.org/10.1002/hep.28017
7. Lindor KD, Gershwin ME, Poupon R, Kaplan M, Bergasa NV, Heathcote EJ, American Association for Study of Liver Diseases (2009) Primary biliary cirrhosis. Hepatol Baltim Md 50(1):291–308. https://doi.org/10.1002/hep.22906
8. Oertelt S, Rieger R, Selmi C, Invernizzi P, Ansari AA, Coppel RL, Podda M, Leung PSC, Gershwin ME (2007) A sensitive bead assay for antimitochondrial antibodies: chipping away at AMA-negative primary biliary cirrhosis. Hepatol Baltim Md 45(3):659–665. https://doi.org/10.1002/hep.21583
9. Tanaka A, Leung PSC, Young HA, Gershwin ME (2017) Toward solving the etiological mystery of primary biliary cholangitis. Hepatol Commun 1(4):275–287. https://doi.org/10.1002/hep4.1044
10. Nakamura M (2014) Clinical significance of autoantibodies in primary biliary cirrhosis. Semin Liver Dis 34(03):334–340. https://doi.org/10.1055/s-0034-1383732

11. Invernizzi P, Podda M, Battezzati PM, Crosignani A, Zuin M, Hitchman E, Maggioni M, Meroni PL, Penner E, Wesierska-Gadek J (2001) Autoantibodies against nuclear pore complexes are associated with more active and severe liver disease in primary biliary cirrhosis. J Hepatol 34(3):366–372. https://doi.org/10.1016/S0168-8278(00)00040-4
12. Nakamura M, Kondo H, Mori T, Komori A, Matsuyama M, Ito M, Takii Y, Koyabu M, Yokoyama T, Migita K, Daikoku M, Abiru S, Yatsuhashi H, Takezaki E, Masaki N, Sugi K, Honda K, Adachi H, Nishi H, Watanabe Y, Nakamura Y, Shimada M, Komatsu T, Saito A, Saoshiro T, Harada H, Sodeyama T, Hayashi S, Masumoto A, Sando T, Yamamoto T, Sakai H, Kobayashi M, Muro T, Koga M, Shums Z, Norman GL, Ishibashi H (2007) Anti-gp210 and anti-centromere antibodies are different risk factors for the progression of primary biliary cirrhosis. Hepatol Baltim Md 45(1):118–127. https://doi.org/10.1002/hep.21472
13. Wesierska-Gadek J, Penner E, Battezzati PM, Selmi C, Zuin M, Hitchman E, Worman HJ, Gershwin ME, Podda M, Invernizzi P (2006) Correlation of initial autoantibody profile and clinical outcome in primary biliary cirrhosis. Hepatol Baltim Md 43(5):1135–1144. https://doi.org/10.1002/hep.21172
14. Griffiths L, Dyson JK, Jones DEJ (2014) The new epidemiology of primary biliary cirrhosis. Semin Liver Dis 34(03):318–328. https://doi.org/10.1055/s-0034-1383730
15. Prince MI, Chetwynd A, Diggle P, Jarner M, Metcalf JV, James OF (2001) The geographical distribution of primary biliary cirrhosis in a well-defined cohort. Hepatol Baltim Md 34(6):1083–1088. https://doi.org/10.1053/jhep.2001.29760
16. Ala A, Stanca CM, Bu-Ghanim M, Ahmado I, Branch AD, Schiano TD, Odin JA, Bach N (2006) Increased prevalence of primary biliary cirrhosis near superfund toxic waste sites. Hepatol Baltim Md 43(3):525–531. https://doi.org/10.1002/hep.21076
17. Triger DR (1980) Primary biliary cirrhosis: an epidemiological study. Br Med J 281(6243):772–775. https://doi.org/10.1136/bmj.281.6243.772
18. Carbone M, Mells GF, Pells G et al (2013) Sex and age are determinants of the clinical phenotype of primary biliary cirrhosis and response to ursodeoxycholic acid. Gastroenterology 144:560–569. https://doi.org/10.1053/j.gastro.2012.12.005
19. Zhang H, Carbone M, Lleo A, Invernizzi P (2015) Geoepidemiology, genetic and environmental risk factors for PBC. Dig Dis Basel Switz 33(Suppl 2):94–101. https://doi.org/10.1159/000440754
20. Boonstra K, Beuers U, Ponsioen CY (2012) Epidemiology of primary sclerosing cholangitis and primary biliary cirrhosis: a systematic review. J Hepatol 56(5):1181–1188. https://doi.org/10.1016/j.jhep.2011.10.025
21. Koulentaki M, Mantaka A, Sifaki-Pistolla D, Thalassinos E, Tzanakis N, Kouroumalis E (2014) Geoepidemiology and space-time analysis of primary biliary cirrhosis in Crete, Greece. Liver Int Off J Int Assoc Study Liver 34(7):e200–e207. https://doi.org/10.1111/liv.12479
22. Metcalf JV, Bhopal RS, Gray J et al (1997) Incidence and prevalence of primary biliary cirrhosis in the city of Newcastle upon Tyne, England. Int J Epidemiol 26(4):830–836. https://doi.org/10.1093/ije/26.4.830
23. Dahlqvist G, Gaouar F, Carrat F, Meurisse S, Chazouillères O, Poupon R, Johanet C, Corpechot C, the French network of Immunology Laboratories (2017) Large-scale characterization study of patients with antimitochondrial antibodies but nonestablished primary biliary cholangitis. Hepatol Baltim Md 65(1):152–163. https://doi.org/10.1002/hep.28859
24. Selmi C, Mackay IR, Gershwin ME (2014) Chapter 62—primary biliary cirrhosis. In: Autoimmune Dis. Fifth Ed. Academic Press, Boston, pp 909–924, https://doi.org/10.1016/B978-0-12-384929-8.00062-9
25. Floreani A, Franceschet I, Cazzagon N, Spinazzè A, Buja A, Furlan P, Baldo V, Gershwin ME (2015) Extrahepatic autoimmune conditions associated with primary biliary cirrhosis. Clin Rev Allergy Immunol 48(2-3):192–197. https://doi.org/10.1007/s12016-014-8427-x
26. Muratori P, Fabbri A, Lalanne C, Lenzi M, Muratori L (2015) Autoimmune liver disease and concomitant extrahepatic autoimmune disease. Eur J Gastroenterol Hepatol 27(10):1175–1179. https://doi.org/10.1097/MEG.0000000000000424
27. Gershwin ME, Selmi C, Worman HJ, Gold EB, Watnik M, Utts J, Lindor KD, Kaplan MM, Vierling JM, USA PBC Epidemiology Group (2005) Risk factors and comorbidities in primary biliary cirrhosis: a controlled interview-based study of 1032 patients. Hepatol Baltim Md 42(5):1194–1202. https://doi.org/10.1002/hep.20907
28. Invernizzi P, Selmi C, Ranftler C, Podda M, Wesierska-Gadek J (2005) Antinuclear antibodies in primary biliary cirrhosis. Semin Liver Dis 25(03):298–310. https://doi.org/10.1055/s-2005-916321
29 Boonstra K, Kunst AE, Stadhouders PH, Tuynman HA, Poen AC, van Nieuwkerk KMJ, Witteman EM, Hamann D, Witteman BJ, Beuers U, Ponsioen CY, the Epi PSC PBC study group (2014) Rising incidence and prevalence of primary biliary cirrhosis: a large population-based study. Liver Int Off J Int Assoc Study Liver 34(6):e31–e38. https://doi.org/10.1111/liv.12434
30 Corpechot C, Abenavoli L, Rabahi N, Chrétien Y, Andréani T, Johanet C, Chazouillères O, Poupon R (2008) Biochemical response to ursodeoxycholic acid and long-term prognosis in primary biliary cirrhosis. Hepatol Baltim Md 48(3):871–877. https://doi.org/10.1002/hep.22428
31 Corpechot C, Chazouillères O, Poupon R (2011) Early primary biliary cirrhosis: biochemical response to treatment and prediction of long-term outcome. J Hepatol 55(6):1361–1367. https://doi.org/10.1016/j.jhep.2011.02.031
32 Kumagi T, Guindi M, Fischer SE, Arenovich T, Abdalian R, Coltescu C, Heathcote EJ, Hirschfield GM (2010) Baseline ductopenia and treatment response predict long-term histological progression in primary biliary cirrhosis. Am J Gastroenterol 105(10):2186–2194. https://doi.org/10.1038/ajg.2010.216
33 Cheung K-S, Seto W-K, Fung J, Lai CL, Yuen MF (2017) Epidemiology and natural history of primary biliary cholangitis in the Chinese: a territory-based study in Hong Kong between 2000 and 2015. Clin Transl Gastroenterol 8(8):e116. https://doi.org/10.1038/ctg.2017.43
34 Kanth R, Shrestha RB, Rai I, Van Wormer JJ, Roy PK (2017) Incidence of primary biliary cholangitis in a rural midwestern population. Clin Med Res 15(1-2):13–18. https://doi.org/10.3121/cmr.2017.1351
35 Lleo A, Jepsen P, Morenghi E, Carbone M, Moroni L, Battezzati PM, Podda M, Mackay IR, Gershwin ME, Invernizzi P (2016) Evolving trends in female to male incidence and male mortality of primary biliary cholangitis. Sci Rep 6(1). https://doi.org/10.1038/srep25906
36 Kim K-A, Ki M, Choi HY, Kim BH, Jang ES, Jeong SH (2016) Population-based epidemiology of primary biliary cirrhosis in South Korea. Aliment Pharmacol Ther 43(1):154–162. https://doi.org/10.1111/apt.13448
37 McNally RJQ, James PW, Ducker S, Norman PD, James OFW (2014) No rise in incidence but geographical heterogeneity in the occurrence of primary biliary cirrhosis in North East England. Am J Epidemiol 179(4):492–498. https://doi.org/10.1093/aje/kwt308
38 Delgado J-S, Vodonos A, Delgado B, Jotkowitz A, Rosenthal A, Fich A, Novack V (2012) Primary biliary cirrhosis in Southern Israel: a 20 year follow up study. Eur J Intern Med 23(8):e193–e198. https://doi.org/10.1016/j.ejim.2012.09.004
39 Baldursdottir TR, Bergmann OM, Jonasson JG, Ludviksson BR, Axelsson TA, Björnsson ES (2012) The epidemiology and natural

history of primary biliary cirrhosis: a nationwide population-based study. Eur J Gastroenterol Hepatol 24(7):824–830. https://doi.org/10.1097/MEG.0b013e328353753d

40 Ngu JH, Gearry RB, Wright AJ, Stedman CAM (2012) Low incidence and prevalence of primary biliary cirrhosis in Canterbury, New Zealand: a population-based study. Hepatol Int 6(4):796–800. https://doi.org/10.1007/s12072-011-9329-0

41 Chong VH, Telisinghe PU, Jalihal A (2010) Primary biliary cirrhosis in Brunei Darussalam. Hepatobiliary Pancreat Dis Int HBPD INT 9(6):622–628

42 Wong R-K, Lim S-G, Wee A, Chan YH, Aung MO, Wai CT (2008) Primary biliary cirrhosis in Singapore: evaluation of demography, prognostic factors and natural course in a multi-ethnic population. J Gastroenterol Hepatol 23(4):599–605. https://doi.org/10.1111/j.1440-1746.2007.05058.x

43 Tanaka A, Ma X, Yokosuka O, Weltman M, You H, Amarapurkar DN, Kim YJ, Abbas Z, Payawal DA, Chang ML, Efe C, Ozaslan E, Abe M, Mitchell-Thain R, Zeniya M, Han KH, Vierling JM, Takikawa H (2016) Autoimmune liver diseases in the Asia-Pacific region: proceedings of APASL symposium on AIH and PBC 2016. Hepatol Int 10(6):909–915. https://doi.org/10.1007/s12072-016-9767-9

44 Liu H, Liu Y, Wang L, Xu D, Lin B, Zhong R, Gong S, Podda M, Invernizzi P (2010) Prevalence of primary biliary cirrhosis in adults referring hospital for annual health check-up in Southern China. BMC Gastroenterol 10(1):100. https://doi.org/10.1186/1471-230X-10-100

45 Myers RP, Shaheen AAM, Fong A, Burak KW, Wan A, Swain MG, Hilsden RJ, Sutherland L, Quan H (2009) Epidemiology and natural history of primary biliary cirrhosis in a Canadian health region: a population-based study. Hepatol Baltim Md 50(6):1884–1892. https://doi.org/10.1002/hep.23210

46 Mytilinaiou MG, Meyer W, Scheper T, Rigopoulou EI, Probst C, Koutsoumpas AL, Abeles D, Burroughs AK, Komorowski L, Vergani D, Bogdanos DP (2012) Diagnostic and clinical utility of antibodies against the nuclear body promyelocytic leukaemia and Sp100 antigens in patients with primary biliary cirrhosis. Clin Chim Acta Int J Clin Chem 413(15-16):1211–1216. https://doi.org/10.1016/j.cca.2012.03.020

47 Czaja AJ (1998) Frequency and nature of the variant syndromes of autoimmune liver disease. Hepatol Baltim Md 28(2):360–365. https://doi.org/10.1002/hep.510280210

48 Chazouillères O, Wendum D, Serfaty L, Montembault S, Rosmorduc O, Poupon R (1998) Primary biliary cirrhosis-autoimmune hepatitis overlap syndrome: clinical features and response to therapy. Hepatol Baltim Md 28(2):296–301. https://doi.org/10.1002/hep.510280203

49 Ngu JH, Bechly K, Chapman BA, Burt MJ, Barclay ML, Gearry RB, Stedman CAM (2010) Population-based epidemiology study of autoimmune hepatitis: a disease of older women? J Gastroenterol Hepatol 25(10):1681–1686. https://doi.org/10.1111/j.1440-1746.2010.06384.x