Review Article

THAI J GASTROENTEROL 2013
Vol. 14 No. 1
Jan. - Apr. 2013

Effects of Alcohol Consumption on Alcoholic Liver Disease

Leelanuntakul W
Werawatganon D

ABSTRACT

More than 95% of heavy drinkers develop fatty liver, but only up to 35% of this population develops more severe forms of alcoholic liver disease (ALD), including fibrosis, alcoholic hepatitis, cirrhosis, and HCC. Many risk factors have been proposed for the severe forms of ALD. Alcohol consumption and comorbid factors act in synergy to accelerate the progression of ALD. Alcohol consumption can directly (via acetaldehyde) or indirectly (via regulation of multiple factors) up-regulate the expression of SREBP-1c and down-regulate the expression of PPARα, leading to the induction of fatty acid synthesis and inhibition of fatty liver β-oxidation, which results in the development of alcoholic fatty liver. Alcohol consumption can also modify many factors, including HIF-1, C3, C1qa, PKC□, and iNOS, that subsequently contribute to the development of liver injury. The mechanisms underlying the effects of these factors remain unclear necessary for further investigation.

Key words : Alcoholic liver disease, alcoholic hepatitis, alcoholic fatty liver

[Thai J Gastroenterol 2013; 14(1):43-48.]

Alcoholic liver disease (ALD) presents as a broad spectrum of disorders, ranging from simple fatty liver to more severe forms of liver injury, including alcoholic hepatitis (AH), cirrhosis, and superimposed hepatocellular carcinoma (HCC)[1]. Fatty liver, an early response to alcohol consumption, develops in most (more than 90%) heavy drinkers, with early-mild steatosis in zone 3 (perivenular) hepatocytes; it can also affect zone 2 and even zone 1 (periportal) hepatocytes when liver injury is more severe. Interestingly, only about 30% of heavy drinkers develop more severe forms of ALD, such as advanced fibrosis and cirrhosis. In patients with underlying ALD and heavy alcohol intake, episodes of superimposed AH may occur. In severe cases and in patients with liver cirrhosis, AH leads to severe complications related to liver failure and portal hypertension and has high short-term mortality[1-3].

The fact that only about 35% of heavy drinkers develop advanced ALD indicates that other factors are involved. Several risk factors for ALD have been identified. These include sex, obesity, drinking patterns, dietary factors, non-sex-linked genetic factors, and cigarette smoking (Figure 1)[1-3]. Female sex is a well-documented risk factor for susceptibility to ALD; the

Department of Physiology, Faculty of Medicine, Chulalongkorn University, Bangkok 10330, Thailand.
Address for Correspondence: *Assoc. Prof. Duangporn Werawatganon, M.D. Department of Physiology, Faculty of Medicine, Chulalongkorn University, Bangkok 10330, Thailand.*

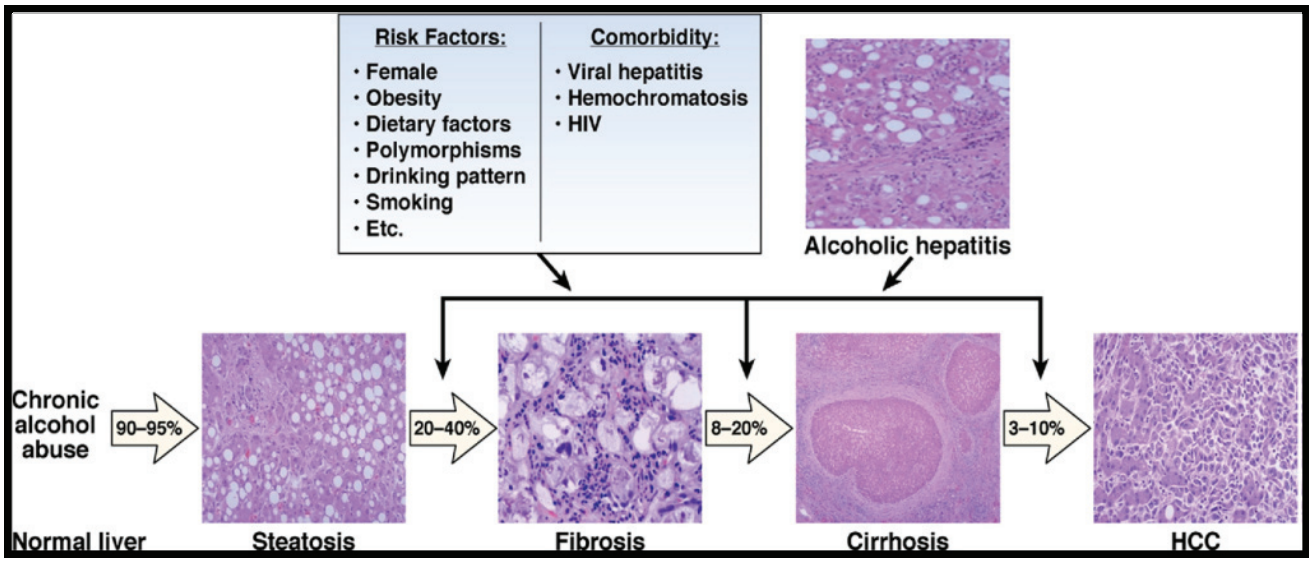

Figure 1. Spectrum of ALD, risk factors, and comorbidity[1].

increased risk among women likely results from lower levels of gastric alcohol dehydrogenase, a higher proportion of body fat, and the presence of estrogens. Obesity represents another important risk factor that accelerates fibrosis progression and the development of cirrhosis in ALD[4,5]. Experimental studies indicate that the synergistic effects of obesity and alcohol abuse involve the endoplasmic reticulum response to cell stress, type I macrophage activation, and adiponectin resistance[6]. Daily or near-daily heavy drinking,begun at an early age, increases the risk of the development of severe forms of ALD compared with episodic or binge drinking[7].

Genetic factors might also influence susceptibility to advanced ALD, but little data are available. Variations in genes that encode antioxidant enzymes, cytokines and other inflammatory mediators, and alcohol- metabolizing enzymes could have a role[3]. Also, recent studies indicate that variations in *patatin-like phospholipase domain-containing protein 3 (PNPLA3)* affect development of alcoholic cirrhosis in white alcoholic subjects[8-9]. Despite the strong link between the *PNPLA3* polymorphisms and fatty liver diseases, deletion of this gene did not affect obesity-associated fatty liver or levels of liver enzymes in mice fed a high-fat diet[11]. Further studies are required to clarify the role of *PNPLA3* variants in the pathogenesis of ALD.

Finally, long-term alcohol drinking has synergistic effects with hepatitis virus B or C and/or human immunodeficiency virus infection, nonalcoholic fatty liver disease, and disorders such as hemochromatosis to accelerate progression of liver diseases. For example, many patients with viral hepatitis consume alcohol, which accelerates progression of liver fibrosis, cirrhosis, and HCC, likely via multiple mechanisms[12,13]. A greater understanding of the interaction between alcohol and these comorbid factors could help us design better therapies for the treatment of chronic liver disease.

Pathogenesis of alcoholic liver disease

Steatosis, the earliest response of the liver to alcohol abuse, is characterized by the accumulation of fat (mainly triglycerides, phospholipids, and cholesterol esters) in hepatocytes. Early studies indicated that alcohol consumption increases the ratio of reduced nicotinamide adenine dinucleotide/oxidized nicotinamide adenine dinucleotide in hepatocytes, which disrupts mitochondrial β-oxidation of fatty acids and results in steatosis[14]. Alcohol intake has also been shown to augment the supply of lipids to the liver from the small intestine, increasing mobilization of fatty acids from adipose tissue and uptake of fatty acids by the liver[14]. However, the contribution of these mechanisms to the development of steatosis after long-term alcohol consumption is not clear and requires further investigation.

Recent studies indicate that alcohol exposure, directly or indirectly, regulates lipid metabolism-associated transcription factors; this stimulates lipogenesis and inhibits fatty acid oxidation (Figure 2). Ethanol increases fatty acid synthesis in hepatocytes via up-

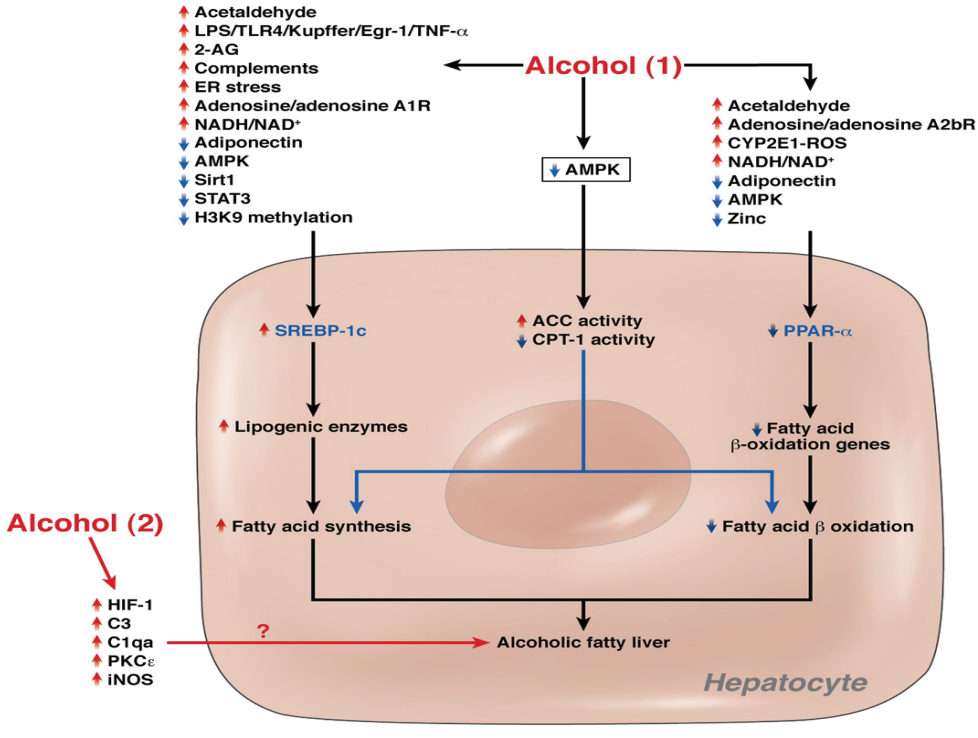

Figure 2. Mechanisms of alcoholic fatty liver[15].

regulation of sterol regulatory element-binding protein 1c (SREBP-1c), a transcription factor that promotes fatty acid synthesis via up-regulation of lipogenic genes. Alcohol consumption could directly increase transcription of SREBP-1c gene via its metabolite acetaldehyde[15] or indirectly up-regulate SREBP-1c expression by activating processes and factors that stimulate SREBP-1c expression, such as the endoplasmic reticulum response to cell stress[16,17], adenosine[18], endocannabinoids[19], LPS signaling via Toll-like receptor (TLR) 4, and its downstream proteins, including IRF-3, Egr-1, or tumor necrosis factor (TNF)-α[17,20-24]. Alcohol also down-regulates factors that reduce SREBP-1c expression, such as AMP-activated protein kinase (AMPK)[25], Sirtuinl[26], adiponectin[27], and signal transducer and activator of transcription 3 (STAT3)[28]. Disruption of *SREBP-1c* in mice reduced ethanol-induced fatty liver, indicating its role in ALD[29].

Alcohol consumption inhibits fatty acid oxidation in hepatocytes mainly via inactivation of the peroxisome proliferator-activated receptor (PPAR) ∞, a nuclear hormone receptor that controls transcription of a range of genes involved in free fatty acid transport and oxidation[30,31]. The ethanol metabolite acetaldehyde, but not ethanol itself, directly inhibits the transcriptional activation activity and DNA-binding ability of PPAR∞ in hepatocytes[32]. Ethanol consumption can also indirectly inhibit PPAR∞ via up-regulation of cytochrome P450 2E1-derived oxidative stress[33] and adenosine[18], both of which inhibit PPAR∞, or via down-regulation of adiponectin[34] and zinc[35], which each activate PPAR∞.

In addition to regulating fat metabolism-associated transcription factors, ethanol can also affect the activities of enzymes involved in fat metabolism by inhibiting AMPK, which reduces fat metabolism and fatty liver. AMPK is a serine-threonine kinase that can phosphorylate and subsequently inactivate acetyl-CoA carboxylase (ACC), a rate-limiting enzyme for fatty acid synthesis. Inactivation of ACC also reduces levels of malonyl-CoA, a precursor in fatty acid synthesis and an inhibitor of carnitine palmitoyltransferase 1, a rate-limiting enzyme for fatty acid oxidation[36]. In addition, AMPK directly phosphorylates and inhibits its SREBP activity in hepatocytes, thereby attenuating steatosis[37]. In this manner, AMPK inhibits fatty acid synthesis but promotes fatty acid oxidation via the inactivation of ACC enzyme activity. Alcohol consumption inhibits AMPK activity in the liver, leading to decreased phosphorylation and increased activity of ACC and decreased activity of carnitine palmitoyltrans-

ferase1; each has an important role in the development of alcoholic fatty liver[25].

Ethanol-induced steatosis is markedly reduced in many strains of mice, including *HIF-1*$^{-/-}$[38], *C3*$^{-/-}$[39], *C1qa*$^{-/-}$[40], *PKC*$^{-/-}$[41] and *iNOS,*$^{-/-}$[42] indicating that regulation of these genes also contributes to the pathogenesis of alcoholic fatty liver. However, the underlying mechanisms remain to be determined.

Finally, autophagy has an important role in removing lipid droplets in hepatocytes[43]. Long-term alcohol consv˙ption inhibits autophagy[44,45]. However, a recent study showed that short-term ethanol exposure activates autophagy by generating reactive oxygen species and inhibiting the mammalian target of rapamycin, indicating that acute ethanol activation of autophagy could have a compensatory role that prevents development of steatosis during the early stages of alcoholic liver injury[46]. The inhibitory and stimulatory effects of ethanol on autophagy require further studies to clarify.

Alcoholic Hepatitis (AH) is a syndrome characterized by infiltration of the liver by inflammatory cells and hepatocellular injury. AH develops in patients with steatosis and is usually associated with progressive fibrosis. The prevalence of AH has not been accurately determined; it is believed to occur in 10% to 35% of heavy drinkers. AH includes a spectrum of diseases that range from mild injury to severe, life threatening injury[1,47]. The histologic characteristics of AH include centrilobular ballooning of hepatocytes, neutrophilic infiltration, Mallory-Denk hyaline inclusions, steatosis, and a "chicken wire"-like pattern of fibrosis. In many cases, there is underlying cirrhosis[1,47]. A large body of evidence indicates that many factors contribute to alcohol-induced inflammation (figure 3)[47-49].

Mechanisms underlying inflammation in ALD are including (1) activation of innate immunity: parenchymal infiltration of neutrophils and macrophages is a prominent feature of ALD and is likely due to ethanol-mediated activation of innate immunity and subsequent induction of proinflammatory cytokines and chemokines. Alcohol consumption up-regulates a variety of factors that activate Kupffer cells, stellate cells, and hepatocytes, resulting in the production of cytokines and chemokines. Alcohol exposure also decreases proteasome activity and elevates IL-8 expression in hepatocytes and (2) activation of adaptive immunity: ALD is associated with infiltration of CD4_ and CD8_ T cells in the liver. Alcohol consumption induces reactive oxygen species (ROS) and causes the formation of many proteins adducts that might serve as antigens in the adaptive immune response[48,49].

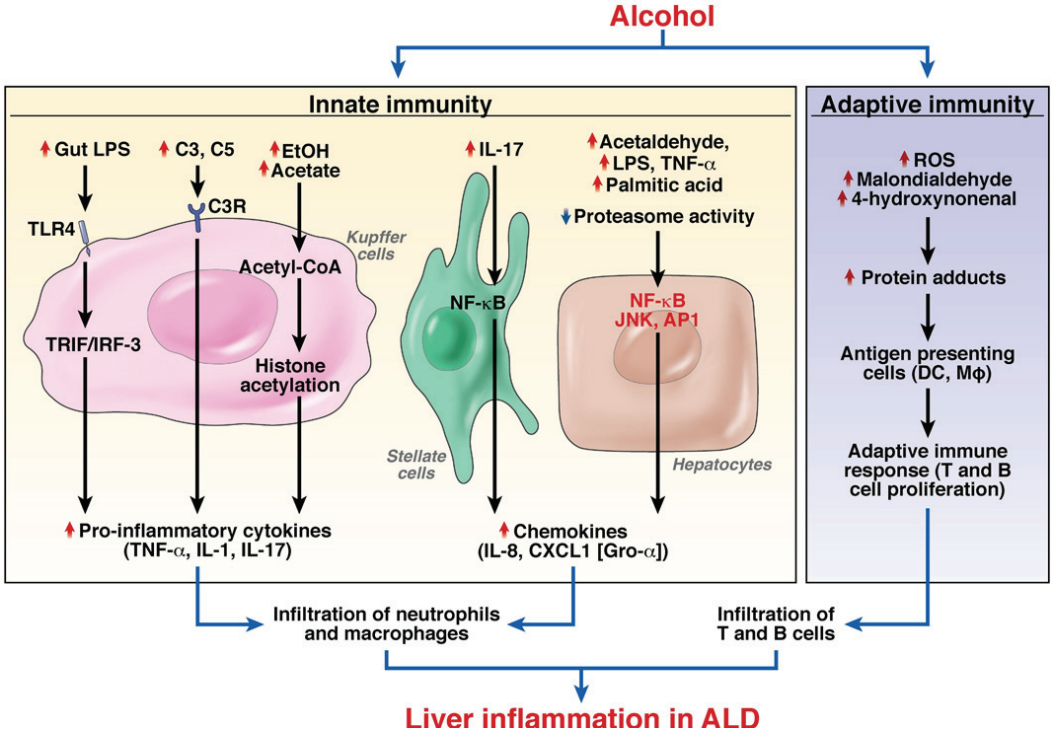

Figure 3. Mechanisms of inflammation in ALD[47].

In hepatocytes, ethanol is primarily metabolized into acetaldehyde by alcohol dehydrogenase in the cytosol, cytochrome P450 in microsomes, and catalase in peroxisomes. Ethanol metabolism generates reactive oxygen species and causes lipid peroxidation, mitochondrial glutathione depletion, and S-adenosylmethionine depletion; all of these products subsequently prime and sensitize hepatocytes to injury. Acetaldehyde is rapidly metabolized into acetate by aldehyde dehydrogenase in mitochondria. Acetaldehyde is a reactive compound; it is highly toxic to hepatocytes because it forms a variety of protein and DNA adducts that promote glutathione depletion, lipid peroxidation, and mitochondrial damage[48,49]. The acetate that results from acetaldehyde breakdown is rapidly released from the liver into the circulation and is then metabolized into CO_2 via the TCA cycle in heart, skeletal muscle, and brain. Although acetate has no direct hepatotoxicity, it is believed to regulate the inflammatory response in patients with AH via the up-regulation of proinflammatory cytokines in macrophages[50,51]. The mechanism pathways of these factors remain unclear that necessary for further investigations.

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