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Original article

Reevaluating the clinical course of AMA-positive patients with normal liver enzymes: A large retrospective cohort study

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ABSTRACT

Introduction and Objectives: The long-term clinical significance of anti-mitochondrial antibody (AMA)-positive patients with normal liver enzymes remains unclear. Despite increasing detection of AMA positivity in asymptomatic individuals, clinicians lack evidence-based protocols for long-term surveillance, and guidelines offer no clear recommendations. This study, the largest of its kind, investigates the natural history, prognostic implications, and risk factors for disease progression in this population.

Materials and Methods: We conducted a retrospective cohort study using a national healthcare database to identify adults (aged 18 years or older) with a positive AMA and normal alkaline phosphatase (ALP) levels between 2002 and 2023. Demographics, laboratory data, and liver-related outcomes were assessed. Multivariate logistic and Cox regression models were used to evaluate predictors of primary biliary cholangitis (PBC), cirrhosis, and hepatic complications.

Results: Among 1018 patients (median follow-up 6.3 years (IQR 2.5–11.5), 76 (7.5 %) developed PBC and 30 (2.9 %) progressed to cirrhosis. Liver-related complications were infrequent: esophageal varices (1.1 %), ascites (1.8 %), hepatocellular carcinoma (0.2 %), and liver transplantation (0.1 %). Higher AMA titers were strongly associated with increased risk of PBC, cirrhosis, and complications, showing a clear titer-dependent gradient. Longitudinal analysis also demonstrated titer-associated increases in ALP and bilirubin over time.

Conclusions: AMA-positive individuals with normal liver enzymes typically experience a benign clinical course. However, high AMA titers identify a subgroup at increased risk for progression to PBC and advanced liver disease. These findings underscore the importance of risk-stratified surveillance strategies in clinical practice, guiding healthcare professionals in the management of their patients.

1. Introduction

Primary biliary cholangitis (PBC) is a chronic autoimmune liver disease characterized by progressive destruction of small intrahepatic bile ducts, leading to cholestasis, fibrosis, and potentially cirrhosis or liver failure. It predominantly affects middle-aged women and is the most common chronic cholestatic liver disease in adults, with an estimated prevalence ranging from 1.9 to 40.2 per 100,000 individuals [1–4]. While many patients are asymptomatic at diagnosis, some develop complications such as portal hypertension, hepatic decompensation, or hepatocellular carcinoma [5,6].

Anti-mitochondrial antibodies (AMA), specifically directed against the E2 component of the pyruvate dehydrogenase complex (PDC-E2), are the hallmark serologic marker of primary biliary cholangitis (PBC), present in 90–95 % of affected individuals [7–10]. AMA

Abbreviations: AMA, anti-mitochondrial antibody; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CHS, clalit health services; EMR, electronic medical record; GGT, gamma-glutamyltransferase; HBV, hepatitis b virus; HCC, hepatocellular carcinoma; HCV, hepatitis c virus; HIV, human immunodeficiency virus; HMO, health maintenance organization; ICD-9, international classification of diseases, ninth revision; INR, International Normalized Ratio; LT, Liver Transplantation; PBC, Primary Biliary Cholangitis; PDC-E2, Pyruvate Dehydrogenase Complex E2; PT, Prothrombin Time; PTT, Partial Thromboplastin Time; PVT, Portal Vein Thrombosis

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positivity alone, however, is not synonymous with PBC. PBC is diagnosed based on the presence of at least two of the following features: a persistent elevation of serum alkaline phosphatase (ALP) for at least six months, anti-mitochondrial antibody (AMA) positivity at a titer of ≥1:40 or AMA-M2 reactivity by ELISA, and liver histology consistent with non-suppurative destructive cholangitis involving the interlobular bile ducts [11,12].

With the increasing use of broad autoantibody screening, AMA positivity is being detected in individuals without liver enzyme abnormalities. AMA are detectable in 0.5 % to 3.8 % of the general population, with high-titer positivity reported in approximately 1 %. Prevalence is notably higher among individuals with autoimmune or rheumatologic diseases, particularly systemic sclerosis, Sjögren's syndrome, and lupus, as well as among first-degree relatives of patients with primary biliary cholangitis, where rates may exceed 10 % [13,14]. Some AMA-positive individuals with normal liver biochemistry have been shown to harbor histological features consistent with early PBC, raising concern for subclinical disease and future risk of progression. However, the natural history, clinical significance, and long-term prognosis of this group remain poorly defined [15].

Small studies have suggested that AMA-positive individuals with normal ALP may represent a heterogeneous population. A minority may progress to PBC, especially those with higher AMA titers or elevated IgM levels [16], while the majority remain clinically stable. Reported rates of PBC development in this population vary widely, from as low as 0.5 % over five years to as high as 76 % over longer follow-up in highly selected cohorts [17]. Nevertheless, most prior studies have been limited by small sample sizes, short follow-up durations, or inconsistent risk stratification [18].

Current guidelines provide limited recommendations for the management or surveillance of AMA-positive individuals with normal liver enzymes [19]. Despite increasing detection of AMA positivity in asymptomatic individuals, clinicians lack evidence-based protocols for long-term management, leading to potential under- or over-surveillance. The present study addresses this gap by evaluating the clinical course of AMA-positive patients with normal liver enzymes over 20 years in a large national healthcare database. We hypothesized that most AMA-positive patients with normal liver enzymes have a benign course, but higher AMA titers identify a subgroup at increased risk for progression. We aimed to assess the incidence of PBC and advanced liver disease and identify predictors that could inform risk-stratified monitoring and clinical decision-making.

2. Materials and methods

2.1. Ethical approval

The study was approved by the Institutional Review Board of Clalit Health Services, which granted a waiver of written informed consent owing to the retrospective, noninterventional design and use of anonymized, encoded data.

2.2. Data source

This retrospective cohort study utilized the comprehensive electronic medical record (EMR) database of Clalit Health Services (CHS), Israel's largest Health Maintenance Organization (HMO), which serves approximately 4.8 million members. CHS operates a nationwide network of 14 hospitals and approximately 1500 outpatient clinics, laboratories, imaging centers, and pharmacies. The EMR system aggregates data from primary care physicians, specialty clinics, hospital admissions, laboratory results, and pharmacy transactions. Chronic disease diagnoses were recorded using validated algorithms based on the International Classification of Diseases, Ninth Revision (ICD-9).

2.3. Study design and population

The study included adult patients (aged 18 years or older) with a positive anti-mitochondrial antibody (AMA) test and normal alkaline phosphatase (ALP) levels between 2002 and 2023. Normal ALP was defined as below 130 IU/L (based on institutional laboratory standards) within three months of the first positive AMA result. To ensure cross-laboratory consistency, a unified ALP upper limit of 130 U/L was applied across sexes. Recognizing the lower reference range typically observed in women, yearly mean ALP values were additionally calculated for female patients with multiple measurements to mitigate transient fluctuation bias. This approach is now acknowledged as a limitation with possible minimal misclassification in women.

The date of the first recorded positive AMA test, from any source, was established as the index date for each patient. To ensure accurate longitudinal analysis, only patients with complete follow-up data were included, defined as having at least one additional ALP measurement and clinical encounter (visit, lab result, or diagnosis code) following the index date. The end of follow-up was defined as the date of the last available ALP measurement in the EMR system.

PBC was diagnosed according to international guidelines, requiring at least two of the following criteria: (1) ALP >1.5 × ULN persisting for >6 months, (2) AMA positivity, and (3) compatible liver histology when available. Cirrhosis was defined based on one or more of the following: (1) imaging findings consistent with cirrhosis or portal hypertension, (2) liver stiffness >16.9 kPa by elastography, (3) histological confirmation when available, or (4) clinical complications such as ascites, variceal bleeding, or hepatic encephalopathy.

2.4. Exclusion criteria

Included patients younger than 18 years, those with negative AMA test results, those with ALP levels greater than 130 IU/L following AMA positivity, and those with a preexisting diagnosis of primary biliary cholangitis (PBC) at or before the index date. In addition, patients with preexisting diagnoses of liver disease due to hepatitis B virus (HBV) infection, hepatitis C virus (HCV) infection, HIV infection, excessive alcohol consumption, hemochromatosis, Wilson's disease, α1-antitrypsin deficiency, celiac disease, uncontrolled hyperthyroidism, autoimmune hepatitis, or cirrhosis and its complications before the index date were excluded.

2.5. Outcomes

The primary outcome was the development of primary biliary cholangitis (PBC) or cirrhosis during the follow-up period. The primary outcome was defined as the development of primary biliary cholangitis (PBC) or cirrhosis, as determined by standardized diagnostic criteria and confirmed by review of electronic medical records. Secondary outcomes included the incidence of liver-related complications, specifically hepatocellular carcinoma (HCC), ascites, hepatic encephalopathy, esophageal varices, spontaneous bacterial peritonitis (SBP), portal vein thrombosis (PVT), hepatorenal syndrome, and liver transplantation (LT).

2.6. Data collection

Data extracted from the CHS database included demographic information (age and gender), baseline and follow-up laboratory parameters (alanine aminotransferase [ALT], aspartate aminotransferase [AST], gamma-glutamyltransferase [GGT], alkaline phosphatase [ALP], albumin, bilirubin, creatinine, urea, international normalized ratio [INR], prothrombin time [PT], partial thromboplastin time [PTT], hemoglobin, white blood cell [WBC] count and platelet count), initial and follow-up AMA titers, and the occurrence of predefined liver-related complications.

2.7. Statistical analysis

Descriptive statistics were used to summarize the study population's demographic and clinical characteristics and calculate the incidence of liver-related complications. Comparative analyses were conducted using chi-square tests for categorical variables and independent *t*-tests for continuous variables to assess differences between patients who developed significant liver disease and those who did not. Multivariate logistic regression analysis was performed to adjust for potential confounders, including age and gender, and to evaluate the independent association between AMA positivity with normal ALP and the development of liver disease. Additionally, survival analyses using Kaplan-Meier curves and Cox proportional hazards models were undertaken to assess time-to-event outcomes for liver-related complications, adjusting for relevant covariates. The statistical analyses were performed using SAS Enterprise Guide 8.3 software (SAS Institute Inc., Cary, NC, USA). P-value<0.05 was considered statistically significant.

3. Results

3.1. Patient characteristics and AMA titer distribution

Between 2022 and 2023, 1018 patients with positive AMA and normal liver enzymes were identified at Clalit Medical Centers, excluding those with concomitant liver disease. Of the 1018 patients, 459 (45.1 %) underwent titer testing. The AMA titer distribution and stratification across demographic subgroups are detailed in Table 2: 288 patients (28.3 %) had a titer of 1:40; 148 (14.5 %) had 1:80; 77 (7.6 %) had 1:160; 44 (4.3 %) had 1:320; and 2 (0.2 %) had titers above 1:320. Notably, the small sample size of only two patients in the >1:320 group limits the statistical reliability of the findings in this subgroup.

The median follow-up period after AMA positivity was 6.3 years (1–22.7 years). The median age at diagnosis was 57 years (range, 18–93), with 85.3 % of patients being female. Only 30 patients (2.9 %) progressed to cirrhosis, with a mean time to development of 7.5 years (median: 6.3 years) (Table 1).

3.2. Liver disease progression and complications

During the follow-up period, liver disease complications were observed in a minority of patients. Among the 571 AMA-positive patients with normal liver enzymes at baseline, 12 (2.1 %) developed cirrhosis-related complications, and LT was required in a single patient (0.1 %) over the follow-up period (Table 2). The incidence of the composite outcome slightly increased with age groups, ranging from 1.4 % to 4.5 %, except in the oldest age group (>80 years), where the incidence reached 50 %. However, this subgroup included only two patients.

The average time to event was 7.1 years (SD 5.1), with a median of 6.1 years (IQR 2.0–10.8).

In the overall cohort ($N = 559$), the median follow-up time was 5.7 years (SD, 5.1), with a median of 3.8 years (IQR, 1.5–8.6). Longer follow-up durations were observed in the older age groups, with a mean time of 11.9 years in patients aged 70–79 and 11.1 years in those aged 80 and above.

3.3. PBC diagnosis and association with AMA titers

During follow-up, 76 patients (7.5 %) were diagnosed with PBC, with a mean time to diagnosis of 7.4 years (median: 6.2 years). Among those who underwent titer testing, higher AMA titers were strongly associated with an increased risk of PBC development.

Table 1
Demographic characteristics of the study population. Demographic distribution of the study cohort, including age at diagnosis, gender and socioeconomic status (SES). Age is reported as mean, standard deviation (SD), quartiles (Q1, median, Q3) and range (minimum–maximum). Gender is shown as male and female. SES is classified as high, medium, low, or unspecified. Data are presented as absolute numbers (N) and percentages (%).

Characteristic	N (%)
Age at diagnosis (Mean)	56.0 ± 15.5 [18–93]
Age group: 18–30	69 (6.8 %)
Age group: 30–40	119 (11.7 %)
Age group: 40–50	150 (14.7 %)
Age group: 50–60	248 (24.4 %)
Age group: 60–70	241 (23.7 %)
Age group: ≥70	191 (18.8 %)
Gender: Female	868 (85.3 %)
Gender: Male	150 (14.7 %)
SES: High	419 (41.2 %)
SES: Medium	293 (28.8 %)
SES: Low	247 (24.3 %)
SES: Unspecified	59 (5.8 %)
Median follow-up (years, IQR)	6.3 (2.5–11.5)
Cirrhosis (incident cases)	30 (2.9 %)
Primary biliary cholangitis (PBC)	76 (7.5 %)
Esophageal varices	11 (1.1 %)
Ascites	18 (1.8 %)
Hepatorenal syndrome	2 (0.2 %)
Portal vein thrombosis	2 (0.2 %)
Bacterial peritonitis	3 (0.3 %)
Malignant neoplasm	2 (0.2 %)
Liver transplantation	1 (0.1 %)
Composite endpoint*	27 (2.7 %)

Table 2
Demographic characteristics stratified by anti-mitochondrial antibody (AMA) titer categories. Demographic variables of the study population stratified by AMA titer. Age at diagnosis is summarized by mean, SD, interquartile range (Q1, median, Q3) and range. Age group distribution is presented in six categories (18–30, 30–40, 40–50, 50–60, 60–70, and ≥70 years). Gender and SES (high, medium, low, unspecified) are provided as N and %.

Parameter	1:40	1:80	1:160	1:320	>1:320
Total Patients (N)	288	148	77	44	2
Age at Diagnosis (Mean)	56.8	56.5	58.0	53.9	58.9
Age at Diagnosis (Std)	15.3	16.1	13.6	15.1	15.4
Age group: 18–30 (%)	5.5	7.3	2.7	5.2	4.5
Age group: 30–40 (%)	11.6	11.5	9.5	19.5	6.8
Age group: 40–50 (%)	14.7	13.9	15.5	13.0	18.2
Age group: 50–60 (%)	24.2	21.9	29.7	24.7	20.5
Age group: 60–70 (%)	23.8	25.7	20.9	22.1	25.0
Age group: ≥70 (%)	20.2	19.8	21.6	15.6	25.0
Gender: Female (%)	89.1	87.8	91.9	88.3	88.6
Gender: Male (%)	10.9	12.2	8.1	11.7	11.4
SES: Low (%)	3.0	2.4	4.7	1.3	4.5
SES: Medium (%)	29.3	32.6	27.7	24.7	20.5
SES: High (%)	52.4	48.3	55.4	57.1	61.4
Mean Years since AMA positivity	7	5.7	7.9	7.6	11.3
Standard Deviation (Years)	5.5	5	4.9	5.3	7.3
Cirrhosis Follow-up (%)	2.7	0.7	2	5.2	11.4
Esophageal Varices Follow-up (%)	0.7	0	0.7	1.3	4.5
Ascites Follow-up (Ascites%)	1.6	0.7	0.7	3.9	4.5
HRS Follow-up (%)	0.2	0.0	0.0	0.0	2.3
Liver Transplant Follow-up (%)	0.2	0.0	0.0	0.0	2.3
Malignant Neoplasm Follow-up (%)	0.2	0.0	0.0	0.0	0
PV Thrombosis Follow-up (Thrombosis%)	0.4	0.7	0.0	0.0	0.0
SBP Follow-up (Bacterial Peritonitis%)	0.2	0.0	0.7	1.3	2.3
PBC Follow-up (PBC%)	8.4	5.6	9.5	11.7	18.2
Composite (Composite%)	2.1	1.4	1.4	3.9	4.5

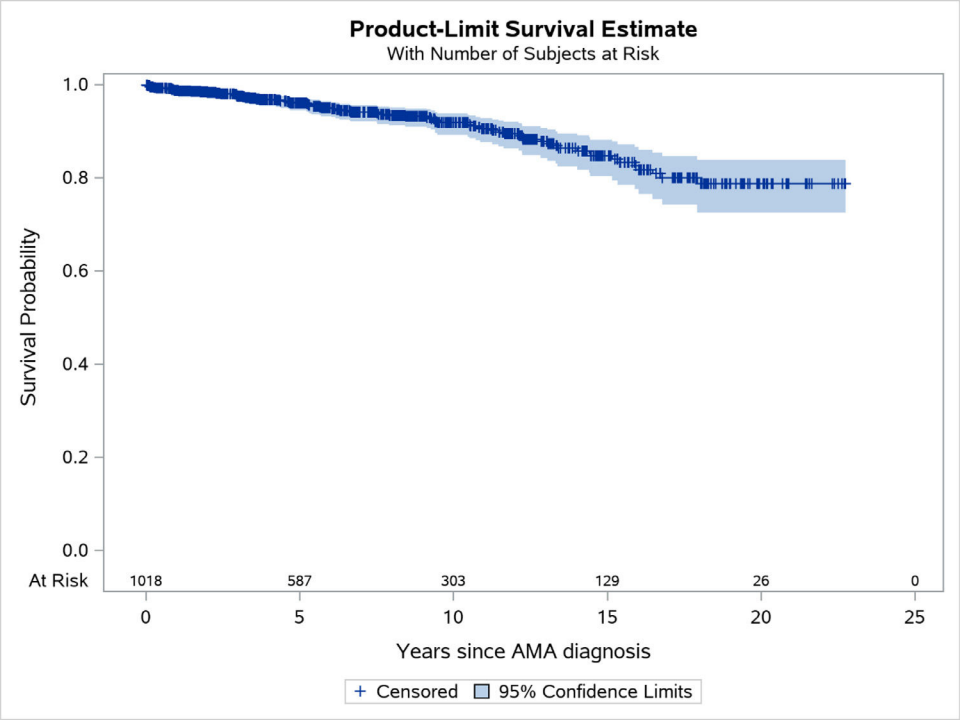

Fig. 1. Kaplan-Meier estimates of the probability of remaining free of primary biliary cholangitis (PBC) among AMA-positive patients with normal liver enzymes. Kaplan-Meier survival curve illustrating the cumulative probability of remaining free of PBC in AMA-positive patients with normal liver enzymes over a 25-year follow-up period. The solid blue line denotes survival probability; censored observations are indicated by plus signs (+). The shaded region represents the 95 % confidence interval. Clarification: Analysis includes AMA-positive patients with normal liver enzymes at baseline; incident PBC during follow-up was adjudicated per the criteria detailed in Methods.

(Fig. 1) displays the cumulative incidence of PBC stratified by AMA titer groups, highlighting a significantly increased risk among patients with higher titers. The PBC by AMA titer level was as follows: 5.6 % at 1:40, 9.5 % at 1:80, 11.7 % at 1:160, and 18.2 % at 1:320. Notably, neither developed PBC among the two patients with titers greater than 1:320. However, given the minimal sample size, no meaningful conclusions regarding risk in this subgroup can be drawn. The mean time to PBC diagnosis also varied by AMA titer, ranging from 5.5 years in those with titers of 1:40 to 11.6 years in those with titers of 1:320.

These findings reinforce that while most AMA-positive patients with normal liver enzymes remain stable over time, individuals with higher titers exhibit a greater risk of PBC development**.** None of the analyzed variables, including age, gender, socioeconomic status (SES), and titers, are significantly associated with PBC in the time-to-event analysis (Fine and Gray's model). The univariate raw analysis identified SES ($P = 0.0096$) and titers ($p = 0.0381$) as potentially significant, but this was not confirmed in the time-dependent analysis.

3.4. AMA titers and cirrhosis complications

The study's findings on the relationship between AMA titers and the progression of liver disease are significant. Higher AMA titers were found to be associated with an increased incidence of cirrhosis-related complications. The severity and frequency of hepatic complications increased in parallel with rising titer levels. (Fig. 2)

(Fig. 3), which features Kaplan-Meier curves, further illustrates the progression to cirrhosis in patients with high titers compared to those with lower titers. Among the 559 patients (45.1 %) who underwent titer testing, liver-related complications varied according to AMA titer levels. The prevalence of cirrhosis increased with increasing titers, affecting 0.7 % of patients with a titer of 1:40, 2.0 % at 1:80, 5.2 % at 1:160, 11.4 % at 1:320, and 50.0 % in the group with a titer of greater than 1:320.

A similar trend was observed for esophageal varices, which were absent in patients with a titer of 1:40 but present in 0.7 % of those at 1:80, 1.3 % at 1:160, and 4.5 % at 1:320. The occurrence of ascites also increased with higher titers, affecting 0.7 % of patients with a titer of 1:40, 0.7 % at 1:80, 3.9 % at 1:160, 4.5 % at 1:320, and 50.0 % in the >1:320 category. (Fig. 4)

In the multivariate analysis, the association between AMA titers and liver-related complications remained significant after adjusting for age, gender, and SES.

3.5. ALP trends and long-term survival analysis

Analysis of longitudinal ALP trends following AMA positivity demonstrated a gradual increase in mean ALP levels over time. Notably, higher AMA titers were associated with a more significant rise in ALP levels (Fig. 5). Patients with lower AMA titers (1:40 and 1:80) exhibited relatively stable ALP levels with minimal progression over time, while patients with higher AMA titers (1:160 and 1:320) exhibited higher baseline ALP levels and a more pronounced increase over time than those with lower titers. This pattern suggests a potential relationship between AMA titers and biochemical disease progression, with higher titers correlating with a greater likelihood of ALP elevation.

3.6. Bilirubin trends and long-term survival analysis

(Fig. 6) illustrates the longitudinal trend of bilirubin levels in anti-mitochondrial antibody (AMA)-positive patients over a follow-up period of up to 15 years, stratified by AMA titer levels at baseline. Patients with lower AMA titers (1:40 and 1:80) had relatively stable bilirubin levels over time, with minimal fluctuation. Patients with higher AMA titers (1:160 and 1:320)

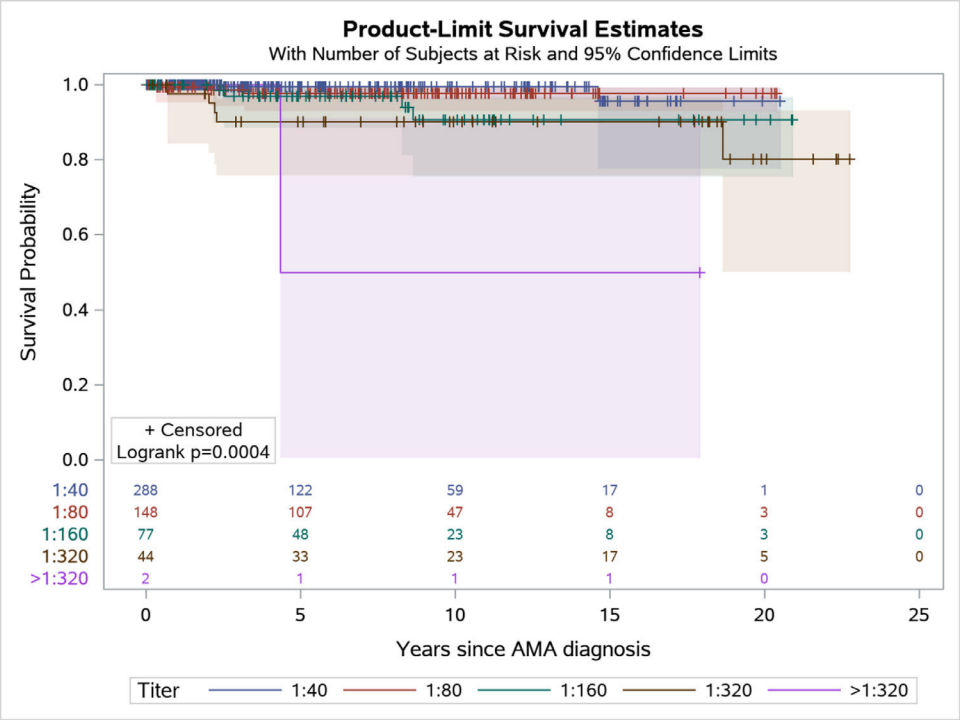

Fig. 2. Kaplan-Meier estimates of cirrhosis incidence by AMA titer group. Kaplan-Meier curves showing the probability of remaining free of cirrhosis, stratified by AMA titer groups: 1:40 (blue); 1:80 (red); 1:160 (teal); 1:320 (brown); >1:320 (purple). Censored data are marked by plus signs (+). Shaded regions indicate 95 % confidence intervals. Log-rank test p-value=0.0004.

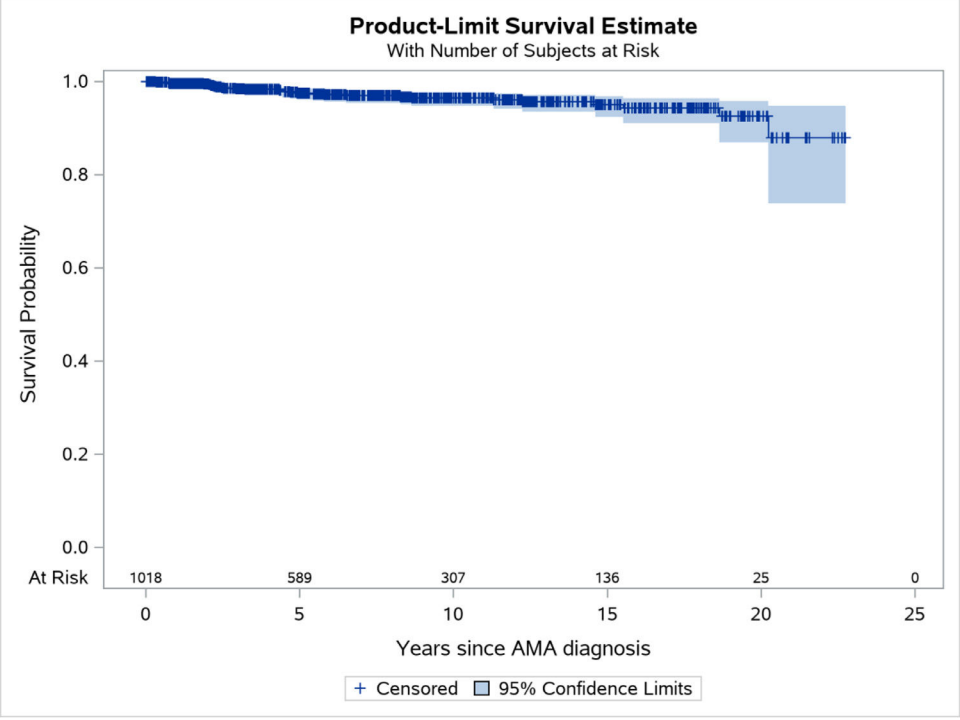

Fig. 3. Kaplan-Meier estimates of the probability of remaining free of cirrhosis among AMA-positive patients with normal liver enzymes. Kaplan-Meier survival analysis showing the probability of remaining free of cirrhosis over a 25-year follow-up period. The solid blue line denotes survival probability; censored cases are indicated by plus signs (+). The shaded area represents the 95 % confidence interval.

exhibited a more pronounced increase in bilirubin levels over time, suggesting a potential for subclinical progression. The 1:320 group demonstrated the most significant increase, peaking around years 8–10, followed by a decline, while the 1:160 group showed a steady upward trajectory. These findings indicate that higher AMA titers at baseline may be associated with a greater risk of bilirubin elevation over time, potentially reflecting progressive liver dysfunction.

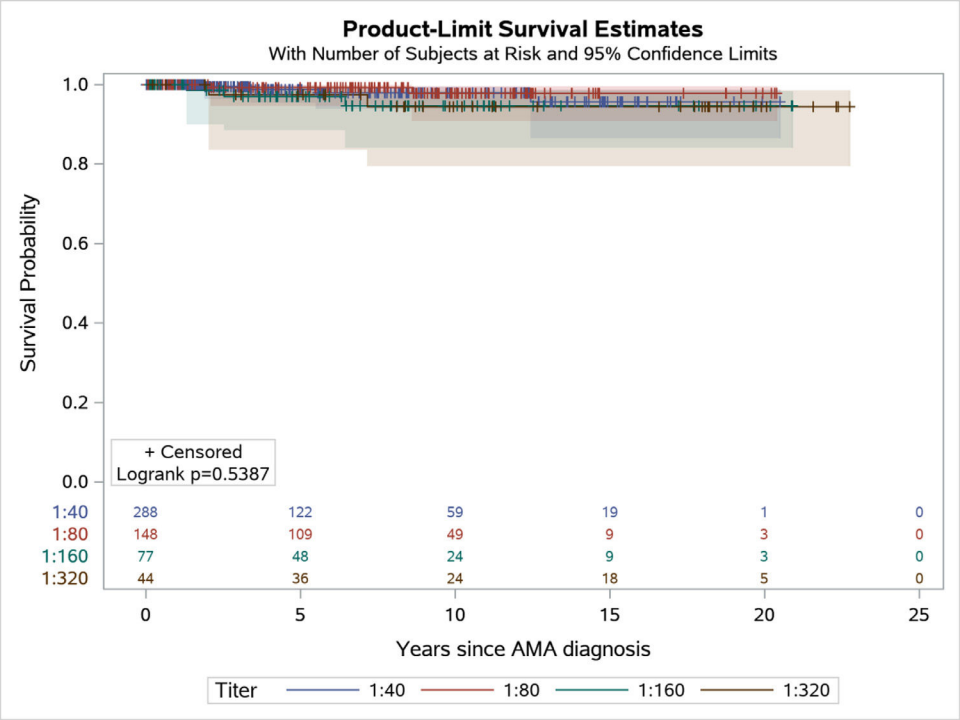

Fig. 4. Longitudinal changes in composite cirrhosis complications in AMA-positive patients by baseline antibody titer. Longitudinal trends in composite cirrhosis complications among AMA-positive patients over a 15-year follow-up period, stratified by baseline titer (1:40, 1:80, 1:160, 1:320). Solid lines denote group means; shaded areas indicate 95 % confidence intervals.

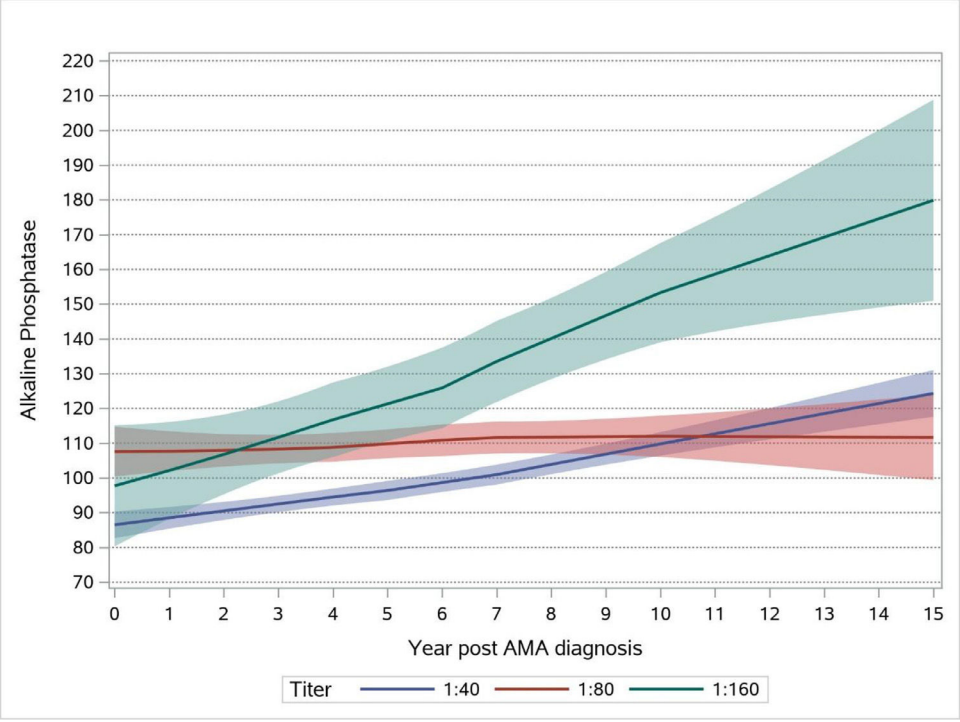

Fig. 5. Longitudinal trends of alkaline phosphatase (ALP) levels in AMA-positive patients by baseline antibody titer. Longitudinal analysis of ALP levels over a 15-year follow-up period, stratified by baseline AMA titer (1:40, 1:80, 1:160, 1:320). Solid lines denote mean ALP values for each group; shaded areas indicate 95 % confidence intervals. Note: The 1:320 subgroup was small; results for high titers are presented descriptively without inferring heterogeneity and should be interpreted cautiously.

4. Discussion

This large, retrospective cohort study demonstrates that AMA-positive patients with normal liver enzymes have a low risk of progression to PBC or cirrhosis. Still, higher AMA titers are associated with increased risk. Our findings strongly support two key conclusions. First, AMA-positive patients with normal liver enzymes generally have a low risk of developing PBC or cirrhosis. Over a median follow-up of

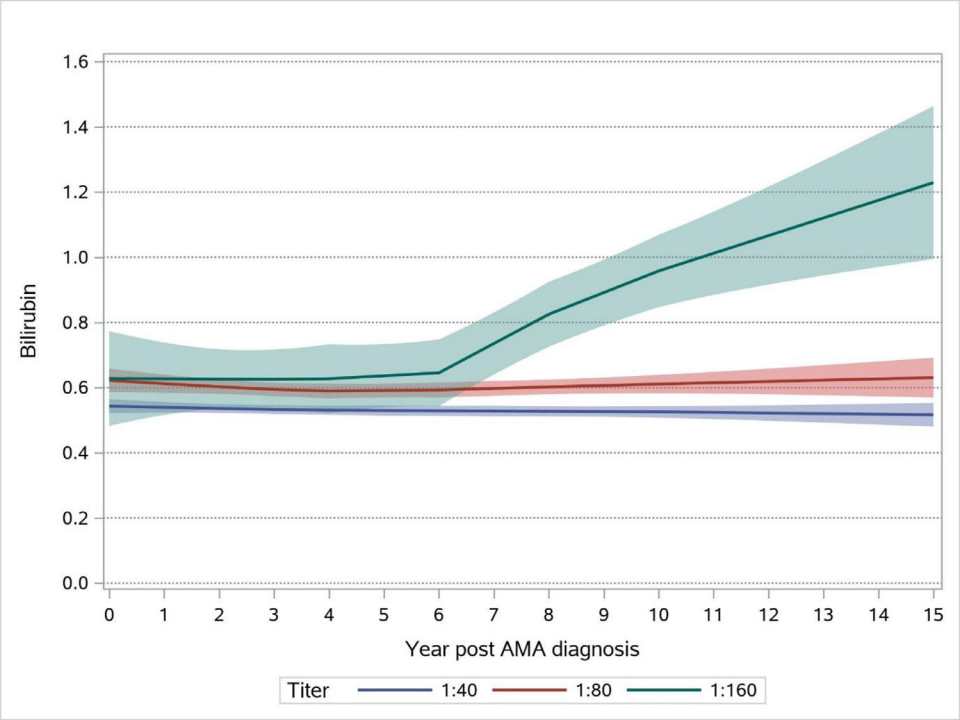

Fig. 6. Longitudinal changes in bilirubin levels in AMA-positive patients by baseline antibody titer. Longitudinal trends in bilirubin levels over a 15-year follow-up period, stratified by baseline AMA titer (1:40, 1:80, 1:160, 1:320). Solid lines denote group means; shaded areas indicate 95 % confidence intervals. Note: The 1:320 subgroup was small; results for high titers are presented descriptively without inferring heterogeneity and should be interpreted cautiously.

6.3 years, only 7.5 % developed PBC and 2.9 % progressed to cirrhosis. Second, higher AMA titers were found to be a strong predictor of disease progression. Patients with higher AMA titers had a significantly higher risk of developing PBC and cirrhosis. The incidence of cirrhosis and other liver-related complications increased proportionally with AMA titers, emphasizing the prognostic value of AMA levels.

These results support a risk-adapted approach to surveillance, reserving intensive monitoring for patients with high AMA titers, while reassuring the majority with low titers and normal enzymes.

Dahlqvist et al. conducted a large-scale characterization study that examined AMA-positive patients without established PBC diagnoses. They found that some of these individuals exhibited biochemical and histological features suggestive of early-stage PBC, with a subset eventually progressing to a confirmed diagnosis of PBC [20]. However, their study was limited by a relatively short follow-up duration and a lack of systematic risk stratification.

Similarly, Metcalf et al. followed a small cohort of 29 AMA-positive patients with normal liver enzymes for a median of 17.8 years. They reported that 76 % eventually developed PBC, indicating a high conversion rate. However, despite this high conversion rate, no patient developed cirrhosis or clinically apparent portal hypertension, reinforcing an overall indolent disease course [21]. In contrast, our study found a much lower incidence of PBC (7.5 %) over a median follow-up of 6.3 years. While differences in patient selection, population genetics, and diagnostic criteria used at different time points may account for this discrepancy, both studies ultimately support that AMA-positive individuals with normal liver enzymes generally exhibit slow disease progression, with cirrhosis and severe hepatic complications remaining rare.

In recent data, Li et al. examined a large cohort of AMA-M2-positive individuals in a health check-up setting in China and reported a 5-year cumulative PBC incidence of only 0.5 % [22]. These findings align more closely with our study, further supporting that the overall risk of PBC development in AMA-positive individuals with normal liver enzymes may be lower than previously estimated.

Ellez et al. investigated 26 AMA-positive patients with normal ALP levels and found that 43.75 % of those who underwent biopsies exhibited findings and histological features consistent with PBC [23]. While these findings suggest that a considerable proportion of AMA-positive individuals may harbor early histological abnormalities, the limited sample size and lack of long-term follow-up restrict the generalizability of these conclusions.

Similarly, the Swiss PBC Cohort Study by Terziroli Beretta-Piccoli et al. demonstrated that a high proportion of AMA-positive patients with normal ALP already exhibited histological evidence of PBC, even in the absence of biochemical cholestasis [24]. However, this study also lacked long-term outcome data, making it unclear whether histological abnormalities in this population invariably translate into clinically significant diseases.

Duan et al. conducted a retrospective study analyzing 139 AMA-positive patients without a baseline diagnosis of PBC. They found that only 4.3 % progressed to PBC over a median follow-up period of 4.6 years. The study identified lower alanine aminotransferase (ALT) and higher IgM levels as independent predictors of PBC development. [25]. Consistent with our findings, AMA positivity does not appear to be an isolated marker of inevitable disease but instead defines a heterogeneous population in which only a subset exhibits clinically meaningful progression.

4.1. Limitations

This study has several limitations. First, as a retrospective, noninterventional analysis, it is inherently subject to potential biases characteristic of observational research, including missing data, residual confounding, and incomplete variable ascertainment. Second, although a strong correlation was observed between higher AMA titers and disease progression, causality cannot be inferred from these associations. Third, the study population was derived from a single national healthcare system and may not fully represent other geographic or ethnic populations, thereby limiting generalizability.

Fourth, liver biopsies were not performed systematically, which may have led to underestimation of subclinical or histologically mild disease. Additionally, applying a unified ALP upper limit of normal (130 U/L) across sexes may have introduced minimal misclassification among women, particularly given lower female reference ranges. Finally, serologic data completeness for markers such as IgM, ANA, gp210, and sp100 was limited, and the number of patients within the highest-titer subgroup was relatively small, restricting the statistical power for inference in this subset.

Future prospective studies are needed to validate these findings and refine guidelines for managing AMA-positive individuals with normal liver enzymes.

5. Conclusions

In conclusion, this study, the largest of its kind, demonstrates that most AMA-positive patients with normal liver enzymes do not develop significant liver disease, challenging the need for intensive monitoring. While higher AMA titers indicate increased risk, overall progression remains slow, with severe complications being rare. A risk-stratified follow-up approach may be more appropriate than universal surveillance, warranting further prospective validation.

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Author contributions

All contributors participated in the conception and design, data handling and analysis, and drafting and critical revision of the manuscript, and approved the final version with full responsibility for the work.

Declaration of interests

None.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.aohep.2025.102147.

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