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Comparative study on the impact on mouse livers of different amounts of Chinese Baijiu, beer, and wine consumption

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Abstract

Due to chronic alcohol consumption, alcoholic liver disease (ALD) has become the second most common liver disease in the world. The study was to explore the effects of consuming different amounts of various alcoholic beverages on the liver of mice. Results showed wine contained far more polyphenols and organic acids than beer and Chinese Baijiu. When daily alcohol consumption was low (less than 100 mL/60 kg/day), the liver and serum indicators of mice treated with alcohol, beer, Chinese Baijiu, and wine were not significantly different from those in saline group. At 250 mL/60 kg/day, beer used significantly damaged the liver and caused adverse changes in serum components, while wine did not cause changes in serum, indicating that it may cause less damage due to the existence of a large number of polyphenols. When the daily consumption exceeded 500 mL/60 kg/day, all four alcoholic beverages significantly increased serum markers of hepatic injury and decreased serum total antioxidant capacity and liver antioxidant enzyme levels, resulting in liver fat accumulation and serious pathological damage. At this level, beer used caused the most damage and wine the least. This study explores the appropriate drinking amount of different alcoholic beverages and provides a scientific reference for alcohol consumption.

Keywords: wine; beer; Chinese Baijiu; liver; moderate drinking.

Practical Application: It could provide a scientific reference for alcohol consumption.

1 Introduction

Alcohol has a vital role in dining culture in China and around the world (Sun et al., 2019). The types and quantities of alcohol consumption in China are constantly changing as this culture develops (Yang et al., 2019). Chinese Baijiu, beer, and wine are widely consumed by a large range of consumers, have a high market share, and may dominate the Chinese alcohol market for a long time. However, excessive ethyl alcohol intake can negatively impact the human body, especially the liver (Griswold et al., 2018), and long-term drinking can lead to alcoholic liver disease (ALD) (Griswold et al., 2018). More than 90% of alcohol is absorbed in the intestine, and most of it is metabolized in the liver (Tian et al., 2020). Alcohol and its metabolites, acetaldehydes, can directly disrupt intestinal functioning and impair hepatic proteins and DNA, resulting in hepatocyte injury (Tian et al., 2020). Ethyl alcohol initially causes triglycerides to accumulate in hepatocytes, leading to hepatic steatosis, which is clinically manifested as alcoholic fatty liver (steatosis). Further deterioration leads to hepatitis and cirrhosis, which may eventually lead to hepatocellular carcinoma (Bajaj, 2019; Zheng & Wang, 2020).

Of all alcoholic beverages, wine is an interesting and unique product. Wine may be distinguished from the others by the presence of grape-derived phenolic compounds (Deroover et al., 2021; Miele, 2021; Zhang et al., 2022). Somani et al. (2015) suggested that grape polyphenols (including anthocyanins, flavonols, favan-3-ols, stilbene, and phenolic acids) could have anti-inflammatory activities that regulate multiple molecular targets and pathways. Yin et al. (2017) found that grape seed procyanidin B2 could have a positive effect on hepatic lipid metabolism disorders by reducing hepatic lipid synthesis in db/db mice. Gerardi et al. (2020) showed that some grape polyphenols, such as resveratrol or epigallocatechin gallate, could reduce PPARγ, thus inhibiting the differentiation and proliferation of adipocytes. Meanwhile, a report on 219,279 Norwegians aged 30-67 found that drinking wine was significantly negatively correlated with mortality from ALD (Tverdal et al., 2018). Of course, these health benefits are based on moderate wine drinking. Our team had previously confirmed through *in vitro* gastrointestinal digestion that the bioaccessibility of most polyphenols decreased as the amount consumed increased (Sun et al., 2020). The World Health Organization (WHO) has stated that excessive drinking is harmful and associated with a range of negative social and health consequences.

In China, people are more comfortable choosing the type and amount of drinking according to their own preferences and habits when drinking at home, but in social situations, Chinese people are more likely to be affected by social factors such as drinking occasions, local culture, and atmosphere. The Chinese culture has many concepts such as "drink and enjoy yourself", "no wine, no banquet", and "wine is the essence of grain, the

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more you drink, the younger you are", which affects people's choice and consumption of alcoholic beverage (He et al., 2016; Im et al., 2021; Yoon et al., 2015). Because of this, drinking becomes the most important part of the social event, which often leads to overdrinking and even shock or death (He et al., 2016; Im et al., 2021). At present, there have been many reports on the relationship between alcohol consumption and ALD, but the relationship between different alcoholic beverage types or the amount consumed and ALD is unclear. In this study, we established an mice model of chronic hepatic injury similar to the mechanism of human ALD and compared the effects of drinking different amounts of Chinese Baijiu, beer, and wine on the liver of mice under the same alcohol intake. We explored the effects of different alcoholic beverages and drinking amounts on the liver to provide consumers with more scientific and reasonable drinking methods.

2 Materials and methods

2.1 Chemicals

All standard substances including resveratrol, rutin, catechin, etc. (purity HPLC 98%) were purchased from from Merck (Darmstadt, Germany). The diagnostic kits specific for aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin (TBIL), alkaline phosphatase (ALP), total cholesterol (TC), triacylglycerols (TG), the total antioxidant capacity (T-AOC) kit, catalase (CAT), glutathione (GSH), superoxide dismutase (SOD), and lipid peroxidation [expressed as nmol of malondialdehyde equivalents (MDAeq)] were obtained from the Jiancheng Institute of Biotechnology (Nanjing, Jiangsu, China). Haematoxylin and eosin (H&E) were purchased from the Shanghai Lanji Technological Development Co. Ltd. (Shanghai, China). All of the other chemicals and reagents used in the experiments were of analytical grade.

2.2 Intragastric fluid

The actual intragastric fluid was based on the daily intragastric dose of 100 mL/60 kg/day, 250 mL/60 kg/day, 500 mL/60 kg/day, and 750 mL/60 kg/day of wine (14% vol), and the other 3 alcoholic beverages were prepared according to the principle of the same ethyl alcohol intake (Table S1).

Determination of phenolics contents of intragastric fluid

Analysis of the phenolic composition of intragastric fluid was conducted using a Waters Alliance HPLC 2695 (Waters, Milford, USA) equipped with a photodiode array detector 2996 (Waters, Milford, USA). Separation was performed using a Capcell Pak C18 column (250 mm × 4.6 mm, 5 μm) (Shiseido, Osaka, Japan) with the column temperature was 28 °C and the detection wavelength was 280 nm. The injection volume was 10 μL, and the peak area external standard method was used. The flow rate was 0.5 mL/min. The mobile phase consisted of solvent A [V(water): V(acetonitrile) = 19:1, containing 0.3% acetic acid], and solvent B [V(acetonitrile): V(water) = 9:1, containing 0.2% acetic acid], using the following gradient elution program for separation: 0-16 min, 12-14% (B); 16-18 min, 14% (B); 18-30 min, 14-16% (B); 30-36 min, 16-20% (B); 36-46 min, 20-24% (B); 46-56 min, 24-30% (B); 56-66 min, 30-50% (B); 66-70 min, 50% (B); 70-80 min, 50-12% (B); and 80-85 min, 12% (B). The alcoholic beverage sample was filtered by 0.45 μm organic system membrane and tested.

Determination of organic acids of intragastric fluid

Analysis of the organic acid composition of intragastric fluid was conducted using a Waters Alliance HPLC 2695 (Waters, Milford, USA) equipped with a photodiode array detector 2996 (Waters, Milford, USA). Separation was performed using a Capcell Pak C18 column (250 mm × 4.6 mm, 5 μm) (Shiseido, Osaka, Japan) with the column temperature was 45 °C and the detection wavelength was 210 nm. The injection volume was 10 μL, and the peak area external standard method was used. The flow rate was 0.5 mL/min. The mobile phase consisted of solvent A (0.02 mol/L K_2HPO_4, adjusted pH to 2.3 by H_3PO_4 solution), and solvent B (methanol), using the following isocratic elution program for separation (Equation 1):

$$V(A):V(B) = 99:1 \quad (1)$$

2.3 Animals and experimental design

A total of 200 male ICR mice (weight 33 ± 2 g) were obtained from the Beijing Vital River Laboratory Animal Technology Co., Ltd. [license number of the experimental animals: SCXK (SCXK (Beijing) 2012-0001)]. They were allowed free access to tap water and rodent chow. The animals were housed under standard conditions with a 12/12 h light/dark cycle at 25 ± 2 °C and a humidity level of 55 ± 5%. All of the experimental animals were treated in accordance with the guidelines of the Chinese Council for Animal Care. After all the mice had adapted to the environment, the experiment was started.

The 200 male ICR mice were randomly divided into 5 groups: saline group (SG), alcohol group (AG), wine group (WG), beer group (BG) and Chinese Baijiu group (CBG). Each group were divided into four teams according to the gavage dose, a total of 20 teams, 10 mice in each team. The gavage was calculated according to the daily drinking amount equivalent to 60 kg body weight to simulate a human's (Table S2). After 90 days of continuous intragastric administration, all of the animals were sacrificed to collect blood and livers. The livers were immediately removed and washed using ice-cold physiological saline. The blood samples were centrifuged, the serum was stored at 4 °C until use, and the isolated livers were stored at -80 °C until analysis.

2.4 Determination of serum and liver indicators

The liver and spleen indices were calculated by the following equations: liver index = liver weight/body weight × 100% and spleen index = spleen weight/body weight × 100%. AST, ALT, TC, TG, ALP, TBIL, SOD, and T-AOC were evaluated using kits. Liver samples were prepared by homogenization in sterile PBS, centrifuged at 3000 × *g* 4 °C for 15 min, and the supernatant was taken. CAT, GSH, SOD, and MDA were measured strictly in accordance with the kit instructions. Liver sections were prepared for H&E staining and Oil red (OR) staining (Do et al., 2021), and

the percentage of the oil red area was calculated. The flowchart of the experimental design was as Figure S1.

2.5 Statistical analysis

The experimental results were expressed as the mean standard deviation (SD). A one-way analysis of variance (ANOVA) and Duncan's multiple range test was conducted to determine the significance using DPS software (version 7.05). SPSS 26.0 (IBM, Armonk, NY, USA) was used to perform principal component analysis (PCA). The statistical significance for all tests was set at $p < 0.05$.

3 Results

3.1 Analysis of the phenolic compounds and organic acids in the intragastric fluids

Phenolics are considered as the most contributing factor with biological activities in food, including phenolic acids, stilbenes, flavonols, flavanols, and anthocyanins derivatives. These health-promoting phenolics have antioxidant, anticancer and anti-inflammatory and other advantages for human (Zhang et al., 2021). Furthermore, organic acids are also recognized as important nutraceuticals (Robles et al., 2019). Through the analysis of the composition of the intragastric fluids, it was found that in wine, epicatechin (243.20 ± 12.93 mg/L), rutin (175.66 ± 2.06 mg/L) and ethyl 4-hydroxybenzoate (173.97 ± 11.42 mg/L) were the highest phenols; shikimic acid (1332.34 ± 11.65 mg/L) and malic acid (1320.56 ± 21.87 mg/L) were the most abundant organic acids (Table 1). What's more, although wine, beer and Chinese Baijiu all contained some bioactive substances, both the type and content of wine were much higher than those of other two alcoholic beverages, indicating that drinking wine could obtain more beneficial ingredients under the same intake.

3.2 Effects of different alcoholic beverages on serum enzyme indicators of mice

The duration of intragastric administration was 90 days. Initial lavage with a total of 200 mice and there were 190 mice at the end of intragastric administration.

AST and ALT

In the synthesis and catabolism of amino acids, the two hepatocyte enzymes, AST and ALT, play an important role and are the most sensitive indicators of hepatocyte damage in hepatic injury. The results of AST and ALT activities are shown in Figure 1.

By analyzing the serum AST levels (Figure 1A), it was found that when the daily intragastric dose was low (100 mL/60 kg/day and 250 mL/60 kg/day), there were no significant differences in AST values among the teams ($p > 0.05$), which was not enough to cause damage to the liver. The AST value of WG at 250 mL/60 kg/day was significantly lower than SG and other groups ($p < 0.05$). When the dose continued to increase (500 mL/60 kg/day and 750 mL/60 kg/day), the AST values of the 4 alcoholic beverages were significantly higher than SGs. However, the AST values of the WGs were significantly lower than the other alcoholic beverage groups, and there was no significant difference in AST level between AGs and CBGs, but it was significantly lower than BGs, indicating that at high doses, the degree of hepatic injury was ranked as follows: wine < ethyl alcohol and Chinese Baijiu < beer.

ALT levels are shown in Figure 1B. Different from AST, firstly, the ALT value of WG at 100 mL/60 kg/day was significantly higher than SG ($p < 0.05$), and when the dose was 250 mL/60 kg/day, there was no significant difference in ALT value of each group ($p > 0.05$). Secondly, when the dose increased to 750 mL/60 kg/day, there was no significant difference in ALT between AG and BG.

TBIL and ALP

TBIL and ALP are also sensitive indicators of liver and biliary tract diseases. When the liver is damaged or diseased, the serum TBIL and ALP increase (Figure 1C-1D). When the daily intragastric dose was 100 mL/60 kg/day, there were no significant differences in TBIL and ALP levels among SGs and the others ($p > 0.05$). At 250 mL/60 kg/day, the levels of TBIL and ALP in WG were not significantly different from those in SGs, and there was no significant difference among AG, BG, and CBG, but they were all significantly higher than SG in TBIL ($p < 0.05$). When the dose continued to increase (500 mL/60 kg/day and 750 mL/60 kg/day), there was no significant difference in the TBIL values of the four alcoholic beverages and SGs, but WGs were significantly lower than other alcoholic beverage groups.

The trend of the ALP value was slightly different. When the dose was 500 mL/60 kg/day, the ALP value of WG and SG had no significant difference, and AG and CBG also had no significant difference, but it was significantly higher than that of SG and significantly lower than that of BG, indicating that beer was the most harmful to the liver. When the dose was increased to 750 mL/60 kg/day, the ALP value of WG was significantly higher than SG but lower than the other three kinds of alcoholic beverages, indicating that drinking wine causes less damage to the liver at this dose.

3.3 Effects of different alcoholic beverages lipid indicators of mice

As shown in Figure 2A-2B, there were no significant differences in TC and TG values among the 4 alcoholic beverage groups and SG ($p > 0.05$) at low doses (100 mL/60 kg/day and 250 mL/60 kg/day), indicating that drinking various alcoholic beverages at this dose was not enough to affect the liver. When the intragastric dose continued to increase to 500 mL/60 kg/day, the TC value of BG was significantly higher than that of SG and AG, and the TG values of BG and CBG were significantly higher than SG ($p < 0.05$). When the dose was 750 mL/60 kg/day, the TC values of the 4 alcoholic beverage groups were significantly higher than SG, only WG had no significant difference in the TG value with SG.

Table 1. Contents of the phenolic compounds, organic acids and anthocyanins identified in the intragastric fluids (mg/L).

Wine										
Phenolic compounds	Flavanol									
	Epigallocatechin	Catechin	Epigallocatechin 3-gallate	Epicatechin	Epicatechin-3-O-gallate	Myricetin	Isorhamnetin			
	52.32 ± 4.23	149.18 ± 3.65	13.23 ± 0.65	243.20 ± 12.93	35.63 ± 1.79	32.46 ± 0.63	2.66 ± 0.08			
	Flavonol									
	Rutin	Isoquercitrin	Quercitrin	Resveratrol	Quercetin	Luteolin	Apigenin	Kaempferol		
	175.66 ± 2.06	114.52 ± 1.14	69.23 ± 3.27	4.89 ± 0.31	28.64 ± 1.87	1.43 ± 0.04	0.51 ± 0.04	1.34 ± 0.09		
	Hydroxycinnamic acid									
	Trans-fertaric acid	Caffeic acid	Coumaric acid	4-hydroxycinnamic acid	Ferulic acid	3-hydroxycinnamic acid	sinapic acid	Isoferulic acid		
	2.42 ± 0.22	19.45 ± 1.20	25.11 ± 1.58	5.34 ± 0.56	4.18 ± 0.13	3.422 ± 0.32	11.13 ± 0.11	4.41 ± 0.09		
	Hydroxy benzoic acid									
	Gallic acid	Protocatechuic acid	Gentisic acid	Vanillic acid	Syringic acid	Methyl 3,4-dihydroxybenzoate	Methyl vanillate	Ethyl 4-hydroxybenzoate	Vanilic acid ethyl ester	
	33.14 ± 3.10	0.76 ± 0.01	98.15 ± 8.38	38.21 ± 0.88	16.33 ± 0.23	6.43 ± 0.15	1.42 ± 0.08	173.97 ± 11.42	8.21 ± 0.52	
Organic acids	Tartaric acid	Malic acid	Shikimic acid	Acetic acid	Fumaric acid	Succinic acid				
	820.11 ± 11.23	1320.56 ± 21.87	1332.34 ± 11.65	321.44 ± 12.64	5.08 ± 0.02	804.21 ± 5.68				
Beer										
Phenolic compounds	Flavanol									
	Catechin	Epicatechin								
	2.23 ± 0.55	0.51 ± 0.07								
	Flavonol									
	Quercetin									
	5.56 ± 0.03									
	Hydroxycinnamic acid									
	Caffeic acid	Coumaric acid	Ferulic acid							
	0.52 ± 0.02	1.68 ± 0.06	2.78 ± 0.24							
	Hydroxy benzoic acid									
	Gallic acid	Protocatechuic acid	Vanillic acid	Syringic acid						
	6.06 ± 0.79	1.75 ± 0.25	0.82 ± 0.08	2.63 ± 0.31						
Organic acids	Oxalic acid	Tartaric acid	Quinic acid	Pyruvic acid	Shikimic acid	Malic acid	Malonic acid	Lactic acid	Citric acid	Acetic acid
	13.56 ± 1.67	2280.56 ± 132.00	32.23 ± 1.89	82.33 ± 4.78	100.22 ± 6.77	109.56 ± 7.42	7.02 ± 0.50	2109.56 ± 77.42	28.48 ± 3.15	126.32 ±11.23
	Fumaric acid	Succinic acid								
	0.99 ± 0.12	989.26 ± 38.89								
Chinese Baijiu										
Phenolic compounds	Gallic acid	Syringic acid	Coumaric acid	Ferulic acid	2-Methoxy-4-methylphenol	Phenol	4-Vinylguaiacol	p-Cresol	4-Ethylphenol	Vanillic aldehyde
	0.12 ± 0.01	1.83 ± 0.11	3.21 ± 0.52	0.14 ± 0.01	4.56 ± 0.33	3.48 ± 0.26	3.14 ± 0.19	1.67 ± 0.09	2.48 ± 0.16	3.55 ± 0.44
Organic acids	Tartaric acid	Malic acid	Acetic acid	Succinic acid	Lactic acid	Chlorogenic acid				
	0.120 ± 0.01	0.24 ± 0.01	334.88 ± 15.37	0.18 ± 0.02	470.64 ± 23.33	0.12 ± 0.01				

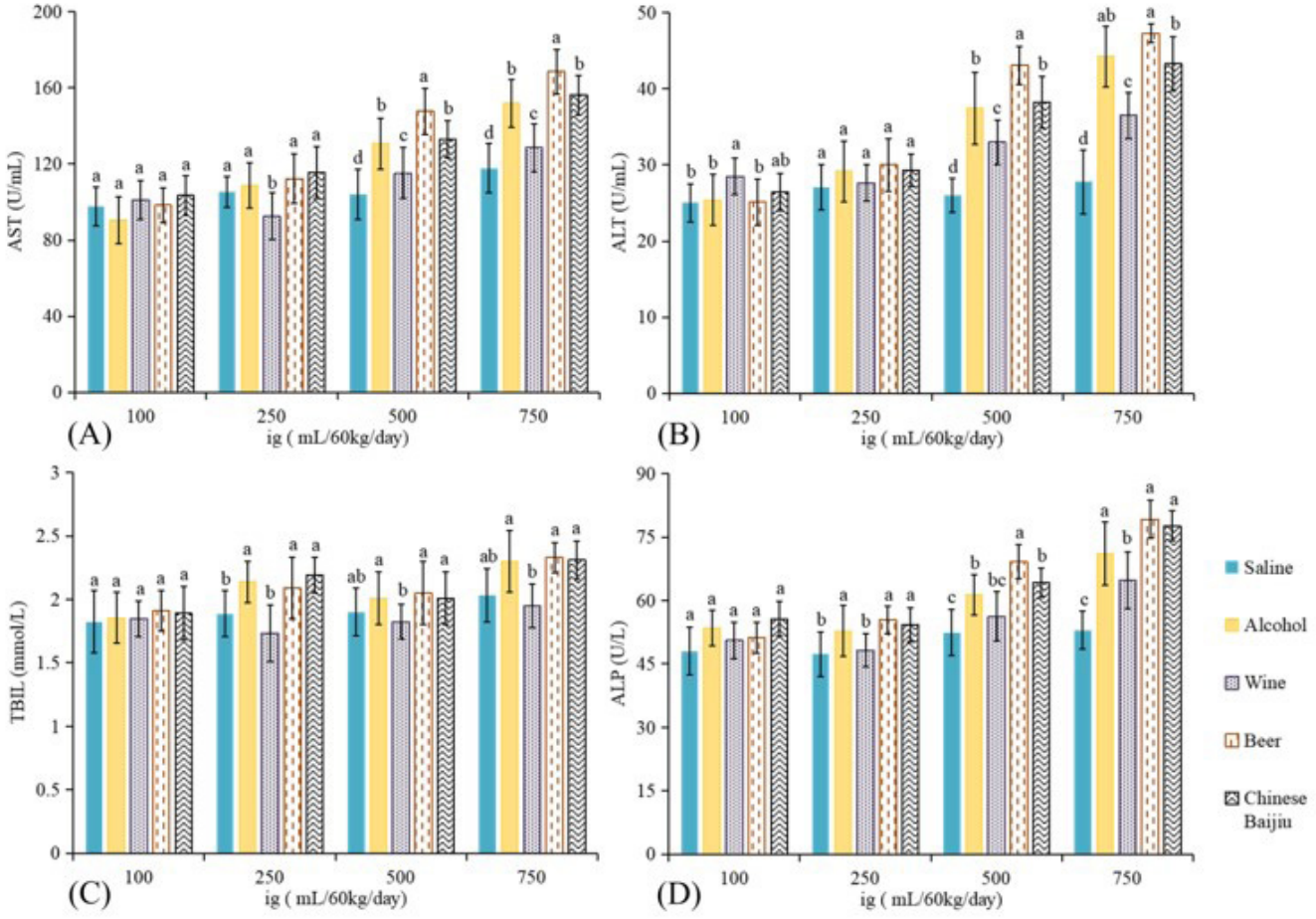

Figure 1. The AST (A), ALT (B), TBIL (C), and ALP (D) levels in serum of 20 team mice (there were five groups SG, AG, WG, BG and CBG, each group was given 100, 250, 500, 750 mL/60 kg/day gavage respectively).

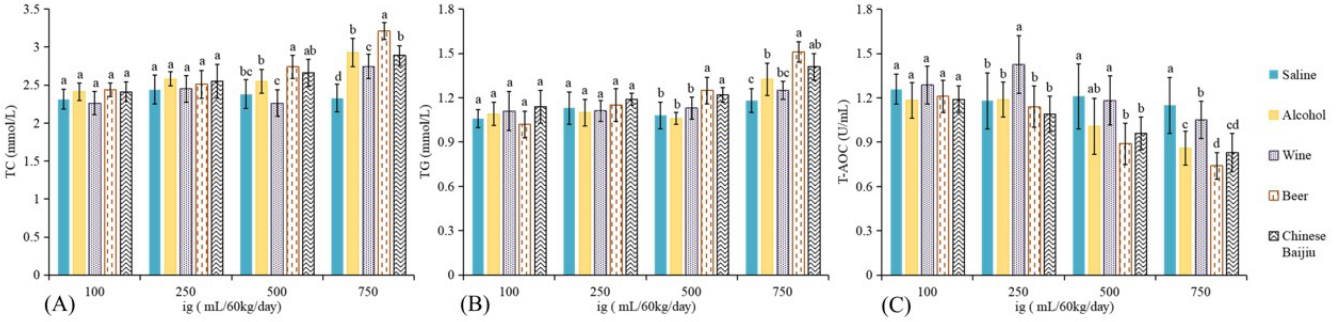

Figure 2. The TC (A), TG (B), and T-AOC (C) levels in serum of mice (there were five groups SG, AG, WG, BG and CBG, each group was given 100, 250, 500, 750 mL/60 kg/day gavage respectively).

3.4 Effects of different alcoholic beverages on T-AOC of mice serum

Analysis of the T-AOC levels (Figure 2C) found that when the intragastric dose increased to 250 mL/60 kg/day, the T-AOC value of WG was increased, which was significantly higher than the other groups ($p < 0.05$), indicating that drinking wine at this dose could enhance the antioxidant capacity of serum to a certain extent. When the dose continued to increase (500 mL/60 kg/day), there was no significant difference in T-AOC among AG, WG, and SG ($p > 0.05$), while BG and CBG were significantly lower than SG ($p < 0.05$). At 750 mL/60 kg/day, the T-AOC values

of the 4 alcoholic beverages were significantly lower than SG, and the value of wine was significantly higher than the other 3 alcoholic beverages. The T-AOC value of AG was significantly higher than BG but was not significantly different from CBG, while the value of CBG and BG had no significant difference.

3.5 Effects of different alcoholic beverages on liver indicators of mice

Liver index and spleen index

Liver index and spleen index are vital indicators of hepatic injury. When the liver is damaged, the liver and spleen swell significantly, increasing the liver index and spleen index (Figure 3A-3B). There was no significant difference in liver index between all the groups at 250 mL/60 kg/day or 500 mL/60 kg/day ($p > 0.05$), and there was no significant difference in spleen index between all the groups at 100 mL/60 kg/day and 250 mL/60 kg/day ($p > 0.05$). When the dose was low (100 mL/60 kg/day) or high (750 mL/60 kg/day), the liver index of the 4 alcoholic beverages was significantly higher than that of the SG ($p < 0.05$). At 500 mL/60 kg/day, the spleen index of BG or CBG was significantly higher than that of the SG. At 750 mL/60 kg/day, the spleen indexes of the 4 alcoholic beverages were all significantly higher than SG, but the spleen index of WG was significantly lower than that of the other three ($p < 0.05$).

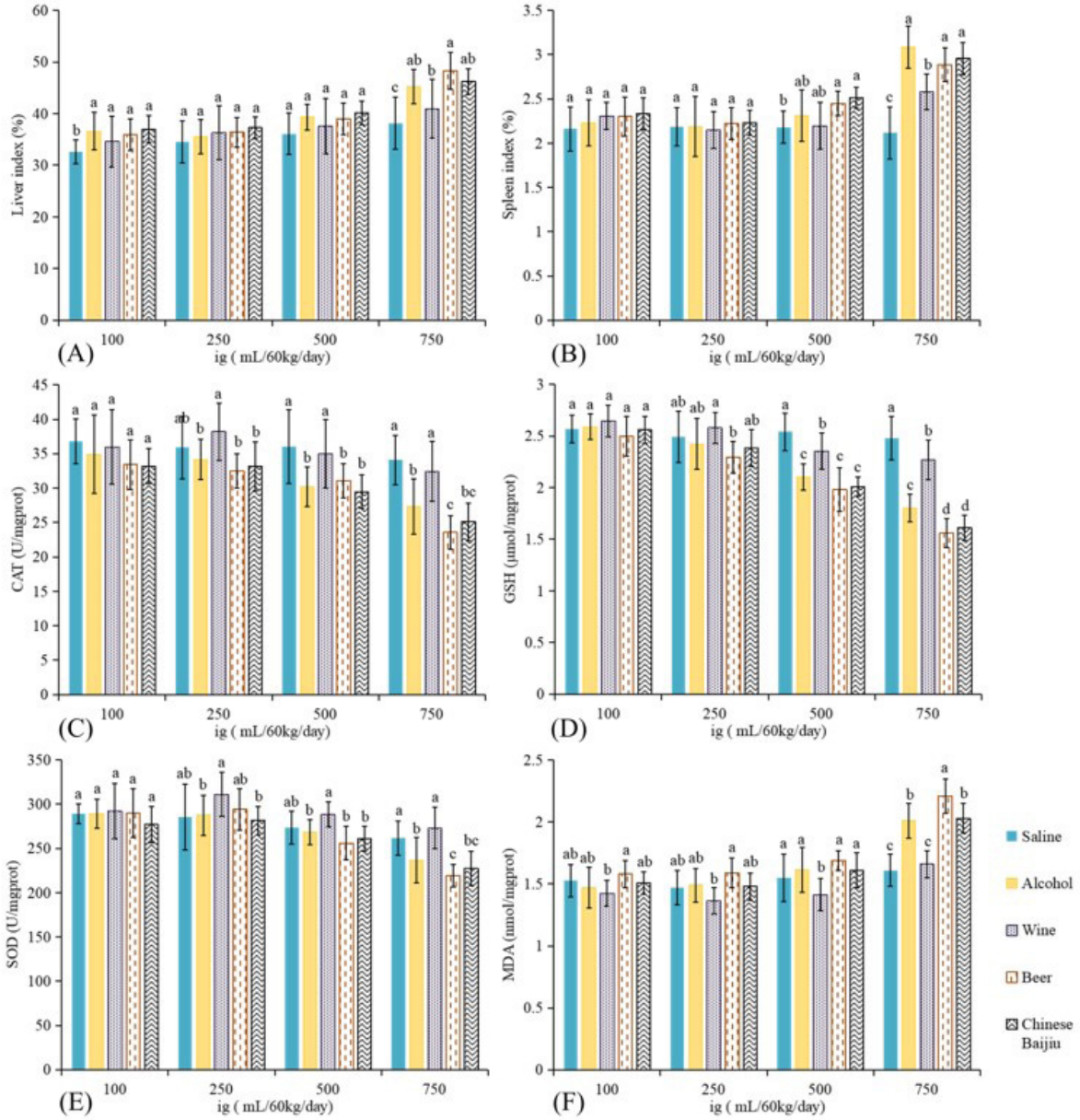

Figure 3. The liver indices (A) and spleen indices (B) of mice; CAT (C), GSH (D), SOD (E), and MDA (F) levels in mouse livers (there were five groups SG, AG, WG, BG and CBG, each group was given 100, 250, 500, 750 mL/60 kg/day gavage respectively).

CAT, GSH, SOD, and MDA

As shown in Figure 3C-3F, when the intragastric dose was ≤ 250 mL/60 kg/day, there was no significant difference in GSH, CAT, SOD, and MDA levels among the four ethyl alcoholic beverages and SG ($p > 0.05$), indicating that low-dose ethyl alcohol consumption would not significantly affect liver antioxidant levels. At 500 mL/60 kg/day, the CAT value of WG was not significantly different from that of SG, while the values of AG, BG, and CBG were significantly reduced ($p < 0.05$). The GSH values of the four alcoholic beverages were significantly lower than SG, but the reduction in GSH value caused by wine was significantly less than that of the other three alcoholic beverages. As for SOD values, the four alcoholic beverages were not significantly different from SG, but wine was significantly higher than the other three. There were no significant differences in MDA level between AGs, BGs, CBGs, and SGs. On the contrary, wine could reduce MDA level at 500 mL/60 kg/day. When the intragastric dose continued to rise to 750 mL/60 kg/day, there was still no significant difference in the CAT value and SOD value between WG and SG, while the values of other groups were significantly lower than SG, and the decrease caused by beer was the most significant. The four types of alcoholic beverages all caused a significant decrease in GSH content, but the GSH value of WG was significantly higher than the other three. Only WG had no significant difference in MDA content with SG. There was no significant difference in the MDA content between AG and CBG. BG had the most significant effect on increasing MDA levels.

3.6 Histopathological observation of mouse liver

H&E staining

H&E staining is one of the most commonly used staining methods for Histopathological observation (Muñoz-Espín & Serrano, 2014). The mice livers were cut into sections for H&E staining observation (Figure 4).

The structures of the hepatocyte cord in all SGs were clear, lymphocytes were visible, and no obvious hepatocyte degeneration was found (Figure 4A1-4A4). At 100 mL/60 kg/day, every group was similar to SG, the cells were arranged tightly and neatly, and there was no obvious necrosis or inflammatory cell infiltration (Figure 4B1-4E1). However, when the intragastric dose increased, the tissue lesions in the AGs, CBGs, and BGs were all aggravated. Taking 750 mL/60 kg/day as an example. In CBGs (Figure 4D4), the structure of hepatic cords was still clear, most hepatocytes were swollen, cytoplasm was loose, and fat vacuoles of different sizes could be seen in the hepatocytes, showing watery degeneration and steatosis, as shown by the black arrows. Megakaryocytes were seen in the tissue, as shown by the red arrow. Lymphocytes were seen in the hepatic sinuses, as indicated by the yellow arrow. In BGs (Figure 4E4), there was extensive hepatocyte edema around the interlobular vein of the liver, and the inner layer of the vein was dropped, as indicated by the black arrow. A large number of hepatocytes were swollen, and there were vacuoles of different sizes, as shown by the red arrow; mild dilatation and congestion could be found in the hepatic sinuses, as shown by the yellow arrow. In contrast, WG had relatively mild lesions. At 750 mL/60 kg/day (Figure 4C4), the arrangement of hepatocyte cords was slightly disordered, and Kupffer cells were seen in the liver sinusoids, as shown by the black arrow. A small number of red blood cells could be seen in the portal area, as shown by the red arrow; fat vacuoles of varying sizes could be seen in some hepatocytes, as shown by the yellow arrow; binuclear hepatocytes could be seen in the tissue, as shown by the blue arrow.

OR staining

Further liver sections were made for OR staining observation (Figure 5 and Table 2). The darker the tissue color, the greater the OR staining area percentage, indicating more lipids.

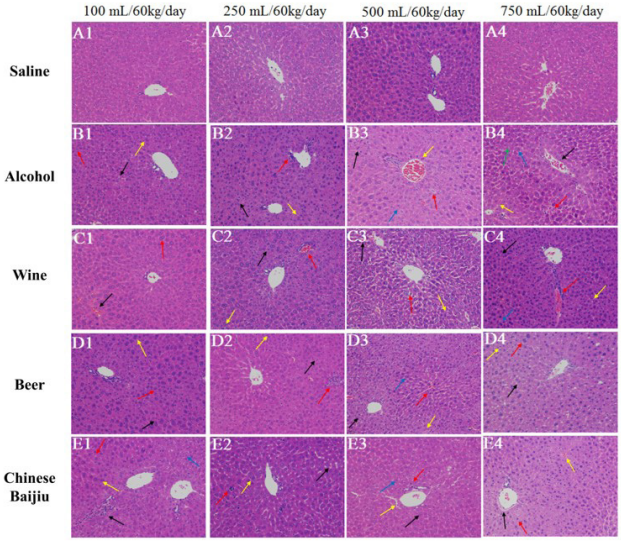

Figure 4. The H&E staining of mouse livers with 200 times magnification (there were five groups SG, AG, WG, BG and CBG, each group was given 100, 250, 500, 750 mL/60 kg/day gavage respectively).

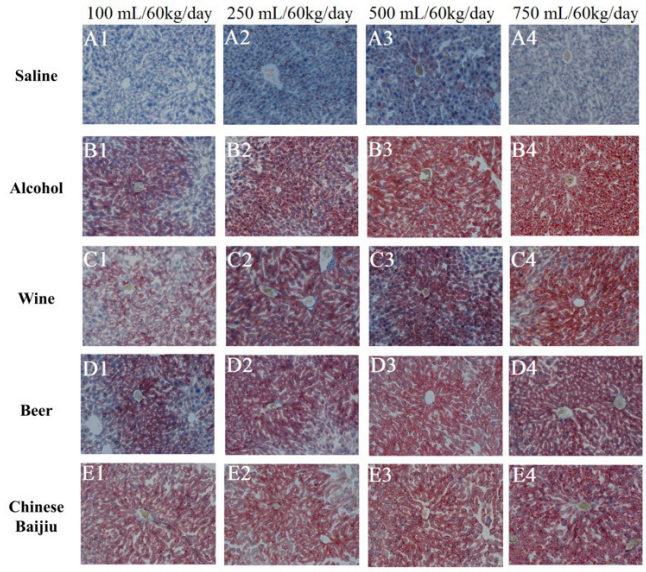

Figure 5. The OR staining of mouse livers with 200 times magnification (there were five groups SG, AG, WG, BG and CBG, each group was given 100, 250, 500, 750 mL/60 kg/day gavage respectively).

As can be seen in Table 2, when the intragastric dose was 100 mL/60 kg/day, the OR staining area of the 4 alcoholic beverage groups increased significantly, and all of them were dose-dependent, and the increase was relatively gentle after 250 mL/60 kg/day. Among them, the area increase caused by wine was the smallest, proving that wine polyphenols could protect the damage of liver lipid metabolism function to a certain extent, while the other three kinds of alcoholic beverages showed similar results in this study.

3.7 PCA for hepatic injury parameters of mice

PCA, which uses the idea of dimension reduction to remove overlapping parts of numerous information and transform multiple variables into a few unrelated comprehensive variables, has been widely used in the comprehensive evaluation of food quality (Wang et al., 2021). Overall, based on liver injury parameters, including serum indicators (AST, ALT, TC, TG, ALP, TBIL, SOD, T-AOC) and liver indicators (CAT, GSH, SOD, MDA, Oil red area), PCA was performed for the treatment of different alcoholic beverages and different drinking amounts (Figure 6). The cumulative variance of the first 2 principal components was over 88.502%, which could represent the overall information of the samples. There was no cross area between the teams, and the distinction was clear, indicating that the overall damage to the liver of mice caused by different treatments was discrepant. SGs at each intragastric dose and the other groups at 100 mL/60 kg/day gavage were all located in the third quadrant, with close distance and similar PCA scores, indicating that each alcoholic beverage had less damage to the liver at this dose. Under the same intragastric dose, compared with other alcoholic beverages, the distribution of WG was closer to SG, indicating that wine had protective effects on the liver. In addition, the mice in BG with 750 mL/60 kg/day intragastric dose were significantly separated from other groups, showing that 750 mL/60 kg/day beer intragastric administration caused the most severe hepatic injury in mice.

4 Discussion

After ingesting a large amount of ethyl alcohol, abundant acetaldehyde will be produced, which disrupts the trichloroacetic acid cycle and affects lipid metabolism (Ma et al., 2015). ALT and AST are crucial aminotransferases in the liver. When hepatocytes are under the pressure of ethyl alcohol, the inflammation of hepatocytes leads to changes in cell membrane permeability, swelling, and necrosis of hepatocytes, resulting in these two transaminases entering the blood due to poor concentration and increasing their content in serum (Tang et al., 2013). TBIL is a metabolite of bile salts and exists in plasma. ALP mainly exists in the sinusoidal side of hepatocytes. When liver inflammation, necrosis, poisoning, and other damage occur, TBIL and ALP in serum can significantly increase (Cha et al., 2013; Negrão et al., 2006). In this study, low dose (≤ 250 mL/60 kg/day) treatment did not increase the levels of ALT, AST, and ALP. Wine at the dose of 250 mL/60 kg/day even significantly reduced AST levels ($p < 0.05$). This may be due to the fact that at low doses, the functional substances in wine, such as some acids and inorganic salts, can improve the internal and external environment of hepatocytes and reduce inflammatory symptoms, thus reducing the loss and conversion between internal and external substances of hepatocytes (Li et al., 2020). With the continuous increase of intragastric dose, the levels of ALT, AST, and ALP in the four alcoholic beverage groups were significantly higher than those in SGs ($p < 0.05$), indicating that high-dose ethyl alcohol intake significantly damages the liver. Wine caused the least liver damage and beer the most. Functional substances in wine, such as polyphenols, can buffer and protect the liver to a certain extent (Deroover et al., 2021). Other salts also buffer and inhibit the radical of cell membrane permeability caused by ethyl alcohol to prevent excessive changes of hepatocytes (Gomulkievich et al., 1980). However, when a large amount of beer is ingested, the abundant bubbles in beer may cause the liver to expand rapidly, thus changing the permeability of the cell membrane, causing rapid changes in internal and external substances, and aggravating the damage of hepatocytes. The results of TBIL were

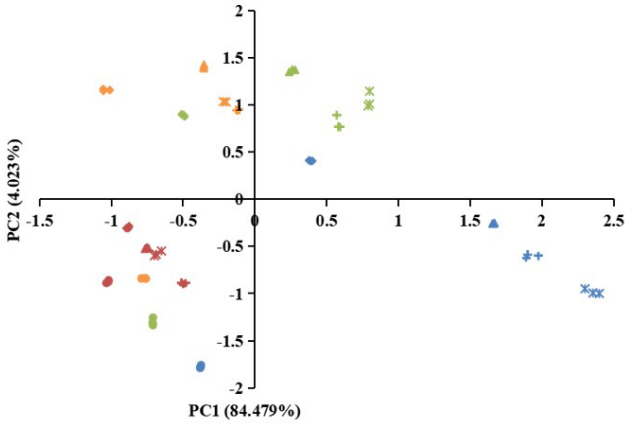

Figure 6. PCA score for liver injury parameters of 20 team mice (●: saline, ▲: alcohol, ◆: wine, Ж: beer, ┼: Chinese Baijiu. Red: 100 mL/60 kg/day; orange: 250 mL/60 kg/day; green: 500 mL/60 kg/day; blue: 750 mL/60 kg/day).

Table 2. The oil red staining area in liver of mice.

Groups	Area (%)			
	100 mL/60 kg/day	250 mL/60 kg/day	500 mL/60 kg/day	750 mL/60 kg/day
Saline	0.009	0.304	0.152	0.350
Alcohol	8.289	36.895	41.242	49.680
Wine	7.809	22.740	25.271	34.886
Beer	8.041	32.834	42.029	49.460
Chinese Baijiu	9.608	34.810	39.440	48.641

slightly different. There was no significant difference between WGs and the corresponding SGs at all doses, showing that test conditions were insufficient to cause a significant increase in TBIL. It indicated that wine could reduce the damage of alcohol to hepatocytes.

Lipid metabolism occurs in the liver, and fat accumulation is the earliest and most common response to excessive ethyl alcohol intake (Namachivayam & Gopalakrishnan, 2021). When the intragastric dose reached 750 mL/60 kg/day, ethyl alcohol intake significantly increased TC and TG levels, but wine was significantly lower than the other three alcoholic beverages ($p < 0.05$). It may be that some substances in wine can reduce the accumulation of lipids. For example, organic acids in wine, such as tartaric acid and malic acid, can maintain the acid-base balance in the body, help digestion, and reduce the energy accumulation and lipid accumulation caused by ethyl alcohol to maintain the role of triglyceride and total cholesterol. Beer was the most likely cause of fatty liver among alcoholic beverages. It is speculated that the possible reason is that beer promotes the speed and amount of ethyl alcohol into the blood during the drinking process, which causes excessive ethyl alcohol in the blood and the formation of lipid droplets (Landmann et al., 2017). However, the specific reason remains to be proved by further experiments.

Oxidative stress is considered to be the key factor leading to the onset of ALD (Reddy et al., 2014). GSH, CAT, and SOD are three important antioxidant enzymes, which support each other to form an antioxidant defense system, which can effectively eliminate various harmful substances generated in the body's metabolism (Gao et al., 2014; Ma et al., 2015). MDA is an important index of oxidative diseases in the body, and its content is positively correlated with the severity of the disease (Stewart & Bewley, 1980). The results showed that the four kinds of alcoholic beverages did not significantly affect GSH, CAT, and SOD when the dose was not higher than 250 mL/60 kg/day ($p > 0.05$). However, when the drinking amount continued to increase, only wine did not significantly reduce the content of antioxidant enzymes, and the oxidative stress reflected by the MDA value was the same as before. The T-AOC of WGs showed good performance under all drinking amounts, indicating that wine had a certain protective effect on liver antioxidant capacity.

High dose (750 mL/60 kg/day) ethyl alcohol exposure could cause significant liver tissue lesions (Figure 5). Beer was the most serious, and wine was the lightest, which was consistent with the results of Kasdallah-Grissa et al. (2007). The results of OR staining also showed that wine resulted in the slowest lipid accumulation and the lowest amount of lipid in tissues. In addition, when alcoholic beverage intake reached 750 mL/60 kg/day, all treatments caused significant spleen enlargement, but wine had the least degree (Figure 3A-3B).

Resveratrol is the main antioxidant in wine that inhibits the accumulation of lipid peroxides in the liver and has antioxidant effects. Kasdallah-Grissa et al. (2006) stated that resveratrol was a good candidate to prevent ethanol-induced lipid peroxidation in the liver and other organs and verified that resveratrol had the physiological effects of reducing liver tissue damage and improving antioxidant enzyme activities. Serrano et al. (2021) found that resveratrol supplementation increased the lipid oxidation capacity of the liver and decreased lipid production capacity. In addition, preclinical studies have shown that resveratrol supplementation in adult animals has a variety of benefits on liver metabolism, including prevention and mitigation of alcohol-induced liver lipid accumulation (Ajmo et al., 2008). Resveratrol can also act as an activator of PPARalpha, enhancing the cell's ability to convert and catabolize fatty acids while inhibiting the expression of lipid-producing genes, including Srbf1 (Serrano et al., 2021). At the same time, the antioxidant activity of resveratrol has been repeatedly confirmed (Miguel et al., 2021). Because it has both hydrophilic and lipophilic properties, it may provide more effective protection than common antioxidants (such as vitamin C and vitamin E). Resveratrol cannot only improve the activity of erythrocyte antioxidant enzymes, but can also inhibit the peroxidation of low-density lipoprotein, liver microsomal, and neuronal cells. It can protect cells even during ethanol intoxication (Kasdallah-Grissa et al., 2007). The antioxidant mechanism of resveratrol may be related to its ability to reduce the superoxide and nitrite anions produced by neutrophils and macrophages, capture peroxy free radicals and/or hydroxyl free radicals, and reduce α-tocopherol radicals to regenerate endogenous tocopherol (Banez et al., 2020; Kasdallah-Grissa et al., 2007; Miguel et al., 2021). The hepatoprotective activity of resveratrol on fibrosis can be explained by its ability to produce more IL-10 to promote the polarization of M(LPS) to M(IL-4)-like macrophages (Yu et al., 2019). Resveratrol can also protect the liver by interfering with the TLR4/NF-κB signaling pathway, inhibiting the production of tumor necrosis factor-α, inducible nitric oxide synthase, and HMGB1 (Lu et al., 2021; Stewart & Bewley, 1980).

However, because the content of resveratrol in wine was low, the protection of wine on the liver in our study might not come from a monomeric substance. Surely the overall polyphenols have some effect, maybe a synergic one. Furthermore, because other substances in wine have anti-inflammatory and antioxidant effects, such as proanthocyanidins (PAs), are currently internationally recognized as the most effective natural antioxidant for scavenging free radicals in the human body (Wei et al., 2021). Another example is tannins, which also have antioxidant effects (Peng et al., 2022). These substances may all play a certain role in protecting the human liver.

In this study, it was found that the liver damage of beer was more serious than of wine and Chinese Baijiu. It should be noted that in the specific consumption environment in China, most of the beer consumed is not craft beer, including the beer used in the intragastric fluid. Differences in brand and type, dilution of nutraceuticals, large amounts of carbon dioxide bubbles, special food matrix, and drinking patterns might all contribute to this phenomenon. However, the specific mechanism remains unclear and needs further study.

5 Conclusion

This study preliminarily explored the appropriate drinking amount of different alcoholic beverages. In conclusion, when the daily drinking amount reached 250 mL/60 kg/day, the four alcoholic beverages caused hepatosplenomegaly and the accumulation of lipids in the liver tissue but did not cause serious damage

to the liver. At low doses (≤ 250 mL/60 kg/day), the protective effect of functional substances (especially phenolics) in wine on the liver could protect it from alcohol damage. When the daily consumption continued to increase, all alcoholic beverages could cause hepatic injury, but wine caused the least damage. Chinese Baijiu and alcohol had the most similar damage to the liver. The specific beer used might be more toxic due to lack of nutrients and high levels of carbon dioxide, but the accurate mechanism needed to be further studied.

Abbreviations

ANOVA: a one-way analysis of variance. ALT: alanine aminotransferase. AG: alcohol group. ALD: alcoholic liver disease. ALP: alkaline phosphatase. AST: aspartate aminotransferase. BG: beer group. CAT: catalase. CBG: Chinese Baijiu group. GSH: glutathione. H&E: haematoxylin and eosin. MDAeq: malondialdehyde equivalents. OR: Oil red. PCA: principal component analysis. Pas: proanthocyanidins. SG: saline group; SD: standard deviation. SOD: superoxide dismutase. T-AOC: total antioxidant capacity. TBIL: total bilirubin. TC: total cholesterol. TG: triacylglycerols. WG: wine group. WHO: World Health Organization.

Ethical approval

The studies were reviewed and approved by the ethics committee of Northwest A&F University.

Conflict of interest

The authors declare no conflict of interest.

Availability of data and material

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding author/s.

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Author contributions

Conceptualization and methodology, T.M. and X.S.; formal analysis, J.W., and C.L.; data curation, J.W. and Q.G.; writing—original draft preparation, J.W.; writing—review and editing, T.M. and Y.F.; visualization, J.W. All authors have read and agreed to the published version of the manuscript.

References

Ajmo, J. M., Liang, X., Rogers, C. Q., Pennock, B., & You, M. (2008). Resveratrol alleviates alcoholic fatty liver in mice. *American Journal of Physiology. Gastrointestinal and Liver Physiology*, 295(4), G833-G842. http://dx.doi.org/10.1152/ajpgi.90358.2008. PMid:18755807.

Bajaj, J. S. (2019). Alcohol, liver disease and the gut microbiota. *Nature Reviews. Gastroenterology & Hepatology*, 16(4), 235-246. http://dx.doi.org/10.1038/s41575-018-0099-1. PMid:30643227.

Banez, M. J., Geluz, M. I., Chandra, A., Hamdan, T., Biswas, O. S., Bryan, N. S., & Von Schwarz, E. R. (2020). A systemic review on the antioxidant and anti-inflammatory effects of resveratrol, curcumin, and dietary nitric oxide supplementation on human cardiovascular health. *Nutrition Research*, 78, 11-26. http://dx.doi.org/10.1016/j.nutres.2020.03.002. PMid:32428778.

Cha, J. Y., Ahn, H. Y., Cho, Y. S., & Je, J. Y. (2013). Protective effect of cordycepin-enriched *Cordyceps militaris* onalcoholic hepatotoxicity in Sprague-Dawley rats. *Food and Chemical Toxicology*, 60, 52-57. http://dx.doi.org/10.1016/j.fct.2013.07.033. PMid:23876821.

Deroover, K., Siegrist, M., Brain, K., McIntyre, J., & Bucher, T. (2021). A scoping review on consumer behaviour related to wine and health. *Trends in Food Science & Technology*, 112, 559-580. http://dx.doi.org/10.1016/j.tifs.2021.03.057.

Do, M. H., Lee, H. L. L., Kim, Y., Lee, H. B., Lee, E., Park, J. H., & Park, H. Y. (2021). *Corchorus olitorius* L. ameliorates alcoholic liver disease by regulating gut-liver axis. *Journal of Functional Foods*, 85, 104648. http://dx.doi.org/10.1016/j.jff.2021.104648.

Gao, C. Y., Tian, C. R., Zhou, R., Zhang, R. G., & Lu, Y. H. (2014). Phenolic composition, DNA damage protective activity and hepatoprotective effect of free phenolic extract from *Sphallerocarpus gracilis* seeds. *International Immunopharmacology*, 20(1), 238-247. http://dx.doi.org/10.1016/j.intimp.2014.03.002. PMid:24657314.

Gerardi, G., Cavia-Saiz, M., Rivero-Pérez, M. D., González-SanJosé, M. L., & Muñiz, P. (2020). Wine pomace product modulates oxidative stress and microbiota in obesity high-fat diet-fed rats. *Journal of Functional Foods*, 68, 103903. http://dx.doi.org/10.1016/j.jff.2020.103903.

Gomulkievich, I., Popdimitrova, N., Kunchev, N., & Zlatarev, G. (1980). Effect of citrate, tartrate, succinate and phosphate ions on the penetration of phosphorus-labelled anions into erythrocytes. *Eksperimentalna Meditsina i Morfologiia*, 19(2), 92-97. PMid:7379734.

Griswold, M. G., Fullman, N., Hawley, C., Arian, N., Zimsen, S. R. M., Tymeson, H. D., Venkateswaran, V., Tapp, A. D., Forouzanfar, M. H., Salama, J. S., Abate, K. H., Abate, D., Abay, S. M., Abbafati, C., Abdulkader, R. S., Abebe, Z., Aboyans, V., Abrar, M. M., Acharya, P., Adetokunboh, O. O., Adhikari, T. B., Adsuar, J. C., Afarideh, M., Agardh, E. E., Agarwal, G., Aghayan, S. A., Agrawal, S., Ahmed, M. B., Akibu, M., Akinyemiju, T., Akseer, N., Asfoor, D. H. A., Al-Aly, Z., Alahdab, F., Alam, K., Albujeer, A., Alene, K. A., Ali, R., Ali, S. D., Alijanzadeh, M., Aljunid, S. M., Alkerwi, A., Allebeck, P., Alvis-Guzman, N., Amare, A. T., Aminde, L. N., Ammar, W., Amoako, Y. A., Amul, G. G. H., Andrei, C. L., Angus, C., Ansha, M. G., Antonio, C. A. T., Aremu, O., Ärnlöv, J., Artaman, A., Aryal, K. K., Assadi, R., Ausloos, M., Avila-Burgos, L., Avokpaho, E. F., Awasthi, A., Ayele, H. T., Ayer, R., Ayuk, T. B., Azzopardi, P. S., Badali, H., Badawi, A., Banach, M., Barker-Collo, S. L., Barrero, L. H., Basaleem, H., Baye, E., Bazargan-Hejazi, S., Bedi, N., Béjot, Y., Belachew, A. B., Belay, S. A., Bennett, D. A., Bensenor, I. M., Bernabe, E., Bernstein, R. S., Beyene, A. S., Beyranvand, T., Bhaumik, S., Bhutta, Z. A., Biadgo, B., Bijani, A., Bililign, N., Birlik, S. M., Birungi, C., Bizuneh, H., Bjerregaard, P., Bjørge, T., Borges, G., Bosetti, C., Boufous, S., Bragazzi, N. L., Brenner, H., Butt, Z. A., Cahuana-Hurtado, L., Calabria, B., Campos-Nonato, I. R., Campuzano, J. C., Carreras, G., Carrero, J. J., Carvalho, F., Castañeda-Orjuela, C. A., Castillo Rivas, J., Catalá-López, F., Chang, J.-C., Charlson, F. J., Chattopadhyay, A., Chaturvedi, P., Chowdhury, R., Christopher, D. J., Chung, S.-C., Ciobanu, L. G., Claro, R. M., Conti, S., Cousin, E., Criqui, M. H., Dachew, B. A., Dargan, P. I., Daryani, A., Das Neves, J., Davletov, K., De Castro, F.,

De Courten, B., De Neve, J.-W., Degenhardt, L., Demoz, G. T., Des Jarlais, D. C., Dey, S., Dhaliwal, R. S., Dharmaratne, S. D., Dhimal, M., Doku, D. T., Doyle, K. E., Dubey, M., Dubljanin, E., Duncan, B. B., Ebrahimi, H., Edessa, D., El Sayed Zaki, M., Ermakov, S. P., Erskine, H. E., Esteghamati, A., Faramarzi, M., Farioli, A., Faro, A., Farvid, M. S., Farzadfar, F., Feigin, V. L., Felisbino-Mendes, M. S., Fernandes, E., Ferrari, A. J., Ferri, C. P., Fijabi, D. O., Filip, I., Finger, J. D., Fischer, F., Flaxman, A. D., Franklin, R. C., Futran, N. D., Gallus, S., Ganji, M., Gankpe, F. G., Gebregergs, G. B., Gebrehiwot, T. T., Geleijnse, J. M., Ghadimi, R., Ghandour, L. A., Ghimire, M., Gill, P. S., Ginawi, I. A., Giref, A. Z. Z., Gona, P. N., Gopalani, S. V., Gotay, C. C., Goulart, A. C., Greaves, F., Grosso, G., Guo, Y., Gupta, R., Gupta, R., Gupta, V., Gutiérrez, R. A., Gvs, M., Hafezi-Nejad, N., Hagos, T. B., Hailu, G. B., Hamadeh, R. R., Hamidi, S., Hankey, G. J., Harb, H. L., Harikrishnan, S., Haro, J. M., Hassen, H. Y., Havmoeller, R., Hay, S. I., Heibati, B., Henok, A., Heredia-Pi, I., Hernández-Llanes, N. F., Herteliu, C., Hibstu, D. T. T., Hoogar, P., Horita, N., Hosgood, H. D., Hosseini, M., Hostiuc, M., Hu, G., Huang, H., Husseini, A., Idrisov, B., Ileanu, B. V., Ilesanmi, O. S., Irvani, S. S. N., Islam, S. M. S., Jackson, M. D., Jakovljevic, M., Jalu, M. T., Jayatilleke, A. U., Jha, R. P., Jonas, J. B., Jozwiak, J. J., Kabir, Z., Kadel, R., Kahsay, A., Kapil, U., Kasaeian, A., Kassa, T. D. D., Katikireddi, S. V., Kawakami, N., Kebede, S., Kefale, A. T., Keiyoro, P. N., Kengne, A. P., Khader, Y., Khafaie, M. A., Khalil, I. A., Khan, M. N., Khang, Y.-H., Khater, M. M., Khubchandani, J., Kim, C.-I., Kim, D., Kim, Y. J., Kimokoti, R. W., Kisa, A., Kivimäki, M., Kochhar, S., Kosen, S., Koul, P. A., Koyanagi, A., Krishan, K., Kuate Defo, B., Kucuk Bicer, B., Kulkarni, V. S., Kumar, P., Lafranconi, A., Lakshmana Balaji, A., Lalloo, R., Lallukka, T., Lam, H., Lami, F. H., Lan, Q., Lang, J. J., Lansky, S., Larsson, A. O., Latifi, A., Leasher, J. L., Lee, P. H., Leigh, J., Leinsalu, M., Leung, J., Levi, M., Li, Y., Lim, L.-L., Linn, S., Liu, S., Lobato-Cordero, A., Lopez, A. D., Lorkowski, S., Lotufo, P. A., Macarayan, E. R. K., Machado, I. E., Madotto, F., Magdy Abd El Razek, H., Magdy Abd El Razek, M., Majdan, M., Majdzadeh, R., Majeed, A., Malekzadeh, R., Malta, D. C., Mapoma, C. C., Martinez-Raga, J., Maulik, P. K., Mazidi, M., Mckee, M., Mehta, V., Meier, T., Mekonen, T., Meles, K. G., Melese, A., Memiah, P. T. N., Mendoza, W., Mengistu, D. T., Mensah, G. A., Meretoja, T. J., Mezgebe, H. B., Miazgowski, T., Miller, T. R., Mini, G., Mirica, A., Mirrakhimov, E. M., Moazen, B., Mohammad, K. A., Mohammadifard, N., Mohammed, S., Monasta, L., Moraga, P., Morawska, L., Mousavi, S. M., Mukhopadhyay, S., Musa, K. I., Naheed, A., Naik, G., Najafi, F., Nangia, V., Nansseu, J. R., Nayak, M. S. D. P., Nejjari, C., Neupane, S., Neupane, S. P., Ngunjiri, J. W., Nguyen, C. T., Nguyen, L. H., Nguyen, T. H., Ningrum, D. N. A., Nirayo, Y. L., Noubiap, J. J., Ofori-Asenso, R., Ogbo, F. A., Oh, I.-H., Oladimeji, O., Olagunju, A. T., Olivares, P. R., Olusanya, B. O., Olusanya, J. O., Oommen, A. M., Oren, E., Orpana, H. M., Ortega-Altamirano, D. D. V., Ortiz, J. R., Ota, E., Owolabi, M. O., Oyekale, A. S., P A, M., Pana, A., Park, E.-K., Parry, C. D. H., Parsian, H., Patle, A., Patton, G. C., Paudel, D., Petzold, M., Phillips, M. R., Pillay, J. D., Postma, M. J., Pourmalek, F., Prabhakaran, D., Qorbani, M., Radfar, A., Rafay, A., Rafiei, A., Rahim, F., Rahimi-Movaghar, A., Rahman, M., Rahman, M. A., Rai, R. K., Rajsic, S., Raju, S. B., Ram, U., Rana, S. M., Ranabhat, C. L., Rawaf, D. L., Rawaf, S., Reiner, R. C., Reis, C., Renzaho, A. M. N., Rezai, M. S., Roever, L., Ronfani, L., Room, R., Roshandel, G., Rostami, A., Roth, G. A., Roy, A., Sabde, Y. D., Saddik, B., Safiri, S., Sahebkar, A., Salama, J. S., Saleem, Z., Salomon, J. A., Salvi, S. S., Sanabria, J., Sanchez-Niño, M. D., Santomauro, D. F., Santos, I. S., Santric Milicevic, M. M. M., Sarker, A. R., Sarmiento-Suárez, R., Sarrafzadegan, N., Sartorius, B., Satpathy, M., Sawhney, M., Saxena, S., Saylan, M., Schaub, M. P., Schmidt, M. I., Schneider, I. J. C., Schöttker, B., Schutte, A. E., Schwendicke, F., Sepanlou, S. G., Shaikh, M. A., Sharif, M., She, J., Sheikh, A., Shen, J., Shiferaw, M. S., Shigematsu, M., Shiri, R., Shishani, K., Shiue, I., Shukla, S. R., Sigfusdottir, I. D., Silva, D. A. S., Silva, N. T. D., Silveira, D. G. A., Sinha, D. N., Sitas, F., Soares Filho, A. M., Soofi, M., Sorensen, R. J. D., Soriano, J. B., Sreeramareddy, C. T., Steckling, N., Stein, D. J., Sufiyan, M. B., Sur, P. J., Sykes, B. L., Tabarés-Seisdedos, R., Tabuchi, T., Tavakkoli, M., Tehrani-Banihashemi, A., Tekle, M. G., Thapa, S., Thomas, N., Topor-Madry, R., Topouzis, F., Tran, B. X., Troeger, C. E., Truelsen, T. C., Tsilimparis, N., Tyrovolas, S., Ukwaja, K. N., Ullah, I., Uthman, O. A., Valdez, P. R., Van Boven, J. F. M., Vasankari, T. J., Venketasubramanian, N., Violante, F. S., Vladimirov, S. K., Vlassov, V., Vollset, S. E., Vos, T., Wagnew, F. W. S., Waheed, Y., Wang, Y.-P., Weiderpass, E., Weldegebreal, F., Weldegwergs, K. G., Werdecker, A., Westerman, R., Whiteford, H. A., Widecka, J., Wijeratne, T., Wyper, G. M. A., Xu, G., Yamada, T., Yano, Y., Ye, P., Yimer, E. M., Yip, P., Yirsaw, B. D., Yisma, E., Yonemoto, N., Yoon, S.-J., Yotebieng, M., Younis, M. Z., Zachariah, G., Zaidi, Z., Zamani, M., Zhang, X., Zodpey, S., Mokdad, A. H., Naghavi, M., Murray, C. J. L., & Gakidou, E. (2018). Alcohol use and burden for 195 countries and territories, 1990-2016: a systematic analysis for the Global Burden of Disease study 2016. *Lancet*, 392(10152), 1015-1035. http://dx.doi.org/10.1016/S0140-6736(18)31310-2. PMid:30146330.

He, J., Assanangkornchai, S., Cai, L., & McNeil, E. (2016). Disparities in drinking patterns and risks among ethnic majority and minority groups in China: the roles of acculturation, religion, family and friends. *Drug and Alcohol Dependence*, 159, 198-206. http://dx.doi.org/10.1016/j.drugalcdep.2015.12.028. PMid:26790824.

Im, P. K., Millwood, I. Y., Kartsonaki, C., Guo, Y., Chen, Y., Turnbull, I., Yu, C., Du, H., Pei, P., Lv, J., Walters, R. G., Li, L., Yang, L., & Chen, Z. (2021). Alcohol drinking and risks of liver cancer and non-neoplastic chronic liver diseases in China: a 10-year prospective study of 0.5 million adults. *BMC Medicine*, 19(1), 216. http://dx.doi.org/10.1186/s12916-021-02079-1. PMid:34530818.

Kasdallah-Grissa, A., Mornagui, B., Aouani, E., Hammami, M., Gharbi, N., Kamoun, A., & El-Fazaa, S. (2006). Protective effect of resveratrol on ethanolinduced lipid peroxidation in rats. *Alcohol and Alcoholism*, 41(3), 236-239. http://dx.doi.org/10.1093/alcalc/agh256. PMid:16517551.

Kasdallah-Grissa, A., Mornagui, B., Aouani, E., Hammami, M., May, M., Gharbi, N., Kamoun, A., & El-Fazaâ, S. (2007). Resveratrol, a red wine polyphenol, attenuates ethanol-induced oxidative stress in rat liver. *Life Sciences*, 80(11), 1033-1039. http://dx.doi.org/10.1016/j.lfs.2006.11.044. PMid:17258234.

Landmann, M., Sellmann, C., Engstler, A. J., Ziegenhardt, D., Jung, F., Brombach, C., & Bergheim, I. (2017). Hops (*Humulus lupulus*) content in beer modulates effects of beer on the liver after acute ingestion in female mice. *Alcohol and Alcoholism*, 52(1), 48-55. http://dx.doi.org/10.1093/alcalc/agw060. PMid:27659607.

Li, R., Wang, G. P., Whitlock, J. A., Zhao, S., Yagiz, Y., & Gu, L. W. (2020). Muscadine grapes (*Vitis rotundifolia*) and dealcoholized muscadine wine alleviated symptoms of colitis and protected against dysbiosis in mice exposed to dextran sulfate sodium. *Journal of Functional Foods*, 65, 103746. http://dx.doi.org/10.1016/j.jff.2019.103746.

Lu, J.-M., Jin, G.-N., Lu, Y.-N., Zhao, X.-D., Lan, H.-W., Mu, S.-R., Shen, X.-Y., Xu, G.-H., Jin, C.-H., Ma, J., Jin, X., Xu, X., & Piao, L.-X. (2021). Resveratrol modulates *Toxoplasma gondii* infection induced liver injury by intervening in the HMGB1/TLR4/NF-κB signaling pathway. *European Journal of Pharmacology*, 910, 174497. http://dx.doi.org/10.1016/j.ejphar.2021.174497. PMid:34508751.

Ma, T., Sun, X., Tian, C., Zheng, Y., Zheng, C., & Zhan, J. (2015). Chemical composition and hepatoprotective effects of polyphenols extracted from the stems and leaves of Sphallerocarpus gracilis. *Journal of Functional Foods*, 18, 673-683. http://dx.doi.org/10.1016/j.jff.2015.09.001.

Miele, A. (2021). Wine composition of Merlot and Cabernet Sauvignon vine clones under the environmental conditions of Serra Gaúcha, Brazil. *Food Science and Technology*, 41(Suppl. 1), 116-122. http://dx.doi.org/10.1590/fst.10520.

Miguel, C. A., Noya-Riobó, M. V., Mazzone, G. L., Villar, M. J., & Coronel, M. F. (2021). Antioxidant, anti-inflammatory and neuroprotective actions of resveratrol after experimental nervous system insults. Special focus on the molecular mechanisms involved. *Neurochemistry International*, 150, 105188. http://dx.doi.org/10.1016/j.neuint.2021.105188. PMid:34536545.

Muñoz-Espín, D., & Serrano, M. (2014). Cellular senescence: from physiologyto pathology. *Nature Reviews. Molecular Cell Biology*, 15(7), 482-496. http://dx.doi.org/10.1038/nrm3823. PMid:24954210.

Namachivayam, A., & Gopalakrishnan, A. V. (2021). A review on molecular mechanism of alcoholic liver disease. *Life Sciences*, 274, 119328. http://dx.doi.org/10.1016/j.lfs.2021.119328. PMid:33711388.

Negrão, M. R., Keating, R., Faria, A., Azevedo, I., & Martins, M. J. (2006). Acute effect of tea, wine, beer, and polyphenols on ecto-alkaline phosphatase activity in human vascular smooth muscle cells. *Journal of Agricultural and Food Chemistry*, 54(14), 4982-4988. http://dx.doi.org/10.1021/jf060505u. PMid:16819906.

Peng, K., Lv, X., Zhao, H., Chen, B., Chen, X., & Huang, W. (2022). Antioxidant and intestinal recovery function of condensed tannins in *Lateolabrax maculatus* responded to *in vivo* and *in vitro* oxidative stress. *Aquaculture*, 547, 737399. http://dx.doi.org/10.1016/j.aquaculture.2021.737399.

Reddy, V. D., Padmavathi, P., Hymavathi, R., Maturu, P., & Varadacharyulu, N. (2014). Alcohol-induced oxidative stress in rat liver microsomes: protective effect of Emblica officinalis. *Pathophysiology*, 21(2), 153-159. http://dx.doi.org/10.1016/j.pathophys.2013.12.001. PMid:24393670.

Robles, A., Fabjanowicz, M., Chmiel, T., & Płotka-Wasylka, J. (2019). Determination and identification of organic acids in wine samples. Problems and challenges. *Trends in Analytical Chemistry*, 120, 115630. http://dx.doi.org/10.1016/j.trac.2019.115630.

Serrano, A., Ribot, J., Palou, A., & Bonet, M. L. (2021). Long-term programming of skeletal muscle and liver lipid and energy metabolism by resveratrol supplementation to suckling mice. *The Journal of Nutritional Biochemistry*, 95, 108770. http://dx.doi.org/10.1016/j.jnutbio.2021.108770. PMid:34000411.

Somani, S. J., Modi, K. P., Majumdar, A. S., & Sadarani, B. N. (2015). Phytochemicals and their potential usefulness in inflammatory bowel disease. *Phytotherapy Research*, 29(3), 339-350. http://dx.doi.org/10.1002/ptr.5271. PMid:25572840.

Stewart, R. R., & Bewley, J. D. (1980). Lipid peroxidation associated with accelerated aging of soybean axes. *Plant Physiology*, 65(2), 245-248. http://dx.doi.org/10.1104/pp.65.2.245. PMid:16661168.

Sun, X. Y., Liu, L. L., Ma, T. T., Yu, J., Huang, W. D., Fang, Y. L., & Zhan, J. C. (2019). Effect of high Cu^{2+} stress on fermentation performance and copper biosorption of Saccharomyces cerevisiae during wine fermentation. *Food Science and Technology*, 39(1), 19-26. http://dx.doi.org/10.1590/1678-457x.24217.

Sun, X., Cheng, X., Zhang, J., Ju, Y., Que, Z., Liao, X., Lao, F., Fang, Y., & Ma, T. (2020). Letting wine polyphenols functional: estimation of wine polyphenols bioaccessibility under different drinking amount and drinking patterns. *Food Research International*, 127, 108704. http://dx.doi.org/10.1016/j.foodres.2019.108704. PMid:31882093.

Tang, C. C., Huang, H. P., Lee, Y. J., Tang, Y. H., & Wang, C. J. (2013). Hepatoprotective effect of mulberry water extracts on ethanol-induced liver injury via anti-inflammation and inhibition of lipogenesis in C57BL/6J mice. *Food and Chemical Toxicology*, 62, 786-796. http://dx.doi.org/10.1016/j.fct.2013.10.011. PMid:24140469.

Tian, X., Li, R., Jiang, Y., Zhao, F., Yu, Z., Wang, Y., Dong, Z., Liu, P., & Li, X. (2020). *Bifidobacterium breve* ATCC15700 pretreatment prevents alcoholic liver disease through modulating gut microbiota in mice exposed to chronic alcohol intake. *Journal of Functional Foods*, 72, 104045. http://dx.doi.org/10.1016/j.jff.2020.104045.

Tverdal, A., Skurtveit, S., Selmer, R., Myhre, R., & Thelle, D. (2018). Coffee and wine consumption is associated with reduced mortality from alcoholic liver disease: follow-up of 219,279 Norwegian men and women aged 30-67 years. *Annals of Epidemiology*, 28(11), 753-758. http://dx.doi.org/10.1016/j.annepidem.2018.08.010. PMid:30241792.

Wang, J., Ma, T., Wang, L., Lan, T., Fang, Y., & Sun, X. (2021). Research on the consumption trend, nutritional value, biological activity evaluation, and sensory properties of mini fruits and vegetables. *Foods*, 10(12), 2966. http://dx.doi.org/10.3390/foods10122966. PMid:34945517.

Wei, X., Ju, Y., Ma, T., Zhang, J., Fang, Y., & Sun, X. (2021). New perspectives on the biosynthesis, transportation, astringency perception and detection methods of grape proanthocyanidins. *Critical Reviews in Food Science and Nutrition*, 61(14), 2372-2398. http://dx.doi.org/10.1080/10408398.2020.1777527. PMid:32551848.

Yang, X., Lau, J. T. F., Wang, Z., & Lau, M. C. M. (2019). Prevalence of binge drinking and relationships between masculine role discrepancy and binge drinking via discrepancy stress among Chinese men. *Drug and Alcohol Dependence*, 196, 57-61. http://dx.doi.org/10.1016/j.drugalcdep.2018.12.013. PMid:30685737.

Yin, M., Zhang, P., Yu, F., Zhang, Z., Cai, Q., Lu, W., Li, B., Qin, W., Cheng, M., Wang, H., & Gao, H. (2017). Grape seed procyanidin B2 ameliorates hepatic lipid metabolism disorders in db/db mice. *Molecular Medicine Reports*, 16(3), 2844-2850. http://dx.doi.org/10.3892/mmr.2017.6900. PMid:28677803.

Yoon, S., Lam, W. W. T., Sham, J. T. L., & Lam, T.-H. (2015). Learning to drink: how Chinese adolescents make decisions about the consumption (or not) of alcohol. *The International Journal on Drug Policy*, 26(12), 1231-1237. http://dx.doi.org/10.1016/j.drugpo.2015.09.001. PMid:26440773.

Yu, B., Qin, S., Hu, B., Qin, Q., Jiang, H., & Luo, W. (2019). Resveratrol improves CCL4-induced liver fibrosis in mouse by upregulating endogenous IL-10 to reprogramme macrophages phenotype from M(LPS) to M(IL-4). *Biomedicine and Pharmacotherapy*, 117, 109110. http://dx.doi.org/10.1016/j.biopha.2019.109110. PMid:31252263.

Zhang, L., Li, X., Pang, Y., Cai, X., Lu, J., Ren, X., & Kong, Q. (2021). Phenolics composition and contents, as the key quality parameters of table grapes, may be influenced obviously and differently in response to short-term high temperature. *LWT*, 149, 111791. http://dx.doi.org/10.1016/j.lwt.2021.111791.

Zhang, M., Xing, L., Wang, Y., Luo, R., Li, X., & Dong, J. (2022). Anti-fatigue activities of anthocyanins from Lycium ruthenicum Murry. *Food Science and Technology*, 42, e31921. http://dx.doi.org/10.1590/fst.242703.

Zheng, F., & Wang, Z. (2020). miRNA-1180 suppresses HCC cell activities via TRAF1/NF-κB signaling pathway. *Food Science and Technology*, 40(Suppl. 2), 626-633. http://dx.doi.org/10.1590/fst.26219.

Supplementary Material

Supplementary material accompanies this paper.

Table S1. Types of intragastric solution.
Table S2. Experimental animal grouping.
Figure S1. The flowchart of the experimental design.

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