Digestive Diseases

Liver: Research Article

Dig Dis 2023;41:632–640
DOI: 10.1159/000530473

Received: November 12, 2022
Accepted: March 28, 2023
Published online: April 5, 2023

Anemia Is Associated with Disease Severity, Hepatic Complications, and Progression of Wilson Disease: A Retrospective Cohort Study

Si-Qi Wang[a] Yong-Qiang Zhan[b] Xuan Hu[a] Yu-Pei Zhuang[a]
Hong-Qian Liu[a] Ming-Fan Hong[c] Hao-Jie Zhong[b]

[a]Department of Gastroenterology, The First Affiliated Hospital of Guangdong Pharmaceutical University, Guangzhou, China; [b]Department of Hepatobiliary and Pancreatic Surgery, The First Affiliated Hospital of Shenzhen University, Shenzhen Second People's Hospital, Shenzhen, China; [c]Department of Neurology, The First Affiliated Hospital of Guangdong Pharmaceutical University, Guangzhou, China

Keywords
Anemia · Disease progression · Hepatic encephalopathy · Liver cirrhosis

Abstract

Introduction: Anemia is a common manifestation of chronic liver diseases. It is a predictor of severe disease, a high risk of complications, and poor outcomes in various liver diseases. However, it remains unclear whether anemia serves as a similar indicator in patients with Wilson disease (WD). Therefore, this study aimed to investigate the relationship between anemia and severity, hepatic complications, and the progression of WD. ***Methods:*** Medical data were collected retrospectively from January 1, 2016, to December 31, 2020. Univariate and multivariate analyses were carried out to investigate the relationship between anemia and liver-associated disease severity, hepatic complications, and the progression of WD. ***Results:*** A total of 288 WD patients (48 with and 240 without anemia) were enrolled in the study. Multivariate linear regression revealed that WD patients with anemia had significantly higher levels of bilirubin, alanine transaminase, prothrombin time, international normalized ratio, type IV collagen, and hyaluronic acid and significantly lower levels of albumin, total cholesterol, and high-density lipoprotein-cholesterol (all $p < 0.05$). Multivariate logistic regression showed that anemia was a risk factor for gastric varices and ascites (all $p < 0.05$). Fully adjusted Cox regression revealed that anemia was an independent risk factor for advanced Child-Pugh classification ($p = 0.034$). ***Conclusions:*** Anemia was common in WD patients and was associated with greater disease severity, a higher risk of hepatic complications, and a faster progression.

Introduction

Wilson disease (WD) is caused by mutations in the ATPase copper-transporting beta gene, which results in copper accumulation in the hepatic and extrahepatic tissues [1]. While WD is rare, with an estimated prevalence of symptomatic cases being 1 in 30,000, genetic variants associated with WD are much more common, affecting approximately 1 in 7,000 individuals [2, 3].

Si-Qi Wang and Yong-Qiang Zhan are co-first authors.

karger@karger.com
www.karger.com/ddi

Correspondence to:
Hao-Jie Zhong, jaxzhong@126.com
Ming-Fan Hong, hmf9001@163.com

Karger

Given that the liver is the primary organ involved in copper excretion, liver injury is the first manifestation in up to 60% of WD patients [4]. Unlike chronic liver diseases of other etiologies, WD progresses rapidly [5], and half of WD patients eventually develop cirrhosis [6]. Once decompensated cirrhosis develops, it is followed by a series of complications such as ascites, esophagogastric varices, and hepatic encephalopathy [7], leading to a marked increase in mortality [1]. Several clinical indicators are widely used nowadays to evaluate the severity of chronic liver diseases and predict their progression [8–10]. However, because patients with WD have significantly different liver features and complications compared to patients with other liver diseases [11, 12], it remains unclear whether these indicators could be used to assess the severity, complications, and progression of WD.

Anemia is common in patients with chronic liver diseases due to chronic bleeding, nutritional deficiencies, and hypersplenism [13]. In these patients, it is also a predictor of severe disease, a high risk of complications, and poor outcomes [14, 15]. Because copper causes oxidative damage in erythrocytes, the resulting hemolysis is one of the causes of anemia in WD patients, with a prevalence of 5–6% [16]. WD patients with hemolytic anemia have higher serum copper levels [17], which is indicative of a more severe and rapidly progressive disease [4]. However, it remains unclear whether anemia could be used as an indicator of disease severity and complications or as a predictor of disease progression in WD patients. Therefore, this study aimed to investigate the relationships between anemia and the severity and progression of WD as well as the hepatic complications associated with it.

Materials and Methods

Study Design and Ethical Approval

This retrospective study was conducted in accordance with the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines. It was also in compliance with the Declaration of Helsinki and approved by the Research Ethics Board of the First Affiliated Hospital of Guangdong Pharmaceutical University (#2021-172). As the study was retrospective in nature, the requirement for informed consent was waived.

Participants

Based on preliminary data, online software (Power and Sample Size Calculators; http://powerandsamplesize.com/) was used to estimate the sample size. Inclusion criteria included consecutive inpatients with WD at the First Affiliated Hospital of Guangdong Pharmaceutical University (Guangzhou, China) from January 1, 2016, to December 31, 2020. Exclusion criteria included the presence of other liver diseases (viral, alcoholic, autoimmune, or drug-induced), current malignancies (except for primary liver cancer), or the absence of important medical data.

Data Collection

The following data were extracted from electronic medical records: demographic variables, body mass index (BMI), comorbidities (hypertension and type 2 diabetes), smoking status, alcohol consumption, duration of WD, anti-copper therapy, imaging findings, anemia etiology, hepatic complications (gastric varices, splenomegaly, ascites, spontaneous bacterial peritonitis [SBP], liver failure, renal impairment, hepatic encephalopathy, and primary liver cancer) and laboratory tests, including hemoglobin levels, liver function parameters (bilirubin, alanine transaminase [ALT], aspartate transaminase, and albumin), coagulation parameters (prothrombin time [PT], international normalized ratio [INR], and platelet count), blood lipid parameters (total cholesterol, triglyceride, high-density lipoprotein-cholesterol [HDL-C], and low-density lipoprotein-cholesterol), liver fibrosis parameters (type IV collagen, procollagen type-III N-terminal propeptide, laminin, and hyaluronic acid), urinary copper, serum creatinine levels, and the Coombs test. The follow-up time was calculated for each patient from the first to the last hospitalization during the study period.

Definitions

Alcoholism is defined as consuming 70 g or more of alcohol per week based on self-reported alcohol consumption. Neurological manifestations in WD patients primarily included movement disorders, speech disturbances, drooling, and gait and balance disturbances [1]. Psychiatric manifestations in WD patients primarily included personality disorders, mood disorders, psychosis, and cognitive impairment [1]. In China, anemia is defined as hemoglobin levels below 110 g/L in women and below 120 g/L in men [18]. Cirrhosis was confirmed using a combination of clinical, biochemical, and imaging examinations. Gastric varices were diagnosed through gastroscopy, computed tomography, or magnetic resonance imaging. Splenomegaly and splenectomy were diagnosed through physical examinations, imaging findings, and medical history. Ascites was diagnosed through physical examination, ultrasound, or computed tomography. SBP was diagnosed based on an ascitic fluid neutrophil count greater than 250/mm^3 in the absence of an evident intra-abdominal, surgically treatable source. Liver failure is defined as the presence of hepatic encephalopathy and an INR greater than 1.5 [19]. Renal impairment is defined as serum creatinine levels greater than 1.5 mg/dL [20]. Hepatic encephalopathy was assessed using the West Haven criteria [21]. Liver cancer was diagnosed through clinical symptoms, pathology, and imaging findings. Hepatic decompensation was confirmed based on the presence of hepatic encephalopathy, ascites, or varices [22]. The aspartate aminotransferase-to-platelet ratio index and fibrosis-4 index were calculated as previously described [23, 24]. The Child-Pugh classification was used to assess liver function [25]. According to the Child-Pugh classification, grades B and C represent advanced hepatic dysfunction.

In China, WD is defined by a combination of age of onset, hepatic and neuropsychiatric manifestations, serum, liver, and urinary copper concentrations, serum ceruloplasmin concentration, Kayser-Fleischer rings, and gene mutation analysis [26],

Table 1. Patient characteristics

Variable	Anemic patients (N = 48)	Non-anemic patients (N = 240)	p value
Age, years	28.00 (20.50–33.00)	24.00 (18.00–30.75)	0.015
Male sex	17 (35.42)	126 (52.50)	0.031
BMI, kg/m^2	19.84 (17.63–21.72) (n = 43)	19.66 (17.92–22.06) (n = 224)	0.984
Diabetes	0 (0)	1 (0.42)	1.000
Hypertension	1 (2.08)	2 (0.83)	0.423
Alcoholism	1 (2.08)	2 (0.83)	0.423
Smoking			
Never	45 (93.75)	222 (92.50)	0.577
Former	0 (0)	3 (1.25)	
Current	3 (6.25)	15 (6.25)	
Anti-copper therapy	33 (68.75)	205 (85.42)	0.005
Cirrhosis	39 (81.25)	171 (71.25)	0.155

Data are expressed as median (interquartile range) or n (%). BMI, body mass index.

which is consistent with the international diagnostic scoring system for WD [1].

Statistical Analyses

Data were analyzed using SPSS Statistics version 22 (IBM Corp., Armonk, NY, USA). Normally distributed continuous variables are expressed as mean ± standard deviation, non-normally distributed continuous variables as median (interquartile range), and categorical variables as frequency (percentage). For univariate comparisons between groups with and without anemia, statistical significance was determined using Student's t tests, Mann-Whitney U tests, χ^2 tests, or Fisher's exact tests, as appropriate. Additionally, multivariate linear regression and multivariate logistic regression were used to adjust for age, sex, BMI, alcoholism, smoking status, anti-copper therapy, and cirrhosis to assess the relationship of anemia with laboratory results and hepatic complications. Kaplan-Meier curves and Cox proportional hazards regression models (adjusted for age, sex, BMI, alcoholism, smoking, anti-copper therapy, and cirrhosis [for the advanced Child-Pugh classification]) were used to determine whether anemia was a risk factor for an advanced Child-Pugh classification (grade B or C) in patients with WD and hepatic decompensation in patients with WD-associated cirrhosis. The results of significant variables in multivariate logistic regression are expressed as odds ratios (ORs) (95% confidence interval [CI]) and p values, while the results of Cox regression are expressed as hazard ratios (95% CI) and p values. A p value <0.05 was considered statistically significant.

Results

Patient Characteristics

The clinical data of 294 patients with WD were reviewed retrospectively. Based on our inclusion and exclusion criteria, a total of 288 patients were enrolled in our study, with 48 (16.67%) having anemia and 240 not having anemia. The demographic and clinical characteristics of the groups with and without anemia are detailed in Table 1. The duration of WD (5.00 [0.00–10.00] vs. 5.00 [1.00–10.00] years, p = 0.909), as well as the rates of neurological manifestation (75.00% vs. 62.08%, p = 0.088), psychiatric manifestation (8.33% vs. 11.25%, p = 0.552), and the Kayser-Fleischer ring (37.50% vs. 30.83%, p = 0.366), were not significantly different between the anemic and non-anemic groups. However, urinary copper levels were significantly higher in patients with anemia than in those without anemia (751.4 [197.3–1,514] vs. 572.2 [226.1–982.4] μg/24 h, p = 0.405).

In patients undergoing anti-copper therapy, 201 (84.45%) were treated with sodium dimercaptosulfonate alone, 6 (2.53%) with penicillamine alone, 29 (12.18%) with sodium dimercaptosulfonate and penicillamine, and 2 (0.84%) with sodium dimercaptosulfonate and dimercaptosuccinic acid. The prevalence of anti-copper therapy was significantly lower in the anemic group compared to the non-anemic group (68.75% vs. 85.42%, p = 0.005). After adjusting for age, sex, BMI, alcoholism, smoking, and cirrhosis, anti-copper therapy was associated with a lower risk of anemia in patients with WD (OR = 0.36, 95% CI: 0.17–0.76; p = 0.008).

Anemia and Liver-Associated Disease Severity

In terms of liver function parameters, WD patients with anemia had significantly higher levels of bilirubin, ALT, PT, and INR and significantly lower levels of albumin and platelet count compared to those without anemia (all p < 0.05). In terms of blood lipid parameters,

Table 2. Liver-related parameters in WD patients with and without anemia

Variable	Anemic patients (N = 48)	Non-anemic patients (N = 240)	p value
Liver function parameters			
Bilirubin, μmol/L	12.65 (8.725–40.85)	10.70 (8.23–15.78)	0.017
ALT, U/L	33.50 (23.50–54.75)	26.00 (20.00–38.75)	0.038
AST, U/L	27.50 (12.50–46.75)	28.00 (16.25–45.00)	0.661
Albumin, g/L	36.74±4.57	39.98±4.17	<0.001
Coagulation parameters			
PT, s	15.15 (14.10–18.78)	14.00 (13.20–14.70)	<0.001
INR	1.20 (1.08–1.58)	1.08 (1.01–1.15)	<0.001
Platelet count, 10^9/L	109.00 (78.25–214.50)	162.00 (115.00–230.80)	0.005
Blood lipid parameters			
Total cholesterol, mmol/L	3.26 (2.78–3.81) n = 38	3.91 (3.38–4.46) n = 210	<0.001
Triglycerides, mmol/L	0.77 (0.56–1.04) n = 38	0.88 (0.70–1.29) n = 210	0.019
HDL-C, mmol/L	1.06 (0.88–1.28) n = 38	1.26 (1.09–1.47) n = 210	<0.001
LDL-C, mmol/L	1.75 (1.45–2.21) n = 38	2.19 (1.75–2.59) n = 210	0.004
Liver fibrosis parameters			
FIB-4	0.97 (0.47–1.88)	0.76 (0.39–1.36)	0.075
APRI	0.61 (0.23–1.32)	0.45 (0.24–0.76)	0.212
Type IV collagen, ng/mL	61.47 (49.35–83.07) n = 36	55.61 (47.47–64.95) n = 224	0.049
PIIINP, μg/mL	94.77 (66.99–117.70) n = 34	81.16 (63.81–110.90) n = 220	0.192
Laminin ng/mL	106.10 (95.22–125.80) n = 36	110.90 (98.18–123.10) n = 224	0.735
Hyaluronic acid, ng/mL	97.10 (33.92–233.10) n = 36	51.45 (31.21–86.41) n = 224	0.002
Portal vein diameter, mm	10.00 (10.00–12.00) n = 32	10.00 (9.00–11.00) n = 191	0.070
Child-Pugh classification			
A	41 (85.42)	234 (97.50)	0.001
B/C	7 (14.58)	6 (2.50)	
Hepatic decompensation	16 (33.33)	13 (5.42)	<0.001

Data are expressed as mean ± standard deviation, median (interquartile range), or n (%). ALT, alanine transaminase; APRI, aspartate aminotransferase-to-platelet ratio index; AST, aspartate transaminase; BMI, body mass index; FIB-4, fibrosis-4; HDL-C, high-density lipoprotein-cholesterol; INR, international normalized ratio; LDL-C, low-density lipoprotein-cholesterol; PIIINP, procollagen type-III N-terminal propeptide; PT, prothrombin time; WD, Wilson disease.

the levels of total cholesterol, triglycerides, and HDL-C were all significantly lower in the anemic group (all p < 0.05). In terms of liver fibrosis markers, the levels of type IV collagen and hyaluronic acid were significantly higher (p = 0.49 and p = 0.002, respectively) in the anemic group. Additionally, anemic WD patients had a higher rate of advanced Child-Pugh classification (p = 0.001) and hepatic decompensation (p < 0.001) (Table 2).

Multivariate linear regression analyses revealed that WD patients with anemia had significantly higher levels of bilirubin, ALT, PT, INR, type IV collagen, and hyaluronic acid and significantly lower levels of albumin, total cholesterol, and HDL-C compared to those without anemia (all p < 0.05) (Table 3). Moreover, multivariate logistic regression analyses revealed that anemia was a risk factor for advanced Child-Pugh classification (OR = 7.56, 95% CI: 2.92–19.59; p < 0.001) and hepatic decompensation (OR = 9.60, 95% CI: 3.48–26.50; p < 0.001).

Anemia and Hepatic Complications

WD patients with anemia had higher rates of gastric varices (18.75% vs. 2.50%, p < 0.001), ascites (22.92% vs. 1.67%, p < 0.001), liver failure (8.33% vs. 0.42%, p = 0.002), and hepatic encephalopathy (8.33% vs. 0.83%, p = 0.006) than those without anemia (Table 4). SBP, renal impairment, and liver cancer were absent in both groups.

According to multivariate logistic regression, anemia was a significant predictor of gastric varices (OR = 6.24, 95% CI: 1.95–19.90; p = 0.002) and ascites (OR = 21.93, 95% CI: 4.21–114.20; p < 0.001).

Anemia and the Progression of WD-Associated Cirrhosis

A total of 208 WD patients without an advanced Child-Pugh classification at baseline had at least one follow-up visit. During the median follow-up of 25.4 (13.1–41.7) months, 11 cases progressed to Child-Pugh classification. According to the fully adjusted Cox proportional hazards regression model, there were 15.08 times the number of cases with an

Table 3. Multivariate linear regression analyses of hepatic features in WD patients with and without anemia

Variable	Beta	Standard error	*p* value
Liver function parameters			
Bilirubin, μmol/L	21.328	8.486	0.013
ALT, U/L	9.294	3.357	0.006
AST, U/L	–	–	–
Albumin, g/L	−2.558	0.675	<0.001
Coagulation parameters			
PT, s	2.349	0.389	<0.001
INR	0.256	0.042	<0.001
Platelet count, 10^9/L	–	–	–
Blood lipid parameters			
Total cholesterol, mmol/L	−0.577	0.177	0.017
Triglycerides, mmol/L	–	–	–
HDL-C, mmol/L	−0.232	0.051	<0.001
LDL-C, mmol/L	–	–	–
Liver fibrosis parameters			
FIB-4	–	–	–
APRI	–	–	–
Type IV collagen, ng/mL	11.315	3.532	0.002
PIIINP, μg/mL	–	–	–
Laminin, ng/mL	–	–	–
Hyaluronic acid, ng/mL	112.933	27.818	<0.001
Portal vein diameter, mm	–	–	–

Analyses were adjusted for age, sex, BMI, alcoholism, smoking, anti-copper therapy use, and cirrhosis. ALT, alanine transaminase; APRI, aspartate aminotransferase-to-platelet ratio index; AST, aspartate transaminase; BMI, body mass index; CI, confidence interval; FIB-4, fibrosis-4; HDL-C, high-density lipoprotein-cholesterol; INR, international normalized ratio; LDL-C, low-density lipoprotein-cholesterol; OR, odds ratio; PIIINP, procollagen type-III N-terminal propeptide; PT, prothrombin time; WD, Wilson disease.

Table 4. Hepatic complications in patients with and without anemia

Variable	Anemic patients (*N* = 48)	Non-anemic patients (*N* = 240)	*p* value
Gastric varices	9 (18.75)	6 (2.50)	<0.001
Splenomegaly/splenectomy	33 (68.75)	135 (56.25)	0.109
Ascites	11 (22.92)	4 (1.67)	<0.001
SBP	0 (0)	0 (0)	–
Liver failure	4 (8.33)	1 (0.42)	0.002
Renal impairment	0 (0)	0 (0)	–
Hepatic encephalopathy	4 (8.33)	2 (0.83)	0.006
Liver cancer	0 (0)	0 (0)	–

Data are expressed as *n* (%).

advanced Child-Pugh classification among WD patients with anemia than among those without anemia (hazard ratio = 15.08, 95% CI: 1.22–186.05; $p = 0.034$).

A total of 155 WD patients with compensated cirrhosis at baseline had at least one follow-up visit. During the median follow-up of 25.9 (14.9–40) months, 13 of them developed decompensated cirrhosis. However, the fully adjusted Cox model revealed that anemia did not increase the risk of decompensated cirrhosis.

Table 5. Linear regression analyses of hepatic features in WD patients with and without hemolytic anemia

Variable	Model 1			Model 2		
	Beta	Standard error	*p* value	Beta	Standard error	*p* value
Total bilirubin, μmol/L	248.95	86.32	0.006	–	–	–
ALT, U/L	88.97	16.65	<0.001	51.59	16.17	0.003
AST, U/L	68.31	12.95	<0.001	38.92	14.60	0.011
PT, s	14.68	4.53	0.002	–	–	–
INR	1.83	0.55	0.002	–	–	–
Triglycerides, mmol/L	–	–	–	1.35	0.38	0.001
Type IV collagen, ng/mL	164.04	19.68	<0.001	101.00	25.04	<0.001
PIIINP, μg/mL	176.81	39.46	<0.001	–	–	–
Laminin, ng/mL	86.63	13.38	<0.001	77.35	19.00	<0.001
Hyaluronic acid, ng/mL	877.83	210.70	0.006	–	–	–

ALT, alanine transaminase; AST, aspartate transaminase; INR, international normalized ratio; PIIINP, procollagen type-III N-terminal propeptide; PT, prothrombin time; WD, Wilson disease. Model 1: non-adjusted. Model 2: adjusted for age, sex, BMI, alcoholism, smoking, anti-copper therapy use, and cirrhosis.

Etiology and Severity of Anemia for the Liver-Associated Disease Severity, Hepatic Complications, and Progression of WD-Associated Cirrhosis

Table 5 shows that patients with hemolytic anemia had significantly higher levels of total bilirubin, ALT, aspartate transaminase, PT, INR, type IV collagen, PIIINP, laminin, and hyaluronic acid than those without hemolytic anemia (all $p < 0.05$). However, hemolysis did not increase the risk of hepatic complications or advanced Child-Pugh classification in the anemic patients.

Among the anemic patients, 38 had mild anemia (Hb less than 9 g/dL) and 10 had moderate anemia (Hb between 6 g/dL and 9 g/dL). Patients with moderate anemia had a higher risk of gastric varices than those with mild anemia (OR = 8.27, 95% CI: 1.41–48.54; $p = 0.019$). However, the levels of liver-associated disease severity parameters and the risk of progression to the advanced Child-Pugh classification did not differ significantly between patients with mild and moderate anemia (all $p > 0.05$).

Discussion

Anemia was found to be common in WD patients, and it was associated with a greater disease severity, a higher risk of hepatic complications, and a faster progression. Moreover, in WD patients with anemia, hemolysis was associated with a more severe liver dysfunction. To the best of our knowledge, this is the first study to investigate the relationship between anemia and disease severity, complications, and progression in WD patients.

Anemia is a common hematologic disorder in chronic liver diseases that has been linked to disease severity [27]. A recent study of 494 patients with the advanced chronic liver disease found that patients with anemia had lower levels of serum albumin and higher levels of INR, hepatic venous pressure, Child-Pugh score, and model for end-stage liver disease points, as well as a higher rate of decompensated liver disease compared to those without anemia [14]. Another study of 239 cirrhotic patients found that those with an advanced Child-Pugh classification were more likely to have low hemoglobin levels [28]. Moreover, anemia was found to be a predictor of liver fibrosis and cirrhosis in patients with chronic hepatitis B infection [29]. In line with these studies, we found that WD patients with anemia, particularly hemolytic anemia, had a more severe liver injury, worse coagulation, more serious lipid synthesis dysfunction, more advanced liver fibrosis, and a more advanced disease stage than those without anemia. Our findings indicate that anemia, hemolytic anemia, in particular, is an important indicator of severity in patients with WD and should be valued accordingly.

The following mechanisms can help explain the above findings: First, patients with more severe WD may be more likely to develop anemia. In patients with severe WD, a higher level of non-ceruloplasmin-bound copper was found to be released into the blood from necrotic hepatocytes, causing oxidative damage in erythrocytes [30]. Anemia is also exacerbated by gastrointestinal bleeding, hypersplenism, and poor nutrition, which are all symptoms of severe liver disease [31]. Furthermore,

copper-transporting ATPase dysfunction impaired ceruloplasmin synthesis, which decreased ferroxidase activity, resulting in iron accumulation in the liver and contributing to iron deficiency anemia [32]. Second, anemia can also aggravate WD. Anemia not only increases gastric blood flow but also worsens hyperdynamic circulation in patients with chronic liver disease, potentially exacerbating portal hypertension [33]. Additionally, higher serum copper levels have been reported in WD patients with hemolytic anemia [17], which can lead to hepatocyte stress and death, worsening liver function [34]. Impaired liver function disrupts bile acid synthesis and secretion, reduces fat absorption, and subsequently leads to low blood lipid levels [35]. Third, some case reports indicate that copper deficiency in WD patients caused by overtreatment may also cause anemia by affecting ferroportin stability [36, 37].

Anemia was also found to be a risk factor for hepatic complications in patients with WD, including gastric varices, ascites, and hepatic encephalopathy. Similarly, Paternostro et al. [33] demonstrated that anemia was associated with a higher incidence of ascites in patients with cirrhosis. Moreover, Scheiner et al. [14] reported that patients with anemia were more likely to have hepatic complications, as evidenced by higher incidence rates of varices and refractory ascites. Notably, our findings revealed that anemia was associated with an increased risk of hepatic encephalopathy. Given that gastrointestinal bleeding is the leading cause of anemia in patients with chronic liver disease [13], the higher incidence of hepatic encephalopathy in WD patients with anemia may be due to hyperammonemia caused by gastrointestinal bleeding. Although anemia has previously been found to be an independent risk factor for intra-abdominal bacterial infections [38], liver failure [39], and liver cancer [40] in patients with chronic liver diseases, no such associations were found in our study because SBP and liver cancer were absent, and liver failure was rare among our enrolled patients.

Anemia is also considered to be a major contributor to disease progression and poor outcomes in many chronic liver diseases [33, 41–43]. For instance, a retrospective cohort study of 1,244 patients indicated that anemia was an independent risk factor for hepatic decompensation in compensated patients [33]. Several studies have also revealed that anemia is associated with a higher mortality rate in patients with alcoholic liver disease, pyogenic liver abscess, acute-on-chronic liver failure, and hepatic encephalopathy [15, 42–44]. In our cohort, WD patients with anemia had a higher risk of developing an advanced Child-Pugh classification, indicating that anemia may promote WD progression. Therefore, treating anemia may delay disease progression and improve clinical outcomes in these patients. However, because no patients died during the follow-up period, it is unclear whether anemia contributes to higher mortality in WD patients.

According to our findings, non-anemic WD patients were more likely to receive anti-copper therapy. Furthermore, multivariate logistic regression revealed that anti-copper therapy was associated with a lower risk of anemia in WD patients. Several studies have reported that early anti-copper therapy can reduce copper accumulation in the body while stabilizing liver function [45, 46]. Moreover, lowering the level of non-ceruloplasmin-bound copper was beneficial in alleviating oxidative damage in erythrocytes and, thus, hemolysis [30]. Therefore, encouraging WD patients to initiate and adhere to anti-copper therapy could lower the incidence of anemia and thereby delay WD progression.

Some limitations are worth mentioning. First, due to the retrospective nature of the study, we were unable to control for some potential confounders, such as dietary copper intake. Second, because several hepatic complications were rare or even absent among the enrolled patients, the relationship between anemia and these complications (liver failure, SBP, renal impairment, and liver cancer) was difficult to assess. Third, while hemolytic anemia is associated with the severity of WD, determining the cause of hemolysis is difficult because it can be caused by WD-associated copper accumulation or cirrhosis-associated hypersplenism. Finally, the median duration of follow-up was only 25 months. Therefore, additional research with larger sample sizes and longer follow-up periods is required to confirm our findings.

In conclusion, anemia was found to be associated with more severe disease, a higher risk of hepatic complications, and a faster progression in WD patients. By identifying anemia in WD patients, early intervention may be able to prevent hepatic complications and delay disease progression.

Statement of Ethics

This study was approved by the Research Ethics Board of the First Affiliated Hospital of Guangdong Pharmaceutical University (#2021-172). Due to the retrospective nature of the study, the need for informed consent was waived.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

Funding Sources

This study was supported by the Postdoctoral Innovation and Practice Research Project of Jiangmen, China (grant number JMBSH2021B09).

Author Contributions

S.Q.W., Y.Q.Z., Y.P.Z., X.H., and H.Q.L. contributed to the collection and analysis of the data. M.F.H. and H.J.Z. contributed to the conception and design of the study. S.Q.W., Y.Q.Z., and H.J.Z. wrote and revised the manuscript. All authors contributed to the article and approved the submitted version.

Data Availability Statement

All the data generated or analyzed during this study are included in this article. Further inquiries can be directed to the corresponding author (Hao-Jie Zhong).

References

1 Członkowska A, Litwin T, Dusek P, Ferenci P, Lutsenko S, Medici V, et al. Wilson disease. Nat Rev Dis Primers. 2018;4(1):21.
2 Coffey AJ, Durkie M, Hague S, McLay K, Emmerson J, Lo C, et al. A genetic study of Wilson's disease in the United Kingdom. Brain. 2013;136(Pt 5):1476–87.
3 Gao J, Brackley S, Mann JP. The global prevalence of Wilson disease from next-generation sequencing data. Genet. Med2019 May;21(5):1155–63.
4 Fernando M, van Mourik I, Wassmer E, Kelly D. Wilson disease in children and adolescents. Arch Dis Child. 2020;105(5):499–505.
5 Roberts EA, Socha P. Wilson disease in children. Handb Clin Neurol. 2017;142:141–56.
6 Ferenci P, Stremmel W, Członkowska A, Szalay F, Viveiros A, Stattermayer AF, et al. Age and sex but not ATP7B genotype effectively influence the clinical phenotype of Wilson disease. Hepatology. 2019;69(4):1464–76.
7 Mulligan C, Bronstein JM. Wilson disease: an overview and approach to management. Neurol Clin. 2020;38(2):417–32.
8 Caraceni P, Tufoni M, Zaccherini G, Riggio O, Angeli P, Alessandria C, et al. On-treatment serum albumin level can guide long-term treatment in patients with cirrhosis and uncomplicated ascites. J Hepatol. 2021;74(2):340–9.
9 Trieb M, Rainer F, Stadlbauer V, Douschan P, Horvath A, Binder L, et al. HDL-related biomarkers are robust predictors of survival in patients with chronic liver failure. J Hepatol. 2020;73(1):113–20.
10 Piano S, Tonon M, Vettore E, Stanco M, Pilutti C, Romano A, et al. Incidence, predictors and outcomes of acute-on-chronic liver failure in outpatients with cirrhosis. J Hepatol. 2017;67(6):1177–84.
11 Zhong HJ, Zhuang YP, Zhang YT, Xu SP, Hong MF, He XX. Distinguishing between the complications of Wilson disease-related cirrhosis and HBV-related cirrhosis. Curr Med Res Opin. 2022;38(1):75–81.
12 Zhong HJ, Sun HH, Xue LF, McGowan EM, Chen Y. Differential hepatic features presenting in Wilson disease-associated cirrhosis and hepatitis B-associated cirrhosis. World J Gastroenterol. 2019;25(3):378–87.
13 Gkamprela E, Deutsch M, Pectasides D. Iron deficiency anemia in chronic liver disease: etiopathogenesis, diagnosis and treatment. Ann Gastroenterol. 2017;30(4):405–13.
14 Scheiner B, Semmler G, Maurer F, Schwabl P, Bucsics TA, Paternostro R, et al. Prevalence of and risk factors for anaemia in patients with advanced chronic liver disease. Liver Int. 2020;40(1):194–204.
15 Novo-Veleiro I, Herrera-Flores J, Roson-Hernandez B, Medina-Garcia JA, Muga R, Fernandez-Sola J, et al. Alcoholic liver disease among patients with Wernicke encephalopathy: a multicenter observational study. Drug Alcohol Depend. 2022;230:109186.
16 Xu Z, Berry BR. Oxidative hemolysis due to Wilson disease. Blood. 2019;134(7):657.
17 Forman SJ, Kumar KS, Redeker AG, Hochstein P. Hemolytic anemia in Wilson disease: clinical findings and biochemical mechanisms. Am J Hematol. 1980;9(3):269–75.
18 Cai J, Wu M, Ren J, Du Y, Long Z, Li G, et al. Evaluation of the efficiency of the reticulocyte hemoglobin content on diagnosis for iron deficiency anemia in Chinese adults. Nutrients. 2017;9(5):450.
19 Driever EG, Stravitz RT, Zhang J, Adelmeijer J, Durkalski V, Lee WM, et al. VWF/ADAMTS13 imbalance, but not global coagulation or fibrinolysis, is associated with outcome and bleeding in acute liver failure. Hepatology. 2021;73(5):1882–91.
20 Piano S, Rosi S, Maresio G, Fasolato S, Cavallin M, Romano A, et al. Evaluation of the acute kidney injury network criteria in hospitalized patients with cirrhosis and ascites. J Hepatol. 2013;59(3):482–9.
21 Riordan SM, Williams R. Treatment of hepatic encephalopathy. N Engl J Med. 1997;337(7):473–9.
22 Jachs M, Hartl L, Simbrunner B, Bauer D, Paternostro R, Scheiner B, et al. Decreasing von Willebrand factor levels upon nonselective beta blocker therapy indicate a decreased risk of further decompensation, acute-on-chronic liver failure, and death. Clin Gastroenterol Hepatol. 2022;20(6):1362–73.e6.
23 Sterling RK, Lissen E, Clumeck N, Sola R, Correa MC, Montaner J, et al. Development of a simple noninvasive index to predict significant fibrosis in patients with HIV/HCV coinfection. Hepatology. 2006;43(6):1317–25.
24 Lin ZH, Xin YN, Dong QJ, Wang Q, Jiang XJ, Zhan SH, et al. Performance of the aspartate aminotransferase-to-platelet ratio index for the staging of hepatitis C-related fibrosis: an updated meta-analysis. Hepatology. 2011;53(3):726–36.
25 Pugh RN, Murray-Lyon IM, Dawson JL, Pietroni MC, Williams R. Transection of the oesophagus for bleeding oesophageal varices. Br J Surg. 1973;60(8):646–9.
26 Zhong HJ, Xiao P, Lin D, Zhou HM, He XX. Cirrhosis in Wilson disease is characterized by impaired hepatic synthesis, leukopenia and thrombocytopenia. Int J Med Sci. 2020;17(10):1345–50.
27 Qamar AA, Grace ND, Groszmann RJ, Garcia-Tsao G, Bosch J, Burroughs AK, et al. Incidence, prevalence, and clinical significance of abnormal hematologic indices in compensated cirrhosis. Clin Gastroenterol Hepatol. 2009;7(6):689–95.
28 Sahin A, Artas H, Tunc N, Yalniz M, Bahcecioglu IH. Hematological indices in portal hypertension: cirrhosis versus noncirrhotic portal hypertension. J Clin Med. 2018;7(8):196.
29 Poon TC, Hui AY, Chan HL, Ang IL, Chow SM, Wong N, et al. Prediction of liver fibrosis and cirrhosis in chronic hepatitis B infection by serum proteomic fingerprinting: a pilot study. Clin Chem. 2005;51(2):328–35.
30 Dzieżyc-Jaworska K, Litwin T, Członkowska A. Clinical manifestations of Wilson disease in organs other than the liver and brain. Ann Transl Med. 2019;7(Suppl 2):S62.

31 Varghese J, Varghese James J, Karthikeyan M, Rasalkar K, Raghavan R, Sukumaran A, et al. Iron homeostasis is dysregulated, but the iron-hepcidin axis is functional, in chronic liver disease. J Trace Elem Med Biol. 2020;58:126442.
32 Jończy A, Lipiński P, Ogorek M, Starzyński RR, Krzysztofik D, Bednarz A, et al. Functional iron deficiency in toxic milk mutant mice (tx-J) despite high hepatic ferroportin: a critical role of decreased GPI-ceruloplasmin expression in liver macrophages. Metallomics. 2019;11(6):1079–92.
33 Paternostro R, Kapzan L, Mandorfer M, Schwarzer R, Benedikt S, Viveiros A, et al. Anemia and iron deficiency in compensated and decompensated cirrhosis: prevalence and impact on clinical outcomes. J Gastroenterol Hepatol. 2020;35(9):1619–27.
34 Wooton-Kee CR, Jain AK, Wagner M, Grusak MA, Finegold MJ, Lutsenko S, et al. Elevated copper impairs hepatic nuclear receptor function in Wilson's disease. J Clin Invest. 2015;125(9):3449–60.
35 Li T, Chiang JY. Bile acid signaling in metabolic disease and drug therapy. Pharmacol Rev. 2014;66(4):948–83.
36 Dzieżyc K, Litwin T, Sobańska A, Członkowska A. Symptomatic copper deficiency in three Wilson's disease patients treated with zinc sulphate. Neurol Neurochir Pol. 2014;48(3):214–8.
37 Harada M, Miyagawa K, Honma Y, Hiura M, Shibata M, Matsuhashi T, et al. Excess copper chelating therapy for Wilson disease induces anemia and liver dysfunction. Intern Med. 2011;50(14):1461–4.
38 Nie K, Ran R, Tan W, Yi B, Luo X, Yu Y, et al. Risk factors of intra-abdominal bacterial infection after liver transplantation in patients with hepatocellular carcinoma. Chin J Cancer Res. 2014;26(3):309–14.
39 Jiang AA, Greenwald HS, Sheikh L, Wooten DA, Malhotra A, Schooley RT, et al. Predictors of acute liver failure in patients with acute hepatitis A: an analysis of the 2016-2018 San Diego county hepatitis A outbreak. Open Forum Infect Dis. 2019;6(11):ofz467.
40 Suzuki A, Lymp J, Donlinger J, Mendes F, Angulo P, Lindor K. Clinical predictors for hepatocellular carcinoma in patients with primary biliary cirrhosis. Clin Gastroenterol Hepatol. 2007;5(2):259–64.
41 Virk ZM, Patel AA, Leaf RK, Al-Samkari H. Predictors of mortality and outcomes of liver transplant in spur cell hemolytic anemia. Am J Hematol. 2021;96(12):1611–20.
42 Chen JF, Weng WZ, Huang M, Peng XH, He JR, Zhang J, et al. Derivation and validation of a nomogram for predicting 90-day survival in patients with HBV-related acute-on-chronic liver failure. Front Med. 2021;8:692669.
43 Xu J, Zhou X, Zheng C. The geriatric nutritional risk index independently predicts adverse outcomes in patients with pyogenic liver abscess. BMC Geriatr. 2019;19(1):14.
44 Hu XP, Gao J. International normalized ratio and Model for End-stage Liver Disease score predict short-term outcome in cirrhotic patients after the resolution of hepatic encephalopathy. World J Gastroenterol. 2019;25(26):3426–37.
45 Cochen De Cock V, Woimant F, Poujois A. Sleep disorders in Wilson's disease. Curr Neurol Neurosci Rep. 2019;19(11):84.
46 Zhuang YP, Zhong HJ. Impact of COVID-19 on the clinical status of patients with Wilson disease. World J Gastroenterol. 2021;27(26):4248–51.