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Research Article

Patterns of liver fibrosis evolution in Chinese HIV/HBV co-infected adults following 5-year antiretroviral treatment: A longitudinal study using non-invasive APRI and Fib-4 scores

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ABSTRACT

The long-term effects of combined antiretroviral therapy (ART) on liver fibrosis patterns in adults living with human immunodeficiency virus (HIV) and chronic hepatitis B virus (HBV) are not well understood. Therefore, this study aimed to investigate the trajectories of liver fibrosis and identify the associations of baseline variables with different patterns of liver fibrosis evolution. A total of 333 individuals with HIV/HBV co-infection and undergoing long-term ART were enrolled in this study. Demographic, clinical, and biochemical data were collected at baseline and during annual visits. Group-based trajectory models (GBTMs) were used to detect the patterns of liver fibrosis evolution based on longitudinal data of fibrosis-4 (Fib-4) and aspartate aminotransferase to platelet ratio index (APRI) scores. Logistic regression analysis was performed to identify baseline predictors of liver fibrosis evolution. The median age of all participants was 33 years. Among them, 89.5% initially received TDF-containing ART. GBTMs identified two distinct patterns of liver fibrosis evolution using either APRI or Fib-4 scores. The majority of individuals (78.5% for APRI and 75.3% for Fib-4; pattern A) showed stable or low fibrosis with no progression, while the remaining participants showed regression from high fibrosis levels (21.5% for APRI and 24.7% for Fib-4; pattern B). Pattern A participants were younger and had higher $CD4^+$ cell counts, higher lymphocyte cell counts, higher white blood cell counts, and lower platelet counts at baseline compared to pattern B participants. For HIV/HBV co-infected patients with varying degrees of initial liver fibrosis, long-term ART has shown distinct patterns of alleviating liver fibrosis.

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INTRODUCTION

Approximately 8.4% of individuals worldwide living with human immunodeficiency virus (HIV) were co-infected with chronic hepatitis B virus (HBV) (Leumi et al., 2020), with a co-infection rate of around 10% in China (Xie et al., 2016). Due to the potent efficacy against both HIV and HBV replication, the combination of tenofovir disproxil fumarate (TDF) with lamivudine (3 TC) or emtricitabine (FTC) is the most widely recommended combined antiretroviral therapy (cART) in the treatment of HIV/HBV co-infected patients (de Vries-Sluijs et al., 2010; Guidelines for the Prevention and Treatment of Opportunistic Infections in Adults and Adolescents, 2022). While tenofovir (TDF)-based antiretroviral therapy (ART) has demonstrated significant clinical benefits, particularly in reducing liver fibrosis in patients with chronic HBV mono-infection (Marcellin et al., 2013), the outcomes regarding the evolution of liver fibrosis in individuals co-infected with HIV and HBV remain inconsistent (Audsley et al., 2016; Boyd et al., 2017; Dezanet et al., 2021; Iacob et al., 2022; Sterling et al., 2019).

Previous studies have revealed varying patterns of liver fibrosis regression that differ from those observed in HBV mono-infected individuals treated with cART. Some studies report a rapid decrease followed by stabilization of liver fibrosis after the first year of ART treatment (Audsley et al., 2016; Boyd et al., 2017), while others observe an increasing trend in liver fibrosis among some HIV-HBV co-infected individuals after treatment (Dezanet et al., 2021; Iacob et al., 2022; Sterling et al., 2019). Given these varied and complex evolutions of liver fibrosis, potentially influenced by short follow-up durations, there remains a notable gap in long-term longitudinal cohort studies examining the impact of ART on liver fibrosis progression in HIV/HBV co-infected patients, particularly in China.

The objective of this study was to identify the evolution of liver fibrosis in co-infected individuals over a long-term TDF-based antiretroviral therapy. Two non-invasive biochemical scores, the aspartate aminotransferase to platelet ratio index (APRI) and fibrosis-4 (Fib-4), were used to assess liver fibrosis, as they are reliable alternatives to liver biopsy in HIV-infected patients (Iacob et al., 2022; Sterling et al., 2006; Yang et al., 2023). This longitudinal cohort study collected data from annual visits conducted at baseline and at 12, 24, 36, 48, and 60 months. Given the varying patterns of liver fibrosis observed in previous studies, we implemented a group-based trajectory approach, which was designed to detect patients who have longitudinally similar medical patterns (Hsu et al., 2018; Kim et al., 2018; Niyonkuru et al., 2013; Song et al., 2016), to identify patients with similar trajectories of liver fibrosis evolution based on two non-invasive scores. Furthermore, we examined the association between baseline characteristics and different patterns of liver fibrosis evolution.

RESULTS

Clinical and laboratory assessment of the study population

In this longitudinal cohort study, a total of 333 adults with HIV and chronic HBV co-infection were followed for at least five years (Fig. 1). The baseline characteristics of all patients are presented in Table 1. The median age of the individuals at inclusion was 33 years. Among the 333 patients, the majority were male (n = 320, 96.1%) and acquired HIV through men who have sex with men (MSM) transmission (n = 262, 78.7%). Of the patients, 89.5% (n = 298) initiated TDF-based ART after the diagnosis of HIV and chronic HBV, while the remaining 34 patients switched to TDF-based ART treatment during the 5-year follow-up period.

The baseline HIV-RNA viral load had a median of 4.18 $\log_{10}$ IU/mL (IQR = 1.11). Accordingly, the baseline $CD4^+$ cell count had a median of 246.89/mm3 (IQR = 223.41). The baseline lymphocyte (LYM), platelet,

Table 1
Description of the study population with two liver fibrosis patterns by using two non-invasive biochemical scores.

	All (n = 333)	APRI scores (n = 333)			Fib-4 scores (n = 330)		
		Pattern A (n = 264)	Pattern B (n = 69)	*P*	Pattern A (n = 248)	Pattern B (n = 82)	*P*
Demographics							
Age (IQR) (years)	33 (11)	32 (11)	35 (12)	0.005	30 (9)	42 (14.75)	<0.001
BMI (IQR) (kg/m^2)	21.37 (3.57)	21.45 (3.44)	20.85 (3.36)	0.134	21.26 (3.03)	21.46 (3.51)	0.332
Gender				0.529			0.03
Male	320 (96.1%)	254 (96.2%)	66 (95.7%)		244 (98.4%)	74 (90.2%)	
HIV-related biomarkers							
Transmission				0.405			0.405
MSM	262 (78.7%)	216 (81.8%)	46 (66.7%)		207 (83.5%)	54 (65.9%)	
Baseline HIV viral load (IQR) ($\log_{10}$ copies/mL)	4.18 (1.11)	4.23 (0.96)	4.11 (1.7)	0.731	4.23 (1.05)	4.01 (1.52)	0.432
Baseline $CD4^+$ cell count (/mm^3)	246.89 (223.41)	266.43 (223.09)	211 (189)	0.01	274 (226.32)	191.87 (177.96)	0.001
HBV-related biomarkers							
Baseline HBV viral load (IQR) ($\log_{10}$IU/L)	3.2 (4.73)	3.05 (4.89)	3.86 (4.31)	0.423	3.48 (5.31)	2.7 (2.21)	0.087
Baseline HBeAg status				0.319			0.046
Negative	162 (48.6%)	136 (51.5%)	26 (37.7%)		118 (47.6%)	42 (51.2%)	
Positive	98 (29.4%)	77 (29.2%)	21 (30.4%)		82 (33.1%)	15 (18.3%)	
HBsAg seroclearance*	43 (12.9%)	34 (12.9%)	9 (13%)	1	27 (10.9%)	16 (19.5%)	0.108
Others							
Baseline WBC cell count (IQR) (10^9/L)	5.04 (2.01)	5.29 (2.02)	4.29 (1.83)	<0.001	5.34 (1.76)	4.25 (1.67)	<0.001
Baseline LYM (IQR) (10^9/L)	1.71 (0.84)	1.76 (0.79)	1.31 (0.96)	<0.001	1.83 (0.84)	1.32 (0.81)	<0.001
Baseline platelets (IQR) (10^9/L)	193 (73)	197 (63.5)	147.5 (87.25)	<0.001	198.5 (65.5)	144 (87.25)	<0.001
Baseline AST (IQR) (IU/L)	28 (13.4)	27 (11.65)	36.5 (29.95)	<0.001	27.5 (12.2)	32.4 (16.75)	0.003
Baseline ALT (IQR) (IU/L)	29.2 (24.1)	28.3 (21.85)	39.7 (43.73)	<0.001	29.25 (24.7)	28.25 (23.63)	0.364
Baseline Hb (IQR) (g/L)	150 (20)	152 (18.5)	146 (25.5)	0.062	153 (17.75)	145 (36)	<0.001
Baseline γ-GT (IQR) (U/L)	24.5 (28.3)	21.6 (20.85)	46.2 (38.3)	<0.001	22.85 (24.28)	31.65 (39.33)	<0.001
Baseline blood glucose (IQR) (mmol/L)	5.01 (0.95)	5 (0.88)	5.07 (1.17)	0.464	4.95 (0.82)	5.3 (1.11)	0.003
Baseline LDL (IQR) (mmol/L)	2.11 (0.91)	2.13 (0.86)	1.9 (0.99)	0.074	2.14 (0.92)	1.93 (0.9)	0.107
Baseline ALP (IQR) (U/L)	72 (25.6)	71.7 (23.75)	74.7 (36.98)	0.501	73.2 (23.8)	69.8 (29.1)	0.36

Notes: Data were presented as counts and percentages for categorical variables and as medians and interquartile ranges (IQRs) for continuous variables. HBsAg seroclearance was calculated at the last visit (60 months). Because age data were missing for three participants, the number of Fib-4 scores was 3 fewer than the number of APRI scores. BMI, body mass index; WBC, white blood cells; LYM, lymphocyte; AST, aspartate transaminase; ALT, alanine transaminase; Hb, hemoglobin; γ-GT, γ-glutamyl transferase; LDL, low density lipoprotein; ALP, alkaline phosphatase.

Table 2
Baseline description of the excluded 139 participants.

	All (n = 333)	Excluded participants (n = 139)	*P* value
Demographics			
Age (IQR) (years)	33 (11)	33 (11)	0.684
BMI (IQR) (kg/m^2)	21.37 (3.57)	22.49 (4.52)	0.035
Gender			0.121
Male	320 (96.1%)	137 (98.6%)	
HIV-related biomarkers			
Transmission			0.805
MSM	262 (78.7%)	117 (84.2%)	
Baseline HIV viral load (IQR) (log_{10} copies/mL)	4.18 (1.11)	4.36 (1.08)	0.324
Baseline CD4$^+$ cell count (IQR) (/mm^3)	246.89 (223.41)	281.3 (254.75)	0.241
HBV-related biomarkers			
Baseline HBV viral load (IQR) (log_{10}IU/L)	3.2 (4.73)	3.06 (4.3)	0.366
Baseline WBC cell count (IQR) (10^9/L)	5.04 (2.01)	5.18 (2.15)	0.678
Baseline LYM (IQR) (10^9/L)	1.71 (0.84)	1.75 (0.81)	0.988
Baseline platelets (IQR) (10^9/L)	193 (73)	198 (76)	0.121
Baseline AST (IQR) (IU/L)	28 (13.4)	26.95 (14.85)	0.318
Baseline ALT (IQR) (IU/L)	29.2 (24.1)	27.75 (25.17)	0.426
Baseline Hb (IQR) (g/L)	150 (20)	148 (21)	0.214
Baseline γ-GT (IQR) (U/L)	24.5 (28.3)	26.6 (27.08)	0.835
Baseline blood glucose (IQR) (mmol/L)	5.01 (0.95)	5.01 (0.74)	0.607
Baseline LDL (IQR) (mmol/L)	2.11 (0.91)	1.9 (0.98)	0.017
Baseline ALP (IQR) (U/L)	72 (25.6)	71.7 (26.27)	0.873
Baseline HBeAg status			0.24
Negative	162 (48.6%)	78 (56.1%)	
Positive	98 (29.4%)	35 (25.2%)	
HBsAg seroclearance	43 (12.9%)	10 (7.2%)	0.035

Notes: Data were presented as counts and percentages for categorical variables and as medians and interquartile ranges (IQRs) for continuous variables. HBsAg seroclearance was calculated at the last visit (60 months). One participant in both the included and excluded populations had missing gender information. BMI, body mass index; WBC, white blood cells; LYM, lymphocyte; AST, aspartate transaminase; ALT, alanine transaminase; Hb, hemoglobin; γ-GT, γ-glutamyl transferase; LDL, low density lipoprotein; ALP, alkaline phosphatase.

and white blood cells (WBC) counts had medians of 1.71 × 10^9/L (IQR = 0.84), 193 × 10^9/L (IQR = 73), and 5.04 × 10^9/L (IQR = 2.01), respectively. The baseline HBV-DNA viral load had a median of 3.2 log_{10} IU/mL (IQR = 4.73). The baseline aspartate transaminase (AST), alanine transaminase (ALT), hemoglobin (Hb), γ-glutamyl transferase (γ-GT), and blood glucose levels had medians of 28 IU/L (IQR = 13.4), 29.2 IU/L (IQR = 24.1), 150 g/L (IQR = 20), 24.5 U/L (IQR = 28.3), and 5.01 mmol/L (IQR = 0.95), respectively. At the beginning of the study, 98 (29.4%) patients were HBeAg positive, and the prevalence of liver fibrosis in all patients was 31.23% and 25.23% using the APRI and Fib-4 scores, respectively. At the last visit (60 months), the prevalence of liver fibrosis in all patients was 9.61% and 10.21% by using the APRI and the Fib-4 scores, respectively. And, 12.9% of participants had HBsAg loss at the last visit.

Furthermore, we described the baseline characteristics of the 139 participants who were excluded from the analysis due to insufficient follow-up time of less than five years or a lack of follow-up between 6 and 12 months (Table 2). A comparison of baseline variables between this excluded subgroup and the 333 individuals included in the longitudinal trend analysis showed no significant differences for most variables. This suggests that the exclusion of this subgroup does not introduce substantial sample selection bias into the analysis.

Patterns of liver fibrosis evolution using APRI score and Fib-4 score

Two distinct patterns of liver fibrosis evolution were observed in the group-based trajectory model and are graphically summarized in Fig. 2. When using the APRI score, 78.5% of patients had no progression in liver and remained stable over time (pattern A, n = 264). The remaining 21.5% of patients had a high level of liver fibrosis at study entry, which decreased over the following five years but did not return to normal

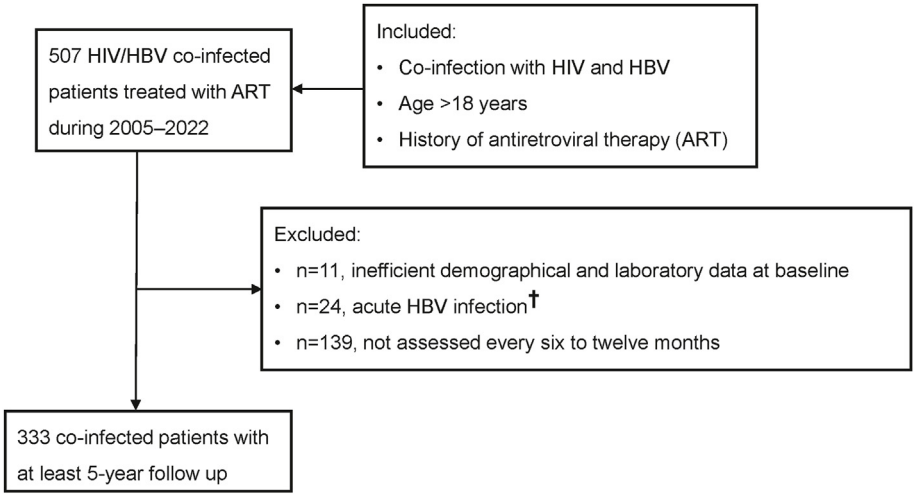

Fig. 1. Flowchart of the study population inclusion. †Acute HBV infection was defined as the clearance of HBsAg within six months.

values (pattern B, n = 69). Similar results were observed when using the Fib-4 score. The first group of patients (75.3%) had a stable low-level Fib-4 score during the 5-year follow-up, indicating no progression in liver fibrosis (pattern A, n = 248). The second group of patients (24.7%) had a high level of liver fibrosis initially, which slightly decreased over the next five years but did not reach a normal level (pattern B, n = 82). Regardless of whether the APRI or Fib-4 score was used, all patients' trajectories of liver fibrosis consistently fell into these two patterns. One pattern showed stable or low fibrosis with no progression, while the other pattern demonstrated regression from high levels of fibrosis (Fig. 2A and B).

Association of baseline predictors with different patterns of liver fibrosis evolution

The baseline characteristics of two evolutionary patterns are detailed in Table 1. Regardless of whether the APRI score or the Fib-4 score was used, patients classified as pattern A were generally younger ($P = 0.005$ and < 0.001, respectively), exhibited higher counts of $CD4^+$ cells ($P = 0.01$ and 0.001, respectively), lymphocytes ($P < 0.001$ in both scenarios), platelets ($P < 0.001$ in both scenarios), and WBC ($P < 0.001$ in both scenarios). Additionally, they had lower levels of AST ($P < 0.001$ and 0.003 respectively) and γ-GT ($P < 0.001$ in both scenarios) compared to patients in pattern B. Specifically, pattern A patients, as defined by the APRI score, also demonstrated significantly lower ALT levels ($P < 0.001$). In contrast, the majority of pattern A patients defined by the Fib-4 score were male ($P = 0.03$), with a higher incidence of HBeAg positivity ($P = 0.046$), increased Hb levels ($P < 0.001$), and lower baseline blood glucose levels ($P < 0.001$).

The results of the univariate logistic regression analysis (Table 3) reveal that different patterns of liver fibrosis evolution, as determined by the APRI and Fib-4 scores, are significantly associated with age ($P = 0.013$ and < 0.001, respectively), $CD4^+$ cell count ($P = 0.026$ and 0.006, respectively), LYM count ($P < 0.001$ in both scenarios), platelet count ($P < 0.001$ in both scenarios), Hb ($P = 0.025$ and < 0.001, respectively), WBC count ($P < 0.001$ in both scenarios), and γ-GT ($P < 0.001$ and $= 0.022$, respectively). Specifically, different patterns defined by the APRI score were associated with AST ($P = 0.001$) and ALT ($P = 0.008$). Different patterns defined by Fib-4 scores were associated with gender

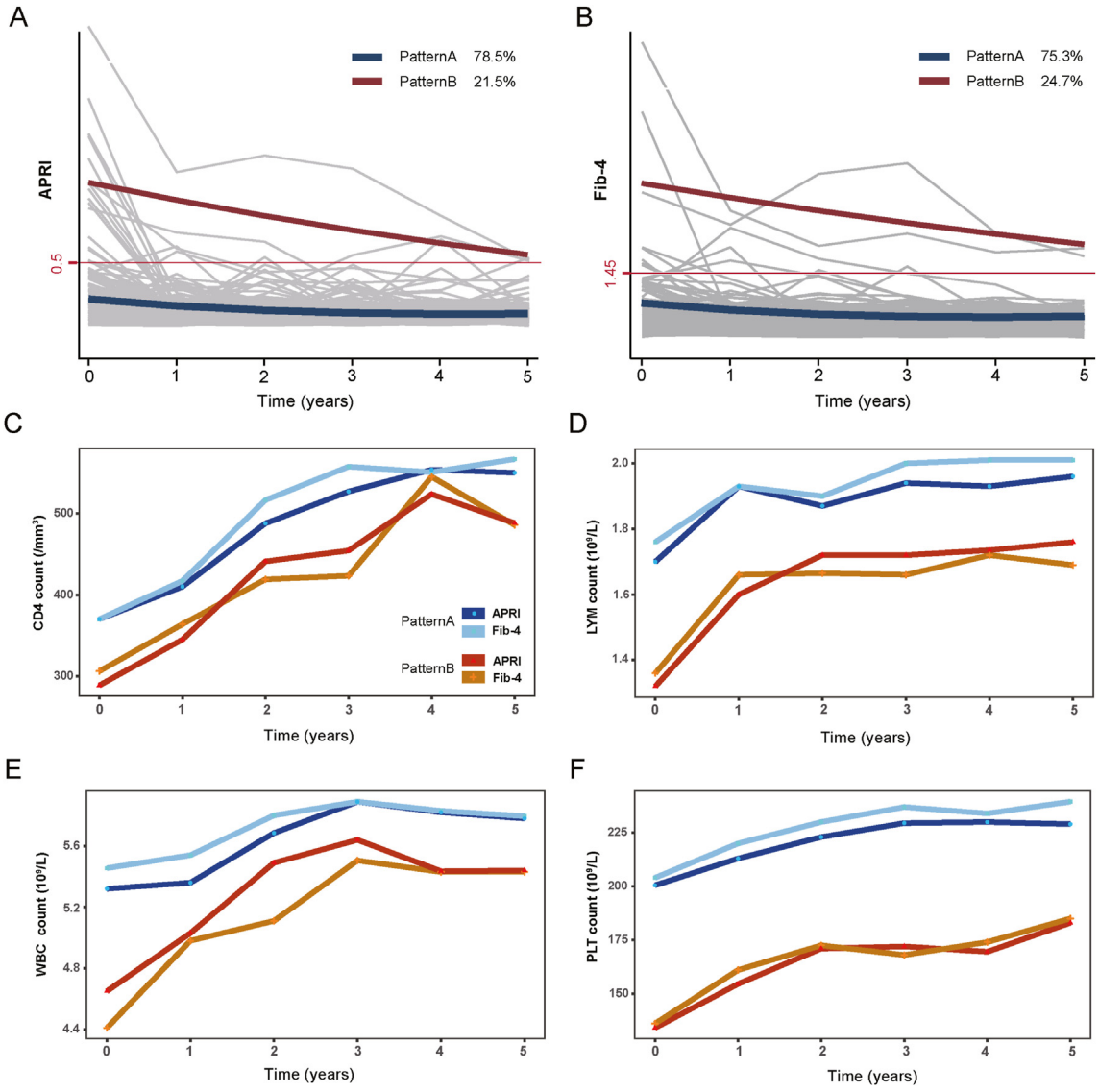

Fig. 2. Patterns of liver fibrosis trajectories throughout long-term ART given by the group-based trajectory model. **A, B** The trajectory pattern of liver fibrosis was defined by APRI scores (**A**) and Fib-4 scores (**B**); the lines of Pattern A and Pattern B were generated by GBTM models and gray lines in the background represented individual's specific fibrosis trajectory. **C–F** $CD4^+$ cell (**C**), Lymphocyte (LYM) cell (**D**), White Blood Cells (WBC) (**E**), and Platelets (PLT) count (**F**) change in different patterns of liver fibrosis over time.

Table 3
Baseline factors associated with the trajectory patterns of liver fibrosis.

	Univariable	*P* Value	Multivariable	*P* Value
	OR (95% CI)		OR (95% CI)	
APRI scores:				
Age	1.035 (1.007–1.063)	0.013		
$CD4^+$ cell count	0.998 (0.996–0.999)	0.026		
AST	1.023 (1.010–1.038)	0.001	1.012 (0.999–1.026)	0.054
ALT	1.010 (1.003–1.019)	0.008		
Platelets	0.985 (0.979–0.999)	< 0.001	0.988 (0.982–0.994)	< 0.001
WBC	0.642 (0.517–0.785)	< 0.001	0.835 (0.657–1.046)	0.128
γ-GT	1.012 (1.006–1.019)	< 0.001	1.010 (1.002–1.019)	0.015
Hb	0.985 (0.972–0.998)	0.025		
LYM	0.438 (0.271–0.686)	< 0.001		
Fib-4 scores:				
Age	1.262 (1.199–1.340)	< 0.001	1.322 (1.238–1.429)	< 0.001
Gender	6.595 (2.018–25.280)	0.003		
CD4 count	0.997 (0.996–0.999)	0.006		
Platelets	0.979 (0.972–0.985)	< 0.001	0.971 (0.961–0.979)	< 0.001
Hb	0.961 (0.947–0.975)	< 0.001	0.978 (0.954–1.002)	0.075
WBC	0.603 (0.486–0.735)	< 0.001		
γ-GT	1.006 (1.001–1.012)	0.022		
LYM	0.375 (0.233–0.582)	< 0.001		
GLu	1.380 (1.142–1.750)	0.003		
Baseline HBeAg status	0.514 (0.261–0.970)	0.046		
HBsAg seroclearance	0.504 (0.258–1.008)	0.047		

Notes: WBC, white blood cells; LYM, lymphocyte; AST, aspartate transaminase; ALT, alanine transaminase; Hb, hemoglobin; γ-GT, γ-glutamyl transferase.

($P = 0.003$), blood glucose ($P = 0.003$), and the presence of HBsAg ($P = 0.047$) and HbeAg status at baseline ($P = 0.046$).

After adjusting for potential confounding variables, multivariable analysis revealed that liver fibrosis trajectory pattern B, as assessed by APRI scores, was significantly associated with higher γ-GT (OR = 1.010, 95% CI: 1.002–1.019, $P = 0.015$) and lower platelet count (OR = 0.988, 95% CI: 0.982–0.994, $P < 0.001$). Additionally, Fib-4 scores for pattern B were associated with older age (OR = 1.332, 95% CI: 1.238–1.429, $P < 0.001$) and lower platelet count (OR = 0.971, 95% CI: 0.961–0.979, $P < 0.001$).

The changes in the median $CD4^+$ count, LYM count, WBC count, and platelet count in two patterns are shown in Fig. 2C–F. Whether we use APRI or FIB-4 scores as indicators of liver fibrosis, the trends of the above four clinical assessments in the pattern A group were similar and higher than those in the pattern B group.

DISCUSSION

This longitudinal study ultimately enrolled 333 participants with HIV-HBV co-infection undergoing long-term ART, with clinical and laboratory data collected at baseline, 12, 24, 36, 48, and 60 months. Using APRI and Fib-4 non-invasive scores to construct group-based trajectory models, we found two distinct patterns in liver fibrosis progression among HIV/HBV co-infected individuals receiving ART. The majority of patients (pattern A) maintained stable and low levels of fibrosis, while those initially with severe fibrosis (pattern B) showed regression in liver fibrosis over long-term ART containing TDF. Moreover, regardless of the non-invasive scores used, participants in pattern A were younger and had higher $CD4^+$ cell counts, higher lymphocyte cell counts, higher white blood cell counts, and lower platelet counts at baseline compared to those in pattern B.

The previous studies on the trajectory pattern of liver fibrosis in patients with HIV-HBV co-infection after ART treatment have yielded conflicting results. Some researchers have reported a regression pattern of liver fibrosis evolution. However, most of these studies had short follow-up periods (≤ 3 years) and only assessed liver fibrosis at baseline and endpoint (Audsley et al., 2016; Grant et al., 2019; Vinikoor et al., 2017; Stockdale et al., 2015). To better understand the trajectory changes of liver fibrosis, longer follow-up times and consecutive assessments of liver fibrosis at multiple time points are necessary for HIV-HBV co-infected patients treated with long-term TDF-containing ART, as suggested by some researchers (Boyd and Lacombe, 2016). However, contradictory results have also been observed in some longitudinal cohort studies with longer follow-up periods (Boyd et al., 2017; Ding et al., 2017; Iacob et al., 2022; Sterling et al., 2021; Malagnino et al., 2019). These inconsistent patterns of liver fibrosis outcomes observed in cohorts with longer follow-ups may be attributed to variations in sample size, population heterogeneity, as well as differences in treatment regimens and medication adherence.

Most previous research has focused on describing or comparing liver fibrosis at the beginning and end of studies, typically relying on the median fibrosis values without conducting detailed trajectory analyses for each participant over time. Exceptionally, one prior study identified population-level trajectories of liver fibrosis in an HIV/HBV co-infected cohort treated with TDF (Dezanet et al., 2021). This French research, involving 169 patients over a median follow-up of 7.6 years, utilized GBTMs to discern four distinct patterns of liver fibrosis evolution. The findings suggested that TDF-inclusive ART did not significantly improve liver fibrosis in co-infected individuals.

Since this trajectory analysis was based on a western TDF-treated co-infected cohort, we were inspired to conduct the first trajectory analysis in a Chinese TDF-treated co-infected cohort, with a follow-up of five years but double the sample size, to investigate liver fibrosis trajectories in China. Our cohort, in contrast to the French study, consisted of a younger Asian population, with a higher proportion initiating TDF-inclusive ART as their first antiviral therapy. These demographic differences might partially account for the variance between our findings and those of the French study. We identified two distinct improvement patterns of liver fibrosis, assessed via non-invasive markers of APRI and Fib-4 scores: a stable pattern with none or low fibrosis showing no progression (pattern A), and a pattern of regression from high fibrosis levels (pattern B). These improvement patterns have primarily been observed in the treatment of chronic hepatitis B mono-infection with TDF (Marcellin et al., 2013; Sun et al., 2020). Our study's results indicate that long-term TDF-inclusive ART is effective in mitigating liver fibrosis progression in Chinese individuals with HIV/HBV co-infection, aligning

with findings from other studies with varying follow-up durations (Audsley et al., 2016; Ding et al., 2017; Grant et al., 2019; Vinikoor et al., 2017; Stockdale et al., 2015).

Consistent with previous studies (Boyd et al., 2017; Ding et al., 2017; Malagnino et al., 2019), our findings indicate that a higher baseline $CD4^+$ cell count, being female, and younger age are protective factors associated with better improvement in maintaining low levels of liver fibrosis in co-infected individuals after long-term ART containing TDF. We also observed that individuals classified as pattern B showed higher levels of ALT and lower platelet counts, which aligns with findings from another co-infection study (Audsley et al., 2016). Additionally, our multi-variable analysis suggests that Hb, WBC, γ-GT, and AST levels at baseline may serve as potential biomarkers for predicting the trajectory pattern of liver fibrosis.

This longitudinal cohort study had a key strength: every participant in our cohort had more than six complete follow-up data points for two non-invasive measurements. This robust dataset allowed us to perform trajectory analysis at multiple time points. However, several limitations need to be addressed. First, due to the retrospective nature of this longitudinal study, the medical records lacked data on certain factors that could potentially influence the score parameters, such as HIV-associated opportunistic infections, cancers, and, especially, the status of HCV infection. Second, although Fib-4 and APRI are commonly used non-invasive indicators for assessing liver fibrosis, their applicability and interpretation in co-infected patients still need further refinement (Sterling et al., 2020). Third, a longer follow-up duration is required to observe the liver fibrosis trajectory in individuals belonging to pattern B. In the end, due to reliance on medical records as the primary data source, we were unable to obtain information on alcohol consumption and clinical outcomes.

CONCLUSIONS

In conclusion, we conducted the first-ever trajectory analyses on liver fibrosis in a Chinese TDF-treated cohort which was one of the largest HIV/HBV co-infection cohorts in Asia. Two distinct patterns of liver fibrosis improvement were observed in our study: most patients demonstrated pattern A, maintaining stable and low levels of fibrosis, while those with severe fibrosis at entry displayed pattern B, showing regression in liver fibrosis during long-term TDF-contained ART. Our findings support the fact that long-term TDF-containing ART has the potential to alleviate liver fibrosis in co-infected individuals. However, it is worth noting that while liver fibrosis levels consistently decreased in individuals of pattern B, they did not fully return to normal levels. Therefore, it is crucial to implement enhanced long-term monitoring and consider timely adjustment of clinical treatment based on individual circumstances for this population.

MATERIALS AND METHODS

Study population

This longitudinal cohort study recruited 507 patients with HIV/HBV co-infection who were undergoing ART at Beijing Youan Hospital between 2005 and 2022. Patients were followed up with every six to twelve months, and data was collected annually during each visit. The inclusion criteria for this study were as follows: 1) enrollment of patients both infected with HIV and chronic HBV; 2) age >18 years; 3) history of TDF-based antiretroviral therapy (ART). We excluded patients with acute HBV infection, as well as those lacking baseline data or regular follow-up, as they were unable to provide the required five-year biochemical data, including age, platelet (PLT) count, alanine transaminase (ALT), and aspartate transaminase (AST), which are necessary for calculating APRI and Fib-4 scores. Finally, a total of 333 patients with HIV/HBV co-infection were included in this study (Fig. 1), all of them having complete APRI values available for assessing liver fibrosis at all time points, with no missing data.

Data collection

Demographical and laboratory data were collected from medical records during annual visits at baseline, 12, 24, 36, 48, and 60 months, respectively. Demographic data included age, gender, body mass index (BMI), HIV transmission route, and treatment regimen. Laboratory measurements included the following: platelet count, alanine transaminase (ALT), aspartate transaminase (AST), $CD4^+$ cell count, hepatitis B e antigen (HBeAg), hepatitis B surface antigen (HBsAg), HBV DNA, HIV RNA viral load, white blood cells (WBC) count, lymphocyte count (LYM), hemoglobin (Hb), γ-glutamyl transferase (γ-GT), blood glucose, low density lipoprotein (LDL), and alkaline phosphatase (ALP).

Liver fibrosis assessment

APRI and Fib-4 scores were used to evaluate liver fibrosis in this study. The APRI score was calculated using the formula: [(AST/upper normal limit of AST) × 100/platelet count], while the Fib-4 score was calculated as [age (years) × AST (U/L)]/[platelet count (10^9/L) × ALT1/2 (U/L)] (Sterling et al., 2006). The presence of liver fibrosis was defined by using the cutoff of APRI >0.5 and Fib-4 ≥ 1.45 (Mendeni et al., 2011).

Statistical analysis

Group-based trajectory models (GBTMs) were employed to identify distinct patterns of liver fibrosis changes among patients receiving 5-year ART. Patients were assigned to specific trajectories based on the posterior probability of belonging to that trajectory, with an average posterior probability exceeding 0.7 considered a good model fit (Nagin, 1999). The Bayesian information criterion (BIC) was utilized to determine the optimal number of trajectory subgroups. The STATA package 'traj' was used to fit the model, employing a beta distribution for APRI and Fib-4 scores (Elmer et al., 2018). GBTMs consistently identified two distinct patterns of liver fibrosis evolution for the APRI and Fib-4 scores based on the final models. Differences in continuous variables between trajectory patterns were evaluated using the Mann-Whitney *U* test. Categorical variables were analyzed using the Chi-square test or Fisher exact test, as appropriate.

Data were presented as counts and percentages for categorical variables and as medians and interquartile ranges (IQRs) for continuous variables. Univariate and multivariate logistic regression models were employed to explore the association between baseline factors and trajectory grouping results. Multivariate logistic regression utilized a stepwise selection process, with only factors demonstrating a $P < 0.05$ in the univariate logistic regression included. A significance level of 0.05 was employed for the stepwise selection procedure and all statistical analyses. All analyses were performed using R Studio Version 4.2.1 (R Foundation for Statistical Computing, Vienna, Austria) and STATA 17 (College Station, Texas, USA). A two-tailed $P < 0.05$ was considered statistically significant.

DATA AVAILABILITY

All the data generated during the current study are included in the manuscript.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Beijing Youan Hospital Research Ethics Committee (with approved No. 2014-024 and 2018-025). The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

Qingrong Zhang: Investigation, formal analysis, Writing-original draft. Lijun Sun: Conceptualization, data curation. Yuxuan Liang: Data curation, validation. Wenlu Zou: Methodology, project administration. Jingtao Huang: Project administration, resources. Yuan Zhang: Software, validation. Yi Jin: Supervision, validation. Na Zhou: Supervision, validation. Jiangzhu Ye: Visualization. Huachun Zou: Writing-review & editing. Hao Wu: Writing-review & editing. Tong Zhang: Writing-review & editing. Bin Su: Funding acquisition. Taiyi Jiang: Funding acquisition, writing-review & editing, Data curation. Haitao Chen: Funding acquisition, writing-review & editing, supervision.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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