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Fatigue related to the severity of liver inflammation in young and middle-aged but not elderly patients with chronic liver disease：A cross-sectional study

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Abbreviations: AFLD, alcoholic liver disease; AIH, autoimmune hepatitis; AILDs, autoimmune liver diseases;AKP, alkaline phosphatase; ALB, albumin; ALD, alcoholic liver disease; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; CHB, chronic hepatitis B; CI, confidence interval; CLD, chronic liver disease; CP,

child Pugh; DBP, diastolic blood pressure; FPG, fasting plasma glucose; GGT, gamma-glutamyl- transferase; HbA1c, glycated haemoglobin; HDL-c/LDL-c, high/low-density lipoprotein cholesterol; HRQL, health-related quality of life; NAFLD, non-alcoholic fatty liver disease; OR, odds ratio; PBC, primary biliary cholangitis; PSC, primary sclerosing cholangitis; PSM, propensity score matching; SBP, systolic blood pressure; STB, serum total bilirubin; TC, total cholesterol; TG, triglycerides.

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Ethics Approval Statement: This study was performed according to the ethical guidelines of the Declaration of Helsinki and was approved by the Ethics Committee of the Affiliated Hospital of Hangzhou Normal University (approval number: 2020(02)-KS-022).

Patient Consent Statement: As this was an observational retrospective study, the requirement for informed consent was waived by the Ethics Committee.

Abstract

Objectives: Fatigue is common in patients with chronic liver disease; however, its pathogenesis is unclear. This study aimed to provide insights into the pathogenesis of chronic liver disease-related fatigue by assessing the relationship between fatigue and the degree of inflammation in chronic liver disease.

Design: We performed a cross-sectional study of 1,374 patients with pathologically proven chronic liver disease diagnosed at the Affiliated Hospital of Hangzhou Normal University in Hangzhou, China.

Setting: Primary single-centre study

Participants:1,374 patients with liver biopsy proven chronic liver disease.

Interventions: The patients were divided into fatigue and non-fatigue groups according to the Chronic Liver Disease Questionnaire. Propensity score matching was used to match the baseline features of the patients in the two groups.

Primary and secondary outcome measures: Liver steatosis, ballooning, inflammation and fibrosis were measured according to the pathological results of liver biopsy. Fatigue was measured using the Chronic Liver Disease Questionnaire. Propensity score

Results: Of the 1,374 patients, 262 (19.67%) experienced fatigue. There were 242 and 482 patients with and without fatigue, respectively, who were successfully matched for sex, age, and classification of chronic liver disease by propensity score matching. After matching, the fatigue group showed higher liver enzyme levels, inflammation grades, and fibrosis stages than the non-fatigue group (P<0.05). Multivariate analysis showed that age (odds ratio: 2.026; P=0.003), autoimmune liver disease (OR: 2.749; P=0.002), and active inflammation (odds ratio: 1.587; P=0.003) were independent risk factors for fatigue after adjusting for confounders. The odds ratio of the risk for fatigue increased in a stepwise manner with

increasing inflammation grade in young and middle-aged patients (P<0.05). This tendency was not observed in elderly patients (P>0.05).

Conclusion: Chronic liver disease patients were burdened by fatigue, which increased progressively with rising liver inflammation severity in young and middle-aged rather than elderly patients.

Strengths and limitations of this study

- Strength: To the best of our knowledge, this is the first study to use liver histopathology to explore the relationship between fatigue and the severity of liver inflammation.
- Limitation: This was a retrospective study design in which we diagnosed fatigue based on responses to the Chronic Liver Disease Questionnaire.
- Limitation: The cross-sectional study design meant that we could not determine a causal relationship between the severity of inflammation and fatigue.

Keywords: hepatology, histopathology, health management

Introduction

Chronic liver disease (CLD) affects approximately 1.5 billion people worldwide. The prevalence of CLD is rising rapidly owing to the ongoing impact of viral hepatitis and the rapidly increasing incidence of non-alcoholic fatty liver disease (NAFLD).[1-3] Fatigue is commonly experienced by patients with CLD and significantly impairs their quality of life.[4] The findings of previous studies suggest that the impact of fatigue on patients with CLD can be substantial,[5] with patients reporting that it interferes with several aspects of their lives, including physical activities, family life, and job performance.[6] These issues add to the

personal and societal burdens associated with CLD and indirectly contribute to financial costs. In addition to affecting quality of life, CLD-related fatigue has a negative impact on survival. In a 4-year follow-up study of patients with primary biliary cholangitis (PBC), fatigue was associated with poor outcomes, as patients with higher fatigue scores at the start of the study period had significantly lower survival rates.[7]

It is difficult to characterize, define, and treat fatigue because it encompasses a complex interaction between biological, psychosocial, and behavioural processes.[8,9] Our understanding of CLD-related fatigue is still incomplete and its pathogenesis remains unclear. The most common view is that there are peripheral pathways between the liver and the brain that, when activated, lead to changes in neurotransmission within the brain and the development of disease-related behaviours, including fatigue.[10-12] Better understanding of the relationship between fatigue and liver histology features in different CLD populations may provide further evidence of the mechanism underlying liver disease-related fatigue and facilitate the development of specific and appropriate treatment for it.

In this study, we explored the risk factors for fatigue in CLD by comparing the clinical and histological features of patients with and without fatigue using a large cohort of patients with biopsy-proven CLD. In addition, we analysed the correlation between the severity of liver histology features and CLD-related fatigue in different CLD populations.

Methods

Patients and Study Design

This was a cross-sectional study of patients with pathologically proven CLD, including NAFLD, alcoholic liver disease (ALD), PBC, primary sclerosing cholangitis (PSC), autoimmune hepatitis (AIH), chronic hepatitis B (CHB), and CHB with fatty liver, diagnosed at the Affiliated Hospital of Hangzhou Normal University in Hangzhou, China between 2011

and 2021. Patients with the following conditions were excluded: psychiatric or mental disorders, or cognitive difficulties that could hinder reliable description of symptoms; CLD combined with any other chronic disorders that may affect fatigue; and causes of CLD other than NAFLD, ALD, autoimmune liver diseases (AILDs) (including PBC, PSC, AIH), and CHB.The included patients were divided into fatigue and non-fatigue groups according to the presence or absence of fatigue.

Clinical Examination and Biochemical Analysis

The clinical examination consisted of a physical examination and a health habit assessment, which were performed by professional physicians. Diastolic blood pressure (DBP), systolic blood pressure (SBP), height, and body weight were measured according to standard protocols. Body mass index (BMI) was calculated as weight (kg) divided by height (m) squared (kg/m^2). Blood samples were collected after 8 h of fasting, and the biochemical tests performed included measurement of fasting plasma glucose (FPG), triglyceride (TG), total cholesterol (TC), high/low-density lipoprotein cholesterol (HDL-c/LDL-c), alanine aminotransferase (ALT), gamma-glutamyl-transferase (GGT), aspartate aminotransferase (AST), alkaline phosphatase(AKP), serum total bilirubin (STB), albumin (ALB), and glycated hemoglobin (HbA1c) levels. The biochemical tests were performed using an automated biochemical analyser according to the manufacturer's instructions (OLYMPUS, Japan AU5821+ISE; OLYMPUS).

Histological Analysis

All liver biopsies were reassessed by three professional histopathologists who were blinded to patient details. The steatosis score (positive if >5%, according to the Brunt classification [S0-S3]), fibrosis stage (based on a meta-analysis of histological data on viral hepatitis score

[S0-S4]), ballooning score (S0-S2), and inflammation grades (G0-G4) of the patients were evaluated.[12-14]. Fibrosis stage ≥2, inflammation grade ≥2, and steatosis score ≥2 were defined as indicative of significant liver fibrosis, active inflammation, and severe steatosis, respectively.

Fatigue Assessment

Fatigue was assessed by a professional physician within 1 week prior to liver biopsy using the Chronic Liver Disease questionnaire.[15]

Statistical Analysis

Continuous variables were compared between the two groups using Student's *t*-test or the Mann–Whitney *U* test. Categorical variables were compared using the Chi-square test. Propensity score matching (PSM) was performed in a ratio of 1:2 and with a calliper value of 0.2 to balance age, sex, and CLD classification between the two patient groups. Univariate and multivariate logistic regression analysis was conducted to analyse the factors that contribute to fatigue. We estimated adjusted odds ratios (ORs) and relevant 95% confidence intervals (CIs) using a parametric proportional hazard model. SPSS 26.0 (IBM, Armonk, NY, USA) was used for statistical analyses. Statistical significance was set at $P<0.05$.

Ethical Considerations

This study was performed according to the ethical guidelines of the Declaration of Helsinki and was approved by the Ethics Committee of the Affiliated Hospital of Hangzhou Normal University (approval number: 2020(02)-KS-022). As this was an observational retrospective study, the requirement for informed consent was waived by the Ethics Committee.

Results

Comparison of the Clinical and Histological Features of Chronic Liver Disease Patients with and without Fatigue

A total of 1,374 patients with biopsy-proven CLD were included in this study. Of these, 262 (19.07%) patients had symptoms of fatigue, while 1,112 (80.93%) had no fatigue. The patients with fatigue were older, had lower BMI and higher HDL-c and GGT levels than the patients without fatigue (P<0.05). There were significant differences between the proportions of patients with NAFLD, CHB with fatty liver, CHB, AFLD, and AILDs in the two groups (respectively, fatigue group: 16.03%, 17.94%, 52.67%, 6.11%, and 7.25% vs non-fatigue 18.44%, 25.45%, 47.75%, 6.20%, and 2.16%; P<0.05). There were no significant differences in SBP, BMI, FPG, LDL-c, TG, TC, STB, ALT, AST, AKP, and ALB levels between the fatigue and non-fatigue groups (P>0.05) (Table 1).

After PSM, 242 patients with fatigue and 482 patients without fatigue were successfully matched, and there were no statistically significant differences between the two groups in terms of sex, age, SBP, BMI, FPG, LDL-c, TG, TC, STB, AKP, and ALB levels. In addition, there were no differences between the proportions of patients with NAFLD, CHB with fatty liver, CHB, AFLD, and AILDs in the two groups (P>0.05) (Table 1). The fatigue group had higher ALT, AST, and GGT levels than the non-fatigue group. Further comparison of the histological features of the two groups is shown in Figure 1. The inflammation grades and liver fibrosis stages of the patients with fatigue were significantly higher than those of the patients without fatigue (P<0.05). There was no difference in hepatic steatosis and ballooning scores between the two groups (P>0.05) (Figure 1, A-D).

Distribution and Risk Factors of Fatigue in Patients with Chronic Liver Disease

The prevalence of fatigue significantly increased with age (R=0.087, *P*=0.001) (Figure 2a). There was no correlation between fatigue and BMI and sex (*P*>0.05) (Figures 2b and c). The prevalence of fatigue among patients with different types of CLD varied. For patients with AILDs, the prevalence of fatigue was 44.19%, which is more than twice that of patients with other CLDs (*P*<0.001) (Figure 2d). The prevalence of fatigue significantly increased with the degree of liver inflammation and fibrosis stage (*P*<0.001) (Figures 2e and f).

Univariate and multivariate analyses were performed to define the association between fatigue using clinical and histological features (Table 2). Univariate analysis showed that old age (OR=2.122, 95% CI: 1.379-3.267, *P*=0.001), AILDs (OR=3.545, 95% CI: 1.911-6.574, *P*<0.001), elevated GGT level (OR=1.356, 95%CI: 1.012-1.816, *P*=0.042), active inflammation (OR=1.768, 95%CI: 1.329-2.353, *P*<0.001), and advanced fibrosis stage (OR=1.743, 95%CI: 1.282-2.370, *P*<0.001) were risk factors for fatigue in CLD. Further multivariate analysis indicated that old age (OR= 2.026, 95% CI: 1.274-3.221, *P*=0.003), AILDs (OR=2.749, 95% CI: 1.446-5.226, *P*=0.002), and active inflammation (OR= 1.587, 95% CI: 1.164-2.164, *P*=0.003) were independent risk factors for fatigue.

Analysis of the Correlation between Histological Features and Fatigue in Chronic Liver Disease

Multivariate analysis was performed to explore the correlation between fatigue and the severity of histological features. Two different models were utilized to estimate the ORs for different outcomes. After adjusting for age, sex, BMI, hypertension, type 2 diabetes mellitus, disease classification, ALT level, AST level, and GGT level, the OR for the risk of fatigue increased in a stepwise manner from inflammation grades G0-G1 (as a reference) and G2 (OR=1.609, 95% CI: 1.085-2.386, *P*=0.018) to G3 (OR=1.745, 95% CI: 1.019-2.986,

P=0.042) (Table 3). The severity of steatosis, ballooning, and fibrosis were not associated with fatigue (*P*>0.05).

Sensitivity Analysis

Subgroup analysis of the risk of fatigue in CLD showed a significant association between fatigue and increasing severity of liver inflammation among patients aged < 60 years old without AILDs (*P*<0.05). However, the severity of inflammation was not associated with fatigue among patients > 60 years old or with AILDs (*P*>0.05) (Table 4).

Discussion

Fatigue is a critical component of CLD.[11] The findings of the present study indicate an association between fatigue and liver inflammation. In the present study, CLD patients with fatigue had significantly higher inflammation grades and liver fibrosis stages than patients without fatigue. In addition, multivariate analysis showed that age, AILDs, and active inflammation were independent risk factors for fatigue, and that the severity of liver inflammation was strongly associated with fatigue after adjustment for confounders. Further sensitivity analysis showed that this association was present in the young and middle-aged population of the present study but not in the elderly population.

The clinical and liver histological features of CLD-related fatigue have not been uniformly demonstrated.[16,17] In the present study, the CLD patients with fatigue were older, had lower BMI, and higher HDL-c and GGT levels than patients without fatigue. In addition, the fatigue group showed significantly higher inflammation grades and liver fibrosis stages than the non-fatigue group after PSM for age and sex (Table 1, Figure 1). The results of the present study are in-line with those of most studies that showed that old age, AILDs, and active inflammation are independent risk factors for fatigue (Table 2).[18-21] The

findings of the present study and previous studies indicate that early evaluation and intervention for fatigue are necessary for patients with CLD. The results of the present study also indicated that the risk of fatigue increased with the severity of inflammation, but not with the severity of hepatic steatosis, ballooning, and liver fibrosis (Table 3). Although the issue of fatigue in patients with CLD, including PBC, PSC, CHB, CHC, and NAFLD, has been extensively studied, the relationship between fatigue and the histological features of CLD remains controversial. Fatigue in NAFLD has been associated with inactivity and excessive daytime sleepiness but not with the severity of liver disease or insulin resistance.[16] However, a recent study indicated that the detection of lobular inflammation in biopsies is correlated with lower health-related quality of life (HRQL) in patients with NAFLD.[17] Data from clinical trials on chronic hepatitis B or C virus infection also support the dominant role of inflammation in fatigue. In these trials, viral elimination or suppression after antiviral therapy was associated with improved HRQL, which suggests an effect of inflammation on fatigue, whereas improvement of fibrosis did not affect HRQL.[22-24]

Further subgroup analysis in the present study revealed a significant association between fatigue and the severity of liver inflammation in patients <60 years old, but not in patients ≥60 years old (Table 4). Our findings suggest that the severity of liver inflammation may play a dominant role in fatigue in young and middle-aged patients with CLD, whereas age-related factors may play dominant roles in fatigue in elderly patients. Previous studies have shown that fatigue is a significant component of the clinical presentation of patients with AIH, often paralleling hepatic inflammation.[25] In line with previous research, the data of the present study showed that patients with AILDs had the highest prevalence of fatigue, and that AILDs was an independent risk factor for CLD-related fatigue. However, fatigue was not correlated with the severity of liver inflammation in patients with AILDs. This may be related to the

relatively small number of AILD cases in the present study. Thus, studies with larger samples are needed to confirm this finding.

The strength of this study is that it is the first study, to the best of our knowledge, in which the relationship between fatigue and the severity of liver inflammation in different CLD populations was explored using liver histopathology features. However, the limitations of this study should be noted as well. First, as this was a retrospective study, we diagnosed fatigue based on responses to the Chronic Liver Disease Questionnaire, which included questions on fatigue. The severity of fatigue cannot be defined; thus, we could only assess the relationship between the severity of inflammation and the presence or absence of fatigue, but could not clarify the relationship between the severity of liver inflammation and the severity of fatigue. Second, as this was a cross-sectional study, we could not determine the causal relationship between the severity of inflammation and fatigue. Further studies with longitudinal cohorts are needed to confirm the effects of the severity of inflammation on fatigue in patients with CLD.

In conclusion, the present study demonstrates that fatigue is correlated with the severity of liver inflammation in young and middle-aged patients with CLD. However, this correlation was not observed in elderly patients. These findings contradict the perception that fatigue is not associated with the severity of liver disease. Since age is an important factor that influences fatigue, our findings highlight the need for age stratification during the evaluation and treatment of CLD patients with fatigue.

Author contributions

Research idea and study design: Junping Shi; data acquisition: Jing Liu, Xiying Gong, Shiyi Liu, Yanming Jiang, Geli Zhu, Xiaojie Ma, Jie Wang, Xiaoping Ye, Yidan Gao, Dian Wang, Gongying Chen; data analysis/interpretation:Jing Liu, Haifeng Lv; statistical analysis: Jing

Liu, Haifeng Lv; supervision or mentorship: Junping Shi. Each author contributed important intellectual content during manuscript drafting and revision, agreed to be personally accountable for the individual's contributions, and ensured questions about the accuracy or integrity of any portion of the work, even one in which the author was not directly involved, are appropriately investigated and resolved.

References

1 Moon AM, Singal AG, Tapper EB. Contemporary epidemiology of chronic liver disease and cirrhosis. *Clin Gastroenterol Hepatol* 2020;18:2650–66.

2 Huang DQ, El-Serag HB, Loomba R. Global epidemiology of NAFLD-related HCC: trends, predictions, risk factors and prevention. *Nat Rev Gastroenterol Hepatol* 2021;18:223–38.

3 Locarnini S, Hatzakis A, Chen DS et al. Strategies to control hepatitis B: public policy, epidemiology, vaccine and drugs. *J Hepatol* 2015;62;Supplement:S76–S86.

4 Swain MG, Jones DEJ. Fatigue in chronic liver disease: new insights and therapeutic approaches. *Liver Int* 2019;39:6–19.

5 Jopson L, Dyson JK, Jones DE. Understanding and treating fatigue in primary biliary cirrhosis and primary sclerosing cholangitis. *Clin Liver Dis* 2016;20:131–42.

6 Cauch-Dudek K, Abbey S, Stewart DE et al. Fatigue in primary biliary cirrhosis. *Gut* 1998;43:705–10.

7 Jones DE, Bhala N, Burt J et al. Four year follow up of fatigue in a geographically defined primary biliary cirrhosis patient cohort. *Gut* 2006;55:536–41.

8 Bower JE. Cancer-related fatigue--mechanisms, risk factors, and treatments. *Nat Rev Clin Oncol* 2014;11:597–609.

9 Gerber LH, Weinstein AA, Mehta R et al. Importance of fatigue and its measurement in chronic liver disease. *World J Gastroenterol* 2019;25:3669–83.

10 D'Mello C, Swain MG. Liver-brain inflammation axis. *Am J Physiol Gastrointest Liver Physiol* 2011;301:G749–61.

11 Kośnik A, Wójcicki M. Fatigue in chronic liver disease patients: prevalence, pathophysiology, and management. *Prz Gastroenterol* 2022;17:21–7.

12 Brunt EM, Janney CG, Di Bisceglie AM et al. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. *Am J Gastroenterol* 1999;94:2467–74.

13 Bedossa P, Poynard T. An algorithm for the grading of activity in chronic hepatitis C. The METAVIR Cooperative Study Group. *Hepatology* 1996;24:289–93.

14 Batts KP, Ludwig J. Chronic hepatitis. An update on terminology and reporting. *Am J Surg Pathol* 1995;19:1409–17.

15 Younossi ZM, Guyatt G, Kiwi M et al. Development of a disease specific questionnaire to measure health related quality of life in patients with chronic liver disease. *Gut* 1999;45:295–300.

16 Newton JL, Jones DE, Henderson E, et al. Fatigue in non-alcoholic fatty liver disease (NAFLD) is significant and associates with inactivity and excessive daytime sleepiness but not with liver disease severity or insulin resistance. *Gut* 2008;57:807–13.

17 Huber Y, Boyle M, Hallsworth K, et al. Health-related quality of life in nonalcoholic fatty liver disease associates with hepatic inflammation. *Clin Gastroenterol Hepatol* 2019;17:2085–2092.e1.

18 Swain MG. Fatigue in liver disease: pathophysiology and clinical management. *Can J Gastroenterol* 2006;20:181–8.

19 Schramm C, Wahl I, Weiler-Normann C, et al. Health-related quality of life, depression, and anxiety in patients with autoimmune hepatitis. *J Hepatol* 2014;60:618–24.

20 Bozzini AB, Neder L, Silva CA et al. Decreased health-related quality of life in children and adolescents with autoimmune hepatitis. *J Pediatr (Rio J)* 2019;95:87–93.

21 Finsterer J, Mahjoub SZ. Fatigue in healthy and diseased individuals. *Am J Hosp Palliat Care* 2014;31:562–75.

22 Younossi ZM, Stepanova M, Janssen HLA, et al. Effects of treatment of chronic hepatitis B virus infection on patient-reported outcomes. *Clin Gastroenterol Hepatol* 2018;16:1641–1649.e6.

23 Younossi ZM, Stepanova M, Afdhal N, et al. Improvement of health-related quality of life and work productivity in chronic hepatitis C patients with early and advanced fibrosis treated with ledipasvir and sofosbuvir. *J Hepatol* 2015;63:337–45.

24 Younossi ZM, Stepanova M, Jacobson IM, et al. Sofosbuvir and velpatasvir with or without voxilaprevir in direct-acting antiviral-naïve chronic hepatitis C: patient-reported outcomes from POLARIS 2 and 3. *Aliment Pharmacol Ther* 2018;47:259–67.

25 Russell AS, Gulliver WP, Irvine EJ et al. Quality of life in patients with immune-mediated inflammatory diseases. *J Rheumatol Suppl* 2011;88:7–19.

TABLE 1 Comparison of clinical and laboratory characteristics between patients with and without fatigue in CLD

Variables	Before propensity score matching			After propensity score matching(1:2)		
	Fatigue	No fatigue	*P* Value	Fatigue	No fatigue	*P* Value
n	262	1112		242	484	
Male(%)	175(66.79)	785(70.59)	0.228‡	167(69.01)	337(69.63)	0.527‡
Age (year)	45.30 ±11.12	41.36 ±11.33	<0.001	44.02 ± 10.48	42.46 ± 11.26	0.071
BMI(kg/m^2)	24.05 ±3.86	24.82 ±4.62	0.032	24.26 ± 3.88	24.18 ±4.02	0.840
BP(S)(mmHg)	124.33 ±15.26	125.70 ±15.52	0.204	123.86 ± 15.43	125.96 ± 16.01	0.098
FPG (mmol/L)	5.56 ± 1.66	5.65 ±1.54	0.493	5.56 ± 1.65	5.61 ± 1.40	0.718
HbA1c(%)	6.18 ± 1.54	6.12 ± 1.52	0.823	6.24 ±1.61	6.09 ± 1.52	0.639
TG (mmol/L)	1.23(0.95-1.85)	1.08(1.55-2.18)	0.793†	1.19(0.93-1.84)	1.31(0.90-2.00)	0.543
TC (mmol/L)	4.74 ± 1.20	4.68 ± 1.11	0.504	4.67 ± 1.16	4.63 ± 0.95	0.733
LDL-c (mmol/L)	2.76 ± 0.88	2.75 ± 0.82	0.875	2.72 ± 0.88	2.70 ± 0.75	0.788
HDL-c(mmol/L)	1.30 ± 0.36	1.24 ± 0.33	0.035	1.28 ± 0.35	1.25 ± 0.35	0.298
STB(μmmol/l)	16.65(12.82-22.45)	16.20(12.90-21.10)	0.511†	16.60(12.85-22.00)	15.50(12.50-20.80)	0.153†
ALT (U/L)	51.00(32.00-83.00)	49.00(30.00-80.00)	0.432†	51.00 (32.00-82.00)	41.00(26.00-66.00)	<0.001†
AST (U/L)	35.00(26.00-52.00)	33.00(25.00-49.00)	0.093†	34.00 (26.00-51.00)	28.00(23.00-41.00)	<0.001†
GGT(U/L)	37.00(23.00-77.00)	33.00(20.00-60.00)	0.005†	35.00(21.00-66.00)	27.00(17.00-49.00)	<0.001†
AKP(U/L)	110.00(88.00-137.00	108.00(87.00-136.00)	0.650†	107.00(87.0-129.00)	106.00(83.00-133.00)	0.797†
ALB (g/L)	44.08 ± 4.15	44.51 ± 5.85	0.297	4437 ± 3.95	44.70 ± 5.77	0.460
CLD Category						
NAFLD n(%)	42(16.03)	205(18.44)	<0.001‡	42(17.36)	70(14.46)	0.190‡
CHB with fatty liver n(%)	47(17.94)	283(25.45)		47(19.42)	103(21.28)	
CHB n(%)	138(52.67)	531(47.75)		137(56.61)	254(52.78)	
AFLD n(%)	16(6.11)	69(6.20)		12(4.96)	46(9.51)	
AILDs n(%)	19(7.25)	24(2.16)		4(1.65)	11(2.27)	

*P-value calculated using the χ^2 test.

†P-value calculated using the Mann–Whitney U-test.

Abbreviations: Data are expressed as mean ± standard deviation or median (interquartile range).

Abbreviations: AFLD, alcoholic fatty liver disease; AILD, autoimmune liver disease; ALB, albumin; AKP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; BP, blood pressure; CHB, chronic hepatitis B; CLD, chronic liver disease; DBP, diastolic blood pressure; FPG, fasting plasma glucose; GGT, gamma-glutamyl-transferase; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; SBP, systolic blood pressure; STB, serum total bilirubin; SUA, serum uric acid; TC, total cholesterol; TG, triglycerides.

Table 2. Univariate and multivariable regression analysis of risk factors for fatigue in chronic liver disease

Variables	Univariate Analysis		Multivariate Analysis	
	OR (95% CI)	*P* Value	OR (95% CI)	*P* Value
Old Age				
No	Ref.		Ref.	
Yes	2.122(1.379-3.267)	0.001	2.026(1.274-3.221)	0.003
Male				
No	Ref.			
Yes	0.838(0.628-1.117)	0.228	--	--
AILDs				
No	Ref.			
Yes	3.545(1.911-6.574)	<0.001	2.749(1.446-5.226)	0.002
Metabolic factors				
Obesity				
No	Ref.			
Yes	0.813(0.591-1.121)	0.207	--	--
Hypertension				
No	Ref.			
Yes	0.735(0.536-1.008)	0.056	--	--
T2DM				
No	Ref.			
Yes	0.694(0.436-1.103)	0.122	--	--
Hypertriglyceridemia				
No	Ref.			
Yes	0.804(0.577-1.121)	0.198	--	--
Hypercholesterolemia				
No	Ref.			
Yes	1.448(0.974-2.153)	0.068	--	--
Hyperuricemia				
No	Ref.			
Yes	0.835(0.570-1.222)	0.353	--	--
High LDL-c				
No	Ref.			
Yes	0.943(0.675-1.316)	0.728	--	--
Low HDL-c				
No	Ref.			
Yes	1.104(0.764-1.569)	0.599	--	--
Liver enzymes				
Elevated ALT				
No	Ref.			
Yes	1.164(0.662-2.441)	0.306	--	--
Elevated AST				
No	Ref.			
Yes	1.205(0.906-1.602)	0.199	--	--
Elevated GGT				
No	Ref.			

Yes	1.356(1.012-1.816)	0.042	--	--
Histopathology				
Severe ballooning				
No	Ref			
Yes	1.049(0.606-1.815)	0.865	--	--
Severe steatosis				
No	Ref			
Yes	0.850(0.488-1.482)	0.567	--	--
Active inflammation				
No	Ref.			
Yes	1.768(1.329-2.353)	<0.001	1.587(1.164-2.164)	0.003
Advanced fibrosis				
No	Ref			
Yes	1.743(1.282-2.370)	<0.001	--	--

Abbreviations: AILDs, autoimmune liver diseases; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; T2DM, type 2 diabetes mellitus; OR, odds ratio; CI, confidence intervals.

Table 3 Odds ratio of liver histological severity for fatigure in chronic liver disease

	Crude		Model 1		Model 2	
	OR(95% CI)	*P* Value	OR(95% CI)	*P* Value	OR(95% CI)	*P* Value
Steatosis score						
S0	Ref.		Ref.		Ref.	
S1-2	1.205(0.793-1.833)	0.383	1.204(0.787-1.841)	0.392	1.360(0.761-2.428)	0.299
S3	0.870(0.498-1.519)	0.624	0.918(0.522-1.615)	0.767	1.328(0.648-2.724)	0.493
Ballooning score						
S0	Ref.		Ref.		Ref.	
S1	0.665(0.405-1.090)	0.979	0.620(0.376-1.023)		0.942(0.521-1.703)	0.468
S2	1.007(0.581-1.747)	0.106	0.871(0.498-1.525)		1.310(0.632-2.716)	0.843
Inflammation grade						
G0-1	Ref.		Ref.		Ref.	
G2	1.618(1.190-2.200)	0.002	1.570(1.152-2.140)	0.004	1.609(1.085-2.386)	0.018
G≥3	2.170(1.486-3.169)	<0.001	2.014(1.372-2.056)	<0.001	1.745(1.019-2.986)	0.042
Liver fibrosis						
S0-1	Ref.		Ref.		Ref.	
S2	1.087(0.751-1.574)	0.657	1.006(0.692-1.462)	0.975	0.691(0.420-1.135)	0.145
S≥3	1.777(1.291-2.447)	<0.001	1.608(1.160-2.226)	0.004	1.371(0.897-2.096)	0.144

Note: Model 1 adjusted for age and sex; Model 2 was adjusted for model 1 plus body mass index hypertension, type 2 diabetes mellitus, alanine aminotransferase, aspartate aminotransferase, gamma-glutamyl-transferase and disease classification.

Abbreviations: OR,odds ratio; CI,confidence intervals.

Table 4. Sensitivity analysis in the presence of risk factors for fatigue in chronic liver disease

Variables	No.of Participants	OR(95%CI)	*P* Value
Age			
≥60 years	107		
G0-1	37	Ref.	
G2	45	1.813(0.668-4.919)	0.632
G≥3	25	2.848(0.937-8.659)	0.870
<60 years	1267		
G0-1	556	Ref.	
G2	517	1.566(1.134-2.163)	0.006
G≥3	194	1.989(1.324-2.988)	0.001
Elevated GGT			
Yes	451		
G0-1	141	Ref.	
G2	191	1.919(1.077-3.420)	0.027
G≥3	119	2.420(1.304-4.490)	0.005
No	923		
G0-1	452	Ref.	
G2	371	1.458(1.011-2.105)	0.019
G≥3	100	1.880(1.108-3.190)	0.044
AILDs			
Yes	43		
G0-1	9	Ref.	
G2	13	1.714(0.294-9.999)	0.549
G≥3	21	1.818(0.357-9.272)	0.472
No	1331		
G0-1	584	Ref	
G2	549	1.587(1.161-2.167)	0.004
G≥3	198	1.959(1.313-2.923)	0.001

Abbreviations: AILDs, autoimmune liver diseases; GGT, gamma-glutamyl-transferase; OR, odds ratio; CI, confidence intervals.

Figure 1. Comparison of the histopathological characteristics of chronic liver disease patients with and without fatigue. (**A**) Comparison of the inflammation grades of patients with CLD stratified according to the presence or absence of fatigue. (**B**) Comparison of the fibrosis stages of patients with CLD stratified according to the presence or absence of fatigue. (**C**) Comparison of hepatic steatosis scores of patients with CLD stratified according to the presence or absence of fatigue. (**D**) Comparison of the ballooning scores of the patients with CLD stratified according to the presence or absence of fatigue.

Figure 2. Prevalence of fatigue in different chronic liver disease populations. (**A**) Prevalence of fatigue stratified according to age. (**B**) Prevalence of fatigue stratified according to body mass index. (**C**) Prevalence of fatigue stratified according to sex. (**D**) Prevalence of fatigue stratified according to CLD classification. (**E**) Prevalence of fatigue stratified according to inflammation grade. (**F**) Prevalence of fatigue stratified according to fibrosis stage.

Abbreviations: CHB, chronic hepatitis B; NAFLD, non-alcoholic fatty liver disease; AFLD, alcoholic fatty liver disease; AILD, autoimmune liver diseases.

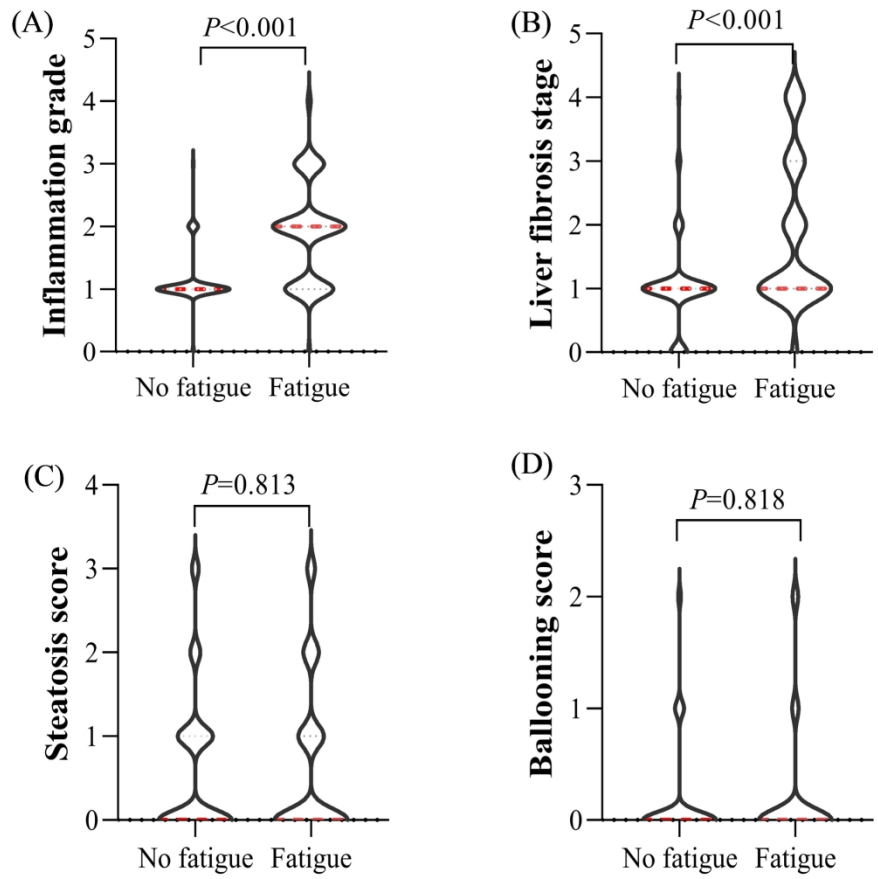

Figure 1. Comparison of the histopathological characteristics of chronic liver disease patients with and without fatigue. (A) Comparison of the inflammation grades of patients with CLD stratified according to the presence or absence of fatigue. (B) Comparison of the fibrosis stages of patients with CLD stratified according to the presence or absence of fatigue. (C) Comparison of hepatic steatosis scores of patients with CLD stratified according to the presence or absence of fatigue. (D) Comparison of the ballooning scores of the patients with CLD stratified according to the presence or absence of fatigue.

193x197mm (300 x 300 DPI)

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STROBE Statement—Checklist of items that should be included in reports of ***cross-sectional studies***

	Item No	Recommendation	Page No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	3
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	4-5
Objectives	3	State specific objectives, including any prespecified hypotheses	5
Methods			
Study design	4	Present key elements of study design early in the paper	5-6
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	5-7
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	5-6
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	6-7
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	*6-7*
Bias	9	Describe any efforts to address potential sources of bias	6-7
Study size	10	Explain how the study size was arrived at	5-6
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	7
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	7
		(*b*) Describe any methods used to examine subgroups and interactions	7
		(*c*) Explain how missing data were addressed	7
		(*d*) If applicable, describe analytical methods taking account of sampling strategy	7
		(*e*) Describe any sensitivity analyses	7
Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	8
		(b) Give reasons for non-participation at each stage	8
		(c) Consider use of a flow diagram	0
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	8-9
		(b) Indicate number of participants with missing data for each variable of interest	0
Outcome data	15*	Report numbers of outcome events or summary measures	8

Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	9-10
		(*b*) Report category boundaries when continuous variables were categorized	7
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	9-10
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	10
Discussion			
Key results	18	Summarise key results with reference to study objectives	10
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	12
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	10-12
Generalisability	21	Discuss the generalisability (external validity) of the study results	12
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	2

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

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Is fatigue related to the severity of liver inflammation in patients with chronic liver disease? A cross-sectional study

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Abbreviations: AFLD, alcoholic liver disease; AIH, autoimmune hepatitis; AILDs, autoimmune liver diseases; AKP, alkaline phosphatase; ALB, albumin; ALD, alcoholic liver disease; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; CHB, chronic hepatitis B; CI, confidence interval; CLD, chronic liver disease; CP, child Pugh; DBP, diastolic blood pressure; FPG, fasting plasma glucose; GGT, gamma-glutamyl-

transferase; HbA1c, glycated hemoglobin; HDL-c/LDL-c, high/low-density lipoprotein cholesterol; HRQL, health-related quality of life; NAFLD, non-alcoholic fatty liver disease; OR, odds ratio; PBC, primary biliary cholangitis; PSC, primary sclerosing cholangitis; PSM, propensity score matching; SBP, systolic blood pressure; STB, serum total bilirubin; TC, total cholesterol; TG, triglycerides.

Competing Interests Statement: The authors have no competing interests or conflicts of interest to disclose.

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Data Availability Statement: The datasets used and/or analyzed during this study are available from the corresponding author upon reasonable request.

Ethics Approval Statement: This study was performed according to the ethical guidelines of the Declaration of Helsinki and was approved by the Ethics Committee of the Affiliated Hospital of Hangzhou Normal University (approval number: 2020(02)-KS-022).

Patient Consent Statement: As this was an observational retrospective study, the requirement for informed consent was waived by the Ethics Committee.

Abstract

Objectives: Fatigue is common in patients with chronic liver disease; however, its pathogenesis is unclear. This study aimed to provide insights into the pathogenesis of chronic liver disease-related fatigue by assessing the relationship between fatigue and the degree of inflammation in chronic liver disease.

Design: We performed a cross-sectional study of 1,374 patients with pathologically proven chronic liver disease diagnosed at the Affiliated Hospital of Hangzhou Normal University in Hangzhou, China.

Setting: Primary single-center study

Participants: One thousand three hundred and seventy-four patients with liver biopsy-proven chronic liver disease.

Interventions: The patients were divided into fatigue and non-fatigue groups according to the Chronic Liver Disease Questionnaire. Propensity score matching was used to match the baseline features of the patients in the two groups.

Primary and secondary outcome measures: Liver steatosis, ballooning, inflammation, and fibrosis were measured according to the pathological results of liver biopsy. Fatigue was measured using the Chronic Liver Disease Questionnaire.

Results: Of the 1,374 patients, 262 (19.67%) experienced fatigue. There were 242 and 482 patients with and without fatigue, respectively, who were successfully matched for sex, age, and classification of chronic liver disease by propensity score matching. After matching, the fatigue group showed higher liver enzyme levels, inflammation grades, and fibrosis stages than the non-fatigue group (P<0.05). Multivariate analysis showed that age (odds ratio: 2.026; P=0.003), autoimmune liver disease (OR: 2.749; P=0.002), and active inflammation (odds ratio: 1.587; P=0.003) were independent risk factors for fatigue after adjusting for confounders. The odds ratio of the risk for fatigue increased in a stepwise manner with increasing inflammation

grade in young and middle-aged patients (P<0.05). This tendency was not observed in elderly patients (P>0.05).

Conclusion: Chronic liver disease patients were burdened by fatigue, which increased progressively with rising liver inflammation severity in young and middle-aged rather than elderly patients.

Strengths and limitations of this study

- This study was the first comprehensive assessment of the relationship between fatigue and the severity of liver inflammation in a large sample of liver biopsy-proven chronic liver disease.
- Propensity score matching was used to exclude the influence of gender, age, blood pressure, blood glucose, liver function, and composition ratio of patients with chronic liver disease.
- Since this is a retrospective study, some data that could contribute to the development of fatigue, namely plasma iron level, markers of thyroid gland function, and blood oxygen tension, are missing.
- An important limitation is the dichotomic division of the chronic liver diseases population into suffering and not-suffering from fatigue with no self-assessment of fatigue severity.
- Due to the cross-sectional study design, we have not been able determine the causal relationship between the severity of inflammation and fatigue.

Keywords: hepatology, histopathology, health management

Introduction

Chronic liver disease (CLD) affects approximately 1.5 billion people worldwide. The prevalence of CLD is rising rapidly owing to the ongoing impact of viral hepatitis and the rapidly increasing incidence of non-alcoholic fatty liver disease (NAFLD).[1-3] Fatigue is commonly experienced by patients with CLD and significantly impairs their quality of life.[4] The findings of previous studies suggest that the impact of fatigue on patients with CLD can be substantial,[5] with patients reporting that it interferes with several aspects of their lives, including physical activities, family life, and job performance.[6] These issues add to the personal and societal burdens associated with CLD and indirectly contribute to financial costs. In addition to affecting quality of life, CLD-related fatigue has a negative impact on survival. In a 4-year follow-up study of patients with primary biliary cholangitis (PBC), fatigue was associated with poor outcomes, as patients with higher fatigue scores at the start of the study period had significantly lower survival rates.[7]

It is difficult to characterize, define, and treat fatigue because it encompasses a complex interaction between biological, psychosocial, and behavioral processes.[8,9] Our understanding of CLD-related fatigue is still incomplete and its pathogenesis remains unclear. The most common view is that there are peripheral pathways between the liver and the brain that, when activated, lead to changes in neurotransmission within the brain and the development of disease-related behaviors, including fatigue.[10-12] Better understanding of the relationship between fatigue and liver histology features in different CLD populations may provide further evidence of the mechanism underlying liver disease-related fatigue and facilitate the development of specific and appropriate treatment for it.

In this study, we explored the risk factors for fatigue in CLD by comparing the clinical and histological features of patients with and without fatigue using a large cohort of patients with biopsy-proven CLD. In addition, we analyzed the correlation between the severity of liver histology features and CLD-related fatigue in different CLD populations.

Methods

Patients and Study Design

This was a cross-sectional study of patients with pathologically proven CLD, including NAFLD, alcoholic liver disease (ALD), PBC, primary sclerosing cholangitis (PSC), autoimmune hepatitis (AIH), chronic hepatitis B (CHB), and CHB with fatty liver, diagnosed at the Affiliated Hospital of Hangzhou Normal University in Hangzhou, China between 2011 and 2021. Patients with the following conditions were excluded: psychiatric or mental disorders, or cognitive difficulties that could hinder reliable description of symptoms; CLD combined with any other chronic disorders that may affect fatigue; causes of CLD other than NAFLD, ALD, autoimmune liver diseases (AILDs) (including PBC, PSC, AIH), and CHB; and past COVID-19 infection. The included patients were divided into fatigue and non-fatigue groups according to the presence or absence of fatigue.

Clinical Examination and Biochemical Analysis

The clinical examination consisted of a physical examination and a health habit assessment, which were performed by professional physicians. Diastolic blood pressure (DBP), systolic blood pressure (SBP), height, and body weight were measured according to standard protocols. Body mass index (BMI) was calculated as weight (kg) divided by height (m) squared (kg/m^2). Blood samples were collected after 8 h of fasting within 1 week before liver biopsy, and the biochemical tests performed included measurement of fasting plasma glucose (FPG), triglyceride (TG), total cholesterol (TC), high/low-density lipoprotein cholesterol (HDL-c/LDL-c), alanine aminotransferase (ALT), gamma-glutamyl-transferase (GGT), aspartate aminotransferase (AST), alkaline phosphatase(AKP), serum total bilirubin (STB), albumin (ALB), and glycated hemoglobin (HbA1c) levels. The biochemical tests were performed using

an automated biochemical analyzer according to the manufacturer's instructions (OLYMPUS, Japan AU5821+ISE; OLYMPUS).

Histological Analysis

All liver biopsies were reassessed by three professional histopathologists who were blinded to patient details. The steatosis score (positive if >5%, according to the Brunt classification [S0-S3]), fibrosis stage (based on a meta-analysis of histological data on viral hepatitis score [S0-S4]), ballooning score (S0-S2), and inflammation grades (G0-G4) of the patients were evaluated.[12-14]. Fibrosis stage ≥2, inflammation grade ≥2, and steatosis score ≥2 were defined as indicative of significant liver fibrosis, active inflammation, and severe steatosis, respectively.

Fatigue Assessment

Fatigue was assessed by a professional physician within 1 week prior to liver biopsy using the Chronic Liver Disease Questionnaire (CLDQ), which defined fatigue as a score of less than 20 according to the items 2, 4, 8, 11, and 13.[15]

Statistical Analysis

Continuous variables were compared between the two groups using Student's *t*-test or the Mann–Whitney *U* test. Categorical variables were compared using the Chi-square test. Propensity score matching (PSM) was performed in a ratio of 1:2 and with a caliper value of 0.2 to balance age, sex, and CLD classification between the two patient groups. Univariate and multivariate logistic regression analysis was conducted to analyze the factors that contribute to fatigue. We estimated adjusted odds ratios (ORs) and relevant 95% confidence intervals (CIs)

using a parametric proportional hazard model. SPSS 26.0 (IBM, Armonk, NY, USA) was used for statistical analyses. Statistical significance was set at $P<0.05$.

Ethical Considerations

This study was performed according to the ethical guidelines of the Declaration of Helsinki and was approved by the Ethics Committee of the Affiliated Hospital of Hangzhou Normal University (approval number: 2020(02)-KS-022). As this was an observational retrospective study, the requirement for informed consent was waived by the Ethics Committee.

Patient and public involvement

Patients or the public were not involved in the design, or conduct, or reporting, or dissemination plans of the current work.

Results

Comparison of the Clinical and Histological Features of Chronic Liver Disease Patients with and without Fatigue

A total of 1,374 patients with biopsy-proven CLD were included in this study. Of these, 262 (19.07%) patients had symptoms of fatigue, while 1,112 (80.93%) had no fatigue. The patients with fatigue were older, had lower BMI and higher HDL-c and GGT levels than the patients without fatigue ($P<0.05$). There were significant differences between the proportions of patients with NAFLD, CHB with fatty liver, CHB, AFLD, and AILDs in the two groups ($P<0.05$) (Table 1).

After PSM, 242 patients with fatigue and 482 patients without fatigue were successfully matched, and there were no statistically significant differences between the two groups in terms

of sex, age, BMI, blood pressure, fasting blood glucose, lipid profile, and liver function (P>0.05) (Table 1). The fatigue group had higher ALT, AST, and GGT levels than the non-fatigue group. Further comparison of the histological features of the two groups is shown in Figure 1. The inflammation grades and liver fibrosis stages of the patients with fatigue were significantly higher than those of the patients without fatigue (P<0.05). There was no difference in hepatic steatosis and ballooning scores between the two groups (P>0.05) (Figure 1, A-D).

Distribution and Risk Factors of Fatigue in Patients with Chronic Liver Disease

The prevalence of fatigue significantly increased with age (R=0.087, P=0.001) (Figure 2a). There was no correlation between fatigue and BMI and sex (P>0.05) (Figures 2b and c). The prevalence of fatigue among patients with different types of CLD varied. For patients with AILDs, the prevalence of fatigue was 44.19%, which is more than twice that of patients with other CLDs (P<0.001) (Figure 2d). The prevalence of fatigue significantly increased with the degree of liver inflammation and fibrosis stage (P<0.001) (Figures 2e and f).

Univariate and multivariate analyses were performed to define the association between fatigue using clinical and histological features (Table 2). Univariate analysis showed that old age (OR=2.122, 95% CI: 1.379-3.267, P=0.001), AILDs (OR=3.545, 95% CI: 1.911-6.574, P<0.001), elevated GGT level (OR=1.356, 95%CI: 1.012-1.816, P=0.042), active inflammation (OR=1.768, 95%CI: 1.329-2.353, P<0.001), and advanced fibrosis stage (OR=1.743, 95%CI: 1.282-2.370, P<0.001) were risk factors for fatigue in CLD. Further multivariate analysis indicated that old age (OR= 2.026, 95% CI: 1.274-3.221, P=0.003), AILDs (OR=2.749, 95% CI: 1.446-5.226, P=0.002), and active inflammation (OR= 1.587, 95% CI: 1.164-2.164, P=0.003) were independent risk factors for fatigue.

Analysis of the Correlation between Histological Features and Fatigue in Chronic Liver Disease

Multivariate analysis was performed to explore the correlation between fatigue and the severity of histological features. Two different models were utilized to estimate the ORs for different outcomes. After adjusting for age, sex, BMI, hypertension, type 2 diabetes mellitus, disease classification, ALT level, AST level, and GGT level, the OR for the risk of fatigue increased in a stepwise manner from inflammation grades G0-G1 (as a reference) and G2 (OR=1.609, 95% CI: 1.085-2.386, *P*=0.018) to G3 (OR=1.745, 95% CI: 1.019-2.986, *P*=0.042) (Table 3). The severity of steatosis, ballooning, and fibrosis were not associated with fatigue (*P*>0.05).

Sensitivity Analysis

Subgroup analysis of the risk of fatigue in CLD showed a significant association between fatigue and increasing severity of liver inflammation among patients aged < 60 years old without AILDs (*P*<0.05). However, the severity of inflammation was not associated with fatigue among patients > 60 years old or with AILDs (*P*>0.05) (Table 4).

Discussion

Fatigue is a critical component of CLD.[11] The findings of the present study indicate an association between fatigue and liver inflammation. In the present study, CLD patients with fatigue had significantly higher inflammation grades and liver fibrosis stages than patients without fatigue. In addition, multivariate analysis showed that age, AILDs, and active inflammation were independent risk factors for fatigue, and that the severity of liver inflammation was strongly associated with fatigue after adjustment for confounders. Further

sensitivity analysis showed that this association was present in the young and middle-aged population of the present study but not in the elderly population.

The clinical features of CLD-related fatigue have not been uniformly demonstrated.[16,17] In the present study, the CLD patients with fatigue were older, had lower BMI, and higher HDL-c and GGT levels than patients without fatigue. In addition, the fatigue group showed significantly higher inflammation grades and liver fibrosis stages than the non-fatigue group after PSM for age and sex (Table 1, Figure 1). The results of the present study are in line with those of most studies that showed that old age, AILDs, and active inflammation are independent risk factors for fatigue (Table 2).[18-21] Notably, our research suggested that liver inflammation caused by elevated GGT, and not elevated ALT or AST, was implicated in fatigue. Elevated GGT is usually a sign of cholestasis, and animal studies in bile duct-ligated rats have demonstrated cholestasis-disordered neurotransmission and the development of fatigue. This is suggested to be due to central nervous system damage caused by manganese accumulation. However, further studies are needed to understand the exact mechanism. [22]

The results of the present study showed that the risk of fatigue increased with the severity of inflammation, but not with the severity of hepatic steatosis, ballooning, and liver fibrosis (Table 3). Although the issue of fatigue in patients with CLD, including PBC, PSC, CHB, CHC, and NAFLD, has been extensively studied, the relationship between fatigue and the histological features of CLD remains controversial. Fatigue in NAFLD has been associated with inactivity and excessive daytime sleepiness but not with the severity of liver disease or insulin resistance.[16] However, a recent study indicated that the detection of lobular inflammation in biopsies is correlated with lower health-related quality of life (HRQL) in patients with NAFLD.[17] Data from clinical trials on chronic hepatitis B or C virus infection also support the dominant role of inflammation in fatigue. In these trials, viral elimination or suppression after antiviral therapy was associated with improved HRQL, which suggests an

effect of inflammation on fatigue, whereas improvement of fibrosis did not affect HRQL.[23-25]

Further subgroup analysis in the present study revealed a significant association between fatigue and the severity of liver inflammation in patients <60 years old, but not in patients ≥60 years old (Table 4). Our findings suggest that the severity of liver inflammation may play a dominant role in fatigue in young and middle-aged patients with CLD, whereas age-related factors may play dominant roles in fatigue in elderly patients. Previous studies have shown that fatigue is a significant component of the clinical presentation of patients with AIH, often paralleling hepatic inflammation.[26] In line with previous research, the data of the present study showed that patients with AILDs had the highest prevalence of fatigue, and that AILDs was an independent risk factor for CLD-related fatigue. Though fatigue was associated with an AILD diagnosis, it was not correlated with the severity of liver inflammation in patients with AILDs. This may be related to the relatively small number of AILD cases in the present study. Thus, studies with larger samples are needed to confirm this finding.

The strength of this study is that it is the first study, to the best of our knowledge, in which the relationship between fatigue and the severity of liver inflammation in different CLD populations was explored using liver histopathology features. However, the limitations of this study should be noted as well. First, since this was a retrospective study, PSM was used to minimize the influence of available factors. However, some of the retrieved data that could contribute to the development of fatigue, namely plasma iron level, markers of thyroid gland function, and blood oxygen tension, were unavailable. Second, although we diagnosed fatigue based on responses to the CLDQ, an important limitation is the dichotomic division of the CLD population into suffering and not-suffering from fatigue with no self-assessment of fatigue severity. Therefore, we could only assess the relationship between the severity of inflammation and the presence or absence of fatigue, but could not clarify the relationship between the

severity of liver inflammation and the severity of fatigue. Third, since the study was based on the liver biopsy, there was no control group composed of sex and aged-matched people with healthy livers, which is especially important in the older population. Fourth, since only 43 patients in this study had AILDs, it is difficult to perform statistical analysis after subdividing. Therefore, the AILDs were grouped together irrespective of whether they were parenchymatic or cholestatic, even though it is known that the pathophysiology of fatigue is different in primary biliary cholangitis, and autoimmune hepatitis may affect the results of AILDs to some extent. Fifth, as this was a cross-sectional study, we could not determine the causal relationship between the severity of inflammation and fatigue. Further studies with longitudinal cohorts are needed to confirm the effects of the severity of inflammation on fatigue in patients with CLD.

In conclusion, the impact of fatigue on the perceived quality of life can be profound for patients with CLD. Since the pathophysiology of fatigue is complex and poorly understood, developing therapeutic trials of symptom-directed therapies is challenging. For fatigue in CLD, the 'TrACE' method of Treating the treatable (co-morbid causes), Ameliorate the ameliorable causes (sleep, autonomic, and mood disorders), Coping strategies (lifestyle changes such as pacing the day, avoiding shift work) and Empathizing is generally suggested.[4,27] The present study demonstrates that fatigue is correlated with the severity of liver inflammation in young and middle-aged patients with CLD. However, this correlation was not observed in elderly patients. These findings contradict the perception that fatigue is not associated with the severity of liver disease. Since age is an important factor that influences fatigue, our findings highlight the need for age stratification during the evaluation and treatment of CLD patients with fatigue, which will provide new evidence for the management and treatment of fatigue in patients with CLD.

Author contributions

Research idea and study design: Junping Shi; data acquisition: Jing Liu, Xiying Gong, Shiyi Liu, Yanming Jiang, Geli Zhu, Xiaojie Ma, Jie Wang, Xiaoping Ye, Yidan Gao, Dian Wang, Gongying Chen; data analysis/interpretation: Jing Liu, Haifeng Lv; statistical analysis: Jing Liu, Haifeng Lv; supervision or mentorship: Junping Shi. Each author contributed important intellectual content during manuscript drafting and revision, agreed to be personally accountable for the individual's contributions, and ensured questions about the accuracy or integrity of any portion of the work, even one in which the author was not directly involved, are appropriately investigated and resolved.

References

1 Moon AM, Singal AG, Tapper EB. Contemporary epidemiology of chronic liver disease and cirrhosis. *Clin Gastroenterol Hepatol* 2020;18:2650–66.

2 Huang DQ, El-Serag HB, Loomba R. Global epidemiology of NAFLD-related HCC: trends, predictions, risk factors and prevention. *Nat Rev Gastroenterol Hepatol* 2021;18:223–38.

3 Locarnini S, Hatzakis A, Chen DS et al. Strategies to control hepatitis B: public policy, epidemiology, vaccine and drugs. *J Hepatol* 2015;62;Supplement:S76–S86.

4 Swain MG, Jones DEJ. Fatigue in chronic liver disease: new insights and therapeutic approaches. *Liver Int* 2019;39:6–19.

5 Jopson L, Dyson JK, Jones DE. Understanding and treating fatigue in primary biliary cirrhosis and primary sclerosing cholangitis. *Clin Liver Dis* 2016;20:131–42.

6 Cauch-Dudek K, Abbey S, Stewart DE et al. Fatigue in primary biliary cirrhosis. *Gut* 1998;43:705–10.

7 Jones DE, Bhala N, Burt J et al. Four year follow up of fatigue in a geographically defined primary biliary cirrhosis patient cohort. *Gut* 2006;55:536–41.

8 Bower JE. Cancer-related fatigue--mechanisms, risk factors, and treatments. *Nat Rev Clin Oncol* 2014;11:597–609.

9 Gerber LH, Weinstein AA, Mehta R et al. Importance of fatigue and its measurement in chronic liver disease. *World J Gastroenterol* 2019;25:3669–83.

10 D'Mello C, Swain MG. Liver-brain inflammation axis. *Am J Physiol Gastrointest Liver Physiol* 2011;301:G749–61.

11 Kośnik A, Wójcicki M. Fatigue in chronic liver disease patients: prevalence, pathophysiology, and management. *Prz Gastroenterol* 2022;17:21–7.

12 Brunt EM, Janney CG, Di Bisceglie AM et al. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. *Am J Gastroenterol* 1999;94:2467–74.

13 Bedossa P, Poynard T. An algorithm for the grading of activity in chronic hepatitis C. The METAVIR Cooperative Study Group. *Hepatology* 1996;24:289–93.

14 Batts KP, Ludwig J. Chronic hepatitis. An update on terminology and reporting. *Am J Surg Pathol* 1995;19:1409–17.

15 Younossi ZM, Guyatt G, Kiwi M et al. Development of a disease specific questionnaire to measure health related quality of life in patients with chronic liver disease. *Gut* 1999;45:295–300.

16 Newton JL, Jones DE, Henderson E, et al. Fatigue in non-alcoholic fatty liver disease (NAFLD) is significant and associates with inactivity and excessive daytime sleepiness but not with liver disease severity or insulin resistance. *Gut* 2008;57:807–13.

17 Huber Y, Boyle M, Hallsworth K, et al. Health-related quality of life in nonalcoholic fatty liver disease associates with hepatic inflammation. *Clin Gastroenterol Hepatol* 2019;17:2085–2092.e1.

18 Swain MG. Fatigue in liver disease: pathophysiology and clinical management. *Can J Gastroenterol* 2006;20:181–8.

19 Schramm C, Wahl I, Weiler-Normann C, et al. Health-related quality of life, depression, and anxiety in patients with autoimmune hepatitis. *J Hepatol* 2014;60:618–24.

20 Bozzini AB, Neder L, Silva CA et al. Decreased health-related quality of life in children and adolescents with autoimmune hepatitis. *J Pediatr (Rio J)* 2019;95:87–93.

21 Finsterer J, Mahjoub SZ. Fatigue in healthy and diseased individuals. *Am J Hosp Palliat Care* 2014;31:562–75.

22. Milkiewicz P, Heathcote EJ. Fatigue in chronic cholestasis. *Gut* 2004;53(4):475–7.

23 Younossi ZM, Stepanova M, Janssen HLA, et al. Effects of treatment of chronic hepatitis B virus infection on patient-reported outcomes. *Clin Gastroenterol Hepatol* 2018;16:1641–1649.e6.

24 Younossi ZM, Stepanova M, Afdhal N, et al. Improvement of health-related quality of life and work productivity in chronic hepatitis C patients with early and advanced fibrosis treated with ledipasvir and sofosbuvir. *J Hepatol* 2015;63:337–45.

25Younossi ZM, Stepanova M, Jacobson IM, et al. Sofosbuvir and velpatasvir with or without voxilaprevir in direct-acting antiviral-naïve chronic hepatitis C: patient-reported outcomes from POLARIS 2 and 3. *Aliment Pharmacol Ther* 2018;47:259–67.

26 Russell AS, Gulliver WP, Irvine EJ et al. Quality of life in patients with immune-mediated inflammatory diseases. *J Rheumatol Suppl* 2011;88:7–19.

27. Phaw NA, Leighton J, Dyson JK, Jones DE. Managing cognitive symptoms and fatigue in cholestatic liver disease. *Expert Rev Gastroenterol Hepatol* 2021;15(3):235–41.

TABLE 1 Comparison of clinical and laboratory characteristics between patients with and without fatigue in CLD

Variables	Before propensity score matching			After propensity score matching(1:2)		
	Fatigue	No fatigue	*P* Value	Fatigue	No fatigue	*P* Value
n	262	1112		242	484	
Male(%)	175(66.79)	785(70.59)	0.228‡	167(69.01)	337(69.63)	0.527‡
Age (year)	45.30 ±11.12	41.36 ±11.33	<0.001	44.02 ± 10.48	42.46 ± 11.26	0.071
BMI(kg/m^2)	24.05 ±3.86	24.82 ±4.62	0.032	24.26 ± 3.88	24.18 ±4.02	0.840
BP(S)(mmHg)	124.33 ±15.26	125.70 ±15.52	0.204	123.86 ± 15.43	125.96 ± 16.01	0.098
FPG (mmol/L)	5.56 ± 1.66	5.65 ±1.54	0.493	5.56 ± 1.65	5.61 ± 1.40	0.718
HbA1c(%)	6.18 ± 1.54	6.12 ± 1.52	0.823	6.24 ±1.61	6.09 ± 1.52	0.639
TG (mmol/L)	1.23(0.95-1.85)	1.08(1.55-2.18)	0.793†	1.19(0.93-1.84)	1.31(0.90-2.00)	0.543
TC (mmol/L)	4.74 ± 1.20	4.68 ± 1.11	0.504	4.67 ± 1.16	4.63 ± 0.95	0.733
LDL-c (mmol/L)	2.76 ± 0.88	2.75 ± 0.82	0.875	2.72 ± 0.88	2.70 ± 0.75	0.788
HDL-c(mmol/L)	1.30 ± 0.36	1.24 ± 0.33	0.035	1.28 ± 0.35	1.25 ± 0.35	0.298
STB(μmmol/l)	16.65(12.82-22.45)	16.20(12.90-21.10)	0.511†	16.60(12.85-22.00)	15.50(12.50-20.80)	0.153†
ALT (U/L)	51.00(32.00-83.00)	49.00(30.00-80.00)	0.432†	51.00 (32.00-82.00)	41.00(26.00-66.00)	<0.001†
AST (U/L)	35.00(26.00-52.00)	33.00(25.00-49.00)	0.093†	34.00 (26.00-51.00)	28.00(23.00-41.00)	<0.001†
GGT(U/L)	37.00(23.00-77.00)	33.00(20.00-60.00)	0.005†	35.00(21.00-66.00)	27.00(17.00-49.00)	<0.001†
AKP(U/L)	110.00(88.00-	108.00(87.00-	0.650†	107.00(87.0-	106.00(83.00-133.00)	0.797†
ALB (g/L)	137.00	136.00)	0.297	129.00)	44.70 ± 5.77	0.460
CLD Category	44.08 ± 4.15	44.51 ± 5.85		4437 ± 3.95		
NAFLD n(%)			<0.001‡		70(14.46)	0.190‡
CHB with fatty liver n(%)	42(16.03)	205(18.44)		42(17.36)	103(21.28)	
CHB n(%)	47(17.94)	283(25.45)		47(19.42)	254(52.78)	
AFLD n(%)	138(52.67)	531(47.75)		137(56.61)	46(9.51)	
AILDs n(%)	16(6.11)	69(6.20)		12(4.96)	11(2.27)	
	19(7.25)	24(2.16)		4(1.65)		

*P-value calculated using the χ^2 test.

†P-value calculated using the Mann–Whitney U-test.

Abbreviations: Data are expressed as mean ± standard deviation or median (interquartile range).

Abbreviations: AFLD, alcoholic fatty liver disease; AILD, autoimmune liver disease; ALB, albumin; AKP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; BP, blood pressure; CHB, chronic hepatitis B; CLD, chronic liver disease; DBP, diastolic blood pressure; FPG, fasting plasma glucose; GGT, gamma-glutamyl-transferase; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; SBP, systolic blood pressure; STB, serum total bilirubin; SUA, serum uric acid; TC, total cholesterol; TG, triglycerides.

Table 2. Univariate and multivariable regression analysis of risk factors for fatigue in chronic liver disease

Variables	Univariate Analysis		Multivariate Analysis	
	OR (95% CI)	*P* Value	OR (95% CI)	*P* Value
Old Age				
No	Ref.		Ref.	
Yes	2.122(1.379-3.267)	0.001	2.026(1.274-3.221)	0.003
Male				
No	Ref.			
Yes	0.838(0.628-1.117)	0.228	--	--
AILDs				
No	Ref.			
Yes	3.545(1.911-6.574)	<0.001	2.749(1.446-5.226)	0.002
Metabolic factors				
Obesity				
No	Ref.			
Yes	0.813(0.591-1.121)	0.207	--	--
Hypertension				
No	Ref.			
Yes	0.735(0.536-1.008)	0.056	--	--
T2DM				
No	Ref.			
Yes	0.694(0.436-1.103)	0.122	--	--
Hypertriglyceridemia				
No	Ref.			
Yes	0.804(0.577-1.121)	0.198	--	--
Hypercholesterolemia				
No	Ref.			
Yes	1.448(0.974-2.153)	0.068	--	--
Hyperuricemia				
No	Ref.			
Yes	0.835(0.570-1.222)	0.353	--	--
High LDL-c				
No	Ref.			
Yes	0.943(0.675-1.316)	0.728	--	--
Low HDL-c				
No	Ref.			
Yes	1.104(0.764-1.569)	0.599	--	--
Liver enzymes				
Elevated ALT				
No	Ref.			
Yes	1.164(0.662-2.441)	0.306	--	--
Elevated AST				
No	Ref.			
Yes	1.205(0.906-1.602)	0.199	--	--
Elevated GGT				
No	Ref.			

Yes	1.356(1.012-1.816)	0.042	--	--
Histopathology				
Severe ballooning				
No	Ref			
Yes	1.049(0.606-1.815)	0.865	--	--
Severe steatosis				
No	Ref			
Yes	0.850(0.488-1.482)	0.567	--	--
Active inflammation				
No	Ref.			
Yes	1.768(1.329-2.353)	<0.001	1.587(1.164-2.164)	0.003
Advanced fibrosis				
No	Ref			
Yes	1.743(1.282-2.370)	<0.001	--	--

Abbreviations: AILDs, autoimmune liver diseases; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; T2DM, type 2 diabetes mellitus; OR, odds ratio; CI, confidence intervals.

Table 3 Odds ratio of liver histological severity for fatigue in chronic liver disease

	Crude		Model 1		Model 2	
	OR(95% CI)	***P* Value**	**OR(95% CI)**	***P* Value**	**OR(95% CI)**	***P* Value**
Steatosis score						
S0	Ref.		Ref.		Ref.	
S1-2	1.205(0.793-1.833)	0.383	1.204(0.787-1.841)	0.392	1.360(0.761-2.428)	0.299
S3	0.870(0.498-1.519)	0.624	0.918(0.522-1.615)	0.767	1.328(0.648-2.724)	0.493
Ballooning score						
S0	Ref.		Ref.		Ref.	
S1	0.665(0.405-1.090)	0.979	0.620(0.376-1.023)		0.942(0.521-1.703)	0.468
S2	1.007(0.581-1.747)	0.106	0.871(0.498-1.525)		1.310(0.632-2.716)	0.843
Inflammation grade						
G0-1	Ref.		Ref.		Ref.	
G2	1.618(1.190-2.200)	0.002	1.570(1.152-2.140)	0.004	1.609(1.085-2.386)	0.018
G≥3	2.170(1.486-3.169)	<0.001	2.014(1.372-2.056)	<0.001	1.745(1.019-2.986)	0.042
Liver fibrosis						
S0-1	Ref.		Ref.		Ref.	
S2	1.087(0.751-1.574)	0.657	1.006(0.692-1.462)	0.975	0.691(0.420-1.135)	0.145
S≥3	1.777(1.291-2.447)	<0.001	1.608(1.160-2.226)	0.004	1.371(0.897-2.096)	0.144

Note: Model 1 adjusted for age and sex; Model 2 was adjusted for model 1 plus body mass index hypertension, type 2 diabetes mellitus, alanine aminotransferase, aspartate aminotransferase, gamma-glutamyl-transferase and disease classification.

Abbreviations: OR, odds ratio; CI, confidence intervals.

Table 4. Sensitivity analysis in the presence of risk factors for fatigue in chronic liver disease

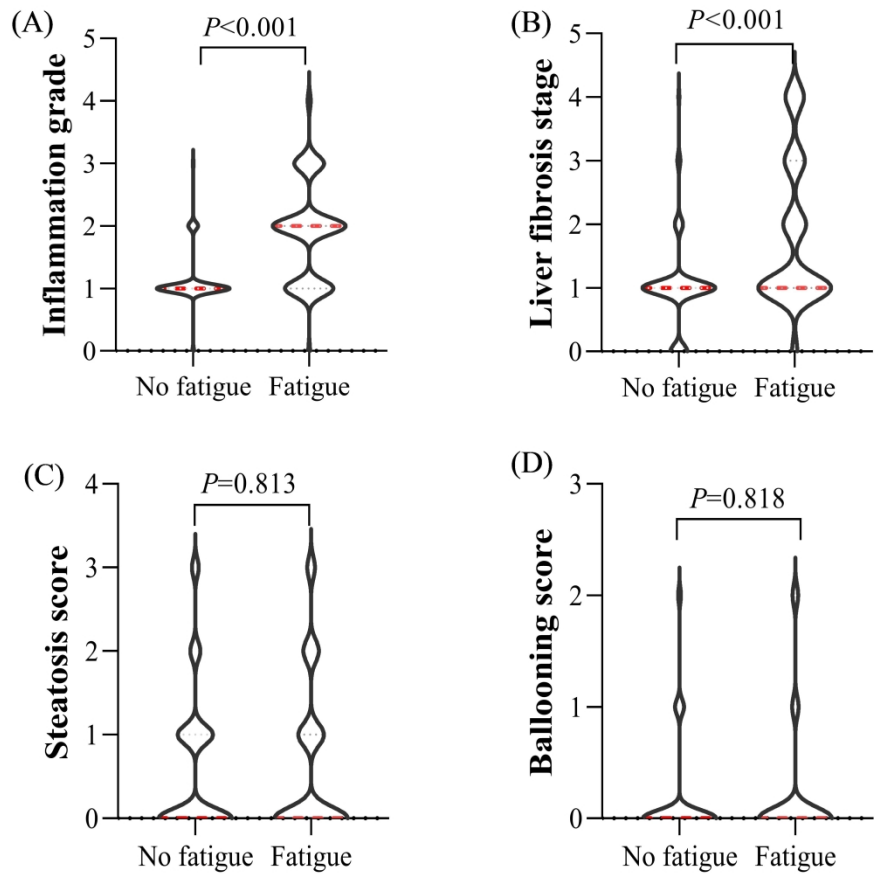
Variables	No. of Participants	OR(95%CI)	*P* Value
Age			
≥60 years	107		
G0-1	37	Ref.	
G2	45	1.813(0.668-4.919)	0.632
G≥3	25	2.848(0.937-8.659)	0.870
<60 years	1267		
G0-1	556	Ref.	
G2	517	1.566(1.134-2.163)	0.006
G≥3	194	1.989(1.324-2.988)	0.001
Elevated GGT			
Yes	451		
G0-1	141	Ref.	
G2	191	1.919(1.077-3.420)	0.027
G≥3	119	2.420(1.304-4.490)	0.005
No	923		
G0-1	452	Ref.	
G2	371	1.458(1.011-2.105)	0.019
G≥3	100	1.880(1.108-3.190)	0.044
AILDs			
Yes	43		
G0-1	9	Ref.	
G2	13	1.714(0.294-9.999)	0.549
G≥3	21	1.818(0.357-9.272)	0.472
No	1331		
G0-1	584	Ref	
G2	549	1.587(1.161-2.167)	0.004
G≥3	198	1.959(1.313-2.923)	0.001

Abbreviations: AILDs, autoimmune liver diseases; GGT, gamma-glutamyl-transferase; OR, odds ratio; CI, confidence intervals.

Figure 1. Comparison of the histopathological characteristics of chronic liver disease patients with and without fatigue. (**A**) Comparison of the inflammation grades of patients with CLD stratified according to the presence or absence of fatigue. (**B**) Comparison of the fibrosis stages of patients with CLD stratified according to the presence or absence of fatigue. (**C**) Comparison of hepatic steatosis scores of patients with CLD stratified according to the presence or absence of fatigue. (**D**) Comparison of the ballooning scores of the patients with CLD stratified according to the presence or absence of fatigue.

Figure 2. Prevalence of fatigue in different chronic liver disease populations. (**A**) Prevalence of fatigue stratified according to age. (**B**) Prevalence of fatigue stratified according to body mass index. (**C**) Prevalence of fatigue stratified according to sex. (**D**) Prevalence of fatigue stratified according to CLD classification. (**E**) Prevalence of fatigue stratified according to inflammation grade. (**F**) Prevalence of fatigue stratified according to fibrosis stage.

Abbreviations: CHB, chronic hepatitis B; NAFLD, non-alcoholic fatty liver disease; AFLD, alcoholic fatty liver disease; AILD, autoimmune liver diseases.



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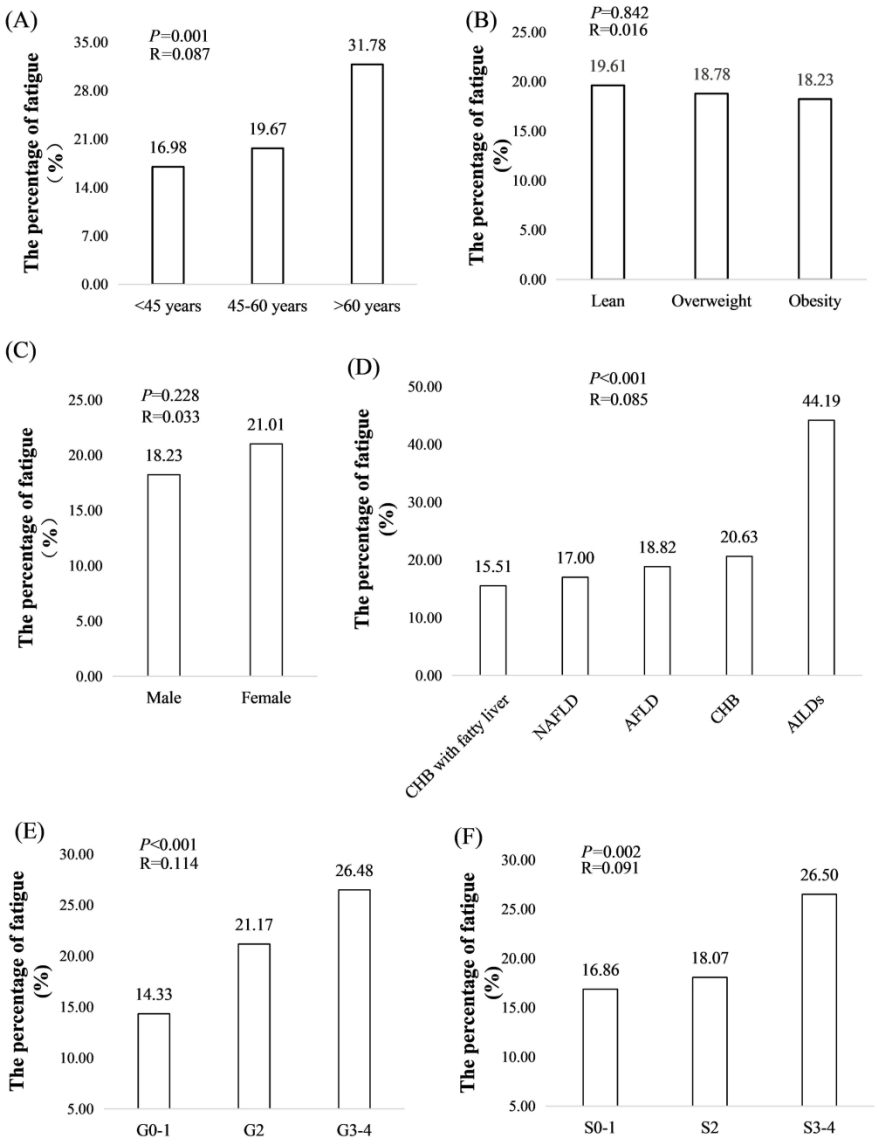

Prevalence of fatigue in different chronic liver disease populations. (A) Prevalence of fatigue stratified according to age. (B) Prevalence of fatigue stratified according to body mass index. (C) Prevalence of fatigue stratified according to sex. (D) Prevalence of fatigue stratified according to CLD classification. (E) Prevalence of fatigue stratified according to inflammation grade. (F) Prevalence of fatigue stratified according to fibrosis stage.

Abbreviations: CHB, chronic hepatitis B; NAFLD, non-alcoholic fatty liver disease; AFLD, alcoholic fatty liver disease; AILD, autoimmune liver diseases.

193x247mm (300 x 300 DPI)

STROBE Statement—Checklist of items that should be included in reports of ***cross-sectional studies***

	Item No	Recommendation	Page No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	3
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	4-5
Objectives	3	State specific objectives, including any prespecified hypotheses	5
Methods			
Study design	4	Present key elements of study design early in the paper	5-6
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	5-7
Participants	6	(*a*) Give the eligibility criteria, and the sources and methods of selection of participants	5-6
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	6-7
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	*6-7*
Bias	9	Describe any efforts to address potential sources of bias	6-7
Study size	10	Explain how the study size was arrived at	5-6
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	7
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	7
		(*b*) Describe any methods used to examine subgroups and interactions	7
		(*c*) Explain how missing data were addressed	7
		(*d*) If applicable, describe analytical methods taking account of sampling strategy	7
		(*e*) Describe any sensitivity analyses	7
Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	8
		(b) Give reasons for non-participation at each stage	8
		(c) Consider use of a flow diagram	0
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	8-9
		(b) Indicate number of participants with missing data for each variable of interest	0
Outcome data	15*	Report numbers of outcome events or summary measures	8

Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	9-10
		(*b*) Report category boundaries when continuous variables were categorized	7
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	9-10
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	10
Discussion			
Key results	18	Summarise key results with reference to study objectives	10
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	12
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	10-12
Generalisability	21	Discuss the generalisability (external validity) of the study results	12
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	2

*Give information separately for exposed and unexposed groups.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.