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Alcoholic liver disease: A current molecular and clinical perspective☆

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Abstract

Heavy alcohol use is the cause of alcoholic liver disease (ALD). The ALD spectrum ranges from alcoholic steatosis to steatohepatitis, fibrosis, and cirrhosis. In Western countries, approximately 50% of cirrhosis-related deaths are due to alcohol use. While alcoholic cirrhosis is no longer considered a completely irreversible condition, no effective anti-fibrotic therapies are currently available. Another significant clinical aspect of ALD is alcoholic hepatitis (AH). AH is an acute inflammatory condition that is often comorbid with cirrhosis, and severe AH has a high mortality rate. Therapeutic options for ALD are limited. The established treatment for AH is corticosteroids, which improve short-term survival but do not affect long-term survival. Liver transplantation is a curative treatment option for alcoholic cirrhosis and AH, but patients must abstain from alcohol use for 6 months to qualify. Additional effective therapies are needed. The molecular mechanisms underlying ALD are complex and have not been fully elucidated. Various molecules, signaling pathways, and crosstalk between multiple hepatic and extrahepatic cells contribute to ALD progression. This review highlights established and emerging concepts in ALD clinicopathology, their underlying molecular mechanisms, and current and future ALD treatment options.

Keywords

Alcoholic liver disease (ALD); Alcoholic hepatitis (AH); Alcoholic cirrhosis; Corticosteroids; Liver transplantation

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Authors' contributions

K. Ohashi, M. Pimienta: writing of the manuscript, contributing equally to this work. E. Seki: writing of the manuscript, critical revision of the manuscript for important intellectual content, and obtained funding.

Conflict of interest

The authors declare that they have no conflict of interest.

1 Introduction

Excessive or chronic alcohol intake causes serious health problems that affect the brain, heart, liver, pancreas, gastrointestinal tract, and immune system. In the United States (US) and Europe, alcohol use disorder (AUD) is the fifth leading cause of death. Worldwide, alcohol use kills 3.3 million people annually, which accounts for 5.9% of all deaths.[1–3] Although low alcohol consumption might have a beneficial effect on ischemic heart disease, alcohol consumption dose-dependently increases the risk of alcoholic liver disease (ALD).[4] In the last two decades, alcohol consumption has decreased slightly in some European countries but increased in China and the US.[5,6] Concomitantly, the prevalence of ALD has increased and is expected to increase further.[7]

ALD is a spectrum of conditions that ranges from alcoholic steatosis to steatohepatitis, fibrosis, and cirrhosis. Up to 50% of cirrhosis-associated deaths are due to alcohol abuse in the US.[8] To date, there are no US Food and Drug Administration (FDA)-approved anti-fibrotic agents for cirrhosis. Cirrhosis treatments rely on supportive care measures, such as ascites control and the treatment of esophageal varices. Liver transplantation is a potential curative treatment, but it is only indicated for end-stage decompensated cirrhosis, and patients must abstain from alcohol use for 6 months prior to transplantation.

Excessive and prolonged alcohol use can also cause a distinct clinical syndrome called alcoholic hepatitis (AH), which produces severe clinical symptoms including signs of liver decompensation (*e.g.*, jaundice, infection, bleeding from esophageal varices, ascites, hepatic encephalopathy). Currently, the primary therapy for AH comprises corticosteroids, but the 6-month-mortality of severe AH is still high (approximately 40%).[9] While liver transplantation is a treatment option, severe AH patients often die before meeting the transplantation criteria. Therefore, only a limited number of severe AH patients can undergo liver transplantation.[10,11] A better under-standing of molecular mechanisms underlying ALD is urgently needed to develop effective therapies. This review highlights the established and emerging concepts in ALD clinicopathology and the associated molecular mechanisms as well as current and future treatment options for ALD.

2. Risk factors for ALD

Chronic alcohol consumption, the consumption of large quantities of alcohol, and specific drinking patterns are associated with progression from steatosis to steatohepatitis, liver fibrosis, and cirrhosis (Fig. 1).[12] Most patients with ALD do not develop cirrhosis even with long-term alcohol use (Fig. 1). Various factors influencing disease progression include gender, ethnicity, genetic variants, viral hepatitis, and obesity.[13]

2.1 Gender and ALD

Women tend to use alcohol less than men; therefore, women have a lower risk for AUD than men.[14] Large national longitudinal surveys found AUD prevalence to be three-fold greater for men than women in the 2001e2002 survey and two-fold greater in the 2012e2013 survey. [15] Despite lower levels of alcohol consumption, women are more susceptible to the hepatotoxic effects of alcohol. Women progress rapidly to fibrosis and cirrhosis compared

with men, and fibrosis persists even after cessation.[16] Women have less gastric alcohol dehydrogenase (ADH) activity than men. The reduced gastric alcohol breakdown in women allows larger amounts of alcohol to enter the bloodstream and increases alcohol bioavailability.[17] This alcohol bioavailability has downstream effects on hormone activity.

The liver is the site of steroid hormone metabolism and a target organ of hormonal actions. Estrogen receptors are expressed in both parenchymal and non-parenchymal cells of the liver. Alcohol consumption increases estrogen receptor expression in human and animal livers.[18] Hormone activity also affects ALD. For example, estrogen treatment increases but ovariectomy reduces alcohol-induced hepatic steatosis. Moreover, estrogen treatment increased and ovariectomy decreased tumor necrosis factor (TNF) α production in Kupffer cells and plasma endotoxin levels in alcoholfed rats.[19] These estrogen-induced changes in portal endotoxin, TNFα, and CD14 levels were diminished by treatment with oral antibiotics,[20] suggesting that estrogen affects Kupffer cell sensitivity and intestinal permeability in ALD. Indeed, treatment of human intestinal cells with estrogen in doses equivalent to those found in women enhanced alcohol-induced apoptosis.[21] These studies show that estrogen enhances the sensitivity of Kupffer cells to alcohol and endotoxin, and increases alcohol-induced gut permeability.

On the other hand, basal levels of hepatoprotective betaine-homocysteine methyltransferase are increased in male mice compared with female mice after ethanol administration.[22,23] The ratio of pro-inflammatory ω−6 and anti-inflammatory ω−3 fatty acids (FAs), which affects ALD development, is also different between genders. This ratio was shifted towards a pro-inflammatory state in female drinkers but not male drinkers. Levels of the anti-inflammatory FAs docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) were higher in male drinkers but not female drinkers.[24] These studies show that differences in hormone activity and levels of hepatoprotective factors between females and males may account for the increase of the susceptibility of females to alcohol-induced liver injury.

2.2. Drinking pattern as a risk for ALD

Recently, there has been the shift in high-risk drinking patterns, such as heavy drinking and binge drinking.[25] Binge drinking, defined by the National Institute on Alcohol Abuse and Alcoholism (NIAAA) as drinking episodes of five or more drinks in men, or four or more drinks in women, is on the rise. A 2010 survey by the Centers for Diseases Control reported that approximately 38 million US adults (1 in 6) engage in binge drinking. Binge drinking is particularly concerning in young adults. Approximately 50% of college students reported engaging in binge drinking.[26] Binge drinking in young adulthood is a risk factor for alcohol abuse and dependence later in life, with consequent risks for developing ALD.[27] Because women are more susceptible to ALD and the consumption gender gap is narrowing, younger women who are more likely to binge drink than drink chronically are particularly vulnerable to the deleterious effects of alcohol. Interestingly, experimental animal models suggest that female hormones may contribute to high levels of binge drinking in female mice.[28] These results are consistent with previous studies showing that depleting circulating female hormones in rodents reduces alcohol intake.[29]

Epidemiological data suggest that binge drinking is partially responsible for increasing rates of cirrhosis and cirrhosis-related death, although this conclusion is controversial.[30] Experimental data has shown intra- and extrahepatic changes that acute alcohol intoxication and repeated binge drinking exacerbate liver injury, such as Kupffer cell activation, increased intestinal permeability, elevated cytokine production, increased oxidative stress, mitochondrial dysfunction, and hepatic apoptosis.[31–34] The studies investigating the pathophysiological effects of binge drinking on the liver have their limitations. Further studies investigating the quality of alcohol consumed per binge and binge frequency are needed to evaluate how extensively this drinking pattern exacerbates liver injury. Table 1 shows the various alcohol contents of different alcoholic beverages, which helps to calculate the consumption of quantities of alcohol by drinking different beverages.

2.3. Genetic variants

Many common diseases have heritable traits that confer protective or susceptibility effects. ALD is a complex disease because both environmental and host factors modify disease progression. For example, Hispanics are more prone to ALD, and twin studies showed that alcoholic cirrhosis prevalence was increased in monozygotic versus dizygotic twins.[35] Few heavy drinkers progress to severe ALD, supporting the hypothesis that genetic background influences the course of the disease. Aldehyde dehydrogenase 2 (ALDH2) is an enzyme that degrades the toxic acetaldehyde resulting from ethanol metabolism. The inactive ALDH2*2 variant (E487K) is associated with an alcohol flush reaction, and approximately 40% of East Asians have this variant.[36] The ALDH2*2 variant promoted chemically-induced hepatocellular carcinoma (HCC) development when knocked in to a mouse model.[36] While several reports have studied the relationship between ALDH2*2 and HCC, the evidence of this variant as an independent risk factor for HCC is weak to date.[37,38]

The patatin-like phospholipase domain-containing protein 3 (PNPLA3) I148M variant, a known risk factor for non-alcoholic steatohepatitis (NASH), is strongly associated with the development of ALD to cirrhosis.[39] A genome-wide association study evaluating two independent cohorts of European descent showed that variants of membrane-bound O-acyltransferase domain-containing 7 (MBOAT7) and transmembrane 6 superfamily member 2 (TM6SF2) are also risk factors for alcohol-related cirrhosis.[38] Unlike the PNPLA3, MBOAT7 and TM6SF2 variants, which increase the risk for alcoholic cirrhosis, a recent study has revealed that a variant of hydroxysteroid 17-beta dehydrogenase 13 (HSD17B13) is associated with reduced alcoholic cirrhosis.[40] All four genes are associated with lipid metabolism, suggesting that molecules produced during lipid metabolism may play a more important role in ALD progression than those produced during alcohol metabolism.

2.4. Obesity

The World Health Organization defines overweight and obesity as having a body mass index (BMI) greater than 25 kg/m^2 and 30 kg/m^2, respectively. Given the rising prevalence of obesity and metabolic syndrome in the US, weight control is among the top public health concerns. The earliest derangement in the ALD spectrum is steatosis, an excessive accumulation of triglycerides in hepatocytes. In fact, up to 90% of alcoholics have histological evidence of fatty liver.[41] The interaction between adipose tissue and alcohol

consumption is complex. Epidemiological data shows strong independent associations between alcohol intake and BMI, with individuals who consume more alcohol having higher BMIs.[42] Results from the Third National Health and Nutrition Examination Survey (NHANES III) showed that ALD patients had an obesity prevalence of 44.5% and increased liver-related mortality.[43] Obesity and high alcohol intake synergistically elevated liver enzymes. This interaction had multiplicative effects, raising serum alanine aminotransferase (ALT) and aspartate transaminase (AST) levels 8.9- and 21-fold, respectively. Obese individuals were more susceptible to alcohol-induced liver injury at lower doses than healthy-weight counterparts.[44] To date, it is unclear if NAFLD is associated with ALD progression because of additive injury or if it intensifies alcohol-mediated hepatotoxicity. Studies investigating the combined effects of alcohol and body fat on extrahepatic mechanisms involved in ALD progression are discussed later in this review.

2.5. Hepatitis C virus (HCV)

An estimated 170 million people are infected with HCV world-wide, and chronic HCV infection is a major cause of chronic liver disease.[45] Alcohol intake negatively modifies the course and outcome of HCV infection. A study of liver biopsies from 1574 HCV patients showed that patients consuming over 50 g of alcohol per day had a 34% increase in the rate of fibrosis progression per year compared with non-drinkers.[46] Another study showed dose-dependent increases in liver injury at even lower consumption levels among patients with HCV. This study showed that as little as 20 g per day in women and 30 g per day in men increased histological activity and fibrosis, illustrating the impact of moderate alcohol intake on liver injury and steatosis.[47] Furthermore, in patients with HCV, alcohol intake increases viremia.[47]

The mechanism underlying the synergistic effect of alcohol and HCV on liver injury remains elusive. However, studies implicated altered immune responses, increased oxidative stress, viral replication, and fatty changes of the liver in this synergistic effect.[48–53] HCV patients who drink alcohol develop HCC 2–3 times more frequently than those who do not drink.[54] Studies have suggested that toll-like receptor 4 (TLR4) is one of the factors implicated in the synergistic effect of alcohol and HCV on hepatic oncogenesis.[55] Despite improvements in available HCV treatments, alcohol consumption still increases mortality in patients with HCV.[56] Among HCV patients who completed anti-HCV interferon therapy, the sustained virologic response (SVR) of those who consumed alcohol was comparable to those who did not drink; however, alcohol use was associated with treatment discontinuation and a subsequent reduction in SVR.[57] The effect of direct acting antivirals on liver disease mediated by HCV and alcohol needs further investigations.

3. Clinicopathology and spectrum of ALD

3.1. Alcoholic fatty liver

As mentioned above, alcoholic liver steatosis is the earliest stage of ALD and is developed in 90% of heavy drinkers. While alcoholic steatosis does not present significant clinical symptoms, patients have a slight elevation in the blood levels of AST, ALT, and gamma-glutamyl transferase as well as an AST/ALT ratio, >2. ALD is often comorbid with

metabolic syndrome, which includes hyperlipidemia, diabetes, hypertension, and obesity. The presence of metabolic syndrome and a prior history of heavy alcohol consumption independently affect ALD progression. Histology of tissues with alcoholic steatosis has numerous large- and small-sized lipid droplets in the hepatocyte cytosol. These changes begin in zone 3 (centrilobular zone) and subsequently extend into zone 2 and zone 1 (periportal zone).[58] These changes can be reversed by 4e6 weeks of abstinence.[59]

3.2. Alcoholic steatohepatitis, fibrosis, and cirrhosis

Approximately 20%e40% of heavy drinkers progress from alcoholic steatosis to steatohepatitis and fibrosis. Alcoholic steatohepatitis and fibrosis are characterized histologically by neutrophil infiltration, hepatocyte ballooning, necrosis, the appearance of Mallory-Denk bodies, cholestatic changes, megamitochondria, and perivenular and pericellular fibrosis (Fig. 1).[60] These pathological changes start in zone 3 due to the higher cytochrome P450 2E1 (CYP2E1) expression compared with other zones and progress towards the portal vein area (zone 1) or neighboring central vein. Patients with alcoholic steatohepatitis can be asymptomatic (sub-clinical alcoholic steatohepatitis) or present with severe clinical symptoms, defined as AH. Among patients with fibrosis, including those who are asymptomatic, 8%–20% will develop cirrhosis.[41] Alcohol abuse is the leading cause of cirrhosis-mediated death in the US (44%–48% of all cirrhosis-mediated deaths), higher even than that caused by HCV.[41] Because direct acting antivirals are highly effective treatments for hepatitis B and C virus, ALD and NAFLD are likely to become the leading indications for liver transplantation in the near future. Alcoholic cirrhosis is a significant risk factor for the development of HCC, which is associated with the consumption of large quantities of alcohol. The 10-year cumulative incidence of HCC ranges from 6.8% to 28.7%.[61–64]

3.3. AH

Consuming large quantities of ethanol (>100 g/day) can cause AH, an acute clinical syndrome of ALD. Patients with severe AH present with severe clinical symptoms, including fever, jaundice, ascites, hepatic encephalopathy, gastrointestinal tract bleeding from esophageal varices and gastro-duodenal ulcers. While AH can develop at any stage of ALD, 40% of alcoholic cirrhosis may develop AH and 80% of severe AH occurs in patients with alcoholic cirrhosis (acute-on-chronic condition) (Fig. 1). The prognosis of these patients is very poor compared with that of AH patients with steatosis alone.[65] The American Association for the Study of Liver Diseases (AASLD) guidelines demonstrated correlations between AH severity and serum bilirubin levels, prothrombin time (PT)/ international normalized ratio (INR), Maddrey's discriminant function (MDF) score, serum creatinine levels, and model for end-stage liver disease (MELD) score. Severe AH is defined by an MDF score >32 or MELD score >18. The 1-month mortality rate of this condition is as high as 30%–50%.[66,67] Of the patients who survive to 6 months, 70% will progress to cirrhosis (Fig. 1).

Several histological features are associated with AH outcomes. Neutrophil accumulation was associated with better outcomes in severe AH patients despite neutrophils playing a prominent role in promoting alcohol-induced liver inflammation.[65] Reduced regenerative response and the presence of proliferating hepatocytes were associated with poorer and

better prognosis, respectively.[68,69] In addition, the presence of proliferative hepatic progenitor cells and ductular reactions were associated with poorer prognosis.[70] A recent study identified 123 genes associated with survival in severe AH patients.[71] Among the 123 dysregulated genes, 51 were associated with patients with severe AH and poor prognosis, and 72 were associated with patients with alcoholic cirrhosis or non-severe AH. This study showed that *lipocalin-2 (LCN2), interleukin 1 receptor like 1 (IL1RL1), C-X-C motif chemokine ligand (CXCL) 1, CXCL2, and keratin 19 (KRT19)* were associated with poorer prognosis, whereas *interleukin (IL)-33* and fi*broblast growth factor (FGF) 21* were associated with better prognosis.[71]

4. Established and emerging molecular mechanisms of ALD

4.1. Oxidative stress in ALD

Hepatocytes are the primary cell type that metabolizes ethanol. Ethanol is primarily metabolized to acetaldehyde by ADH (Fig. 2). Acetaldehyde is then metabolized to non-toxic acetate by cytosolic ALDH1 and mitochondrial ALDH2. When ethanol concentrations are high, CYP2E1, another alcohol-metabolizing enzyme, metabolizes ethanol to acetaldehyde and generates reactive oxygen species (ROS).[72] While both ethanol and acetaldehyde are direct hepatotoxins, excessive ROS production and the subsequent production of inflammatory cytokines can promote alcohol-induced liver injury and inflammation (Fig. 2). Chronic alcohol consumption leads to the upregulation of hepatic CYP2E1 levels, which enhances ROS production.[72] In addition, ethanol and acetaldehyde directly injure hepatocyte mitochondria, upregulating mitochondrial ROS production and further promoting liver injury and inflammation.

Another source of ROS is neutrophil that plays a key role in AH. The presence of neutrophils impacts AH disease severity.[65] Ethanol upregulates intercellular adhesion molecule-1 (ICAM-1) expression on the surface of neutrophils and E-selectin expression on sinu-soidal endothelial cells, enhancing the trafficking of circulating neutrophils to the liver. Additionally, the secretion of chemokines (CXCL1, C-C motif chemokine ligand (CCL2), and CXCL8) produced by Kupffer cells and hepatic stellate cells promote neutrophils migration and infiltration to damaged liver tissues.[73,74] ROS from neutrophils, as well as IL-1β and TNFα from Kupffer cells, promote hepatocyte apoptosis and local inflammation.[75] Thus, ROS produced by excessive alcohol metabolism, damaged mitochondria and neutrophils mediates ethanol-induced liver injury and inflammation.

4.2. The gut-liver axis and hepatic inflammation in ALD

Excessive alcohol consumption can cause bacterial overgrowth and change the composition of the intestinal microbiome (*e.g.* decreased *Lactobacillus* and *Bacteroides*).[76–78] Alcohol abuse also increases intestinal permeability by disrupting intestinal barrier function and tight junction integrity through decreased expression of occludins and zonula occludens. This disruption facilitates the translocation of bacterial products from the intestine to the liver through the portal vein (Fig. 3).[79,80] Bacterial products include lipopolysaccharide (LPS, a.k.a. endotoxin), a Gram-negative bacterial cell-wall component. LPS translocation activates TLR4 in Kupffer cells and hepatic stellate cells, inducing the production of pro-

inflammatory cytokines and mediators (*e.g.*, IL-1, IL-6, TNFα, and ROS) and subsequently promoting liver inflammation and fibrosis.[81] Intestinal fungi also play a role in ALD. Ethanol consumption increased the population of fungi in the intestine and β-D-glucan, a fungal cell wall component, in plasma.[82] Importantly, mice treated with antifungals and those with a knockout of Dectin-1, a pattern recognition receptor for β-D-glucan, had less alcohol-induced steatosis and injury compared with control mice, indicating that intestinal fungi play a detrimental role in ALD development.[82]

Similar to microbe-derived molecules, host-derived alarmins, called damaged-associated molecular patterns (DAMPs), can activate liver-disease-promoting inflammatory signals. In ethanol- and acetaldehyde-damaged hepatocytes, the nuclear protein high mobility group box 1 (HMGB1) is translocated to the cytosol and released into systemic circulation.[83,84] Hepatocyte-specific *HMGB1* knockout mice had reduced alcohol-induced liver injury compared with controls, indicating the detrimental effect of HMGB1 in ALD.[83] Thus, gut-derived pathogen-associated molecular patterns (PAMPs) and damaged-liver-derived DAMPs contribute to ALD progression.

4.3. Altered lipid metabolism in ALD

Alcohol-induced steatosis is characterized by the formation of lipid droplets containing triglyceride and esterified cholesterol in the cytosol of hepatocytes, because of ethanol-induced alteration of hepatic lipid metabolism. Ethanol reduces the activity of adenosine monophosphate-activated kinase (AMPK), peroxisome proliferator-activated receptor (PPAR) a, and sirtuin 1 (SIRT1), which reduces FA β-oxidation (Fig. 2).[85–87] Reduced β-oxidation promotes steatosis. Reductions in AMPK activity increases mammalian target of rapamycin complex 1 (mTORC1) activity, which triggers the transcription and activation of sterol regulatory element-binding protein-1c (SREBP-1c) and PPARg.[88] Reduction of AMPK also directly enhances SREBP-1c by increasing its stability.[86,87] Further, when AMPK is activated, it phosphorylates and inactivates acetyl-Co A carboxylase 1 (ACC1). Thus, ethanol upregulates ACC1 activity through the reduction of AMPK activity.[87] The ethanol-induced reduction in hepatic SIRT1 activity also enhances the transcriptional activity of SREBP-1c.[86,87] The reduced SIRT1 activity by ethanol is associated with reduced DEP domain-containing mTOR-interacting protein (DEPTOR), a negative regulator of mTORC1, which enhances SREBP-1c transcription and cytoplasmic translocation of lipin-1, and inhibits transcriptional activity of PPARα.[89] Together, ethanol exposure reduces AMPK, SIRT1 and PPARα activity and upregulates the expression and activity of SREBP-1c, ACC1, and PPARγ, which promotes lipogenesis.[85,86,89–91] The pivotal role of lipin-1 has been implicated in ALD. Ethanol upregulated hepatic lipin-1 expression but blocked lipin-1 nuclear translocation, which suppresses FA β-oxidation, promoting alcohol-induced fatty liver.[92] Decreased very-low-density lipoproteins (VLDL) secretion is also associated with alcohol-induced steatosis. Microsomal triglyceride transfer protein (MTP) assembles VLDL for the lipid secretion. Hepatic MTP levels were decreased in ethanol-fed animals and the PPARα agonist can increase VLDL secretion by upregulating MTP.[93] Decreased VLDL secretion is also mediated by increased lipin-1 in ALD.[94] In addition, ethanol upregulates lipolysis in peripheral and visceral fat tissues, increasing the overload of circulating FAs in

the liver, which promotes alcoholic steatosis (Fig. 2).[11,95,96] Circulating FAs can also activate TLR4 signaling, promoting liver inflammation (Fig. 3).[97]

Alcohol abuse also impairs lipid-droplet catabolism and lipolysis in hepatocytes. Lipolysis is regulated by cytosolic neutral lipases, such as adipose triglyceride lipase (ATGL), and lipophagy, a specialized form of autophagy associated with lysosomal degradation of lipid droplets (Fig. 2). In hepatocytes, alcohol impairs the b-adrenergic-mediated breakdown of lipid droplets by inhibiting protein-kinase A-mediated phosphorylation of hormone-sensitive lipase and ATGL recruitment to lipid droplets.[98] Autophagy is upregulated by increased AMPK and/or reduced mTORC1 activity. Ethanol exposure decreases AMPK and increases mTORC1 activity, thereby reducing autophagy activity.[93,99] Reduced autophagy could enhance lipid accumulation through impaired lipophagy. Enhanced autophagy by autophagy inducers, rapamycin and carbamazepine, suppressed ethanol-induced hepatic steatosis and injury.[100] Recent studies of alcohol-induced steatosis showed that Rab7 and dynamin 2 (Dyn2) play roles in lipophagy.[101,102] Rab7 is a Rab family guanosine triphosphate-binding protein that mediates the fusion of autophagosomes and lysosomes with lipid droplets. Rab7 activity is reduced in hepatocytes following ethanol exposure.[101] In hepatocytes, Dyn2, a guanosine triphosphatase, is associated with autophagic lysosomal reformation, the terminal step of autophagy. Ethanol impairs Dyn2 activity.[102] These studies implicated that reduced Rab7 and Dyn2 activities by ethanol exposure impair lipophagy, promoting the accumulation of lipid droplets.

With respect to FA b-oxidation, ethanol and acetaldehyde suppress this activity by directly damaging mitochondria. Damaged mitochondria are eliminated via mitophagy. Mitophagy is an autophagy-mediated mitochondrial regulation mechanism that plays a role in maintaining mitochondrial functions, including β-oxidation. Parkin is an E3 ubiquitin ligase that regulates mitophagy through ubiquitination of damaged mitochondrial proteins. Mice deficient in Parkin had increased alcohol-induced liver injury, steatosis, and inflammation compared with controls because mitochondria-mediated β-oxidation was suppressed and ROS production increased.[103] A very recent study demonstrated that chronic ethanol exposure induced the mTORC1 translocation to lysosome, in which mTORC1 inhibited transcription factor EB (TFEB) activity through the phosphorylation of TFEB. TFEB plays a crucial role in lysosomal biogenesis and the induction of autophagy-related gene expression. Additionally, TFEB controls mitochondrial biogenesis and FA β-oxidation through peroxisome proliferator-activated receptor gamma coactivator (PGC)-1α regulation.[99] Thus, the inhibited TFEB activity by ethanol enhances alcohol-mediated steatosis and injury through the inhibition of lysosomal and mitochondrial biogenesis and autophagy. p62/sequestosome 1 (SQSTM1) is an adaptor protein of autophagosome and binds to ubiquitinated damaged proteins.[104] These damaged proteins are degraded through proteasome and/or autolysosome. In ALD, both autophagy and proteasome functions are impaired. p62 is accumulated in hepatocytes.[99] Interestingly, p62 is a component of Mallory-Denk body. The accumulation of p62, ubiquitinated proteins, and cytokeratin 8/18 by impaired autophagy and proteasome is associated with the formation of Mallory-Denk body in ALD.[105] Because p62 can activate mTORC1, accumulated p62 may contribute to hepatic steatosis in ALD.[106] Taken together, ethanol and acetaldehyde promote hepatic lipid accumulation by inhibiting lipid degradation through the suppression of autophagy activity,

mitochondrial and liposomal dysfunctions, impaired FA β-oxidation and lipid secretion, and by increasing lipogenesis in the liver.

4.4. The crosstalk between adipose tissue and the liver

Alcohol consumption promotes adipose-tissue lipolysis via ATGL and inhibits the uptake of circulating free FAs for storage in adipose tissue. The result is an increase in circulating non-esterified FA levels, which increases FA flux to the liver and promotes alcohol-induced fatty liver.[107] Experimental evidence suggests that adipose tissue has roles in regulating immunity and inflammation, and recent data support a role for adipose-tissue dysfunction in ALD pathogenesis. Studies have demonstrated that alcohol mediates oxidative stress, inflammation, and cell death in adipose tissue. ALD severity and adipose-tissue inflammation have been correlated in humans.[44] In rats, chronic ethanol administration increased adipocyte CYP2E1 expression. This increased CYP2E1 induced oxidative stress and adipocyte death, provoking inflammatory responses.[108] Another study showed alcohol-mediated adipocyte death was facilitated by CYP2E1, Bcl-2 homology 3 (BH3)-interacting domain death agonist, and complement component C1Q, causing adipose-tissue inflammation.[109] Alcohol consumption also alters adipokine production. Alcohol abuse increased serum levels of leptin, a pro-inflammatory and pro-fibrogenic adipokine that promotes inflammation in adipose tissue and the liver.[110,111] Adiponectin is an anti-inflammatory adipokine that inhibits TNFα production in Kupffer cells via AMPK.[112] Acute and moderate ethanol consumption increased serum adiponectin levels, whereas chronic alcohol abuse decreased them.[113,114] Together, these data show that FAs, inflammatory cytokines, and adipokines derived from adipose tissue affect ALD development.

4.5. Extracellular vesicles (EVs)

Exosomes are small EVs (50e150 nm) that are shed from most cell types, including hepatocytes, macrophages, and hepatic stellate cells. They contain various macromolecules, including proteins, messenger ribonucleic acids (mRNAs), microRNAs (miRNAs), and other non-coding RNAs.[115] Ethanol exposure increases EV production in hepatocytes. The cargo contained in ethanol-mediated EVs are thought to regulate ALD pathogenesis. Ethanol-mediated EV release was mediated through caspase-3 activation in damaged hepatocytes.[116] Ethanol-induced EVs contained CD40 ligand, which stimulated macrophages and subsequently promoted ALD.[116] EVs derived from damaged hepatocyte also contained mitochondrial deoxyribonucleic acid (DNA) that promoted ALD through activation of TLR9.[117–119] The miRNAs present in EV cargos also contributed to ALD development. One study demonstrated that hepatocyte-derived EVs horizontally transfer miR-122 to monocytes, promoting ALD.[120] ALD-mediated circulating EVs also contain heat shock protein 90 (Hsp90), which enhances monocyte chemotactic protein (MCP-1) production in macrophages and reduces the number of M2 macrophages in ALD.[121] Further, ethanol-exposed human monocytes secrete EVs containing miR-27a that polarize naïve monocytes to M2 macrophages. These M2 polarized macrophages have increased IL-10 and transforming growth factor (TGF)-β production and phagocytic activity.[122] In ALD, EV-mediated cargos released from damaged hepatocytes and monocytes regulate liver inflammation by modulating macrophage activation and polarization.

4.6. Impaired liver regeneration in ALD

Although hepatocytes have the profound capacity to regenerate after liver injury or loss of liver tissues, the regenerative capacity of hepatocytes is significantly impaired in ALD. This was observed in rodent models with chronic ethanol exposure and patients with AH.[68,123] In rodent models, chronic ethanol-feeding impairs regenerative response by lacking an induction of cell cycle genes and altered hepatic miRNA profile after partial hepatectomy.[124] MiR-21 was significantly upregulated in the ALD liver and suppressed regenerative responses after hepatectomy in ethanol-treated rats.[125] In AH patients, p21 and p27, cell cycle inhibitors, were upregulated. p27 upregulation might be induced by miR-34a that is upregulated in AH patients.[126] These factors could contribute to the inhibition of liver regeneration in AH patients. Impaired hepatocyte regeneration is associated with the poor prognosis of AH patients.[68] IL-1 inhibits liver regeneration and is upregulated in ALD.[127] Inhibition of IL-1 signaling by IL-1 receptor antagonist recovered regenerative capacity in ALD.[128] IL-22 has a capacity to promote liver regeneration.[129] IL-22 treatment and IL-1 inhibition might be good therapeutic strategies, not only to inhibit inflammation, but also to promote regeneration in ALD.

4.7. Animal models for preclinical studies of ALD

To elucidate the numerous mechanisms of ALD, we need animal models that mimic the broad spectrum of ALD in humans. Currently, there are several rodent ALD models, with each having different feeding durations and methods as well as the presence or absence of binge ethanol gavage. Different models present with different degrees of hepatocyte injury, fatty changes, inflammatory cell infiltration, and fibrosis. The acute single-binge injection model can be used to study of acute ethanol response or mild steatosis. Experimental conditions for this model are easily performed, but the mice do not develop fibrosis.[130] The most widely used ALD model involves chronically feeding an ethanol-containing Lieber-DeCarli diet to mice for 4e8 weeks. This model produces a mild elevation in serum ALT levels, hepatic fat accumulation, and mild liver inflammation but no fibrosis.[131] A chronic ethanol-containing Lieber-DeCarli diet model modified to include ethanol binges for 3 days results in severe liver damage and some degree of fibrosis; however, this model is associated with high mortality.[127] NIAAA researchers developed a 10 day-chronic ethanol feeding with binge ethanol injection model (a.k.a. NIAAA model or Gao-Binge model) that presents with increased serum ALT levels, steatosis, and neutrophil accumulation, resembling early AH pathophysiology in humans. However, this model again does not develop fibrosis.[74] The experimental conditions of this model are easy to perform, and it is widely used in the basic research field. The Tsukamoto-French intra-gastric ethanol-feeding model is one of the best models of alcoholic steatohepatitis, recapitulating most of the human pathophysiology. The combination of the Tsukamoto-French intra-gastric ethanol-feeding model with weekly ethanol binges and *ad libitum* feeding of a high-fat diet presents with robust neutrophil infiltration and liver fibrosis, which mimic severe AH and alcoholic fibrosis, respectively, as well as steatosis, inflammation, and elevated serum ALT levels. However, the use of this model is limited due to the advanced surgical skills required and the associated animal maintenance.[132]

5. Existing and potential therapies for ALD

5.1. Currently available management for ALD

5.1.1. Abstinence and supportive care—Abstinence is the most common preventative measure for ALD patients. Abstinence can improve liver steatosis, injury, and unfavorable outcomes in patients with early-stage ALD. However, some patients with the progressive ALD can still progress to cirrhosis despite sobriety.[133] There are several FDA-approved medications (*e.g.*, disulfiram, naltrexone) for AUD, but these medications often have hepatoxic properties; therefore, their use is limited for ALD patients.[134] Baclofen and metadoxine can effectively preventing alcohol relapse with fewer hepatotoxic effects; however, these agents are not approved for this indication by the FDA.[134] Because obesity, sarcopenia, and malnutrition are associated with ALD, weight management and nutritional support (*e.g.*, ~2000 kcal with 1.2–1.5 g/kg/d protein and supplementation with amino acids (branched, leucine), zinc, vitamin D, thiamine, folate, cyanocobalamin, and selenium) can improve the course of ALD.[11,135]

5.1.2. Corticosteroids—Corticosteroids have been used to treat patients with AH for several decades. Corticosteroids downregulate TNFα production and upregulate IL-10 production in AH, reducing short-term mortality and incidence of encephalopathy.[136] However, these measures do not improve long-term survival.[66,137] Because corticosteroids can improve short-term survival, it is critical to identify severe AH that responds to corticosteroids early. The Lille model was developed to evaluate the response to corticosteroids in severe AH patients following 7 days of treatment.[138] The Lille model is useful for predicting short-term survival in patients with severe AH. Based on this model, 40% of severe AH patients do not respond to corticosteroids, and their 6-month mortality is approximately 75%.[138] The high mortality may be associated with the increased risk of infection (spontaneous bacterial peritonitis, urinary tract infection, pneumonia) due to corticosteroid use; corticosteroid-treated AH patients with an infection have significantly lower survival compared with those without an infection.[139]

5.1.3. N-acetylcysteine (NAC)—NAC, a glutathione precursor antioxidant, is widely used in clinical settings to treat acetaminophen-induced acute liver failure.[140] Because ROS plays a central role in ALD progression, NAC has been investigated as a treatment for ALD. However, treatment of severe AH with NAC alone did not improve the short-term survival compared with corticosteroids alone. By contrast, combination therapy using NAC and corticosteroids significantly improved 28-day-survival, but there was no observed long-term survival benefit.[141,142]

5.1.4. Pentoxifylline—Pentoxifylline, an antioxidant with an anti-TNFα effect, has been examined in patients with severe AH. Similar to NAC, treatment of severe AH with pentoxifylline alone showed no significant long-term survival benefit compared with corticosteroids. Even when combined with corticosteroids, pentoxifylline did not produce significant survival improvement. Accordingly, pentoxifylline is no longer considered a viable treatment for severe AH.[9]

5.1.5. Anti-TNFα antibodies—TNFα is one of the most critical inflammatory cytokines for ALD development. Anti-TNFα therapy such as infliximab, a chimeric monoclonal anti-TNFα antibody, is commonly used to treat arthritis and inflammatory bowel disease and could have therapeutic properties in ALD. Clinical studies showed that treatment of severe AH patients with anti-TNFα antibody improved disease severity and survival.[143] Furthermore, a randomized controlled pilot study of infliximab plus corticosteroids reported improvements in AH severity at 28 days.[144] However, this combination unexpectedly showed higher incidences of infection and mortality among patients with acute AH.[145]

5.1.6. Liver transplantation—Alcoholic cirrhosis is the second leading indication for liver transplantation, accounting for 25% of all procedures.[146] Liver transplantation is still the best treatment option for patients with severe AH who do not respond to corticosteroids.[147] However, most AH patients cannot apply for liver transplantation because of ethical dilemmas, a high potential for alcohol relapse, and the 6-month-abstinence rule. Recent reports evaluated liver transplantation performed prior to completing the 6 months of abstinence. Early liver transplantation demonstrated a better long-term survival rate (1e3 years) compared with matched AH patients who did not undergo transplantation and a similar survival rate to patients with alcoholic cirrhosis who underwent liver transplantation after 6 months of abstinence.[147–149] Studies did not find differences in alcohol relapse in patients who completed or did not complete the 6 months of abstinence. These studies suggest that clinicians may need to reconsider the selection process for early liver transplantation in severe AH patients.

5.2. Emerging treatment options for ALD

5.2.1. IL-22—IL-22, an IL-10-family cytokine produced by immune cells (*e.g.*, T helper (Th) 17, Th22 cells), has anti-inflammatory and regenerative properties. In animal models of ALD and ALD patients, IL-22 receptor expression is upregulated, but IL-22 expression is unchanged.[150,151] In animal studies, recombinant IL-22 treatment ameliorated alcoholic liver steatosis, injury, and fibrosis by activating signal transducer and activator of transcription 3 (STAT3).[150,151] By contrast, IL-22 has pro-inflammatory effects in patients with hepatitis B virus infections and may promote hepatocarcinogenesis.[152,153] However, because IL-22 promotes liver regeneration in addition to its anti-inflammatory effect,[153] treatment with IL-22 could yield large benefits for severe AH patients. F-652, a recombinant fusion protein containing human IL-22 and human immunoglobulin G2-Fc, is currently being evaluated for use in human AH (ClinicalTrials.gov, NCT02655510).

5.2.2. IL-1 receptor antagonist—IL-1b is initially produced in a pro-form that is processed by the inflammasome complex, consisting of caspase-1, apoptosis-associated speck-like protein containing a caspase recruitment domain (ASC), and nucleotide binding and oligomerization domain-like receptor family pyrin domain-containing 3 (NLRP3). The inflammasome converts it to the active form. IL-1β and inflammasome activation are crucial for ALD development.[127] Anakinra is an IL-1 receptor antagonist that inhibits the binding of active IL-1β to the IL-1 receptor. Anakinra has been shown to have therapeutic effects in an animal model.[127] Treatment of severe AH with anakinra is currently being examined in a

clinical trial. This trial is comparing supplementation with a combination of anakinra, pentoxifylline, and zinc in patients being treated with methylprednisolone or placebo (ClinicalTrials.gov, AH/NCT01809132).

5.2.3. Targeting the gut microbiome—Intestinal dysbiosis and bacterial overgrowth are often seen with ALD and contribute to alcohol-induced liver damage.[154] In mice, gut sterilization using orally administered non-absorbable antibiotics prevented alcohol-induced hepatic steatosis and injury and decreased serum endotoxin levels.[80] Probiotics have been reported to ameliorate ALD in both animal and human studies.[155–157] Treatment with prebiotic fructooligosaccharides or pectin prevented ALD development in mice.[78,158] Clinical trials testing the therapeutic effects of gut sterilization using a combination of vancomycin, gentamycin and meropenem (Clinical.gov, NCT03157388) and the effects of probiotics (*Lactobacillus rhamnosus* GG) for AH patients are currently underway (Clinical.gov, NCT01922895). Notably, fecal microbiota transplantation (FMT) from ALD-resistant mice to ALD-sensitive mice improved ALD.[78] FMT from healthy individuals to patients with ALD potentially could be a novel therapeutic approach. In ALD, levels of saturated long-chain FA (LCFA) were reduced in the intestine, and saturated LCFA metabolism is required for the growth of intestinal *Lactobacillus.*[159] Dietary supplementation of saturated LCFA was shown to improve ALD and gut leakiness in mice, indicating that saturated LCFA could maintain intestinal homeostasis and prevent ALD development.[159]

5.2.4. Farnesoid X receptor (FXR) and FGF15/19—The FXR is a nuclear receptor that regulates bile-acid metabolism by inhibiting hepatic CYP7A1. CYP7A1 regulation occurs directly through FXR activity and indirectly through intestine-derived FGF15/19 signaling. FXR signaling can also regulate lipid and glucose metabolism.[160] In animal models and patients with NASH fibrosis, an FXR agonist improved hepatic steatosis, inflammation, and fibrosis.[161,162] FXR signaling can suppress liver inflammation and cancer, improve intestinal barrier integrity, and promote liver regeneration.[163] A clinical trial evaluating the effect of obeticholic acid, a semi-synthetic bile-acid FXR agonist, in patients with severe AH is underway (Clinical.gov, NCT0239219). In a trial evaluating the use of obeticholic acid for NASH fibrosis, the intervention produced side effects. Given this result, researchers hypothesized that an intestine-restricted approach might reduce unfavorable effects. The intestine-restricted FXR agonist fexaramine mitigated alcohol-induced liver injury without affecting the systemic bile-acid pool in mice.[164] With respect to FGF19, over-expression of the FGF19 variant M52 attenuated alcohol-induced liver injury in mice.[164] These findings suggest that targeting intestinal FXR or FGF15/19 could be safer approaches for treating ALD than targeting systemic FXR.

5.2.5. S-adenosyl methionine (SAMe)—Long-term ethanol consumption decreases hepatic levels of SAMe, a major methyl donor, and its synthesizing enzyme methionine adenosyltransferase (MAT) α1. This reduction affects DNA and histone methylation in hepatocytes.[165] SAMe supplementation has antioxidative effects that maintain mitochondrial function and downregulate TNFα, which produces protective effects in ALD.[166] This result suggests that long-term treatment with SAMe could improve long-term survival or extend

the timing for liver transplantation in patients with alcoholic liver cirrhosis.[167] However, a previous randomized control study did not find SAMe treatment to be effective in patients with ALD.[168] SAMe potentially could be safe agent that can be delivered orally, but more evidence demon-strating the benefit in ALD requires further investigations.[157]

6. Conclusions and future perspectives

Here, we have discussed the established molecular mechanisms and those currently emerging, such as EVs and the crosstalk between liver and adipose tissues, and reviewed potential targets, such as IL-22, IL-1, and FXR signaling, for effective therapies. To develop effective future therapies for ALD, a precise understanding of its molecular mechanisms is required, and translational research using human specimens will be crucial. Testing new therapies also requires the use of consistent animal models. Unfortunately, currently available animal models of ALD do not fully recapitulate all the features of ALD, including AH and alcoholic cirrhosis. Therefore, improved animal models are similarly crucial for the development of effective therapies. It has been several decades since the current therapeutic strategies for ALD have been developed. Although corticosteroids and liver transplantation continue to be the mainstay of therapy, new therapeutic approaches should be considered. Currently, an extracorporeal human-cell-based liver support system is being tested under a clinical trial for alcohol-induced liver decompensation and severe AH (Clinical.gov, NCT02612428). In this trial, improved survival was observed only in patients who had a MELD score <28 and were <46.9 years of age.[169] Although additional prospective, randomized, controlled clinical studies in patients with lower MELD score and age are needed to evaluate the reproducibility of this observation, this approach could have a survival benefit for patients with decompensated ALD who cannot undergo liver transplantation and do not respond to corticosteroids. In the future, the combination of effective anti-inflammatory therapies and liver support systems could improve survival for high-mortality AH and alcoholic cirrhosis. If therapies to enhance liver regeneration could be added to this combination, the survival rate would increase further. There is also evidence for reconsidering the selection process for early transplantation in patients with severe AH because recent studies for of early liver transplantation showed excellent outcomes.[147–149] While there is still a long way to go to fully understand the mechanisms under-lying ALD, these promising results suggest that therapeutic advances are on the horizon.

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References

1. Nutt DJ, Rehm J. Doing it by numbers: A simple approach to reducing the harms of alcohol. J Psychopharmacol 2014;28:3–7. [PubMed: 24399337]
2. Mokdad AH, Marks JS, Stroup DF, Gerberding JL. Actual causes of death in the United States, 2000. JAMA 2004;291:1238–1245. [PubMed: 15010446]
3. World Health Organization. Management of substance abuse team. In: Global Status Report on Alcohol and Health Geneva, Switzerland: World Health Organization; 2014.

4. 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;380:2224–2260. [PubMed: 23245609]
5. Haughwout SP, LaVallee RA, Castle IP. Apparent Per capita alcohol consumption: national, state, and regional trends, 1977–2014. In: Surveillance Report 2016.
6. Jiang H, Room R, Hao W. Alcohol and related health issues in China: Action needed. Lancet Glob Health 2015;3:e190–e191. [PubMed: 25794669]
7. Guirguis J, Chhatwal J, Dasarathy J, et al. Clinical impact of alcohol-related cirrhosis in the next decade: Estimates based on current epidemiological trends in the United States. Alcohol Clin Exp Res 2015;39:2085–2094. [PubMed: 26500036]
8. Rehm J, Samokhvalov AV, Shield KD. Global burden of alcoholic liver diseases. J Hepatol 2013;59:160–168. [PubMed: 23511777]
9. Thursz MR, Richardson P, Allison M, et al. Prednisolone or pentoxifylline for alcoholic hepatitis. N Engl J Med 2015;372:1619–1628. [PubMed: 25901427]
10. Di Martino V, Sheppard F, Vanlemmens C. Early liver transplantation for severe alcoholic hepatitis. N Engl J Med 2012;366:478–479.
11. European Association for the Study of Liver. EASL clinical practical guidelines: Management of alcoholic liver disease. J Hepatol 2012;57:399–420. [PubMed: 22633836]
12. Savolainen V, Perola M, Lalu K, Penttila€ A, Virtanen I, Karhunen PJ. Early perivenular fibrogenesis–precirrhotic lesions among moderate alcohol consumers and chronic alcoholics. J Hepatol 1995;23:524–531. [PubMed: 8583139]
13. Osna NA, Donohue TM Jr, Kharbanda KK. Alcoholic liver disease: Pathogenesis and current management. Alcohol Res 2017;38:147–161. [PubMed: 28988570]
14. Nolen-Hoeksema S Gender differences in risk factors and consequences for alcohol use and problems. Clin Psychol Rev 2004;24:981–1010. [PubMed: 15533281]
15. Grant BF, Chou SP, Saha TD, et al. Prevalence of 12-month alcohol use, high-risk drinking, and DSM-IV alcohol use disorder in the United States, 2001–2002 to 2012–2013: Results from the national epidemiologic survey on alcohol and related conditions. JAMA Psychiatr 2017;74:911–923.
16. Poynard T, Mathurin P, Lai CL, et al. A comparison of fibrosis progression in chronic liver diseases. J Hepatol 2003;38:257–265. [PubMed: 12586290]
17. Seitz HK, Egerer G, Simanowski UA, et al. Human gastric alcohol dehydrogenase activity: Effect of age, sex, and alcoholism. Gut 1993;34:1433–1437. [PubMed: 8244116]
18. Colantoni A, Emanuele MA, Kovacs EJ, Villa E, Van Thiel DH. Hepatic estrogen receptors and alcohol intake. Mol Cell Endocrinol 2002;193:101–104. [PubMed: 12161008]
19. Yin M, Ikejima K, Wheeler MD, et al. Estrogen is involved in early alcohol-induced liver injury in a rat enteral feeding model. Hepatology 2000;31:117–123. [PubMed: 10613736]
20. Enomoto N, Yamashina S, Schemmer P, et al. Estriol sensitizes rat Kupffer cells via gut-derived endotoxin. Am J Physiol 1999;277:G671–G677. [PubMed: 10484393]
21. Asai K, Buurman WA, Reutelingsperger CP, Schutte B, Kaminishi M. Modular effects of estradiol on ethanol-induced apoptosis in human intestinal epithelial cells. Scand J Gastroenterol 2005;40:326–335. [PubMed: 15932173]
22. Donohue TM, Curry-McCoy TV, Nanji AA, et al. Lysosomal leakage and lack of adaptation of hepatoprotective enzyme contribute to enhanced susceptibility to ethanol-induced liver injury in female rats. Alcohol Clin Exp Res 2007;31: 1944–1952. [PubMed: 17850215]
23. Tadic SD, Elm MS, Li HS, et al. Sex differences in hepatic gene expression in a rat model of ethanol-induced liver injury. J Appl Physiol (1985) 2002;93: 1057–1068. [PubMed: 12183503]
24. Vatsalya V, Song M, Schwandt ML, et al. Effects of sex, drinking history, and Omega-3 and Omega-6 fatty acids dysregulation on the onset of liver injury in very heavy drinking alcohol-dependent patients. Alcohol Clin Exp Res 2016;40:2085–2093. [PubMed: 27589090]
25. Kerr WC, Mulia N, Zemore SE. U.S. trends in light, moderate, and heavy drinking episodes from 2000 to 2010. Alcohol Clin Exp Res 2014;38:2496–2501. [PubMed: 25257297]

26. Llerena S, Arias-Loste MT, Puente A, Cabezas J, Crespo J, Fábrega E. Binge drinking: Burden of liver disease and beyond. World J Hepatol 2015;7:2703–2715.
27. Chassin L, Pitts SC, Prost J. Binge drinking trajectories from adolescence to emerging adulthood in a high-risk sample: Predictors and substance abuse outcomes. J Consult Clin Psychol 2002;70:67–78. [PubMed: 11860058]
28. Satta R, Hilderbrand ER, Lasek AW. Ovarian hormones contribute to high levels of binge-like drinking by female mice. Alcohol Clin Exp Res 2018;42: 286–294. [PubMed: 29205408]
29. Ford MM, Eldridge JC, Samson HH. Ethanol consumption in the female Long-Evans rat: A modulatory role of estradiol. Alcohol 2002;26:103–113. [PubMed: 12007585]
30. Stokkeland K, Hilm G, Spak F, Franck J, Hultcrantz R. Different drinking patterns for women and men with alcohol dependence with and without alcoholic cirrhosis. Alcohol Alcohol 2008;43:39–45. [PubMed: 17942440]
31. Carmiel-Haggai M, Cederbaum AI, Nieto N. Binge ethanol exposure increases liver injury in obese rats. Gastroenterology 2003;125:1818–1833. [PubMed: 14724834]
32. Demeilliers C, Maisonneuve C, Grodet A, et al. Impaired adaptive resynthesis and prolonged depletion of hepatic mitochondrial DNA after repeated alcohol binges in mice. Gastroenterology 2002;123:1278–1290. [PubMed: 12360488]
33. Mathurin P, Deng QG, Keshavarzian A, Choudhary S, Holmes EW, Tsukamoto H. Exacerbation of alcoholic liver injury by enteral endotoxin in rats. Hepatology 2000;32:1008–1017. [PubMed: 11050051]
34. Nieto N, Rojkind M. Repeated whiskey binges promote liver injury in rats fed a choline-deficient diet. J Hepatol 2007;46:330–339. [PubMed: 17156887]
35. Stinson FS, Grant BF, Dufour MC. The critical dimension of ethnicity in liver cirrhosis mortality statistics. Alcohol Clin Exp Res 2001;25:1181–1187. [PubMed: 11505049]
36. Jin S, Chen J, Chen L, et al. ALDH2(E487K) mutation increases protein turnover and promotes murine hepatocarcinogenesis. Proc Natl Acad Sci U S A 2015;112:90889093.
37. Chang JS, Hsiao JR, Chen CH. ALDH2 polymorphism and alcohol-related cancers in Asians: A public health perspective. J Biomed Sci 2017;24:19. [PubMed: 28253921]
38. Buch S, Stickel F, Trépo E, et al. A genome-wide association study confirms PNPLA3 and identifies TM6SF2 and MBOAT7 as risk loci for alcohol-related cirrhosis. Nat Genet 2015;47:1443–1448. [PubMed: 26482880]
39. Tian C, Stokowski RP, Kershenobich D, Ballinger DG, Hinds DA. Variant in PNPLA3 is associated with alcoholic liver disease. Nat Genet 2010;42:21–23. [PubMed: 19946271]
40. Abul-Husn NS, Cheng X, Li AH, et al. A protein-truncating HSD17B13 variant and protection from chronic liver disease. N Engl J Med 2018;378:1096–1106. [PubMed: 29562163]
41. Gao B, Bataller R. Alcoholic liver disease: Pathogenesis and new therapeutic targets. Gastroenterology 2011;141:1572–1585. [PubMed: 21920463]
42. Breslow RA, Smothers BA. Drinking patterns and body mass index in never smokers: National Health Interview Survey, 1997–2001. Am J Epidemiol 2005;161:368–376. [PubMed: 15692081]
43. Stepanova M, Rafiq N, Younossi ZM. Components of metabolic syndrome are independent predictors of mortality in patients with chronic liver disease: A population-based study. Gut 2010;59:1410–1415. [PubMed: 20660697]
44. Loomba R, Bettencourt R, Barrett-Connor E. Synergistic association between alcohol intake and body mass index with serum alanine and aspartate aminotransferase levels in older adults: The Rancho Bernardo Study. Aliment Pharmacol Ther 2009;30:1137–1149. [PubMed: 19737152]
45. Konstantinou D, Deutsch M. The spectrum of HBV/HCV coinfection: Epidemiology, clinical characteristics, viralinteractions and management. Ann Gastroenterol 2015;28:221–228. [PubMed: 25830779]
46. Poynard T, Bedossa P, Opolon P. Natural history of liver fibrosis progression in patients with chronic hepatitis C. The OBSVIRC, METAVIR, CLINIVIR, and DOSVIRC groups. Lancet 1997;349:825–832. [PubMed: 9121257]
47. Hézode C, Lonjon I, Roudot-Thoraval F, Pawlotsky JM, Zafrani ES, Dhumeaux D. Impact of moderate alcohol consumption on histological activity and fibrosis in patients with chronic

hepatitis C, and specific influence of steatosis: A prospective study. Aliment Pharmacol Ther 2003;17:1031–1037. [PubMed: 12694085]

48. Aloman C, Gehring S, Wintermeyer P, Kuzushita N, Wands JR. Chronic ethanol consumption impairs cellular immune responses against HCV NS5 protein due to dendritic cell dysfunction. Gastroenterology 2007;132:698–708. [PubMed: 17258730]
49. Bedogni G, Miglioli L, Masutti F, et al. Natural course of chronic HCV and HBV infection and role of alcohol in the general population: The Dionysos Study. Am J Gastroenterol 2008;103:2248–2253. [PubMed: 18637095]
50. Novo-Veleiro I, Alvela-Suarez L, Chamorro AJ, González-Sarmiento R, Laso FJ, Marcos M. Alcoholic liver disease and hepatitis C virus infection. World J Gastroenterol 2016;22:1411–1420. [PubMed: 26819510]
51. Bukong TN, Hou W, Kodys K, Szabo G. Ethanol facilitates hepatitis C virus replication via up-regulation of GW182 and heat shock protein 90 in human hepatoma cells. Hepatology 2013;57:70–80. [PubMed: 22898980]
52. 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;30:709–719. [PubMed: 16573590]
53. Tikhanovich I, Kuravi S, Campbell RV, et al. Regulation of FOXO3 by phosphorylation and methylation in hepatitis C virus infection and alcohol exposure. Hepatology 2014;59:58–70. [PubMed: 23857333]
54. Iida-Ueno A, Enomoto M, Tamori A, Kawada N. Hepatitis B virus infection and alcohol consumption. World J Gastroenterol 2017;23:2651–2659. [PubMed: 28487602]
55. Machida K, Tsukamoto H, Mkrtchyan H, et al. Toll-like receptor 4 mediates synergism between alcohol and HCV in hepatic oncogenesis involving stem cell marker Nanog. Proc Natl Acad Sci U S A 2009;106:1548–1553. [PubMed: 19171902]
56. Chen CM, Yoon YH, Yi HY, Lucas DL. Alcohol and hepatitis C mortality among males and females in the United States: A life table analysis. Alcohol Clin Exp Res 2007;31:285–292. [PubMed: 17250621]
57. Anand BS, Currie S, Dieperink E, et al. Alcohol use and treatment of hepatitis C virus: Results of a national multicenter study. Gastroenterology 2006;130: 1607–1616. [PubMed: 16697724]
58. Yeh MM, Brunt EM. Pathological features of fatty liver disease. Gastroenterology 2014;147:754–764. [PubMed: 25109884]
59. Mendenhall CL. Anabolic steroid therapy as an adjunct to diet in alcoholic hepatic steatosis. Am J Dig Dis 1968;13:783–791. [PubMed: 5672729]
60. Fleming KA, McGee JO. Alcohol induced liver disease. J Clin Pathol 1984;37: 721–733. [PubMed: 6086722]
61. Toshikuni N, Izumi A, Nishino K, et al. Comparison of outcomes between patients with alcoholic cirrhosis and those with hepatitis C virus-related cirrhosis. J Gastroenterol Hepatol 2009;24:1276–1283. [PubMed: 19486451]
62. Jepsen P, Ott P, Andersen PK, Sørensen HT, Vilstrup H. Risk for hepatocellular carcinoma in patients with alcoholic cirrhosis: A Danish nationwide cohort study. Ann Intern Med 2012;156:841–847. W295. [PubMed: 22711076]
63. Mancebo A, González-Diéguez ML, Cadahía V, et al. Annual incidence of hepatocellular carcinoma among patients with alcoholic cirrhosis and identification of risk groups. Clin Gastroenterol Hepatol 2013;11:95–101. [PubMed: 22982095]
64. Lin CW, Lin CC, Mo LR, et al. Heavy alcohol consumption increases the incidence of hepatocellular carcinoma in hepatitis B virus-related cirrhosis. J Hepatol 2013;58:730–735. [PubMed: 23220252]
65. Altamirano J, Miquel R, Katoonizadeh A, et al. A histologic scoring system for prognosis of patients with alcoholic hepatitis. Gastroenterology 2014;146: 1231–1239 (e1-e6). [PubMed: 24440674]
66. O'Shea RS, Dasarathy S, McCullough AJ, et al. Alcoholic liver disease. Hepatology 2010;51:307–328. [PubMed: 20034030]

67. Srikureja W, Kyulo NL, Runyon BA, Hu KQ. MELD score is a better prognostic model than Child-Turcotte-Pugh score or Discriminant Function score in patients with alcoholic hepatitis. J Hepatol 2005;42:700–706. [PubMed: 15826720]
68. Dubuquoy L, Louvet A, Lassailly G, et al. Progenitor cell expansion and impaired hepatocyte regeneration in explanted livers from alcoholic hepatitis. Gut 2015;64:1949–1960. [PubMed: 25731872]
69. Lanthier N, Rubbia-Brandt L, Lin-Marq N, et al. Hepatic cell proliferation plays a pivotal role in the prognosis of alcoholic hepatitis. J Hepatol 2015;63: 609–621. [PubMed: 25872168]
70. Sancho-Bru P, Altamirano J, Rodrigo-Torres D, et al. Liver progenitor cell markers correlate with liver damage and predict short-term mortality in patients with alcoholic hepatitis. Hepatology 2012;55:1931–1941. [PubMed: 22278680]
71. Trépo E, Goossens N, Fujiwara N, et al. Combination of gene expression signature and model for end-stage liver disease score predicts survival of patients with severe alcoholic hepatitis. Gastroenterology 2018;154:965–975. [PubMed: 29158192]
72. Nagy LE, Ding WX, Cresci G, Saikia P, Shah VH. Linking pathogenic mechanisms of alcoholic liver disease with clinical phenotypes. Gastroenterology 2016;150:1756–1768. [PubMed: 26919968]
73. Woodfin A, Voisin MB, Nourshargh S. Recent developments and complexities in neutrophil transmigration. Curr Opin Hematol 2010;17:9–17. [PubMed: 19864945]
74. Bertola A, Park O, Gao B. Chronic plus binge ethanol feeding synergistically induces neutrophil infiltration and liver injury in mice: A critical role for E-selectin. Hepatology 2013;58:1814–1823. [PubMed: 23532958]
75. Xu R, Huang H, Zhang Z, Wang FS. The role of neutrophils in the development of liver diseases. Cell Mol Immunol 2014;11:224–231. [PubMed: 24633014]
76. Hartmann P, Chen WC, Schnabl B. The intestinal microbiome and the leaky gut as therapeutic targets in alcoholic liver disease. Front Physiol 2012;3:402. [PubMed: 23087650]
77. Hartmann P, Seebauer CT, Schnabl B. Alcoholic liver disease: The gut microbiome and liver cross talk. Alcohol Clin Exp Res 2015;39:763–775. [PubMed: 25872593]
78. Ferrere G, Wrzosek L, Cailleux F, et al. Fecal microbiota manipulation prevents dysbiosis and alcohol-induced liver injury in mice. J Hepatol 2017;66: 806–815. [PubMed: 27890791]
79. Keshavarzian A, Fields JZ, Vaeth J, Holmes EW. The differing effects of acute and chronic alcohol on gastric and intestinal permeability. Am J Gastroenterol 1994;89:2205–2211. [PubMed: 7977243]
80. Adachi Y, Moore LE, Bradford BU, Gao W, Thurman RG. Antibiotics prevent liver injury in rats following long-term exposure to ethanol. Gastroenterology 1995;108:218–224. [PubMed: 7806045]
81. Inokuchi S, Tsukamoto H, Park E, Liu ZX, Brenner DA, Seki E. Toll-like receptor 4 mediates alcohol-induced steatohepatitis through bone marrow-derived and endogenous liver cells in mice. Alcohol Clin Exp Res 2011;35:1509–1518. [PubMed: 21463341]
82. Yang AM, Inamine T, Hochrath K, et al. Intestinal fungi contribute to development of alcoholic liver disease. J Clin Invest 2017;127:2829–2841. [PubMed: 28530644]
83. Ge X, Antoine DJ, Lu Y, et al. High mobility group box-1 (HMGB1) participates in the pathogenesis of alcoholic liver disease (ALD). J Biol Chem 2014;289: 22672–22691. [PubMed: 24928512]
84. Laursen TL, Støy S, Deleuran B, Vilstrup H, Grønbaek H, Sandahl TD. The damage-associated molecular pattern HMGB1 is elevated in human alcoholic hepatitis, but does not seem to be a primary driver of inflammation. APMIS 2016;124:741–747. [PubMed: 27357188]
85. Nakajima T, Kamijo Y, Tanaka N, et al. Peroxisome proliferator-activated receptor alpha protects against alcohol-induced liver damage. Hepatology 2004;40:972–980. [PubMed: 15382117]
86. You M, Liang X, Ajmo JM, Ness GC. Involvement of mammalian sirtuin 1 in the action of ethanol in the liver. Am J Physiol Gastrointest Liver Physiol 2008;294: G892–G898. [PubMed: 18239056]
87. Crabb DW, Liangpunsakul S. Alcohol and lipid metabolism. J Gastroenterol Hepatol 2006;3:S56–S60.

88. Zoncu R, Efeyan A, Sabatini DM. mTOR: From growth signal integration to cancer, diabetes and ageing. Nat Rev Mol Cell Biol 2011;12:21–35. [PubMed: 21157483]

89. Chen H, Shen F, Sherban A, et al. DEP domain-containing mTOR-interacting protein suppresses lipogenesis and ameliorates hepatic steatosis and acute-on-chronic liver injury in alcoholic liver disease. Hepatology 2018;68: 496–514. [PubMed: 29457836]

90. 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;277:29342–29347. [PubMed: 12036955]

91. Zhang W, Sun Q, Zhong W, Sun X, Zhou Z. Hepatic peroxisome proliferator-activated receptor gamma signaling contributes to alcohol-induced hepatic steatosis and inflammation in mice. Alcohol Clin Exp Res 2016;40:988–999. [PubMed: 27062444]

92. You M, Jogasuria A, Lee K, et al. Signal transduction mechanisms of alcoholic fatty liver disease: Emerging role of lipin-1. Curr Mol Pharmacol 2017;10: 226–236. [PubMed: 26278388]

93. Sozio M, Crabb DW. Alcohol and lipid metabolism. Am J Physiol Endocrinol Metab 2008;295:E10–E16. [PubMed: 18349117]

94. Bi L, Jiang Z, Zhou J. The role of lipin-1 in the pathogenesis of alcoholic fatty liver. Alcohol Alcohol 2015;50:146–151. [PubMed: 25595739]

95. Ramirez T, Longato L, Dostalek M, Tong M, Wands JR, de la Monte SM. Insulin resistance, ceramide accumulation and endoplasmic reticulum stress in experimental chronic alcohol-induced steatohepatitis. Alcohol Alcohol 2013;48:39–52. [PubMed: 22997409]

96. Zhong W, Zhao Y, Tang Y, et al. Chronic alcohol exposure stimulates adipose tissue lipolysis in mice: Role of reverse triglyceride transport in the pathogenesis of alcoholic steatosis. Am J Pathol 2012;180:998–1007. [PubMed: 22234172]

97. Shi H, Kokoeva MV, Inouye K, Tzameli I, Yin H, Flier JS. TLR4 links innate immunity and fatty acid-induced insulin resistance. J Clin Invest 2006;116: 3015–3025. [PubMed: 17053832]

98. Schott MB, Rasineni K, Weller SG, et al. beta-Adrenergic induction of lipolysis in hepatocytes is inhibited by ethanol exposure. J Biol Chem 2017;292: 11815–11828. [PubMed: 28515323]

99. Chao X, Wang S, Zhao K, et al. Impaired TFEB-mediated lysosome biogenesis and autophagy promote chronic ethanol-induced liver injury and steatosis in mice. Gastroenterology 2018;155:865–879 (e12). [PubMed: 29782848]

100. Lin CW, Zhang H, Li M, et al. Pharmacological promotion of autophagy alleviates steatosis and injury in alcoholic and non-alcoholic fatty liver conditions in mice. J Hepatol 2013;58:993–999. [PubMed: 23339953]

101. Schulze RJ, Drižyte K, Casey CA, McNiven MA. Hepatic lipophagy: New insights into autophagic catabolism of lipid droplets in the liver. Hepatol Commun 2017;1:359–369. [PubMed: 29109982]

102. Rasineni K, Donohue TM Jr, Thomes PG, et al. Ethanol-induced steatosis involves impairment of lipophagy, associated with reduced Dynamin2 activity. Hepatol Commun 2017;1:501–512. [PubMed: 29152606]

103. Williams JA, Ni HM, Ding Y, Ding WX. Parkin regulates mitophagy and mitochondrial function to protect against alcohol-induced liver injury and steatosis in mice. Am J Physiol Gastrointest Liver Physiol 2015;309: G324–G340. [PubMed: 26159696]

104. Manley S, Williams JA, Ding WX. Role of p62/SQSTM1 in liver physiology and pathogenesis. Exp Biol Med (Maywood) 2013;238:525–538. [PubMed: 23856904]

105. Bardag-Gorce F, Francis T, Nan L, et al. Modifications in P62 occur due to proteasome inhibition in alcoholic liver disease. Life Sci 2005;77:2594–2602. [PubMed: 15964033]

106. Taniguchi K, Yamachika S, He F, Karin M. p62/SQSTM1-Dr. Jekyll and Mr. Hyde that prevents oxidative stress but promotes liver cancer. FEBS Lett 2016;590: 2375–2397. [PubMed: 27404485]

107. Parker R, Kim SJ, Gao B. Alcohol, adipose tissue and liver disease: mechanistic links and clinical considerations. Nat Rev Gastroenterol Hepatol 2018;15: 50–59. [PubMed: 28930290]

108. Song Z, Zhou Z, Deaciuc I, Chen T, McClain CJ. Inhibition of adiponectin production by homocysteine: a potential mechanism for alcoholic liver disease. Hepatology 2008;47:867–879. [PubMed: 18167065]

109. Sebastian BM, Roychowdhury S, Tang H, et al. Identification of a cytochrome P4502E1/Bid/C1q-dependent axis mediating inflammation in adipose tissue after chronic ethanol feeding to mice. J Biol Chem 2011;286:35989–35997. [PubMed: 21856753]
110. Ikejima K, Honda H, Yoshikawa M, et al. Leptin augments inflammatory and profibrogenic responses in the murine liver induced by hepatotoxic chemicals. Hepatology 2001;34:288–297. [PubMed: 11481614]
111. Shen J, Sakaida I, Uchida K, Terai S, Okita K. Leptin enhances TNF-alpha production via p38 and JNK MAPK in LPS-stimulated Kupffer cells. Life Sci 2005;77:1502–1515. [PubMed: 15979653]
112. 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;127:1798–1808. [PubMed: 15578517]
113. Naveau S, Perlemuter G, Chaillet M, et al. Serum leptin in patients with alcoholic liver disease. Alcohol Clin Exp Res 2006;30:1422–1428. [PubMed: 16899046]
114. Tang H, Sebastian BM, Axhemi A, et al. Ethanol-induced oxidative stress via the CYP2E1 pathway disrupts adiponectin secretion from adipocytes. Alcohol Clin Exp Res 2012;36:214–222. [PubMed: 21895711]
115. Momen-Heravi F, Saha B, Kodys K, Catalano D, Satishchandran A, Szabo G. Increased number of circulating exosomes and their microRNA cargos are potential novel biomarkers in alcoholic hepatitis. J Transl Med 2015;13:261. [PubMed: 26264599]
116. Verma VK, Li H, Wang R, et al. Alcohol stimulates macrophage activation through caspase-dependent hepatocyte derived release of CD40L containing extracellular vesicles. J Hepatol 2016;64:651–660. [PubMed: 26632633]
117. Cai Y, Xu MJ, Koritzinsky EH, et al. Mitochondrial DNA-enriched microparticles promote acute-on-chronic alcoholic neutrophilia and hepatotoxicity. JCI Insight 2017;2 pii: 92634.
118. He Y, Feng D, Li M, et al. Hepatic mitochondrial DNA/Toll-like receptor 9/ MicroRNA-223 forms a negative feedback loop to limit neutrophil over-activation and acetaminophen hepatotoxicity in mice. Hepatology 2017;66: 220–234. [PubMed: 28295449]
119. Garcia-Martinez I, Santoro N, Chen Y, et al. Hepatocyte mitochondrial DNA drives nonalcoholic steatohepatitis by activation of TLR9. J Clin Invest 2016;126:859–864. [PubMed: 26808498]
120. Momen-Heravi F, Bala S, Kodys K, Szabo G. Exosomes derived from alcohol-treated hepatocytes horizontally transfer liver specific miRNA-122 and sensitize monocytes to LPS. Sci Rep 2015;5:9991. [PubMed: 25973575]
121. Saha B, Momen-Heravi F, Furi I, et al. Extracellular vesicles from mice with alcoholic liver disease carry a distinct protein cargo and induce macrophage activation via Hsp90. Hepatology 2018;67:1986–2000. [PubMed: 29251792]
122. Saha B, Momen-Heravi F, Kodys K, Szabo G. MicroRNA cargo of extracellular vesicles from alcohol-exposed monocytes signals naive monocytes to differentiate into M2 macrophages. J Biol Chem 2016;291:149–159. [PubMed: 26527689]
123. Diehl AM, Thorgeirsson SS, Steer CJ. Ethanol inhibits liver regeneration in rats without reducing transcripts of key protooncogenes. Gastroenterology 1990;99:1105–1112. [PubMed: 2394331]
124. Dippold RP, Vadigepalli R, Gonye GE, Patra B, Hoek JB. Chronic ethanol feeding alters miRNA expression dynamics during liver regeneration. Alcohol Clin Exp Res 2013;37:E59–E69. [PubMed: 22823254]
125. Juskeviciute E, Dippold RP, Antony AN, Swarup A, Vadigepalli R, Hoek JB. Inhibition of miR-21 rescues liver regeneration after partial hepatectomy in ethanol-fed rats. Am J Physiol Gastrointest Liver Physiol 2016;311:G794–G806. [PubMed: 27634014]
126. French SW, Liao G, Li J, et al. What are the mechanisms of regeneration in-hibition in alcoholic hepatitis? Exp Mol Pathol 2016;100:502–505. [PubMed: 27189521]
127. Petrasek J, Bala S, Csak T, et al. IL-1 receptor antagonist ameliorates inflammasome-dependent alcoholic steatohepatitis in mice. J Clin Invest 2012;122:3476–3489. [PubMed: 22945633]
128. Iracheta-Vellve A, Petrasek J, Gyogyosi B, et al. Interleukin-1 inhibition facilitates recovery from liver injury and promotes regeneration of hepatocytes in alcoholic hepatitis in mice. Liver Int 2017;37:968–973. [PubMed: 28345165]

129. Ren X, Hu B, Colletti LM. IL-22 is involved in liver regeneration after hepatectomy. Am J Physiol Gastrointest Liver Physiol 2010;298:G74–G80. [PubMed: 19875704]
130. Shukla SD, Pruett SB, Szabo G, Arteel GE. Binge ethanol and liver: new molecular developments. Alcohol Clin Exp Res 2013;37:550–557. [PubMed: 23347137]
131. Lieber CS, DeCarli LM, Sorrell MF. Experimental methods of ethanol administration. Hepatology 1989;10:501–510. [PubMed: 2673971]
132. Ueno A, Lazaro R, Wang PY, Higashiyama R, Machida K, Tsukamoto H. Mouse intragastric infusion (iG) model. Nat Protoc 2012;7:771–781. [PubMed: 22461066]
133. Sorensen TI, Orholm M, Bentsen KD, Hoybye G, Eghoje K, Christoffersen P. Prospective evaluation of alcohol abuse and alcoholic liver injury in men as predictors of development of cirrhosis. Lancet 1984;2:241–244. [PubMed: 6146805]
134. Addolorato G, Mirijello A, Barrio P, Gual A. Treatment of alcohol use disorders in patients with alcoholic liver disease. J Hepatol 2016;65:618–630. [PubMed: 27155530]
135. Chao A, Waitzberg D, de Jesus RP, et al. Malnutrition and nutritional support in alcoholic liver disease: A review. Curr Gastroenterol Rep 2016;18:65. [PubMed: 27787787]
136. Taïeb J, Mathurin P, Elbim C, et al. Blood neutrophil functions and cytokine release in severe alcoholic hepatitis: effect of corticosteroids. J Hepatol 2000;32:579–586. [PubMed: 10782906]
137. Mathurin P, O'Grady J, Carithers RL, et al. Corticosteroids improve short-term survival in patients with severe alcoholic hepatitis: Meta-analysis of individual patient data. Gut 2011;60:255–260. [PubMed: 20940288]
138. Louvet A, Naveau S, Abdelnour M, et al. The Lille model: A new tool for therapeutic strategy in patients with severe alcoholic hepatitis treated with steroids. Hepatology 2007;45:1348–1354. [PubMed: 17518367]
139. Saberi B, Dadabhai AS, Jang YY, Gurakar A, Mezey E. Current management of alcoholic hepatitis and future therapies. J Clin Transl Hepatol 2016;4:113–122. [PubMed: 27350941]
140. Smilkstein MJ, Bronstein AC, Linden C, Augenstein WL, Kulig KW, Rumack BH. Acetaminophen overdose: A 48-hour intravenous N-acetylcysteine treatment protocol. Ann Emerg Med 1991;20:1058–1063. [PubMed: 1928874]
141. Phillips M, Curtis H, Portmann B, Donaldson N, Bomford A, O'Grady J. Anti-oxidants versus corticosteroids in the treatment of severe alcoholic hepatitis–a randomised clinical trial. J Hepatol 2006;44:784–790. [PubMed: 16469404]
142. Nguyen-Khac E, Thevenot T, Piquet MA, et al. Glucocorticoids plus N-acetylcysteine in severe alcoholic hepatitis. N Engl J Med 2011;365:1781–1789. [PubMed: 22070475]
143. Tilg H, Jalan R, Kaser A, et al. Anti-tumor necrosis factor-alpha monoclonal antibody therapy in severe alcoholic hepatitis. J Hepatol 2003;38:419–425. [PubMed: 12663232]
144. Spahr L, Rubbia-Brandt L, Frossard JL, et al. Combination of steroids with infliximab or placebo in severe alcoholic hepatitis: a randomized controlled pilot study. J Hepatol 2002;37:448–455. [PubMed: 12217597]
145. Naveau S, Chollet-Martin S, Dharancy S, et al. A double-blind randomized controlled trial of infliximab associated with prednisolone in acute alcoholic hepatitis. Hepatology 2004;39:1390–1397. [PubMed: 15122768]
146. Kim WR, Lake JR, Smith JM, et al. OPTN/SRTR 2015 annual data report: Liver. Am J Transplant 2017;1:174–251.
147. Mathurin P, Moreno C, Samuel D, et al. Early liver transplantation for severe alcoholic hepatitis. N Engl J Med 2011;365:1790–1800. [PubMed: 22070476]
148. Weeks SR, Sun Z, McCaul ME, et al. Liver transplantation for severe alcoholic hepatitis, updated lessons from the world's largest series. J Am Coll Surg 2018;226:549–557. [PubMed: 29409981]
149. Lee BP, Mehta N, Platt L, et al. Outcomes of early liver transplantation for patients with severe alcoholic hepatitis. Gastroenterology 2018;155:422–430 (e1). [PubMed: 29655837]
150. Kong X, Feng D, Mathews S, Gao B. Hepatoprotective and anti-fibrotic functions of interleukin-22: Therapeutic potential for the treatment of alcoholic liver disease. J Gastroenterol Hepatol 2013;1:56–60.

151. Ki SH, Park O, Zheng M, et al. Interleukin-22 treatment ameliorates alcoholic liver injury in a murine model of chronic-binge ethanol feeding: Role of signal transducer and activator of transcription 3. Hepatology 2010;52: 1291–1300. [PubMed: 20842630]
152. Zhang Y, Cobleigh MA, Lian JQ, et al. A proinflammatory role for interleukin-22 in the immune response to hepatitis B virus. Gastroenterology 2011;141: 1897–1906. [PubMed: 21708106]
153. Park O, Wang H, Weng H, et al. In vivo consequences of liver-specific interleukin-22 expression in mice: Implications for human liver disease progression. Hepatology 2011;54:252–261. [PubMed: 21465510]
154. Schnabl B, Brenner DA. Interactions between the intestinal microbiome and liver diseases. Gastroenterology 2014;146:1513–1524. [PubMed: 24440671]
155. Forsyth CB, Farhadi A, Jakate SM, Tang Y, Shaikh M, Keshavarzian A. Lactobacillus GG treatment ameliorates alcohol-induced intestinal oxidative stress, gut leakiness, and liver injury in a rat model of alcoholic steatohepatitis. Alcohol 2009;43:163–172. [PubMed: 19251117]
156. Kirpich IA, Solovieva NV, Leikhter SN, et al. Probiotics restore bowel flora and improve liver enzymes in human alcohol-induced liver injury: A pilot study. Alcohol 2008;42:675–682. [PubMed: 19038698]
157. Guo T, Chang L, Xiao Y, Liu Q. S-adenosyl-L-methionine for the treatment of chronic liver disease: A systematic review and meta-analysis. PLoS One 2015;10:e0122124. [PubMed: 25774783]
158. Yan AW, Fouts DE, Brandl J, et al. Enteric dysbiosis associated with a mouse model of alcoholic liver disease. Hepatology 2011;53:96–105. [PubMed: 21254165]
159. Chen P, Torralba M, Tan J, et al. Supplementation of saturated long-chain fatty acids maintains intestinal eubiosis and reduces ethanol-induced liver injury in mice. Gastroenterology 2015;148:203–214 (e16). [PubMed: 25239591]
160. Thomas C, Pellicciari R, Pruzanski M, Auwerx J, Schoonjans K. Targeting bile-acid signalling for metabolic diseases. Nat Rev Drug Discov 2008;7:678–693. [PubMed: 18670431]
161. Mudaliar S, Henry RR, Sanyal AJ, et al. Efficacy and safety of the farnesoid X receptor agonist obeticholic acid in patients with type 2 diabetes and nonalcoholic fatty liver disease. Gastroenterology 2013;145:574–582 (e1). [PubMed: 23727264]
162. Neuschwander-Tetri BA, Loomba R, Sanyal AJ, et al. Farnesoid X nuclear receptor ligand obeticholic acid for non-cirrhotic, non-alcoholic steatohepatitis (FLINT): A multicentre, randomised, placebo-controlled trial. Lancet 2015;385:956–965. [PubMed: 25468160]
163. Gadaleta RM, van Mil SW, Oldenburg B, Siersema PD, Klomp LW, van Erpecum KJ. Bile acids and their nuclear receptor FXR: Relevance for hepatobiliary and gastrointestinal disease. Biochim Biophys Acta 2010;1801: 683–692. [PubMed: 20399894]
164. Hartmann P, Hochrath K, Horvath A, et al. Modulation of the intestinal bile acid/farnesoid X receptor/fibroblast growth factor 15 axis improves alcoholic liver disease in mice. Hepatology 2018;67:2150–2166. [PubMed: 29159825]
165. Esfandiari F, Medici V, Wong DH, et al. Epigenetic regulation of hepatic endoplasmic reticulum stress pathways in the ethanol-fed cystathionine beta synthase-deficient mouse. Hepatology 2010;51:932–941. [PubMed: 19957376]
166. Lu SC, Martinez-Chantar ML, Mato JM. Methionine adenosyltransferase and S-adenosylmethionine in alcoholic liver disease. J Gastroenterol Hepatol 2006;3: S61–S64.
167. Mato JM, Cámara J, Fernandez de Paz J, et al. S-adenosylmethionine in alcoholic liver cirrhosis: A randomized, placebo-controlled, double-blind, multi-center clinical trial. J Hepatol 1999;30:1081–1089. [PubMed: 10406187]
168. Medici V, Virata MC, Peerson JM, et al. S-adenosyl-L-methionine treatment for alcoholic liver disease: A double-blinded, randomized, placebo-controlled trial. Alcohol Clin Exp Res 2011;35:1960–1965. [PubMed: 22044287]
169. Thompson J, Jones N, Al-Khafaji A, et al. Extracorporeal cellular therapy (ELAD) in severe alcoholic hepatitis: A multinational, prospective, controlled, randomized trial. Liver Transpl 2018;24:380–393. [PubMed: 29171941]

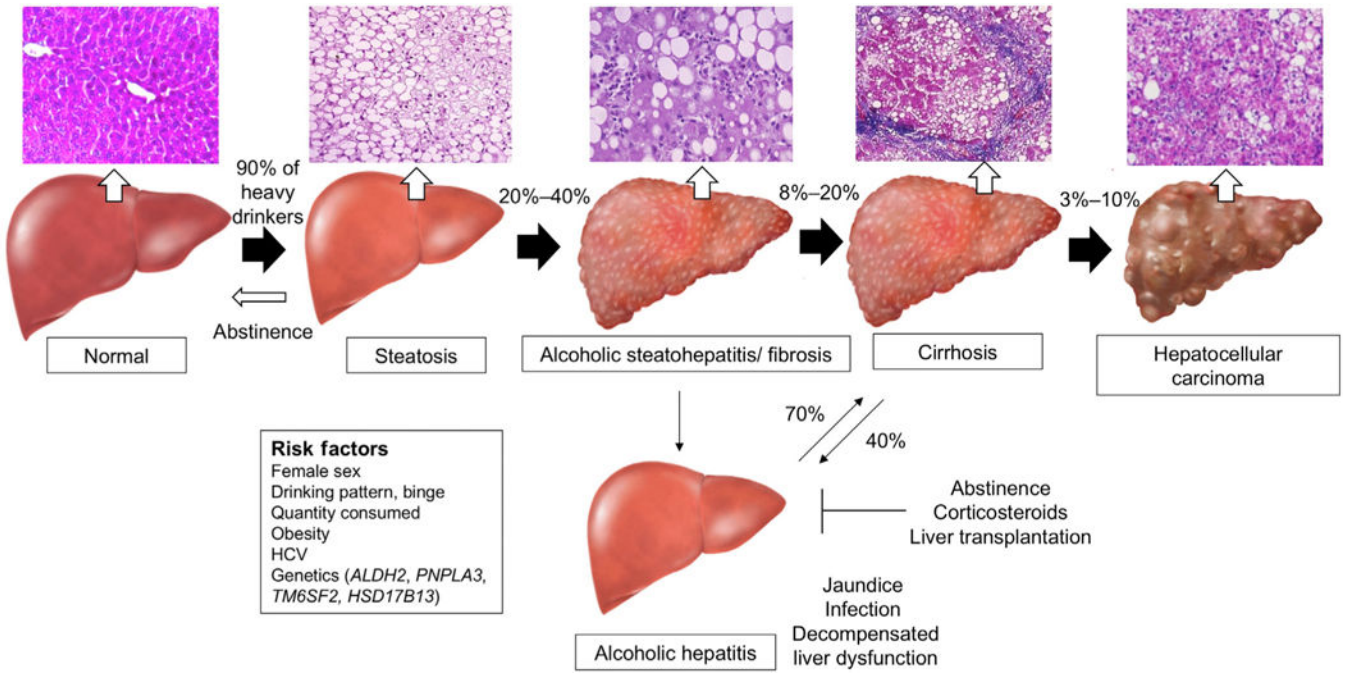

Fig. 1. The progression of ALD.

The spectrum of ALD ranges from steatosis to fibrosis, cirrhosis, and then hepatocellular carcinoma (HCC). Approximately 90% of heavy drinkers develop alcoholic steatosis. This stage is reversible when alcohol use ceases. Risk factors, such as gender, drinking pattern, obesity, viral hepatitis, and genetics, can contribute to ALD progression. About 20%–40% of patients with alcoholic steatosis will progress to alcoholic steatohepatitis, which is histologically characterized by the infiltration of inflammatory cells, especially neutrophils, the appearance of Mallory-Denk bodies, ballooning degeneration, and hepatocyte death in the liver parenchyma. Some of those patients will develop liver fibrosis and subsequently cirrhosis. Fibrosis begins at perivenular region (zone 3) and extends to the neighboring central or portal areas (bridging fibrosis). The surface of cirrhotic liver is irregular. Cirrhosis may further progress to HCC. AH, an acute-on-chronic condition of ALD, presents with clinical symptoms, such as jaundice, infection, and decompensation. AH can occur at any stage of ALD. Treatments for AH include abstinence and corticosteroids, but they are not always effective. However, liver transplantation can be a curative therapy. Abbreviations: ALD, alcoholic liver disease; HCV, hepatitis C virus; AH, alcoholic hepatitis; ALDH2, aldehyde dehydrogenase 2; PNPLA3, patatin-like phospholipase domain-containing protein 3; TM6SF2, transmembrane 6 superfamily member 2; HSD17B13, hydroxysteroid 17-beta dehydrogenase 13.

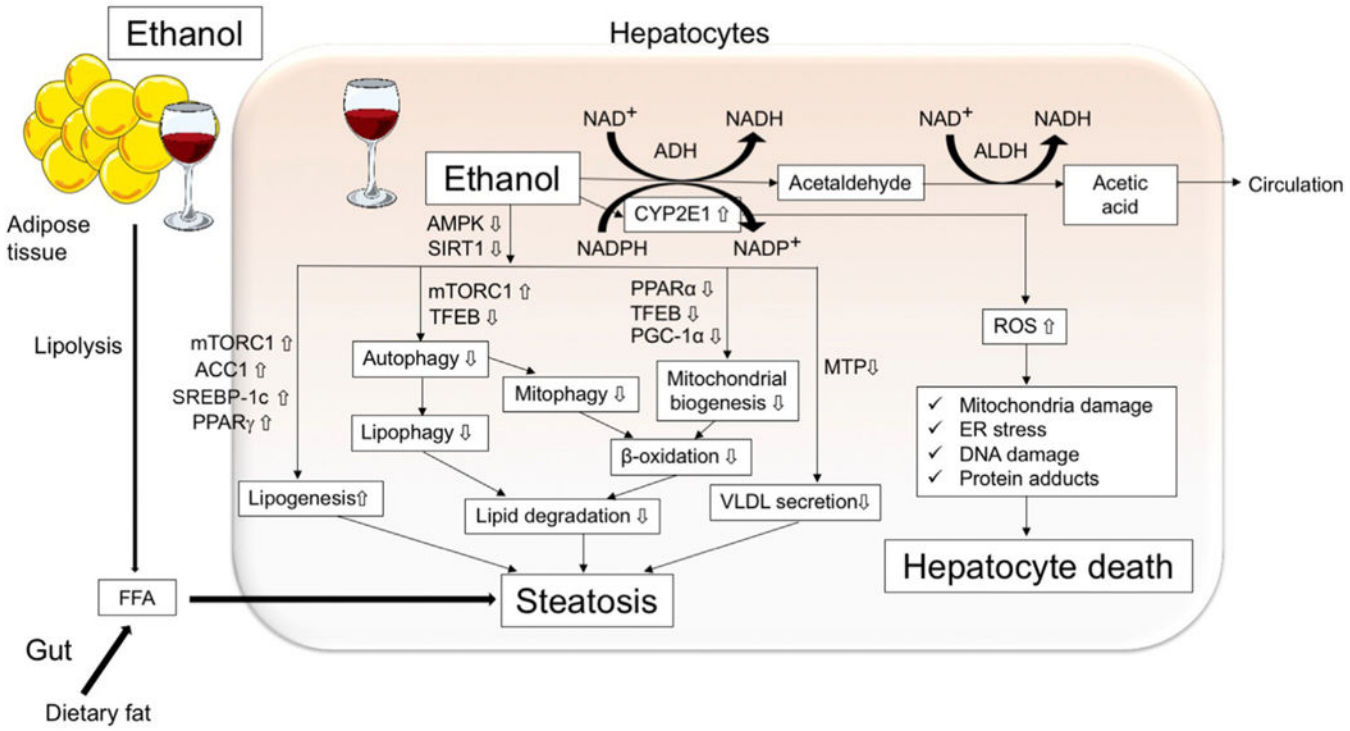

Fig. 2. Ethanol consumption increases hepatic steatosis.

In hepatocytes, ADH oxidizes ethanol to acetaldehyde and converts NAD^+ to NADH. Acetaldehyde entering the mitochondria is converted to acetate and NADH by ALDH through the reduction of NAD^+. Ethanol is also degraded by CYP2E1 through the conversion of NADPH to $NADP^+$. CYP2E1 upregulates ROS production, leading to mitochondria damage, ER stress, DNA damage and the production of protein adducts, resulting in apoptosis. Ethanol reduces AMPK levels, which increases ACC1 activity, decreases PPARa levels, and increases mTORC1 activity. Increased mTORC1 further increases SREBP-1c activity and decreases autophagy. These signaling pathways lead to increased fatty acid synthesis, decreased fatty acid b oxidation, and lipophagy as well as the induction of steatosis. Ethanol also impairs VLDL secretion by inhibiting MTP. Ethanol promotes lipolysis in adipose tissues, resulting in FFA flux to the liver. Excessive intake of dietary fat also promotes alcohol-induced steatosis. Abbreviations: ADH, alcohol dehydrogenase; ALDH, aldehyde dehydrogenase; NAD^+, nicotinamide adenine dinucleotide; NADH, reduced nicotinamide adenine dinucleotide; NADPH, reduced nicotinamide adenine dinucleotide phosphate; $NADP^+$, nicotinamide adenine dinucleotide phosphate; CYP2E1, cytochrome P450 2E1; ROS, reactive oxygen species; ER, endoplasmic reticulum; AMPK, adenosine monophosphate-activated protein kinase; ACC1, acetyl-Co A carboxylase 1; PPAR, peroxisome proliferator-activated receptor; mTORC1, mammalian target of rapamycin complex 1; SREBP-1c, sterol regulatory element-binding protein-1c; VLDL, very-low-density lipoproteins; MTP, microsomal triglyceride transfer protein; FFA, free fatty acid; PGC, peroxisome proliferator-activated receptor gamma coactivator; SIRT1, sirtuin 1; TFEB, transcription factor EB; DNA, deoxyribonucleic acid.

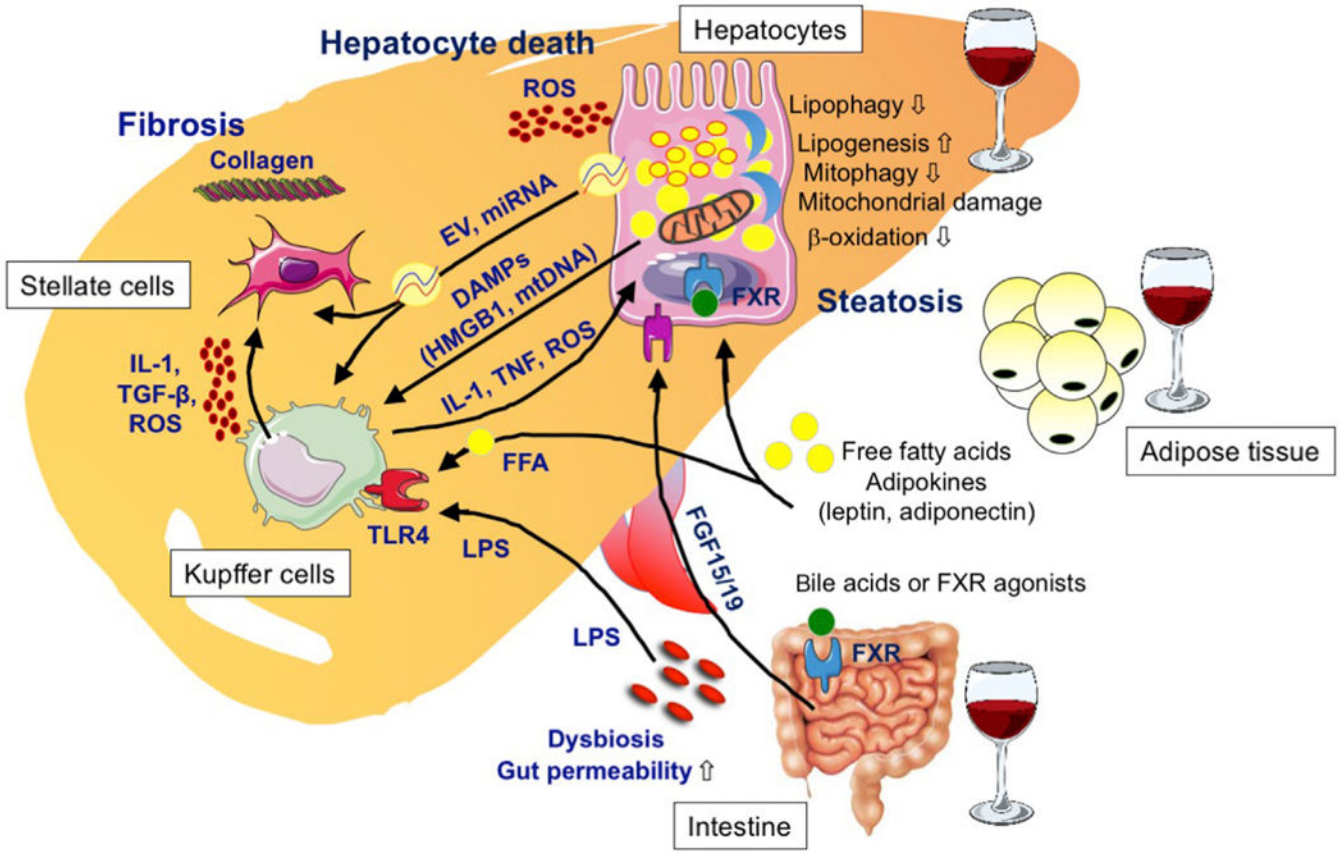

Fig. 3. Gut-adipose tissue-liver network in ALD.

Excessive alcohol consumption can affect the composition of intestinal microbiota and increase intestinal permeability by disrupting intestinal epithelial barrier functions. Intestine-derived PAMPs, such as LPS, translocates to the liver via portal veins. In the liver, translocated LPS binds TLR4 to stimulate neutrophils. Kupffer cells and HSCs produce ROS and pro-inflammatory cytokines, such as TNFα, IL-1, and chemokines, leading to hepatocyte damage and liver inflammation. Chronic LPS stimulation facilitates liver fibrosis by causing Kupffer cells and HSCs to downregulate MMPs and produce extracellular matrix, including collagen. Ethanol and acetaldehyde can damage hepatocytes, leading to release of DAMPs, such as HMGB1, and EVs that contain mitochondrial DNA. Ethanol can promote lipogenesis and inhibit lipid degradation by suppressing β-oxidation and autophagy. Hepatic FXR and intestinal FXR that induces FGF15/19 production regulate bile acid and lipid homeostasis in the liver. Ethanol induces lipolysis and adipokine production in adipose tissues. Fatty acids released from adipocytes promote hepatic steatosis. Adipose-tissue-derived free fatty acids also activate Toll-like receptor 4 (TLR4) signaling. Abbreviations: PAMPs, pathogen-associated molecular patterns; LPS, lipopolysaccharide; HSCs, hepatic stellate cells; MMPs, matrix metalloproteinases; DAMPs, damaged-associated molecular patterns; EVs, extracellular vesicles; FGF, fibroblast growth factor; FXR, farnesoid X receptor; HMGB1, high mobility group box 1; IL, interleukin; miRNA, microRNA; mtDNA, mitochondrial DNA; FFA, free fatty acid; ROS, reactive oxygen species; TGF, transforming

growth factor; TLR, toll-like receptor; TNF, tumor necrosis factor; DNA, deoxyribonucleic acid.

Table 1

Alcohol contents of various alcoholic beverages.

Beverage type	Serving size (fl oz)	ABV (%)	Energy (Cal)
Beer			
Light	12	5	103
Regular			153
Malt	8–9	7	93
Table wine	5	12	121–125
Champagne	4	12	84
Sake	3.5–4.0	16	140
Fortified wine	3–4	17	Varies
Cordial, Liqueur, Aperitif	2–3	24	Varies
Distilled spirits			
Vodka, Rum, Tequila, Gin, Cognac, Brandy	1.5	40	97–98

Abbreviations: fl oz, fluid ounce; ABV, alcohol by volume.