Malays J Pathol 2025; 47(3) : 377 – 400

REVIEW ARTICLE

Pathogenesis of alcoholic fatty liver disease: Molecular and cellular changes

Siti Norain AZAHAR[1], Chin Kok YONG [1], Nur Azlina MOHD FAHAMI[1,*], Ibrahim Abdel Aziz IBRAHIM[2]

[1]Department of Pharmacology, Faculty of Medicine, Universiti Kebangsaan Malaysia, Jalan Yaacob Latif, Cheras, 56000, Kuala Lumpur, Malaysia; [2]Department of Pharmacology and Toxicology, Faculty of Medicine, Umm Al-Qura University, Saudi Arabia

Abstract

Alcohol-related liver disease (ALD) is a significant public health issue, leading to liver injuries, including fatty liver, hepatitis, and cirrhosis. The COVID-19 pandemic has exacerbated the problem by increasing alcohol abuse and hospitalisations for ALD. Since there are no approved therapies for ALD, promoting abstinence from alcohol is the primary approach. However, the mechanisms by which alcohol induces fat accumulation in the liver, the disease's initial stage, are not fully understood. This knowledge gap hampers the development of new treatments for ALD. This review aims to explore research on alcohol-induced fatty liver and compare it with metabolic-associated fatty liver disease. The goal is to merge these findings with current knowledge of ALD and hepatic lipid metabolism, including fatty acid oxidation, lipogenesis, and very-low-density lipoprotein (VLDL) secretion. Besides lipid metabolism, factors like inflammation, oxidative stress, cellular hypoxia, and autophagy also contribute to ALD's development and progression. By identifying gaps in understanding the molecular mechanisms of ALD progression, this review suggests future research directions. It emphasises how alcohol disrupts hepatic lipid metabolism, highlighting mechanisms leading to alcohol-associated fatty liver disease and other harmful effects of alcohol abuse.

Keywords: autophagy, inflammation, lipogenesis, mitochondrial beta-oxidation, oxidative stress, steatosis

INTRODUCTION

Alcohol is a psychoactive substance with addictive properties. Its consumption and health problems are prevalent all over the world. More than 200 distinct illnesses and adverse effects have been linked to alcohol intake, including mental and behavioural disorders, including alcohol dependence, major noncommunicable diseases, such as liver cirrhosis, some cancers, and cardiovascular diseases, as well as injuries from violence, car accidents, and collisions. Most nations continue to have worrisome rates of alcohol-related illness and death.[1]regional and country consumption of alcohol, patterns of drinking, health consequences and policy responses in Mewomber States. It represents a continuing effort by the World Health Organization (WHO One of the top five risk factors in the world for the onset of disease, disability, and mortality is the harmful use of alcohol.[2] According to the World Health Organisation (WHO), alcohol consumption is responsible for more than 3 million deaths each year, accounting for approximately 5% of all global deaths in 2018.[3]

Alcohol consumption not only affects the drinkers and society as a whole, but also has social and economic repercussions for other individuals. The differences in susceptibility to alcohol-related problems across various societies and individuals can be attributed to a number of factors, such as cultural norms, alcohol availability, governmental policies, age, sex, family history, socioeconomic level, and cultural background.[1,4,5]

Excessive and long-term consumption of alcohol is a prominent contributor to liver disease.[3,6] It is strongly linked to the development of alcohol-associated liver disease (ALD), which encompasses a range of conditions, starting from

**Address for correspondence:* Department of Pharmacology, Faculty of Medicine, Universiti Kebangsaan Malaysia, Jalan Yaacob Latif, Cheras, 56000, Kuala Lumpur, Malaysia. Tel: +603-9145 9574. Email: *nurazlinamf@ukm.edu.my* (N.A.M.F.)

the accumulation of fat in the liver (steatosis) and progressing to cirrhosis and hepatocellular carcinoma.[7] ALD occurs due to heavy or risky drinking habits, which involve consuming more than 3 standard drinks per day for men and more than 2 drinks per day for women, or engaging in binge drinking (defined as consuming more than 5 standard drinks for men and more than 4 for women within 2 hours).[8] These patterns of alcohol consumption increase the likelihood of experiencing health issues related to alcohol.[9] The clinical manifestations of ALD range from being asymptomatic to severe cases of acute alcoholic hepatitis (AH) with or without cirrhosis.[10] ALD affects around 2-2.5% of the general population and is more prevalent in regions with higher alcohol consumption rates.[10] In Western countries, alcohol is a major contributing factor in up to 50% of cases involving end-stage liver disease.[11]

Therefore, this article aims to provide a comprehensive overview of alcohol use disorder (AUD) and alcoholic liver disease, a significant health issue commonly associated with chronic alcoholism. Apart from alcoholic liver disease, the focus of this review is to examine one specific aspect of the disease, namely alcoholic fatty liver disease (AFLD). AFLD is characterised by the accumulation of fat in liver cells, representing the earliest form of liver damage that typically occurs in individuals who engage in long-term heavy drinking. The objective is to analyse the extent of this problem, its prevalence, and to summarise the proposed mechanisms underlying its pathophysiology. This review will delve into both the metabolic and molecular factors that contribute to ethanol-induced steatosis, as well as explore the potential mechanisms through which liver injury is initiated, leading to the development of advanced liver pathology.

1. Epidemiology of alcohol abuse

There is a lack of comprehensive data on the prevalence of ALD. There are regional differences in the amount of alcohol consumed. As an example, North Africa and the Middle East reported the lowest yearly per capita alcohol intake at 1.0 L per person, whereas Eastern Europe recorded the highest at 15.7 L per person.[12] A recent study indicates that global adult per-capita alcohol consumption increased from 5.9 L to 6.5 L between 1990 and 2017 and was projected to reach 7.6 L by 2030. In 2017, 20% of adults were heavy episodic drinkers, compared to an estimated 18.5% in 1990, and this prevalence is expected to rise to 23% in 2030.[13]

In North America, alcohol misuse and usage are pervasive. Over the course of their lives, 85.6% of adults in the US reported drinking alcohol, with 25.8% reporting excessive consumption.[14] 19.1% of people in Canada, reported excessive drinking, and 78.2% of people overall reported consuming alcohol in the previous year.[15] In China, where per capita annual alcohol intake ranges from 4-6 L per person[12], a recent study found a prevalence rate of 4.5% for ALD, and this rate was expected to increase in the future.[16]

In Malaysia, the previous National Health and Morbidity Survey (NHMS) conducted in 2011 and 2015 revealed that the prevalence of current alcohol drinkers (including shandy, beer, stout, brandy, whisky, wine, *samsu, tuak*, and others) among individuals aged 18 years and above was 12.8% and 8.4%, respectively.[17] However, according to the NHMS 2019 data, the prevalence of current alcohol drinkers among Malaysian adults aged 18 years and above decreased to 11.8%, of which 17.6% were categorised as risky drinkers. The prevalence of binge drinking among adults in Malaysia aged 18 years and above was 5.4%, while the prevalence of heavy episodic drinkers (HED) was 1.0%.[18] In East Malaysia, specifically the states of Sabah and Sarawak, the prevalence of alcohol consumption was the highest in the country, with 18.4% of the population currently consuming alcohol in Sabah. Among the indigenous people of East Malaysia, 37% of drinkers had an alcohol use disorder identification (AUDIT) score of 8 or more, indicating risky drinking behaviour.[19]

Several attempts have been made to assess the impact of alcohol use and alcohol use disorders on a global scale, as well as within Europe.[20,21] It was estimated that one in three people worldwide consumes alcohol.[22] Although drinking patterns vary across countries, the overall burden on public health due to alcohol use is significant, ranking among the top ten risk factors for both mortality and disability-adjusted life-years (DALYs).[22] This burden was further intensified during the COVID-19 pandemic, which witnessed an increase in alcohol abuse and a subsequent rise in hospitalisation rates for ALD.[23,24] Globally, alcohol is responsible for 5.1% of the overall burden of disease and injury, as measured in disability-adjusted life years (DALYs) in 2016.[3]

Furthermore, alcohol consumption also imposes a considerable economic burden.[20]

Rehm *et al*. (2009) demonstrated that the weighted average cost of alcohol was 2.5% of gross domestic product based on purchasing power parity (GDP-PPP) among high-income countries such as Canada, France, Scotland, and the United States. They also found that the average social cost was 2.1% of GDP-PPP for certain Asian middle-income countries like the Republic of Korea and Thailand, and it ranged between 0.45% and 5.44% of GDP in another study.[25]

The economic impact of alcohol dependence in Europe was significant, with the annual direct costs ranging from 0.04% to 0.31% of a country's gross domestic product (GDP).[26] In the United States, based on national databases, the economic cost of excessive drinking was estimated to be $223.5 billion in 2006. This cost breakdown included 72.2% from lost productivity, 11.0% from healthcare costs, 9.4% from criminal justice costs, and 7.5% from other effects.[27]

Similar to other developing countries, Malaysia is confronted with a growing issue of alcohol abuse. However, due to limited studies conducted, it is challenging to determine the precise magnitude of the problem. Nevertheless, it is estimated that Malaysians spend over RM 2 billion on alcohol annually, primarily on beer and locally known inexpensive spirits called samsu. This amount was nearly four times the annual budget allocated for health services in the country.[28]

Alcohol is recognised as a pervasive and harmful factor in the United Nations' Sustainable Development Goals (SDG) Agenda 2030, emphasising the need for effective measures to address the disease burden associated with harmful alcohol use.[29]

2. Spectrum of ALD

ALD is a broad category that includes many hepatic diseases, the most prominent of which is fatty liver, or steatosis. When 4-5 standard drinks are consumed daily over several years, almost 90% of those with alcohol-related problems experience steatosis as their first and most common reaction.[30] An alcoholic beverage that has around 0.5 fluid ounces, or about 14 grams, of pure alcohol in it is referred to as a standard drink. But binge drinking, which is defined as consuming four to five drinks in a two-hour period, can also result in steatosis.[31] Steatosis was formerly thought to be a harmless side effect of alcohol abuse. When individuals affected by steatosis abstain from drinking, the condition is reversible with a favourable prognosis. However, if alcohol consumption persists, up to 40% of chronic drinkers may progress to more advanced stages of alcoholic liver disease, including inflammation, fibrosis, cirrhosis, and potentially even liver cancer (refer to FIG 1).[32] Patients with chronic steatosis are susceptible to fibrotic liver disease because the presence of fat likely increases the risk of lipid peroxidation and oxidative damage.[33]

The figure illustrates the full spectrum of ALD which encompasses a range of conditions from steatosis to steatohepatitis, fibrosis, cirrhosis, and hepatocellular carcinoma (HCC). The stepwise progression of ALD, beginning with a normal liver that, in up to 90% of heavy drinkers, develops steatosis due to excessive alcohol

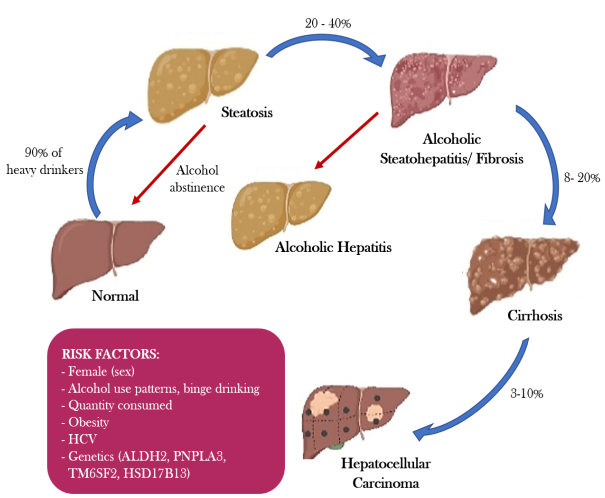

FIG. 1. The Progression of Alcohol-related liver disease (ALD).

intake. While steatosis is reversible with alcohol abstinence, continued drinking can lead to alcoholic hepatitis and subsequently to alcoholic steatohepatitis with fibrosis, affecting about 20–40% of individuals. Alcoholic steatohepatitis is characterised by liver tissue inflammation marked by neutrophil infiltration, Mallory-Denk bodies, ballooning degeneration, and hepatocyte death. Persistent injury may progress to cirrhosis in 8–20% of cases, which may eventually result from liver fibrosis, and is characterised by widespread scarring and impaired liver function. A subset of cirrhotic patients (3–10%) may further advance to hepatocellular carcinoma (HCC). Any stage of ALD can result in alcoholic hepatitis (AH), an acute-on-chronic illness that manifests clinically as jaundice, infection, and decompensation. While corticosteroids and abstinence are treatment options for AH, liver transplantation is a possibility for end-stage liver disease. This clinical trajectory is also heavily influenced by a confluence of key risk factors determining susceptibility and progression—including female sex, binge drinking patterns, total alcohol consumption, obesity, hepatitis C virus (HCV) infection, and specific genetic predispositions such as aldehyde dehydrogenase 2 (ALDH2), patatin-like phospholipase domain-containing protein 3 (PNPLA3), transmembrane 6 superfamily member 2 (TM6SF2), and hydroxysteroid 17-beta dehydrogenase (HSD17B13). These genetic variants are strongly associated with an increased risk of progressing to severe ALD. When assessing alcohol consumption, it is important to note that a serving of 12 fluid ounces of beer, four ounces of wine, or 1.5 fluid ounces of distilled spirits all contain approximately 14 grams or 0.6 fluid ounces of pure alcohol.[34] Based on this information, men have a threshold for developing severe alcohol-associated liver disease when consuming less than 60-80 grams of alcohol per day over 10 years.[35] On the other hand, women appear to be at an increased risk of experiencing a similar degree of liver damage by consuming 20-40 grams of alcohol per day.[36]

TABLE 1 below presents a visual representation of the ascending order of ethanol content in various alcoholic beverages, known as "standard drinks." The National Institute on Alcohol Abuse and Alcoholism defines a standard drink as approximately 14 grams of pure ethanol. The alcohol content, expressed as alcohol by volume (alc/vol), differs across different beverages.

3. Alcoholic fatty liver disease (AFLD)

3.1 Definition

Alcoholic fatty liver disease (AFLD) is characterised histologically by the accumulation of fat molecules within hepatocytes, appearing as a combination of big (macrovesicular) and small (microvesicular) droplets due to increased intracytoplasmic triglyceride formation.[37] Under a microscope, this fat deposition is seen as lipid droplets or fat vacuoles in liver tissue sections. The hepatocytes that surround the liver's central vein (perivenular hepatocytes) are the first to be affected, followed by mid-lobular hepatocytes and, finally, periportal hepatocytes, which surround the liver's portal vein.[31]

3.2. Clinical Signs and Diagnosis

Obesity, bloating, nausea, vomiting, lethargy, and appetite loss are prominent symptoms that indicate the existence of ALD.[38] However, AFLD often exhibits subtle clinical manifestations and is often asymptomatic. Occasionally, when steatohepatitis is suspected, patients with fatty liver may have discomfort in the right upper

TABLE 1: Examples of "Standard Drink" in multiple alcoholic beverages.[31]

1 "Standard Drink" equals to:	
Types of Beverages	**Amount**
Regular beer (contains 5% alcohol)	12 fl oz
Malt liquor (contains 7% alcohol)	1-9 fl oz
Table wine (contains 12% alcohol)	5 fl oz
Fortified wine like sherry or port (contains 17% alcohol)	3-4 fl oz
Cordial, liqueur, or aperitif (contains 24% alcohol)	1-3 fl oz
Brandy or cognac (contains 40% alcohol)	1-5 fl oz (a single shot)
Distilled spirits like gin, rum, tequila, whiskey, vodka (contains 40% alcohol)	1.5 fl oz shot

quadrant, painful hepatomegaly, nausea, and jaundice. If an accurate history of drinking habits, patterns, and quantity is not gathered, it might be difficult to distinguish between alcohol-induced fatty liver and non-alcohol-induced fatty liver (NAFLD), as they have histological and clinical similarities.[39] Underreporting or nonreporting alcohol usage by patients can lead to misdiagnosis of the condition in some situations.[40]

The patient's history of alcohol use, a physical examination, and laboratory results showing liver damage are all used to make the diagnosis of ALD.[41] Since it can be difficult to get a precise history of alcohol use, liver damage and alcohol use are detected using particular biochemical blood indicators. Measurements of aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyltransferase (γ-GT), and mean corpuscular volume (MCV) are part of liver function testing. ALD may be in its early stages if these markers are elevated, but more advanced stages may be indicated by reduced albumin, longer prothrombin time, greater bilirubin levels, or thrombocytopenia.[41]

An indicator that can be used to differentiate between alcoholic and non-alcoholic fatty liver diseases is the aspartate transaminase (AST)/alanine transaminase (ALT) ratio. If the value is 1 or higher, it is indicative of AFLD; if it is less than 1, it strongly suggests NAFLD.[42] Typically, the AST/ALT ratio is twice as high in 70–80% of ALD patients as it is in healthy individuals.[8] Transaminase levels are increased in AFLD by more than 5–10 times the normal value, with AST values typically being greater than ALT values.[43]

The most accurate indicator for evaluating drinking patterns is carbohydrate-deficient transferrin (CDT), which functions as a highly precise biomarker for identifying chronic alcohol consumption. Generally, four to five days following the most recent alcohol misuse incident, the concentration of alcohol metabolites in urine as measured by CDT is assessed.[44]

In addition to liver-related biomarkers, hepatomegaly and lipid deposits seen on computed tomography and ultrasonography can be used as indicators of steatosis.[43] A non-invasive biomarker called FibroTest™ is used to assess fibrosis in chronic liver diseases such as ALD[45] and NAFLD.[46] The World Health Organization (WHO) recommends the FibroTest and SteatoTest for the assessment of individuals with metabolic conditions or excessive alcohol consumption. These tests can be made more accurate in patients with ALD who have liver fibrosis or fatty liver by combining carbohydrate-deficient transferrin (CDT) with other biochemical indicators (e.g., GGT, AST/ALT ratio).[41]

Moreover, recent research has led to the development of various innovative clinical criteria scores and biomarkers, offering improved diagnostic, prognostic, and potential therapeutic options for the management of ALD. The assessment of liver toxicity involves the measurement of different inflammatory mediators such as cytokines, DNA fragmentation, and examination of histopathology.[47] Several scores have been devised, validated, and utilised in clinical practice, including the Discriminant Function Index (DF), Glasgow Alcoholic Hepatitis Score (GAHS), the Age, Bilirubin, INR, and Creatinine (ABIC) score, and the Model of End-Stage Liver Disease (MELD).[48] Importantly, the implementation of screening tools like the Alcohol Use Disorders Inventory Test (AUDIT) has proven to enhance detection rates and the ability to predict long-term clinical outcomes, including hospitalisation for alcohol-related conditions.[49] A score greater than 8 on the AUDIT is indicative of harmful or hazardous alcohol use, while scores greater than 20 suggest alcohol dependence (now referred to as moderate/severe AUD).[50] While liver biopsy is an invasive procedure and usually not necessary for diagnosis, it can be valuable in determining the extent of ALD and ruling out the presence of alcoholic cirrhosis (AC).[43]

3.3 Common Risk Factors for ALD Progression

Several factors have been identified as important determinants or modifiers that can either worsen, slow down, or even prevent the progression of ALD. Notably, women are more vulnerable to the development of ALD compared to men, even with lower alcohol consumption.[51,52] This difference increases susceptibility of women to alcohol-induced liver injury is likely attributed to sex-specific differences in the metabolism of alcohol in the gastrointestinal and hepatic systems.[32] Furthermore, liver damage is exacerbated by the amplification of tumour necrosis factor-alpha (TNF-α) mRNA, which activates macrophages and liver cells, resulting in a pro-inflammatory response and increased reactive oxygen species (ROS) levels.[53] For a comprehensive overview of the risk factors, please refer to TABLE 2.

4. Hepatic Alcohol Metabolism

Alcohol, a polar molecule, can dissolve in both water and fat. When consumed, it is absorbed into the bloodstream through the digestive system. Over 95% of the ingested alcohol is metabolized by the liver, with the remaining tiny amount being expelled by the respiratory system, urine, and perspiration.[65]

Beverage alcohol, such as ethanol, is primarily metabolised in the main liver cells called hepatocytes, which make up about 70% of the liver's mass.[66] The conversion of alcohol into acetaldehyde is mediated by three primary metabolic pathways or systems, according to earlier research. The first system is alcohol dehydrogenase (ADH), located in the cytoplasm of hepatocytes. ADH uses nicotinamide adenine dinucleotide (NAD+) as a co-factor to oxidise ethanol into acetaldehyde. ADH1 is the primary isoenzyme responsible for ethanol metabolism in the liver. Acetaldehyde dehydrogenase (ALDH) is responsible for converting the acetaldehyde that ADH produces into acetate.[67]

The second major metabolic system is cytochrome P450 2E1 (CYP2E1), which is found in the smooth endoplasmic reticulum (ER) and metabolises various chemicals into polar metabolites for excretion. CYP2E1 is the

TABLE 2: General Predictors for the Progression of ALD

Risk Factors	Description
Quantity, type of beverage, and drinking pattern	• Intake of 40–80 g ethanol/day in men and 20-40 g/day in women produces fatty liver • 160 g per day intake for 10–20 years causes more advanced ALD (steatohepatitis, fibrosis, or cirrhosis.[9]
Sex	• Women exhibit a higher susceptibility to alcoholic liver disease (ALD) compared to men, at a quantity exceeding 20 grams per day. • When women consume the same amount of alcohol as males, they appear to be more vulnerable and have greater blood alcohol concentrations. This disparity results from women's bodies having a less percentage of water than men with similar weights do.[54] • Women are suggested to have a lower capacity for ethanol oxidation in the gastrointestinal tract, known as first-pass metabolism.[55] Due to this metabolic deficit, women's livers are exposed to higher doses of ethanol because more ethanol can enter the portal circulation. • Sex-based differences in sensitivity to hepatic inflammatory response and the capacity to metabolise ethanol in the gut have been identified as contributing factors to the increased susceptibility of ALD progression in women compared to men.[55]
Age	• Individuals in the older age group (65 years and above) are at a higher risk and exhibit more pronounced impairments caused by ethanol compared to younger individuals.[56,57]
Genetics	• Specific genetic markers, known as single-nucleotide polymorphisms, within genes that encode alcohol-metabolising enzymes, cytokines, and antioxidant enzymes have been discovered to be associated with the progression of ALD.[58] • The expression of the rs738409 gene is linked to the development of severe liver damage, including liver cirrhosis, in cases of ALD. • Alcoholic cirrhosis is independently associated with an allele of the patatin-like phospholipase domain-containing protein 3 (PNPLA3 I148M), an enzyme that breaks down triglycerides.[59]
Race/Ethnicity	• Alcohol holds a customary significance in the cultural traditions of various ethnic communities. • Ethnicity plays a significant role in determining the age and severity at which different subtypes of ALD manifest.[60]

Nutritional Factors and Obesity	• The consumption of dietary fat, including obesity, contributes to ALD. • There is a significant correlation between alcohol consumption and a higher risk of liver injury in people with a high body mass index.[61] • Dietary unsaturated fat, especially that which is high in linoleic acid, is said to encourage alcohol-induced liver damage in mouse models, but dietary saturated fat appears to protect against it.[62]
Drugs	• Alcohol has the potential to interact with various drugs, both prescription medications and illicit substances, leading to an increased risk of hepatotoxicity. For example, alcohol abuse can worsen the hepatotoxic effects of drugs like paracetamol.[31]
Smoking	• Cigarette smoking increases the incidence of alcoholic cirrhosis in humans and has deleterious effects on some liver functions.[63]
Viral Infections	• Simultaneous infection with hepatitis C (HCV) and hepatitis B (HBV) in individuals with alcoholic liver disease (ALD) is linked to accelerated progression towards fibrosis, cirrhosis, and potentially hepatocellular carcinoma.[64] • Liver disease in individuals with HIV infection can be attributed to viral hepatitis, which occurs within an environment influenced by persistent immune activation and altered cytokine patterns.

main enzyme involved in this system. Under normal conditions, CYP2E1 metabolises a small amount of ethanol (about 10%) into acetaldehyde. However, chronic alcohol abuse leads to an increase in CYP2E1 expression, resulting in enhanced metabolism of ethanol.[7] The endoplasmic reticulum's catalase, the third metabolic system, is dependent on NADPH for its oxidative metabolism. Catalase, an enzyme containing heme and located in peroxisomes, primarily eliminates hydrogen peroxide (H2O2), but also plays a role in oxidising ethanol to acetaldehyde.[7] While catalase is largely involved in the brain's ethanol oxidation process, its involvement in the liver is far smaller.[68] The principal pathway for ethanol metabolism is the ADH pathway when there is low concentration of ethanol in the blood and tissue fluid. But the other two enzyme systems also get involved in the metabolism of ethanol when the concentration rises above 10 mol/L.[7] The overall process of alcohol metabolism in the liver is depicted in FIG 2 below.

The liver's hepatocytes are principally responsible for the metabolism of ethanol, sometimes referred to as ethyl alcohol. This process involves the consecutive catalysis of oxidations that convert ethanol to acetate by ALDH2, an enzyme found in the mitochondria, and ADH, a significant enzyme found in the cytoplasm. Two-mole equivalents of reduced nicotinamide adenine dinucleotide (NADH) are produced by this process. Another significant enzyme involved in ethanol metabolism is cytochrome P450 2E1 (CYP2E1), which is mainly located in the endoplasmic reticulum. In the presence of molecular oxygen (O2), CYP2E1 oxidises ethanol to acetaldehyde and converts reduced NAD phosphate (NADPH) to its oxidised form, generating water. A smaller mechanism of ethanol oxidation occurs in the peroxisomes as well, where catalase converts ethanol to acetaldehyde and water by using hydrogen peroxide (H2O2).

5. Pathogenesis of ALD

Currently, our knowledge of the intricate mechanisms underlying ALD pathogenesis remains significantly limited.[69,70] As a direct hepatotoxin, alcohol consumption sets off a variety of metabolic processes that together affect the final hepatotoxic consequences.[70] When alcohol is metabolised by hepatocytes, it sets off a pathological cascade, involving the formation of protein-aldehyde adducts, immune responses, lipid peroxidation, and the release of cytokines.[71] Alcohol-induced fatty liver, known as steatosis, is characterised by impaired oxidation of hepatic fatty acids, leading to the accumulation of triglycerides, phospholipids, and cholesterol esters in liver cells, thus promoting lipogenesis. Previous studies reveal that excessive alcohol intake modifies the ratio in hepatocytes of reduced NADH to oxidized

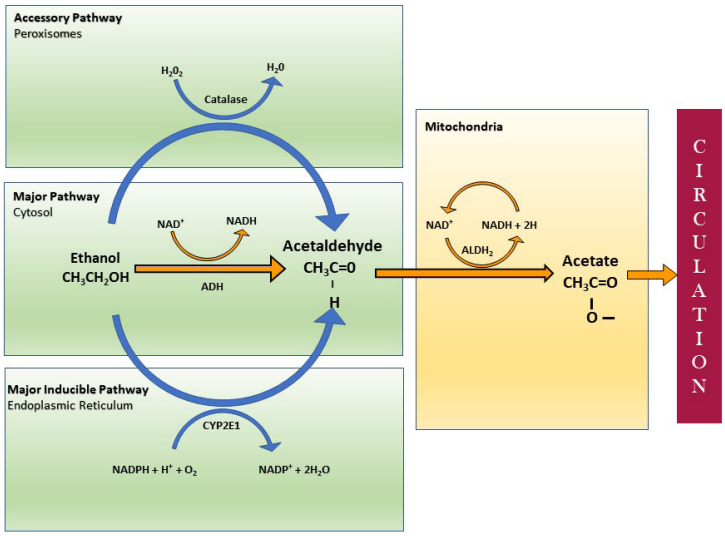

FIG. 2. Major and minor ethanol-oxidizing pathways in the liver.

nicotinamide adenine dinucleotide (NAD+), impairing the mitochondrial beta-oxidation of fatty acids and exacerbating the condition known as alcoholic fatty liver (AFL).[72] Numerous underlying mechanisms, including the direct or indirect regulation of variables linked to lipid metabolism, have been clarified by recent research and have contributed to the development of AFL. Key transcription factors involved in lipid metabolism, such as sterol regulatory element binding protein 1 (SREBP-1), peroxisome proliferator-activated receptor-alpha (PPAR-α), and early growth response-1 (Egr-1), have been implicated. Ethanol increases SREBP-1 expression, stimulating lipogenesis through the activation of target genes involved in fatty acid synthesis. Acetaldehyde, an ethanol byproduct, causes direct overexpression of SREBP-1, while additional inflammatory consequences of ethanol metabolism indirectly contribute to this increase. Moreover, ethanol decreases PPAR-α's transcriptional activity and DNA binding, which results in less fatty acid oxidation.[73] Direct acetaldehyde inhibition takes place, while indirect inhibition is caused by oxidative stress produced by CYP2E1, adenosine, and suppression of adiponectin.[69] The diminished expression of PPAR-α further contributes to lipogenesis and the development of AFLD by promoting fatty acid synthesis (FAS) and inhibiting fatty acid beta-oxidation. Egr-1 is also strongly induced before the onset of steatosis, and all these factors are regulated by the principal regulatory enzyme, adenosine monophosphate (AMP) -activated protein kinase (AMPK). Additionally, alcohol affects various factors, including hypoxia-inducible Factor 1 (HIF-1), which contribute to the development of AFLD.[72–74] In addition to adiponectin, other important humoral factors, such as TNF-α, also play a role in regulating alcohol-induced steatosis (FIG 3).

The figure highlights the interconnected metabolic, inflammatory, and gut–liver axis disturbances that collectively drive the development of alcohol-induced steatosis. It shows how alcohol exposure disrupts hepatocellular homeostasis through multiple signalling pathways, including impaired autophagy, altered lipid-regulatory networks, and enhanced cytokine-mediated stress responses. Through its metabolites, alcohol can cause hepatotoxicity either directly or indirectly. Alcohol is broken down into acetaldehyde by enzymes such as ADH and CYP2E1, which is subsequently transformed into acetate by ALDH and enters the citric acid cycle as acetyl-CoA. Alcohol metabolism through these enzymes increases the NADH/NAD^+ ratio and generates ROS at different stages. ROS are extremely harmful because they lower the antioxidant capacity of hepatocytes, which results in reduced levels of enzymes such as glutathione (GSH), catalase (Cat), and superoxide dismutase (SOD), leading to increased lipid peroxidation and harmful adduct formation. Moreover, alcohol causes the intestinal mucosa to become more fragile, increasing its permeability, and increases the susceptibility of Kupffer cells to endotoxin activation via toll-like receptor 4 (TLR4).

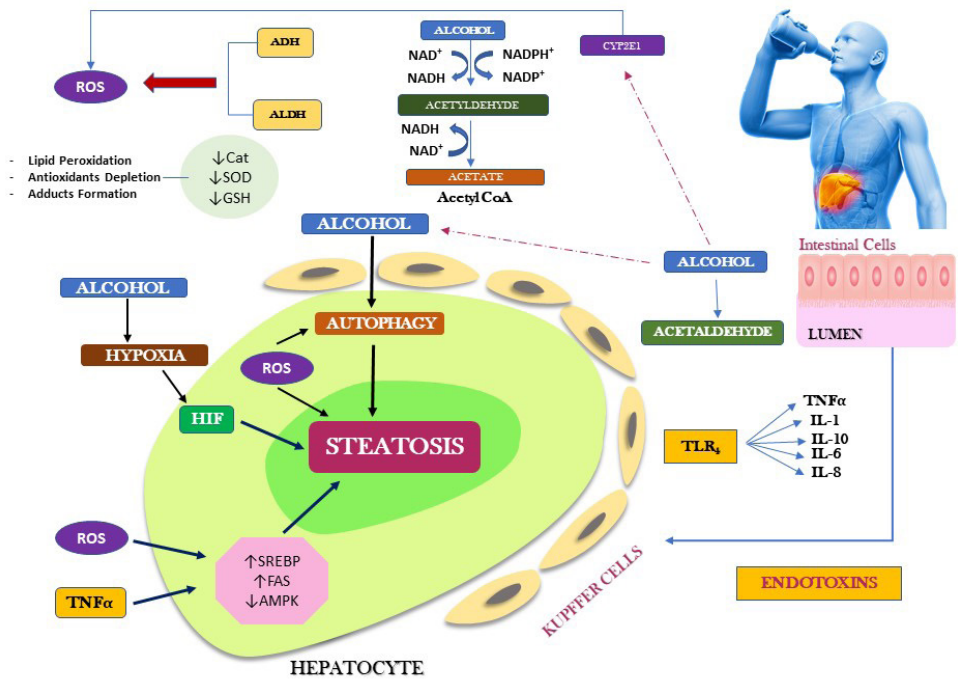

FIG. 3. Molecular basis in the pathogenesis of alcoholic fatty liver disease (AFLD).

As a result, pro-inflammatory cytokines such as tumour necrosis factor-α (TNF-α), ROS, interleukin-1 (IL-1), interleukin-6 (IL-6), interleukin-8 (IL-8), and interleukin-10 (IL-10) are produced at an increased level, which amplifies hepatic injury and hinders the transport and release of triglycerides. Additionally, alcohol can cause steatosis by encouraging the synthesis of fatty acids in hepatocytes and inhibiting their oxidation. SREBP-1, FAS, and AMPK are the two primary lipogenic signalling pathways that are impacted by alcohol. Within hepatocytes, ROS and inflammatory cytokines (notably TNF-α) upregulate lipogenic regulators (↑SREBP, ↑FAS) and suppress AMPK, tipping the balance toward fat synthesis and impaired fatty acid oxidation. Although acute alcohol intake may transiently stimulate autophagy as a protective response, prolonged exposure impairs this process, limiting the clearance of lipid droplets and damaged organelles. Lastly, chronic alcohol intake can induce cellular hypoxia, leading to the activation of hypoxia-inducible factor (HIF), which further contributes to the development of steatosis. Together, these processes converge on the hepatocyte to promote triglyceride accumulation, impaired lipid clearance, and cellular injury, ultimately leading to the steatotic phenotype characteristic of early alcoholic liver disease.

6. Factors involved in multiple mechanisms of Alcoholic Steatosis

As mentioned earlier in the section on ethanol metabolism, the process of oxidizing ethanol and acetaldehyde raises the concentration of NADH, which modifies the cellular redox potential and stimulates lipogenesis or the creation of lipids. But the fast accumulation of fat in the liver brought on by ethanol cannot be fully explained by changes in redox alone. Recent studies provide substantial evidence to suggest that ethanol-induced steatosis is a complex condition influenced by multiple factors, which will be discussed in the following section (refer to FIG 4).

AFLD involves complex interactions between hepatic (liver-related) and extrahepatic (outside the liver) mechanisms contributing to its development, particularly involving increased fatty acid production in the liver, influx of fatty acids from fat tissue, impaired lipid export from the liver, and reduced breakdown of lipids within the liver.

6.1 Alcohol Accelerates Hepatic Lipogenesis

Increased lipid synthesis occurs due to the upregulation of lipogenic enzymes and cytokines

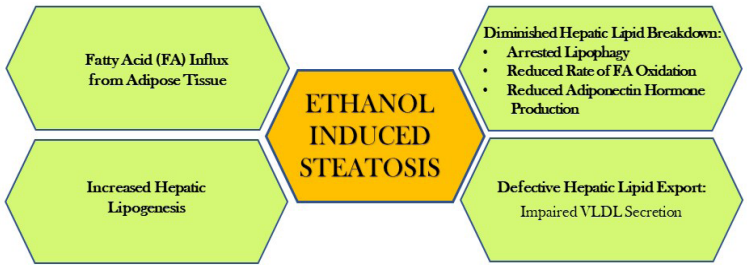

FIG. 4. Mechanisms that contribute to the development of Alcohol-Induced Fatty Liver.

(as listed in TABLE 3), which are controlled by two transcription factors: SREBP-1c and early growth response-1 (Egr-1). Heavy alcohol users experience disruptions in hepatic lipid metabolism, which results in the liver changing from an organ that burns fat to one that stores fat. As such, in abstinent people, SREBP-1c is largely inactive in the ER of hepatocytes. But in binge drinkers or chronic alcohol users, the translocation of SREBP-1c from the ER to the Golgi apparatus is initiated by hepatic ethanol oxidation. In the Golgi apparatus, SREBP-1c undergoes proteolytic maturation, converting it into an active form. This active form of SREBP-1c acts as a transcription factor and enters the nucleus, where it enhances the expression of genes involved in lipogenesis (refer to TABLE 3). Conversely, Egr-1 binds to gene promoter regions linked to alcohol-induced liver damage and steatosis and regulates the expression of genes that react to cellular stress. One significant gene regulated by Egr-1 is TNFα, a cytokine involved in lipogenesis. Additionally, since Egr-1 is activated shortly after ethanol administration, it also regulates the expression of the *SREBP-1c* gene.[75]

6.2 FA influx from adipose tissue

The development of steatosis is facilitated by adipose tissue. Excess calories from diet are stored as fat in adipose tissue, which is an essential location for energy storage. During periods of low nutrition (like fasting) or high calorie consumption (like exercise), fat can be utilised as needed to meet energy needs. Researchers have shown that persistent alcohol feeding of mice results in increased lipolysis (fat breakdown) in adipose tissue, which in turn reduces the overall proportion of adipose tissue.[77,78] Excess fat accumulates in the liver due to the liver's esterification of free fatty acids produced from adipose tissue into triglycerides.[78]

6.3 Alcohol Decelerates Hepatic Lipid Breakdown

Lipid extraction and subsequent oxidation are dependent on the breakdown of lipid droplets, which contain the majority of hepatic lipids. The engulfment of lipid droplets by double-membrane-bound vacuoles known as autophagosomes is the process by which lipophagy, a specialised form of autophagy, accomplishes this target. The lipid cargo is transported from the droplets to the lysosomes by these autophagosomes, where it is broken down by lipases, an enzyme that breaks down lipids. This process releases free fatty acids, which then go through β-oxidation in the mitochondria. Consuming ethanol continuously has been shown to slow down autophagy, in part because ethanol

TABLE 3: SREBP-1c-Regulated Lipogenic Enzymes.[76]

Enzyme	Function
Fatty Acid Synthase (FAS)	Synthesises fatty acids from palmitate and acetyl coenzyme A (CoA).
Acyl CoA Carboxylase (ACC)	Converts acetyl CoA into malonyl CoA.
ATP Citrate Lyase (ACL)	Produces acetyl CoA from CoA and citrate.
Stearoyl CoA Desaturase (SCD)	Converts saturated fatty acids (stearate) into unsaturated fatty acids (oleate).
Malic Enzyme (ME)	Produces NADPH (reducing equivalents) for the synthesis of triglycerides.

is thought to affect lysosome formation, which leaves lysosomes with reduced numbers and dysfunction. Thus, the steatotic liver experiences a slowdown in the breakdown of lipid droplets.[79]

Excessive alcohol intake further decreases the rate of β-oxidation after fatty acids are freed from lipid droplets. There are several reasons for this slowness. First, when ethanol is oxidized, more NADH is produced, which prevents mitochondrial β-oxidation. Second, the acetaldehyde created by metabolism deactivates the transcription factor peroxisome proliferator-activated receptor alpha (PPAR-α), which functions in tandem with the retinoid X receptor (RXR) to control the expression of genes related to fatty acid transport and oxidation. Through the formation of covalent bonds with the transcription factor, acetaldehyde possibly deactivates PPAR-α by blocking its recognition and binding to PPAR-α promoter regions.[74] Third, ethanol oxidation, whether acute or chronic, results in mitochondrial depolarisation, which hinders the production of adenosine triphosphate (ATP) molecules and causes outer membrane leakage, lowers β-oxidation rates and decreases the efficiency of fatty acid import.[80] Fourth, the hormone adiponectin, which is secreted by adipocytes (fat cells), is less produced when ethanol is consumed. Studies have indicated that the restoration of adiponectin levels in animals fed alcohol leads to the normalisation of fatty acid oxidation.[81] There is evidence to suggest that TNFα may also regulate the production of adiponectin, and adiponectin also seems to restrict the production of the cytokine.[82]

6.4 Alcohol Causes Defective Hepatic Lipid Export

It is commonly known that the liver primarily exports cholesterol and triglycerides in the form of very low-density lipoprotein (VLDL) particles. Hepatocyte fat accumulation is a result of any defect in the synthesis or export of VLDL particles. The availability of triglycerides, which make up more than half of the lipids in VLDL and are kept in cytoplasmic lipid droplets, controls the assembly of VLDL. Lipid droplet storage is the source of about 70% of the total triglycerides found in very low-density lipoproteins (VLDLs). To create VLDL triglycerides, these triglycerides are subjected to lipolysis and re-esterification. It is still unclear how exactly alcohol affects lipid droplet lipolysis, VLDL assembly, and subsequent secretion, despite past research pointing to a connection between altered VLDL secretion and the onset of alcoholic steatosis. Yet studies have shown that decreased activity of a crucial protein involved in the assembly of VLDL and decreased synthesis of a key component of VLDL are responsible for alcohol-induced impairment of VLDL secretion.[83,84]

7. Molecular Mechanisms of ALD

Considerable research has been dedicated to understand the molecular mechanisms involved in the pathogenesis of ALD. These mechanisms include direct liver damage, the generation of ROS caused by alcohol and its metabolites, innate immunity activation, and pro-inflammatory cytokines production.[85] The subsequent section will delve into a comprehensive discussion of each of these mechanisms, which are critical to the development of ALD.

7.1 Alcohol and Its Metabolites Damage the Liver

Hepatocyte mitochondria and microtubules are affected by acetaldehyde, a main byproduct of alcohol that damages the liver. Prolonged alcohol misuse usually causes CYP2E1 to be upregulated, which accelerates the breakdown of alcohol into acetaldehyde.[86] Acetaldehyde interferes with the secretion of hepatocytes and mitochondrial fatty acid β-oxidation. Furthermore, SREBP-1c transcription is directly stimulated by it, and this in turn enhances fatty acid production and subsequent accumulation in hepatocytes.[72] The long-term use of alcohol has been linked to the excessive production of ROS by the enzyme CYP2E1, which plays a crucial role as a mediator within cells. These ROS, which include superoxide and hydrogen peroxide ions, are essential for promoting the inflammatory traits linked to alcohol-induced liver injury. They achieve this by increasing the proliferation of pro-inflammatory cytokines and activating immune cells.[86] Furthermore, oxidative stress generated by ROS in fatty liver disease decreases the production of proteins involved in the signalling pathway for energy metabolism, such as AMPK, Sirtuin 1 (SIRT1), and STAT3, the transcription factor signal transducer and activator of transcription 3.[87] In addition, zinc and adiponectin levels are downregulated by this oxidative stress, activating PPARα.[72] This results in the peroxidation of phospholipids in the liver membrane, the production of lipid free radicals, and the activation of acetyl-CoA carboxylase (ACC), adiponectin, and Egr-1 TNF-α. The liver gradually accumulates fatty tissue as a result of these molecular processes.[72]

7.2 Oxidative Stress

Alcohol hepatotoxicity is frequently attributed to the production of free radicals and the ensuing oxidative stress.[88] Oxidative stress can be initiated by free radicals, which are molecules with unpaired electrons.[89]

In contemporary understanding, oxidative stress is characterised as the disturbance of redox signalling and regulatory mechanisms.[90] Hepatocytes possess numerous potential origins of ROS, the activity of which is intensified or influenced by prolonged alcohol consumption, leading to excessive production of oxidants.[90] Among these sources are CYP2E1-mediated oxidation, mitochondrial respiratory chain dysfunction, aldehyde oxidase activity, activation of Kupffer cells, disruption of lipogenesis, and increased cytokine production.[88,91]

Chronic alcohol misuse causes the liver to express more CYP2E1, which in turn causes abnormalities in lipid peroxidation brought on by ROS, or free radicals. Consequently, the antioxidant systems responsible for controlling critical genes involved in antioxidant defence, including nuclear factor erythroid 2-related factor 2 (Nrf-2) in ALD, are disrupted.[92] Superoxide dismutase (SOD), glutathione (GSH), catalase, peroxidase-1, metallothionein, and heme oxygenase (HO-1) are among the other significant antioxidant genes implicated in ROS scavenging that are significantly impacted.[93] (TABLE 4) Research findings have revealed that reactive oxygen species (ROS) produced during alcohol metabolism are rapidly eliminated by peroxiredoxin-1 and CYP2E1 located within the cytoplasmic side of the endoplasmic reticulum membrane.[94] Consequently, impaired function of Nrf-2 disrupts the clearance of ROS. While antioxidant mechanisms play a crucial role in neutralising a significant portion of reactive oxygen species (ROS) to protect cells, some ROS can still inflict cellular damage, such as lipid peroxidation (LPO), enzyme deactivation, and DNA mutations.[95]

Because LPO primarily targets subcellular organelles and biomembranes, it is especially important in alcohol-induced liver injury.[88] According to Lívero et al. (2014), mice with steatosis who were fed a low-protein diet and consumed 10% alcohol for six weeks had increased levels of LPO. Along with lower catalase and SOD activity, the study also discovered increased levels of total ROS, suggesting that oxidative stress contributed to the

TABLE 4: Enzymatic Mechanisms to protect against Liver Damage caused by Free Radicals

Enzyme	Specific area within cells	Role	Impact of long-term ethanol consumption	References
Catalase (Cat)	Peroxisomes	Converts H_2O_2 to H_2O	Boosts activity	97
Glutathione-S-Transferase (GST)	Nuclei, cytosol, mitochondria	Transfers sulphur to acceptor molecules	Boosts activity	97
Glutathione Peroxidase (GPx)	Cytosol/ mitochondria	Scavenges peroxides and free radicals	Unaffected	97
Copper–Zinc-Superoxide Dismutase (Cu/Zn-SOD)	Cytosol	Converts superoxide to H_2O_2	Reduces activity and content	97 98
Manganese-Superoxide Dismutase (Mn-SOD)	Mitochondria	Converts superoxide to H_2O_2	Reduces activity and content	97 98
Glutathione Reductase (GR)	Cytosol	Regenerates reduced GSH from GSSG	Reduces activity	99

animal's development of steatosis.[96] Additionally, acute alcohol consumption was demonstrated to elevate LPO and encourage hepatic steatosis in mice during a binge drinking scenario.

7.3 Centrilobular Hypoxia

Both humans and animals that consume alcohol on a regular basis experience cell death in the oxygen-deficient pericentral areas of the liver.[100] Hepatocytes adjust to cellular hypoxia by using less oxygen and increasing gene transcription, which controls angiogenesis, erythropoiesis, glucose uptake and metabolism, cell death, and proliferation.[101] Low oxygen levels cause hypoxia-inducible factors (HIF) to become activated. HIF-1 is a heterodimeric protein complex made up of three subunits (HIF-1α, -2α, and -3α) among these factors. The principal modulator of genes responsive to variations in intracellular oxygen tension is thought to be the redox-sensitive HIF-1α subunit.[102] The control of hepatic lipid metabolism has been found to be significantly influenced by HIF-2α.[103] HIF-1α and -2α are more highly expressed in the liver of mice consuming alcohol, both acutely and chronically. This could account for the early development of steatosis in zone 3 hepatocytes (perivenular). Zone 2 and even zone 1 hepatocytes (periportal) may be affected in cases of severe liver damage.[72] Additionally, in addition to the hepatocyte changes brought on by toxins, which result in neutrophilic inflammation and coagulative necrosis, sinusoidal cells also play a role in the pathophysiology of liver injury.[104] Interestingly, several major changes including cellular enlargement, blood cell aggregation, and disruptions in microcirculation, happen at the sinusoidal endothelium.[104]

7.4 Microbiome Disruption and Endotoxin Enteric Leakage

Studies show that long-term alcohol consumption can upset the intestinal bacteria's delicate equilibrium, which leads to the buildup of toxins in the intestine. Endotoxins accumulate in the intestines due to the overgrowth of gram-negative bacteria brought on by alcohol exposure. Acetaldehyde buildup is also a result of gram-negative bacteria and intestinal epithelial cells metabolizing alcohol. In tight and adherent junctions, this buildup stimulates tyrosine phosphorylation, which increases intestinal permeability to endotoxins. Endotoxins are consequently transported to the liver, resulting in inflammatory alterations in the liver as well as other organs.[105]

Additionally, studies show that the production of nuclear transcription factors including NF-κB and iNOS is stimulated in the digestive system by alcohol and its metabolites. By attaching to tubulin and triggering intracellular non-specific protease C, these compounds increase intercellular permeability. As a result, this mechanism causes modifications to the microtubule cytoskeleton, cytoskeletal phosphorylation, and alterations in cell structure. The intestinal barrier's integrity is thereby jeopardized.[105,106] Raised amounts of bacterial lipopolysaccharide (LPS) are found in the liver and portal circulation due to bacterial overgrowth and increased intestinal permeability. LPS activates CD14 on the surface of Kupffer cells, which are liver macrophages, by binding to the LPS binding protein. Following CD14's activation of TLR4, cytokines are subsequently activated downstream.

The TLR modulates cytokine production and release at the same time, which causes an overabundance of inflammatory substances to be released. The antiviral response and the systemic immunological regulatory function are hampered by this. As a result, in ALD patients, there is a "leak" that permits endotoxin to enter the bloodstream.[107] Once in the bloodstream, endotoxin stimulates inflammation and Kupffer cells, which prevents macrophage phagocytosis and encourages the production of hepatic stellate cells (HSC). TNF-α, IL-1, IL-17, osteopontin, CXC chemokines, inflammatory compounds, and free radicals are only a few of the many cytokines that these cells generate.[72] Particularly IL-17 stimulates the production of IL-8 and CXCL1 by hepatic stellate cells, which facilitates the recruitment of neutrophils.[69,70,79–88,71,89–98,72,99–107,73–78] Activated Kupffer cells and neutrophils release components associated with fibrosis, such as platelet-derived growth factor and transforming growth factor beta (TGF-β).[108]

Proinflammatory cytokines and TLR4 activation have been implicated with ALD, according to research on mice models. In these models, the likelihood of liver injury following ethanol consumption was decreased by knocking down TLR4, CD14, or the LPS-binding protein.[109–111]

7.5 Inflammation and Immune Exhaustion

Several by products are produced during the metabolism of ethanol, such as hydroxyethyl radicals, acetaldehyde, and components of lipid peroxidation. These metabolites have the ability to adduct with hepatocyte proteins

that are either inside or on their surface. These protein adducts have the potential to disrupt cellular processes or generate novel antigens, which could trigger an immune response that is mediated by T-lymphocytes in the presence of monocytes. The immune response has the potential to kill hepatocytes.[73] While heightened immunological response and inflammation are hallmarks of ALD, multiorgan failure brought on by sepsis from a severe bacterial infection usually accounts for the majority of fatalities in alcoholic hepatitis (AH). Due to the activation of inhibitory receptors on T-lymphocytes, such as T-cell immunoglobulin, mucin protein 3 (TIM-3), and programmed cell death protein 1 (PD-1), prolonged inflammation may result in immunological exhaustion. The inhibitory effects of these receptors on T-lymphocytes can impair neutrophil phagocytosis and the oxidative burst reaction, contributing to immune system failure.

7.6 Apoptotic Signalling Pathway and Autophagy in ALD

Liver cell autophagy and apoptosis are pathological abnormalities linked to ALD. Due to alcohol's detrimental effects on the liver and its metabolism, hepatocyte death and the acceleration of liver disease are caused by oxidative stress and inflammatory reactions. Numerous studies have concentrated on the endoplasmic reticulum (ER) stress brought on by oxidative stress in ALD and the apoptosis caused by mitochondrial damage. An essential part of mitochondria-dependent apoptosis is played by elevated reactive oxygen species (ROS) brought on by alcohol metabolism. As ROS build up in hepatocytes, they suppress the activation of the glycogen synthase kinase 3 beta (GSK3β)/Wnt/β-catenin signalling pathway and down-regulate cyclin D1, a G1 cell cycle protein in hepatocytes. This is accomplished by inhibiting the activation of alpha serine/threonine protein kinase (AKT).[112,113] Cell cycle arrest and the initiation of mitochondrial-dependent apoptosis are brought on by this. ROS have the ability to trigger mitochondrial-dependent apoptosis by activating c-Jun N-terminal kinases (JNK)/P38, nuclear factor kappa-light-chain-enhancer of activated B (NF-κB), and apoptosis signal-regulating kinase (ASK1).[114,115] Moreover, alcohol metabolism in the liver and gut might change gut flora and promote lipopolysaccharide (LPS) synthesis, which intensifies inflammatory reactions and causes disturbances in lipid metabolism and liver inflammation.[116] Both ROS and LPS increase TNF-α production[117], elevate pro-inflammatory factors like interleukin-1 beta (IL-1β) expression[37], trigger apoptotic pathways through ER stress, activate caspase cascades via JNK/STAT3 and P53[118], and recruit neutrophils to trigger Fas ligand-dependent apoptosis.[112]

Autophagy, the self-degradation of apoptotic cells via lysosomes[119], helps to degrade damaged proteins and organelles. Multiple studies have shown that autophagy inhibits steatosis, inflammation, and hepatocyte death in ALD.[120,121]

Several mechanisms have been identified as inducers of autophagy to protect against alcoholic liver damage, including the AMPK-Forkhead box O3A (FOXO3A) axis, sorting nexin 10 (SNX10)/chaperone, ALDH2, and cannabinoid receptor 2.[116,122–125]

8. ALD and NAFLD: Pathogenesis and Interchangeable Characteristics

Although both ALD and NAFLD share the commonality of excessive liver fat accumulation, they have some key differences in their development and other traits.

The amount of alcohol drank is a criterion used to define NAFLD and distinguish it from ALD; nevertheless, this criterion was arbitrarily and without sufficient evidence established. In the United States, alcohol consumption was reported in as many as two-thirds of NAFLD patients.[126]

Numerous studies have demonstrated a link between alcohol consumption and the heightened occurrence and advancement of NASH. For instance, in the Dionysos Study conducted by Bedogni *et al.* in 2007, which tracked 144 individuals without steatosis and 336 with steatosis over 8.5 years, alcohol intake (20 g/day) emerged as the most significant risk factor for both the onset and resolution of steatosis, additionally serving as a mortality indicator for people with fatty livers. Specifically, the study found a 17% rise in fatty liver incidence, a 10% decrease in steatosis resolution, and a 10% increase in mortality within the fatty liver cohort.[127] A study monitoring 71 NAFLD patients through liver biopsy revealed a correlation between fibrosis advancement and occasional drinking (at least once per month) as well as increased weekly alcohol intake.

The authors advise against severe episodic drinking in these patients because the results showed that moderate alcohol use accelerated the advancement of fibrosis in NAFLD.[128] An comprehensive cohort research with 58,927 young and middle-aged Koreans with NAFLD

who had initially low fibrosis scores discovered an independent relationship between declining fibrosis and light and moderate alcohol usage as opposed to abstaining from alcohol. When alcohol consumption was moderate, the effect was most noticeable. The authors also suggested that there might not be a safe limit on alcohol intake.[129] When compared to people who refrained from alcohol, a longitudinal study that examined liver biopsies from NAFLD patients revealed that low to moderate alcohol use was associated with a limited improvement in steatosis, raised AST levels, and reduced resolution of NASH.[130] A separate study indicated that complete alcohol abstinence could potentially halt disease advancement and was the key determinant for survival among patients with confirmed cirrhosis.[131] In individuals with NAFLD, consuming mild to moderate amounts of alcohol might serve as a contributing factor to the development of HCC. A study involving routine health screenings of Korean patients aged between 40 to 80 years found that the incidence of HCC correlated with hepatitis B and C infections. Furthermore, the risk of HCC increased with each additional 20 g/day of alcohol intake, with higher percentages observed in older age groups.[132] The evidence presented in these studies highlights a clear association between alcohol consumption and the progression of steatosis, fibrosis, and the development of cancer.

Furthermore, there is growing evidence of a synergistic relationship between alcohol and obesity in liver-related health issues. Recent research has shown that obesity and metabolic syndrome (MetS) exacerbate the progression of ALD and increase the likelihood of developing HCC as well as mortality rates. In comparison to controls (16.4% [CI, 8–25%]), heavy drinkers (46.4% [95% CI, 34–59%]) and obese people (75.8% [CI, 63–85%]) had greater prevalences of steatosis, according to the results of a cross-sectional investigation. Interestingly, there was an additive effect observed in the significantly higher frequency of NAFLD (94.5% [CI: 85–99%]) among heavy drinkers who were also obese. The study also found that among heavy drinkers, obesity doubles the incidence of steatosis.[133]

An analysis of the Third National Health and Nutrition Examination Survey (NHANES III) data found that MetS and excessive alcohol consumption were independently linked to higher mortality rates in individuals with steatosis. This underscores the significance of acknowledging alcohol consumption as a compounding factor in the context of overweight and obesity.[134] Two population-based studies in Taiwan have established a correlation between body mass index (BMI) and both the incidence and mortality of HCC. These studies highlighted that the combined presence of obesity and alcohol consumption synergistically increases the risk of developing HCC.[135] A comparison between these two prevalent causes of chronic liver disease is presented in TABLE 5.

CONCLUSION AND FUTURE PERSPECTIVES

The pathogenesis of AFLD involves complex interactions between hepatic and extrahepatic factors. This review has examined established molecular mechanisms underlying AFLD, based on current literature and recognised models of ALD. Key pathways include oxidative stress, dysregulation of lipogenic gene expression and inflammatory cytokines, centrilobular hypoxia, and impaired autophagy. However, our understanding of these molecular mediators in humans remains limited. The coexistence of MetS, particularly in individuals with obesity, exacerbates the progression of ALD, increasing the risk of HCC and related mortality. Addressing the dual aetiology of liver disease requires a comprehensive strategy that prioritises early identification and preventive treatments, especially given the global rise in obesity, MetS, and evolving patterns of alcohol consumption. In regions such as Malaysia, per capita alcohol consumption statistics may underestimate actual intake due to the availability of potent, low-cost brews not captured in national data. This discrepancy presents significant public health challenges. The associated socioeconomic burden, compounded by rising disease rates and the progression of ALD to more severe stages, highlights the urgency of targeted interventions. In response, government measures have included public awareness campaigns on the harms of alcohol and stricter enforcement of licensing regulations for alcohol retailers. However, there remains a pressing need to expand health education efforts—particularly around alcohol use disorder—through diverse media platforms and primary healthcare services. These initiatives aim to increase public awareness of the health and social consequences of alcohol misuse and encourage healthier behavioural choices. Acknowledging sex differences is essential, as studies indicate that women develop liver

TABLE 5: Contrasts and similarities of AFLD and NAFLD

	ALD	NAFLD
Public health importance	Associated with morbidity, (including psychiatric comorbidities), healthcare utilisation, and mortality in affected people from various socioeconomic backgrounds.[136]	Arise from the imbalance between caloric intake and expenditure in individuals, due to a global increase in food consumption, higher per capita income, sedentary behaviour and lifestyle, and a rising BMI.[137–139]
Risk factors	Alcohol intake, genetic predisposition, microbiome composition, and endotoxemia.[140]	Metabolic syndrome, sedentary lifestyle, type 2 diabetes, genetic predisposition, and microbiota composition.[140]
Clinical features	Asymptomatic to cirrhosis	
Diagnosis	Patient's alcohol consumption history alongside laboratory and imaging tests, but the accuracy of these methods may vary.[141] Discriminant indices such as the ALD/NAFLD index (ANI) demonstrating high diagnostic accuracy.[142,143]	
	Laboratory biomarkers	
	Increased gamma-glutamyl transpeptidase (GGT) levels, macrocytic anaemia, elevated ESR, thrombocytopenia, and elevated AST/ALT ratio, especially exceeding a value of 2, are highly indicative of ALD.[144]	Typically, the AST/ALT ratio is less than one.[145] An elevated serum ferritin level, exceeding 1.5 times the upper limit of normal (ULN), is linked to increased NAFLD activity score and advanced fibrosis.[146]
	Scoring systems	
	The Glasgow Alcoholic Hepatitis Score (GAHS), Maddrey's discriminant function (mDF), the Age-Bilirubin-International Normalised Ratio (INR)-Creatinine (ABIC) score, and the Lille score are useful for predicting short-term survival in patients with alcoholic hepatitis.[147,148] The Alcohol Use Disorders Identification Test (AUDIT) scores indicate the extent of harmful alcohol consumption and related behaviour.[149]	The Brunt score[150] and the NAFLD activity score (NAS)[151] are effective in distinguishing between simple hepatic steatosis and nonalcoholic steatohepatitis (NASH).[152]
	Histological findings	
	Greater inflammatory cell infiltration.[153]	Higher degree of fatty degeneration in liver cells.[153]
	Imaging studies and non-invasive methods	
	Ultrasound findings may include hyperechogenicity.[140] Validated noninvasive procedures for determining the fibrosis stage in patients with ALD and NAFLD include transient elastographyand fibrosis indices,[154] the AST/platelet ratio index (APRI),[155] the Fibrosis-4 (FIB-4) index,[156] and FibroTest.[157]	

Pathophysiological hallmark	The accumulated fat plays a passive role in an inflammatory process that damages hepatocytes containing fat, with immune cells in the liver triggering the process.[158]	The presence of an enlarged, dysfunctional, insulin-resistant adipose tissue leads to ectopic fat deposition and hepatic storage imbalance, with lipotoxicity being the main cause of hepatocyte injury.[159]
Pathogenic processes	Hepatic Fat Accumulation	
	Ethanol enhances acetyl-CoA carboxylase (ACC) activity, the key enzyme in de novo lipogenesis (DNL), and inhibits palmitic acid oxidation rate, resulting in changes to fatty acid metabolism and the development of steatosis.[160,161]	Excessive calorie intake enlarges adipocytes and increases their number, inducing insulin resistance, unregulated lipolysis and reduced fatty acid uptake, releasing free fatty acids (FFA) into circulation. Hepatocytes subsequently uptake FFA, promoting lipid droplet formation. DNL is frequently increased, which is associated with SREBP1c overexpression and peroxisome proliferator-activated receptor-alpha (PGC1-α) inactivation.[162,163]
	Insulin Resistance	
	Alcohol could induce insulin resistance (IR) and heighten the susceptibility to type 2 diabetes mellitus (T2DM) and advanced liver disease.[164]	Hyperinsulinaemia further activates the transcription factor SREBP-1c, which upregulates genes involved in DNL, thereby exacerbating steatosis.[165] Deteriorating IR is regarded as a potential catalyst for disease advancement in NAFLD.[166]
	Cell Death Pathways	
	Ethanol-induced endoplasmic reticulum (ER) stress and lipotoxicity, alongside reactive oxygen species, activate B-cell lymphoma 2 (Bcl2) apoptosis initiators[167] while suppressing guardian members, leading to cell death via caspase activation in mice.[168]	Aside from apoptosis, there are additional lytic forms of cell death such as necroptosis, pyroptosis, and ferroptosis, which are linked to cell-membrane permeabilization.[169]
	Immune Response	
	Neutrophils play a role in ALD progression by releasing reactive oxygen species (ROS), proteases, and proinflammatory mediators.[170]	Toll-like receptor 4 (TLR4) in NASH is activated by certain damage-associated molecular patterns (DAMPs), such as high-mobility-group protein box 1 (HMGB1), which considerably contributes to the early progression of NAFLD.[171]

	Inflammasome	
	Early stages of ALD exhibit heightened inflammasome activity and interleukin 1 (IL-1) production, contrasting with NASH where this is not typically seen early on.[172] Kupffer cells are the primary site of inflammasome activation in ALD, whereas hepatocytes are the primary site in NASH.[173]	
	Microbiota, Endotoxins, and Oxidative Status Changes in the gut microbiome contribute to increased intestinal permeability, enhanced translocation of bacteria and their byproducts to the liver through the hepatic portal vein, and elevated levels of endotoxins like lipopolysaccharides (LPS).[174]	
	Prolonged alcohol consumption enhances the expression of the NADPH oxidase 4 (NOX4) enzyme, a major ROS source, resulting in elevated mitochondrial superoxide levels, increased apoptotic cell numbers, and lipid accumulation.[175]	Oxidative stress is implicated in hepatic injury progression in NAFLD, spanning from steatosis to steatohepatitis, fibrosis, cirrhosis, and HCC.[176] Lipotoxicity is also linked to this process.[177]
Current management	Alcohol abstinence.	Lifestyle modifications, exercise interventions, and dietary changes.
	No approved treatment available. Treatment primarily aimed at managing disease progression and mitigating risk factors.	

[Abbreviations: ANI: ALD/NAFLD index; GGT: gamma-glutamyl transpeptidase; ESR: Erythrocyte Sedimentation Rate; AST/ALT ratio: serum aspartate to alanine amino-transferase levels ratio; ULN: upper limit of normal; GAHS: the Glasgow Alcoholic Hepatitis Score, mDF: Maddrey's discriminant function, ABIC score: the Age-Bilirubin-International Normalised Ratio (INR)-Creatinine score; The Alcohol Use Disorders Identification Test (AUDIT); NAFLD activity score (NAS); APRI: AST/platelet ratio index; FIB-4 index: Fibrosis-4 index; ACC: acetyl-CoA carboxylase; DNL: de novo lipogenesis; FFA: free fatty acids; PGC1-α: peroxisome proliferator-activated receptor-alpha; IR: insulin resistance; T2DM: type 2 diabetes mellitus; ER: endoplasmic reticulum; ROS: reactive oxygen species; DAMPs: damage-associated molecular patterns; HMGB1: high-mobility-group protein box 1; TLR4: toll-like receptor 4; IL-1: interleukin 1; LPS: lipopolysaccharides; NOX4: NADPH oxidase 4]

damage at lower levels of alcohol exposure and face a higher risk of cirrhosis compared to men. Although previous research has highlighted clinical disparities in ALD presentation between sexes, further investigation is needed to understand the increasing prevalence of this disease in women and the specific mechanisms underlying sex-related differences in alcohol metabolism. AFLD remains a compelling area for research, particularly due to the lack of approved treatments. Various combination therapies have been developed to address hepatocyte injury, with lifestyle modifications—such as reducing alcohol intake, quitting smoking, managing obesity, and adopting a balanced diet—forming key components of treatment for hepatic steatosis. Alcohol abstinence is crucial at all stages of ALD, as it can lead to the resolution of simple alcoholic steatosis and improve survival in individuals with cirrhosis or decompensated liver failure. However, preventing relapse remains a major challenge, underscoring the importance of sustained patient motivation and adherence to treatment plans. Recent studies have also explored the role of antioxidants in mitigating oxidative stress and supporting liver health. For example, vitamin E supplementation, examined in both animal models and clinical trials, has shown potential in reducing hepatic lipid accumulation by restoring redox balance, decreasing apoptosis, and limiting oxidative damage.[178] Future research on the molecular mechanisms underlying AFLD should be prioritised, with a focus on several key areas. Firstly, there is a need to identify novel molecular targets and elucidate specific pathways involved in AFLD development, using advanced techniques such as omics approaches. Secondly, molecular insights should be leveraged to develop targeted therapeutic strategies for AFLD treatment, including personalised medicine based on molecular profiling. Longitudinal studies can provide valuable insights into how molecular

signatures evolve throughout the progression of AFLD, thereby enhancing prognosis and monitoring efforts. Emphasising the investigation of molecular mechanisms—through advanced technologies and targeted therapeutic approaches—is essential for a comprehensive understanding of AFLD pathogenesis. Such knowledge is critical for developing effective prevention and treatment strategies for this increasingly prevalent liver disease.

Acknowledgements: We thank our colleagues and the academic staff at the Department of Pharmacology, UKM, for their support and encouragement. This research was funded by the funding from a UKM Research Grant (Grant number: FF-2021-501), co-sponsored by the Malaysian Palm Oil Board, ExcelVite Sdn. Bhd and Hovid Sdn. Bhd.

Informed Consent Statement: Not applicable.

Authors' contributions: N.M.F (Nur Azlina Mohd Fahami): Conceptualisation, Funding acquisition, project administration, supervision, writing – review & editing, final approval of the manuscript; C.K.Y (Chin Kok Yong): Conceptualization, project administration, supervision, writing – review & editing; S.N.A (Siti Norain Azahar): Investigation, project administration, writing—original draft, writing – review & editing; I.A.A.I (Ibrahim Abdel Aziz Ibrahim): Supervision. All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest: The authors declare no conflicts of interest.

REFERENCES

1. World Health Organization. *Global status report on alcohol and health 2011*. Geneva: World Health Organization; 2012. https://www.who.int/publications/i/item/global-status-report-on-alcohol-and-health-2011. Accessed March 6, 2024.
2. Lim SS, Vos T, Flaxman AD, *et al*. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990–2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet. 2012;380(9859):2224–2260.
3. World Health Organization. *Global status report on alcohol and health 2018: executive summary*. Geneva: World Health Organization; 2018:1–16. https://www.who.int/publications/i/item/9789241565639. Accessed March 7, 2024.
4. Anderson P, Baumberg B. Alcohol in Europe – A public health perspective. A report for the European Commission [Internet]. England; 2006. Available from: https://citeseerx.ist.psu.edu/document?repid=rep1&type=pdf&doi=43691c6e67d4fc66963ed928cb0d8509b9cdb4db
5. Babor TF. Alcohol: No ordinary commodity - A summary of the second edition. Addiction. 2010;105(5):769–79.
6. Bruha R, Dvorak K, Petrtyl J. Alcoholic liver disease. World J Hepatol. 2012;4(3):81–90.
7. Ceni E, Mello T, Galli A. Pathogenesis of alcoholic liver disease: Role of oxidative metabolism. World J Gastroenterol. 2014;20(47):17756–72.
8. Singal AK, Bataller R, Ahn J, Kamath PS, Shah VH. ACG clinical guideline: Alcoholic liver disease. Am J Gastroenterol. 2018;113(2):175–94.
9. Jani BD, McQueenie R, Nicholl BI, *et al*. Association between patterns of alcohol consumption (beverage type, frequency and consumption with food) and risk of adverse health outcomes: a prospective cohort study. BMC Med. 2021;19(1):1–14.
10. Arab JP, Roblero JP, Altamirano J, *et al*. Alcohol-related liver disease: clinical practice guidelines by the Latin American Association for the Study of the Liver (ALEH). Ann Hepatol. 2019;18(3):518–535.
11. Frazier TH, Mcclain CJ, Kershner NA, Marsano LS. Treatment of alcoholic liver disease. Therap Adv Gastroenterol. 2011;4(1):63–81.
12. Rehm J, Samokhvalov A V., Shield KD. Global burden of alcoholic liver diseases. J Hepatol. 2013;59(1):160–8.
13. Manthey J, Shield KD, Rylett M, *et al*. Global alcohol exposure between 1990 and 2017 and forecasts until 2030: a modelling study. Lancet. 2019;393(10190):2493–2502.
14. Substance Abuse and Mental Health Services Administration. Key substance use and mental health indicators in the United States: Results from the 2019 National Survey on Drug Use and Health (HHS Publication No. PEP20-07-01-001, NSDUH Series H-55). Rockville, MD; 2020. https://www.samhsa.gov/data/. Accessed March 7, 2024.
15. Canadian Center on Substance Use and Addiction. Alcohol. 2019:1–17. https://www.ccsa.ca/sites/default/files/2019-09/CCSA-Canadian-Drug-Summary-Alcohol-2019-en.pdf. Accessed March 7, 2024
16. Fan JG. Epidemiology of alcoholic and nonalcoholic fatty liver disease in China. J Gastroenterol Hepatol. 2013;28(SUPPL 1):11–7.
17. Institute for Public Health. *National Health and Morbidity Survey 2011: Non-Communicable Diseases*. Vol. 2. Ministry of Health; 2011:188. https://iku.moh.gov.my/images/IKU/Document/REPORT/NHMS2011-VolumeII.pdf. Accessed March 20, 2022.
18. Institute for Public Health. National Health and Morbidity Survey (NHMS) 2019: NCDs - Non-Communicable Diseases: Risk Factors and other Health Problems [Internet]. Vol. 1, Institute for Public Health. 2019 [cited 2022 Jul 17]. p. 1–392. Available from: http://www.iku.gov.my/nhms-2019
19. Mutalip H, Kamarudin R, A. Manickam M, Abd Hamid HA, Saari R. Alcohol Consumption and

Risky Drinking Patterns in Malaysia: Findings from NHMS 2011. Alcohol Alcohol. 2014;49(5):593–9.
20. Rehm J, Mathers C, Popova S, Thavorncharoensap M, Teerawattananon Y, Patra J. Global burden of disease and injury and economic cost attributable to alcohol use and alcohol-use disorders. Lancet. 2009;373(9682):2223–33.
21. Olesen J, Gustavsson A, Svensson M, *et al*. The economic cost of brain disorders in Europe. Eur J Neurol. 2012 Jan 1;19:155–62.
22. Griswold MG, Fullman N, Hawley C, *et al*. Alcohol use and burden for 195 countries and territories, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet. 2018;392(10152):1015–35.
23. Shaheen AA, Kong K, Ma C, *et al*. Impact of the COVID-19 Pandemic on Hospitalisations for Alcoholic Hepatitis or Cirrhosis in Alberta, Canada. Clin Gastroenterol Hepatol. 2022;20(5):e1170–9.
24. Stockwell T, Andreasson S, Cherpitel C, *et al*. The burden of alcohol on health care during COVID-19. Drug Alcohol Rev. 2021;40(1):3–7.
25. Thavorncharoensap M, Teerawattananon Y, Yothasamut J, Lertpitakpong C, Chaikledkaew U. The economic impact of alcohol consumption: a systematic review. Subst Abuse Treat Prev Policy. 2009 Nov;4:20.
26. Laramée P, Kusel J, Leonard S, Aubin HJ, François C, Daeppen JB. The economic burden of alcohol dependence in Europe. Alcohol Alcohol. 2013;48(3):259–69.
27. Bouchery EE, Harwood HJ, Sacks JJ, Simon CJ, Brewer RD. Economic costs of excessive alcohol consumption in the U.S., 2006. Am J Prev Med. 2011 Nov;41(5):516–524.
28. Yahaya A. Effects of Alcohol Abuse on Human Cognition. J Sci Math Educ. 2008.
29. United Nation. Sustainable Development Goals [Internet]. [cited 2020 May 6]. Available from: http://www.un.org/sustainabledevelopment/sustainable-development-goals/
30. Lieber CS. Alcoholic fatty liver: its pathogenesis and mechanism of progression to inflammation and fibrosis. Alcohol. 2004;34(1):9–19.
31. Osna NA, Donohue TM, Kharbanda KK. Alcoholic liver disease: Pathogenesis and current management. Alcohol Res Curr Rev. 2017;38(2):7–21.
32. Ohashi K, Pimienta M, Seki E. Alcoholic liver disease: A current molecular and clinical perspective. Liver Res. 2018;2(4):161–72.
33. Martín-Fernández M, Arroyo V, Carnicero C, *et al*. Role of Oxidative Stress and Lipid Peroxidation in the Pathophysiology of NAFLD. Antioxidants. 2022;11(11).
34. Kang D, Zhao D, Ryu S, *et al*. Perceived stress and non-alcoholic fatty liver disease in apparently healthy men and women. Sci Rep. 2020 Jan;10(1):38.
35. Mellinger JL. Epidemiology of Alcohol Use and Alcoholic Liver Disease. Clin Liver Dis. 2019;13(5):136–9.
36. Sharma P, Arora A. Clinical presentation of alcoholic liver disease and non-alcoholic fatty liver disease: spectrum and diagnosis. Transl Gastroenterol Hepatol. 2020;5:19.
37. Orman ES, Odena G, Bataller R. Alcoholic liver disease: Pathogenesis, management, and novel targets for therapy. J Gastroenterol Hepatol. 2013;28(SUPPL 1):77–84.
38. Li YM, Fan JG, Wang BY, *et al*. Guidelines for the diagnosis and management of alcoholic liver disease: update 2010: (published in Chinese on Chinese Journal of Hepatology 2010; 18: 167-170). J Dig Dis. 2011 Feb;12(1):45–50.
39. Abd El-Kader SM, El-Den Ashmawy EMS. Non-alcoholic fatty liver disease: The diagnosis and management. World J Hepatol. 2015;7(6):846–58.
40. Lívero FAR, Acco A. Molecular basis of alcoholic fatty liver disease: From incidence to treatment. Hepatol Res. 2016;46(1):111–23.
41. European Association for the Study of the Liver. EASL Clinical Practical Guidelines: Management of Alcoholic Liver Disease. J Hepatol. 2012;57(2):399–420.
42. Zamin I, Mattos A, Perin C, Zatti R. A importância do índice AST/ALT no diagnóstico da esteatohepatite não-alcoólica. Arq Gastroenterol. 2002 Mar 1;39.
43. Menon KVN, Gores GJ, Shah VH. Pathogenesis, diagnosis, and treatment of alcoholic liver disease. Mayo Clin Proc. 2001;76(10):1021–9.
44. Kollmann D, Rasoul-Rockenschaub S, Steiner I, Freundorfer E, *et al*. Good outcome after liver transplantation for ALD without a 6 months abstinence rule prior to transplantation including post-transplant CDT monitoring for alcohol relapse assessment - a retrospective study. Transpl Int Off J Eur Soc Organ Transplant. 2016 May;29(5):559–67.
45. Naveau S, Raynard B, Ratziu V, *et al*. Biomarkers for the prediction of liver fibrosis in patients with chronic alcoholic liver disease. Clin Gastroenterol Hepatol Off Clin Pract J Am Gastroenterol Assoc. 2005 Feb;3(2):167–74.
46. Ratziu V, Massard J, Charlotte F, *et al*. Diagnostic value of biochemical markers (FibroTest-FibroSURE) for the prediction of liver fibrosis in patients with non-alcoholic fatty liver disease. BMC Gastroenterol. 2006 Feb;6:6.
47. Hussain S, Ashafaq M, Alshahrani S, *et al*. Cinnamon oil against acetaminophen-induced acute liver toxicity by attenuating inflammation, oxidative stress and apoptosis. Toxicol reports. 2020;7:1296–304.
48. Gala KS, Vatsalya V. Emerging Noninvasive Biomarkers, and Medical Management Strategies for Alcoholic Hepatitis: Present Understanding and Scope. Cells. 2020 Feb;9(3).
49. Au DH, Kivlahan DR, Bryson CL, Blough D, Bradley KA. Alcohol Screening Scores and Risk of Hospitalisations for GI Conditions in Men. Alcohol Clin Exp Res [Internet]. 2007 Mar 1;31(3):443–51.
50. Kulkarni A V., Singal AK. Screening for alcohol use disorder and monitoring for alcohol use in the liver clinic. Clin Liver Dis. 2023;22(6):219–24.
51. Francque S, Vonghia L. Pharmacological Treatment for Non-alcoholic Fatty Liver Disease. Adv Ther. 2019.

52. Bizzaro D, Becchetti C, Trapani S, *et al*. Influence of sex in alcohol-related liver disease: Pre-clinical and clinical settings. United Eur Gastroenterol J. 2023;11(2):218–27.
53. Diab KA, Ibrahim NE, Fahmy MA, Hassan EM, Omara EA. Inhibitory activity of flaxseed oil against CdCl(2) induced liver and kidney damage: Histopathology, genotoxicity, and gene expression study. Toxicol reports. 2020;7:1127–37.
54. Cederbaum AI. Alcohol Metabolism. Encycl Gastroenterol Second Ed. 2019;16(4):47–55.
55. Caballería J. Current concepts in alcohol metabolism. Ann Hepatol. 2003;2(2):60–8.
56. Masson S, Emmerson I, Henderson E, *et al*. Clinical but not histological factors predict long-term prognosis in patients with histologically advanced non-decompensated alcoholic liver disease. Liver Int. 2014;34(2):235–42.
57. Meier P, Seitz HK. Age, alcohol metabolism and liver disease. Curr Opin Clin Nutr Metab Care. 2008;11(1):21–6.
58. Stickel F, Hampe J. Genetic determinants of alcoholic liver disease. Gut. 2012;61(1):150–9.
59. Burza MA, Molinaro A, Attilia ML, *et al*. PNPLA3 I148M (rs738409) genetic variant and age at onset of at-risk alcohol consumption are independent risk factors for alcoholic cirrhosis. Liver Int Off J Int Assoc Study Liver. 2014 Apr;34(4):514–20.
60. Levy R, Catana AM, Durbin-Johnson B, Halsted CH, Medici V. Ethnic differences in presentation and severity of alcoholic liver disease. Alcohol Clin Exp Res. 2015 Mar;39(3):566–74.
61. Ruhl CE, Everhart JE. Joint effects of body weight and alcohol on elevated serum alanine aminotransferase in the United States population. Clin Gastroenterol Hepatol Off Clin Pract J Am Gastroenterol Assoc. 2005 Dec;3(12):1260–8.
62. Kirpich IA, Miller ME, Cave MC, Joshi-Barve S, McClain CJ. Alcoholic liver disease: Update on the role of dietary fat. Biomolecules. 2016;6(1):2–17.
63. Rutledge SM, Asgharpour A. Smoking and liver disease. Gastroenterol Hepatol. 2020;16(12):617–25.
64. Szabo G, Aloman C, Polyak SJ, Weinman SA, Wands J, Zakhari S. Hepatitis C infection and alcohol use: A dangerous mix for the liver and antiviral immunity. Alcohol Clin Exp Res. 2006;30(4):709–19.
65. Pohl K, Moodley P, Dhanda AD. Alcohol's impact on the gut and liver. Nutrients. 2021;13(9).
66. Yan C, Hu W, Tu J, Li J, Liang Q, Han S. Pathogenic mechanisms and regulatory factors involved in alcoholic liver disease. J Transl Med. 2023;21(1):1–27.
67. Wang W, Wang C, Xu H, Gao Y. Aldehyde dehydrogenase, liver disease and cancer. Int J Biol Sci. 2020;16(6):921–34.
68. Deitrich R, Zimatkin S, Pronko S. Oxidation of ethanol in the brain and its consequences. Alcohol Res Heal. 2006;29(4):266–73.
69. Dunn W, Shah VH. Pathogenesis of Alcoholic Liver Disease. Clin Liver Dis. 2016;20(3):445–56.
70. Andrade RJ, Aithal GP, Björnsson ES, *et al*. EASL Clinical Practice Guidelines: Drug-induced liver injury. J Hepatol. 2019 Jun;70(6):1222–61.
71. Jiang Y, Zhang T, Kusumanchi P, Han S, Yang Z, Liangpunsakul S. Alcohol Metabolizing Enzymes, Microsomal Ethanol Oxidizing System, Cytochrome P450 2E1, Catalase, and Aldehyde Dehydrogenase in Alcohol-Associated Liver Disease. Biomedicines. 2020 Mar;8(3).
72. Gao B, Bataller R. Alcoholic liver disease: pathogenesis and new therapeutic targets. Gastroenterology. 2011 Nov;141(5):1572–85.
73. Li HH, Tyburski JB, Wang YW, *et al*. Modulation of fatty acid and bile acid metabolism by peroxisome proliferator-activated receptor α protects against alcoholic liver disease. Alcohol Clin Exp Res. 2014 Jun;38(6):1520–31.
74. Galli A, Pinaire J, Fischer M, Dorris R, Crabb DW. The transcriptional and DNA binding activity of peroxisome proliferator-activated receptor alpha is inhibited by ethanol metabolism. A novel mechanism for the development of ethanol-induced fatty liver. J Biol Chem. 2001 Jan;276(1):68–75.
75. Thomes PG, Osna NA, Davis JS, Donohue TM. Cellular steatosis in ethanol oxidizing-HepG2 cells is partially controlled by the transcription factor, early growth response-1. Int J Biochem Cell Biol. 2013;45(2):454–63.
76. Carrasco MP, Jiménez-López JM, Segovia JL, Marco C. Comparative study of the effects of short- and long-term ethanol treatment and alcohol withdrawal on phospholipid biosynthesis in rat hepatocytes. Comp Biochem Physiol B Biochem Mol Biol. 2002 Mar;131(3):491–7.
77. Wang ZG, Dou XB, Zhou ZX, Song ZY. Adipose tissue-liver axis in alcoholic liver disease. World J Gastrointest Pathophysiol. 2016 Feb;7(1):17–26.
78. Wei X, Shi X, Zhong W, *et al*. Chronic alcohol exposure disturbs lipid homeostasis at the adipose tissue-liver axis in mice: analysis of triacylglycerols using high-resolution mass spectrometry in combination with in vivo metabolite deuterium labeling. PLoS One. 2013;8(2):e55382.
79. Kharbanda KK, McVicker DL, Zetterman RK, Donohue TMJ. Ethanol consumption reduces the proteolytic capacity and protease activities of hepatic lysosomes. Biochim Biophys Acta. 1995 Dec;1245(3):421–9.
80. Zhong Z, Ramshesh VK, Rehman H, *et al*. Acute ethanol causes hepatic mitochondrial depolarization in mice: role of ethanol metabolism. PLoS One. 2014;9(3):e91308.
81. Xu A, Wang Y, Keshaw H, Xu LY, Lam KSL, Cooper GJS. The fat-derived hormone adiponectin alleviates alcoholic and nonalcoholic fatty liver diseases in mice. J Clin Invest. 2003 Jul;112(1):91–100.
82. You M, Crabb DW. Recent advances in alcoholic liver disease II. Minireview: molecular mechanisms of alcoholic fatty liver. Am J Physiol Gastrointest Liver Physiol. 2004 Jul;287(1):G1-6.
83. Kharbanda KK, Todero SL, Ward BW, Cannella JJ 3rd, Tuma DJ. Betaine administration corrects ethanol-induced defective VLDL secretion. Mol Cell Biochem. 2009 Jul;327(1–2):75–8.
84. Shearn CT, Fritz KS, Shearn AH, *et al*. Deletion of GSTA4-4 results in increased mitochondrial post-

translational modification of proteins by reactive aldehydes following chronic ethanol consumption in mice. Redox Biol. 2016;7:68–77.
85. Breitkopf K, Nagy LE, Beier JI, Mueller S, Weng H, Dooley S. Current experimental perspectives on the clinical progression of alcoholic liver disease. Alcohol Clin Exp Res. 2009 Oct;33(10):1647–55.
86. Seth D, Haber PS, Syn WK, Diehl AM, Day CP. Pathogenesis of alcohol-induced liver disease: Classical concepts and recent advances. J Gastroenterol Hepatol. 2011;26(7):1089–105.
87. Seitz HK, Bataller R, Cortez-Pinto H, *et al*. Alcoholic liver disease. Nat Rev Dis Prim. 2018;4(1).
88. Nagata K, Suzuki H, Sakaguchi S. Common pathogenic mechanism in development progression of liver injury caused by non-alcoholic or alcoholic steatohepatitis. J Toxicol Sci. 2007 Dec;32(5):453–68.
89. Halliwell B, Gutteridge JMC. Free Radicals in Biology and Medicine. 3rd ed. Oxford: Oxford University Press; 1999. 1–25 p.
90. Jones DP. Redefining Oxidative Stress. Antioxid Redox Signal. 2006;8(9-10):1865–79.
91. Bailey SM, Mantena SK, Millender-Swain T, *et al*. Ethanol and tobacco smoke increase hepatic steatosis and hypoxia in the hypercholesterolemic apoE−/− mouse: Implications for a "multihit" hypothesis of fatty liver disease. Free Radic Biol Med. 2009;46(7):928–38.
92. Zhao N, Guo FF, Xie KQ, Zeng T. Targeting Nrf-2 is a promising intervention approach for the prevention of ethanol-induced liver disease. Cell Mol Life Sci. 2018 Sep;75(17):3143–57.
93. Han KH, Hashimoto N, Fukushima M. Relationships among alcoholic liver disease, antioxidants, and antioxidant enzymes. World J Gastroenterol. 2016 Jan;22(1):37–49.
94. Pang C, Sheng YC, Jiang P, Wei H, Ji LL.Chlorogenic acid prevents acetaminophen-induced liver injury: the involvement of CYP450 metabolic enzymes and some antioxidant signals. J Zhejiang Univ Sci B. 2015 Jul;16(7):602–610.
95. Chaudhary P, Janmeda P, Docea AO, *et al*. Oxidative stress, free radicals and antioxidants: potential crosstalk in the pathophysiology of human diseases. Front Chem. 2023;11(May):1–24.
96. Lívero FAR, Stolf AM, Dreifuss AA, *et al*. The FXR agonist 6ECDCA reduces hepatic steatosis and oxidative stress induced by ethanol and low-protein diet in mice. Chem Biol Interact. 2014 Jun;217:19–27.
97. Chen LH, Xi S, Cohen DA. Liver antioxidant defenses in mice fed ethanol and the AIN-76A diet. Alcohol. 1995;12(5):453–7.
98. Zhao M, Matter K, Laissue JA, Zimmermann A. Copper/zinc and manganese superoxide dismutases in alcoholic liver disease: immunohistochemical quantitation. Histol Histopathol. 1996 Oct;11(4):899–907.
99. Dong X, Liu H, Chen F, Li D, Zhao Y. MiR-214 promotes the alcohol-induced oxidative stress via down-regulation of glutathione reductase and cytochrome P450 oxidoreductase in liver cells. Alcohol Clin Exp Res. 2014 Jan;38(1):68–77.
100. Wang S, Pacher P, De Lisle RC, Huang H, Ding WX. A Mechanistic Review of Cell Death in Alcohol-Induced Liver Injury. Alcohol Clin Exp Res. 2016;40(6):1215–23.
101. Ni HM, Bhakta A, Wang S, *et al*. Role of hypoxia inducing factor-1b in alcohol-induced autophagy, steatosis and liver injury in mice. PLoS One. 2014;9(12):1–25.
102. Semenza GL. Hypoxia-inducible factor 1: Master regulator of O2 homeostasis. Curr Opin Genet Dev. 1998;8(5):588–94.
103. Rankin EB, Rha J, Selak MA, *et al*. Hypoxia-inducible factor 2 regulates hepatic lipid metabolism. Mol Cell Biol. 2009 Aug;29(16):4527–38.
104. Tsukamoto H, Lu SC. Current concepts in the pathogenesis of alcoholic liver injury. FASEB J. 2001;15(8):1335–49.
105. Purohit V, Bode JC, Bode C, *et al*. Alcohol, intestinal bacterial growth, intestinal permeability to endotoxin, and medical consequences: summary of a symposium. Vol. 42, Alcohol (Fayetteville, N.Y.). United States; 2008. p. 349–61.
106. Groebner JL, Tuma PL. The altered hepatic tubulin code in alcoholic liver disease. Biomolecules. 2015;5(3):2140–59.
107. Szabo G. Gut-liver axis in alcoholic liver disease. Gastroenterology. 2015;148(1):30–6.
108. Ma HY, Xu J, Liu X, *et al*. The Role of IL-17 Signaling in Regulation of the Liver–Brain Axis and Intestinal Permeability in Alcoholic Liver Disease. Curr Pathobiol Rep. 2016;4(1):27–35.
109. Uesugi T, Froh M, Arteel GE, *et al*. Role of lipopolysaccharide-binding protein in early alcohol-induced liver injury in mice. J Immunol. 2002 Mar;168(6):2963–9.
110. Yin M, Bradford BU, Wheeler MD, *et al*. Reduced early alcohol-induced liver injury in CD14-deficient mice. J Immunol. 2001 Apr;166(7):4737–42.
111. Uesugi T, Froh M, Arteel GE, Bradford BU, Thurman RG. Toll-like receptor 4 is involved in the mechanism of early alcohol-induced liver injury in mice. Hepatology. 2001 Jul;34(1):101–8.
112. Abdelmegeed MA, Banerjee A, Jang S, *et al*. CYP2E1 potentiates binge alcohol-induced gut leakiness, steatohepatitis, and apoptosis. Free Radic Biol Med. 2013 Dec;65:1238–45.
113. Huang CK, Yu T, de la Monte SM, Wands JR, Derdak Z, Kim M. Restoration of Wnt/β-catenin signaling attenuates alcoholic liver disease progression in a rat model. J Hepatol. 2015;63(1):191–198.
114. Ibusuki R, Uto H, Oda K, *et al*. Human neutrophil peptide-1 promotes alcohol-induced hepatic fibrosis and hepatocyte apoptosis. PLoS One. 2017;12(4):e0174913.
115. Lee YJ, Shukla SD. Histone H3 phosphorylation at serine 10 and serine 28 is mediated by p38 MAPK in rat hepatocytes exposed to ethanol and acetaldehyde. Eur J Pharmacol. 2007 Nov;573(1–3):29–38.
116. Kong X, Yang Y, Ren L, *et al*. Activation of autophagy attenuates EtOH-LPS-induced hepatic steatosis and injury through MD2 associated TLR4 signaling. Sci Rep. 2017;7(1):9292.
117. Liu J, Zhou J, Wu Z, Wang X, Liu L, Yao C.

Cyanidin 3-O-β-Glucoside Ameliorates Ethanol-Induced Acute Liver Injury by Attenuating Oxidative Stress and Apoptosis: The Role of SIRT1/FOXO1 Signaling. Alcohol Clin Exp Res. 2016 Mar;40(3):457–66.
118. Pani G, Fusco S, Colavitti R, *et al*. Abrogation of hepatocyte apoptosis and early appearance of liver dysplasia in ethanol-fed p53-deficient mice. Biochem Biophys Res Commun. 2004 Dec;325(1):97–100.
119. Chen G, Ke Z, Xu M, *et al*. Autophagy is a protective response to ethanol neurotoxicity. Autophagy. 2012 Nov;8(11):1577–89.
120. Guo X, Cui R, Zhao J, Mo R, Peng L, Yan M. Corosolic acid protects hepatocytes against ethanol-induced damage by modulating mitogen-activated protein kinases and activating autophagy. Eur J Pharmacol. 2016 Nov;791:578–88.
121. Lu Y, Cederbaum AI. Autophagy protects against CYP2E1/Chronic ethanol-induced hepatotoxicity. Biomolecules. 2015;5(4):2659–74.
122. Denaës T, Lodder J, Chobert MN, *et al*. The Cannabinoid Receptor 2 Protects Against Alcoholic Liver Disease Via a Macrophage Autophagy-Dependent Pathway. Sci Rep. 2016 Jun;6:28806.
123. Guo R, Xu X, Babcock SA, Zhang Y, Ren J. Aldehyde dedydrogenase-2 plays a beneficial role in ameliorating chronic alcohol-induced hepatic steatosis and inflammation through regulation of autophagy. J Hepatol. 2015;62(3):647–56.
124. Nepal S, Park PH. Activation of autophagy by globular adiponectin attenuates ethanol-induced apoptosis in HepG2 cells: Involvement of AMPK/FoxO3A axis. Biochim Biophys Acta - Mol Cell Res. 2013;1833(10):2111–25.
125. You Y, Li WZ, Zhang S, *et al*. SNX10 mediates alcohol-induced liver injury and steatosis by regulating the activation of chaperone-mediated autophagy. J Hepatol. 2018 Jul;69(1):129–41.
126. Eslam M, Sanyal AJ, George J. Toward More Accurate Nomenclature for Fatty Liver Diseases. Gastroenterology. 2019;157(3):590–3.
127. Bedogni G, Miglioli L, Masutti F, *et al*. Incidence and natural course of fatty liver in the general population: The dionysos study. Hepatology. 2007;46(5):1387–91.
128. Ekstedt M, Franzén LE, Holmqvist M, *et al*. Alcohol consumption is associated with progression of hepatic fibrosis in non-alcoholic fatty liver disease. Scand J Gastroenterol. 2009;44(3):366–74.
129. Chang Y, Cho YK, Kim Y, *et al*. Nonheavy Drinking and Worsening of Noninvasive Fibrosis Markers in Nonalcoholic Fatty Liver Disease: A Cohort Study. Hepatology. 2019;69(1):64–75.
130. Ajmera V, Belt P, Wilson LA, *et al*. Among Patients With Nonalcoholic Fatty Liver Disease, Modest Alcohol Use Is Associated With Less Improvement in Histologic Steatosis and Steatohepatitis. Clin Gastroenterol Hepatol. 2018;16(9):1511-1520.e5.
131. Verrill C, Markham H, Templeton A, Carr NJ, Sheron N. Alcohol-related cirrhosis - Early abstinence is a key factor in prognosis, even in the most severe cases. Addiction. 2009;104(5):768–74.
132. Yi SW, Choi JS, Yi JJ, Lee Y ho, Han KJ. Risk factors for hepatocellular carcinoma by age, sex, and liver disorder status: A prospective cohort study in Korea. Cancer. 2018;124(13):2748–57.
133. Bellentani S, Saccoccio G, Masutti F, *et al*. Prevalence of and risk factors for hepatic steatosis in northern Italy. Ann Intern Med. 2000;132(2):112–7.
134. Younossi ZM, Stepanova M, Ong J, *et al*. Effects of Alcohol Consumption and Metabolic Syndrome on Mortality in Patients With Nonalcoholic and Alcohol-Related Fatty Liver Disease. Clin Gastroenterol Hepatol. 2019;17(8):1625-1633.e1.
135. Loomba R, Yang HI, Su J, *et al*. Synergism between obesity and alcohol in increasing the risk of hepatocellular carcinoma: A prospective cohort study. Am J Epidemiol. 2013;177(4):333–42.
136. World Health Organization(WHO). Global status report on alcohol and health 2014 [Internet]. 2014 [cited 2022 Jan 2]. p. 1–392. Available from: http://www.who.int/substance_abuse/publications/global_alcohol_report/msbgsruprofiles.pdf
137. Rinella M, Charlton M. The globalization of nonalcoholic fatty liver disease: Prevalence and impact on world health. Hepatology. 2016;64(1):19–22.
138. Seto WK, Yuen MF. Nonalcoholic fatty liver disease in Asia: emerging perspectives. J Gastroenterol. 2017;52(2):164–74.
139. Estes C, Razavi H, Loomba R, Younossi Z, Sanyal AJ. Modeling the epidemic of nonalcoholic fatty liver disease demonstrates an exponential increase in burden of disease. Hepatology. 2018 Jan;67(1):123–33.
140. Malnick SDH, Alin P, Somin M, Neuman MG. Fatty Liver Disease-Alcoholic and Non-Alcoholic: Similar but Different. Int J Mol Sci. 2022;23(24).
141. McCullough AJ. The clinical features, diagnosis and natural history of nonalcoholic fatty liver disease. Clin Liver Dis. 2004;8(3):521–33.
142. Dunn W, Angulo P, Sanderson S, *et al*. Utility of a New Model to Diagnose an Alcohol Basis for Steatohepatitis. Gastroenterology. 2006;131(4):1057–63.
143. Imbert-Bismut F, Naveau S, Morra R, *et al*. The diagnostic value of combining carbohydrate-deficient transferrin, fibrosis, and steatosis biomarkers for the prediction of excessive alcohol consumption. Eur J Gastroenterol Hepatol. 2009;21(1):18–27.
144. Nyblom H, Berggren U, Balldin J, Olsson R. High AST/ALT ratio may indicate advanced alcoholic liver disease rather than heavy drinking. Alcohol Alcohol. 2004;39(4):336–9.
145. Mofrad P, Contos MJ, Haque M, *et al*. Clinical and histologic spectrum of nonalcoholic fatty liver disease associated with normal ALT values. Hepatology. 2003;37(6):1286–92.
146. Kowdley K V., Belt P, Wilson LA, Yeh MM, Neuschwander-Tetri BA, Chalasani N, *et al*. Serum ferritin is an independent predictor of histologic severity and advanced fibrosis in patients with nonalcoholic fatty liver disease. Hepatology. 2012;55(1):77–85.
147. Lafferty H, Stanley AJ, Forrest EH. The management of alcoholic hepatitis: A prospective comparison

of scoring systems. Aliment Pharmacol Ther. 2013;38(6):603–10.
148. Kadian M, Kakkar R, Dhar M, Kaushik RM. Model for end-stage liver disease score versus maddrey discriminant function score in assessing short-term outcome in alcoholic hepatitis. J Gastroenterol Hepatol. 2014;29(3):581–8.
149. Crabb DW, Im GY, Szabo G, Mellinger JL, Lucey MR. Diagnosis and Treatment of Alcohol-Associated Liver Diseases: 2019 Practice Guidance From the American Association for the Study of Liver Diseases. Hepatology. 2020;71(1):306–33.
150. Brunt E. Nonalcoholic steatohepatitis: a proposal for grading and staging the histological lesions. Am J Gastroenterol. 1999;94(9):2467–74.
151. Kleiner DE, Brunt EM, Van Natta M, *et al*. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. Hepatology. 2005;41(6):1313–21.
152. Hjelkrem M, Stauch C, Shaw J, Harrison SA. Validation of the non-alcoholic fatty liver disease activity score. Aliment Pharmacol Ther. 2011;34(2):214–8.
153. Toshikuni N, Tsutsumi M, Arisawa T. Clinical differences between alcoholic liver disease and nonalcoholic fatty liver disease. World J Gastroenterol. 2014;20(26):8393–406.
154. Baršić N, Lerotić I, Smirčić-Duvnjak L, Tomašić V, Duvnjak M. Overview and developments in noninvasive diagnosis of nonalcoholic fatty liver disease. World J Gastroenterol. 2012;18(30):3945–54.
155. Wai CT, Greenson JK, Fontana RJ, Kalbfleisch JD, Marrero JA, Conjeevaram HS, *et al*. A simple noninvasive index can predict both significant fibrosis and cirrhosis in patients with chronic hepatitis C. Hepatology. 2003;38(2):518–26.
156. 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.
157. Imbert-Bismut F, Ratziu V, Pieroni L, Charlotte F, Benhamou Y, Poynard T. Biochemical markers of liver fibrosis in patients with hepatitis C virus infection: a prospective study. Lancet (London, England). 2001 Apr;357(9262):1069–75.
158. Méndez-Sánchez N, Almeda-Valdés P, Uribe M. Alcoholic liver disease. An update. Ann Hepatol. 2005;4(1):32–42. 159. Diehl AM, Day C. Cause, Pathogenesis, and Treatment of Nonalcoholic Steatohepatitis. N Engl J Med. 2017;377(21):2063–72.
160. Jeon S, Carr R. Alcohol effects on hepatic lipid metabolism. J Lipid Res. 2020;61(4):470–9.
161. You M, Matsumoto M, Pacold CM, Cho WK, Crabb DW. The role of AMP-activated protein kinase in the action of ethanol in the liver. Gastroenterology. 2004;127(6):1798–808.
162. You M, Fischer M, Deeg MA, Crabb DW. Ethanol induces fatty acid synthesis pathways by activation of sterol regulatory element-binding protein (SREBP). J Biol Chem. 2002;277(32):29342–7.
163. Mitsuyoshi H, Yasui K, Harano Y, *et al*. Analysis of hepatic genes involved in the metabolism of fatty acids and iron in nonalcoholic fatty liver disease. Hepatol Res. 2009;39(4):366–73.
164. Carr RM, Correnti J. Insulin resistance in clinical and experimental alcoholic liver disease. Ann N Y Acad Sci. 2015;1353(1):1–20.
165. Jou J, Choi SS, Diehl AM. Mechanisms of disease progression in nonalcoholic fatty liver disease. Semin Liver Dis. 2008;28(4):370–9.
166. Friedman SL, Neuschwander-Tetri BA, Rinella M, Sanyal AJ. Mechanisms of NAFLD development and therapeutic strategies. Nat Med. 2018;24(7):908–22.
167. Malhi H, Bronk SF, Werneburg NW, Gores GJ. Free fatty acids induce JNK-dependent hepatocyte lipoapoptosis. J Biol Chem. 2006;281(17):12093–101.
168. Hirsova P, Gores GJ. Death Receptor-Mediated Cell Death and Proinflammatory Signaling in Nonalcoholic Steatohepatitis. CFmgh. 2015;1(1):17–27.
169. Gautheron J, Gores GJ, Rodrigues CMP. Lytic cell death in metabolic liver disease. J Hepatol. 2020;73(2):394–408.
170. Ramaiah SK, Jaeschke H. Role of Neutrophils in the Pathogenesis of Acute Inflammatory Liver Injury. Toxicol Pathol. 2007;35(6):757–66.
171. Seo YS, Kwon JH, Yaqoob U, *et al*. Hmgb1 recruits hepatic stellate cells and liver endothelial cells to sites of ethanol-induced parenchymal cell injury. Am J Physiol - Gastrointest Liver Physiol. 2013;305(11):838–48.
172. Ganz M, Szabo G. Immune and inflammatory pathways in NASH. Hepatol Int. 2013;7(2013):S771–81.
173. Petrasek J, Csak T, Ganz M, Szabo G. Differences in innate immune signaling between alcoholic and non-alcoholic steatohepatitis. J Gastroenterol Hepatol. 2013;28(Suppl.1):93–8.
174. Kessoku T, Kobayashi T, Imajo K, *et al*. Endotoxins and Non-Alcoholic Fatty Liver Disease. Front Endocrinol (Lausanne). 2021;12:770986.
175. Sun Q, Zhang W, Zhong W, Sun X, Zhou Z. Pharmacological inhibition of NOX4 ameliorates alcohol-induced liver injury in mice through improving oxidative stress and mitochondrial function. Biochim Biophys Acta - Gen Subj. 2017;1861(1):2912–21.
176. Masarone M, Rosato V, Dallio M, *et al*. Role of oxidative stress in pathophysiology of nonalcoholic fatty liver disease. Oxid Med Cell Longev. 2018;2018.
177. Gonzalez A, Huerta-Salgado C, Orozco-Aguilar J, *et al*. Role of Oxidative Stress in Hepatic and Extrahepatic Dysfunctions during Nonalcoholic Fatty Liver Disease (NAFLD). Oxid Med Cell Longev. 2020;2020.
178. Kaur J, Shalini S, Bansal MP. Influence of vitamin e on alcohol-induced changes in antioxidant defenses in mice liver. Toxicol Mech Methods. 2010;20(2):82–9.