Medical Research Archives

ESMED
European Society of Medicine

OPEN ACCESS

**Published:** June 30, 2022

**Citation** Andrea M.P. Romani, 2022 Alcoholic Cardiomyopathy: Where do we stand? Medical Research Archives, [online] 10(6). https://doi.org/10.18103/mra.v10i6.2847



**DOI** https://doi.org/10.18103/mra.v10i6.2847

**ISSN:** 2375-1924

RESEARCH ARTICLE

# Alcoholic Cardiomyopathy: Where do we stand?

**Andrea M.P. Romani***[1]

[1] Department of Physiology and Biophysics, School of Medicine. Case Western Reserve University, Cleveland, OH 44106-4970

* am5@po.cwru.edu

ABSTRACT

The effect of alcohol consumption on cardiac and cardiovascular functions remains a point of contention in the medical field. The consumption of low or moderate amounts of alcohol has been largely associated with having beneficial effect on cardiac contractility and the cardiovascular system as a whole, owing to the detected vasodilatory effect exerted by the alcohol, and the reduction in mortality documented by several studies. In contrast, excessive alcohol consumption results in negative outcomes in both men and women, with cardiac arrhythmias and atrial fibrillation, abnormality in cardiac contraction leading to heart failure, and dilated cardiomyopathy, and overall cardiovascular dysfunctions, including hypertension. The main points of contention are two-fold: the dose of alcohol at which its perceived beneficial effects disappear and proper cardiac and cardiovascular functions become progressively impaired, and how to clinical and therapeutically address cardiac and cardiovascular pathologies in chronic alcoholics to ameliorate their general conditions and their prognosis. The present review aims at providing the reader with a general understanding of the effects of alcohol on the cardiovascular system and the pathophysiological mechanisms that lead to the most common cases of cardiac dysfunction, and highlight the current guidelines for treatment of alcoholic cardiomyopathy to ameliorate clinical presentation and prognosis in alcoholic patients.

**Keywords:** Alcohol, cytP4502E1, alcohol metabolism, cardiac ventricular myocytes, alcoholic cardiomyopathy, acetaldehyde, magnesium

## Introduction

Alcoholic cardiomyopathy has long been recognized as a one of the most serious pathological complications in heavy and chronic drinkers.[1] Historically, the first recognition of the harmful effects of alcohol was by Dr. Bollinger who, in 1886, observed what came to be known as 'Bierherz' or 'Beer heart' in a set of breweries workers, the hearts of whom presented at the autopsy with "*...idiopathic cardiac hypertrophy usually with dilation...*".[2] Clinically, the disorder presents as a heart failure condition characterized by enlarged heart, reduced stroke volume and cardiac output, and arrhythmias, including primarily atrial fibrillation. At the histopathological level, the disorder presents as a cardiomyopathy, with progressive myocardial injury, areas of necrosis and apoptosis, and fibrosis.[3] The gross examination of the beer heart evidences a pale cardiac tissue with marked dilation of all cardiac chambers, especially the left ventricle.[4] Areas of atrophy and degeneration mixed with areas of hypertrophy are observed focally within the cardiac tissue. Electron microscopy of the myocardium underscores loss or damage of contractile myofibrils, enlarged mitochondria, decreased content of oxidative enzymes, saccular dilatation of sarcoplasmic reticulum, and fat deposits primarily constituted by triglycerides within the cardiac myocytes.[3] This picture is in sharp contrast to the limited or no major abnormalities detected by light microscopy,[3] and provides a strong histological rationale to understand the clinical signs of alcoholic cardiomyopathy in the affected individuals.

Presently, the term alcoholic cardiomyopathy (ACM) is currently used to indicate the cardiac dysfunction observed in chronic alcoholics or in patients who intake excessive amounts of alcohol. Despite being long recognized as a complication of large and persistent alcohol consumption and its clinical relevance and incidence, data on how alcohol damages cardiac myocytes and how best treat the conditions are limited.

In the present review, we will attempt to provide a comprehensive overview of how the disease develops and progresses, as well as the most current treatment guidelines.

## Alcohol consumption as a risk factor

Alcohol use is recognized as one of the most common cause of non-communicable disease. Furthermore, the negative implications of excessive alcohol consumption are both preventable and modifiable.[5] The Global Burden of Disease Study identified alcohol as the seventh leading risk factor for death and for disability-adjusted life years, and as responsible for 2.2% and 6.8% death rate in women and men, respectively, after age-standardized adjustment.[6] While drinking no alcohol minimizes health-related risks, alcohol consumption was found to be responsible for almost 3 million deaths world-wide.[6] When assessed for the US alone, the number of deaths conducible to alcohol consumption was in excess of 70,000 (~2.6% of all deaths recorded in the country), more than doubled the 1999 number.[6] Further analysis by the Global Burden of Disease Study indicated that alcohol was the loading risk factor for premature death and disability in individuals up to 49 years of age.[6] In contrast to the negative effects of persistent and excessive alcohol consumption, several epidemiological studies suggest that limited alcohol consumption may actually be beneficial in reducing the cardiovascular insults, in particular coronary artery disease and cerebrovascular ischemic insults.[7] The validity of these studies has been disputed, and it has been argued that the presumed benefits are rather limited in number as compared to the large number of alcohol-related deaths and complications.[7]

Interpretation of the data relative to alcohol-related deaths and health complications is made more difficult by the lack of consensus on what limited alcohol consumption actually means and the amount of alcohol present in a standard alcohol drink. As an example, the 14 g of pure alcohol in a standard drink in the US contrasts to the 8 g in the UK and differing alcohol contents in other countries, making direct comparison of alcohol intake to pathological consequences rather difficult. Furthermore, assessment of the amount of alcohol consumed by patients over a certain period of time is often based, at least in the initial phase of data-collection and sample identification, on self-reported questionnaires, an approach that can easily lead to misrepresentations or underestimation of the type of alcohol and the number of drinks consumed by the reporting individuals.[8] Recently, a certain degree of consensus has been attained in discriminating among 'low', 'moderate', or 'hazardous' risk drinking. Low-risk drinking is defined as two or fewer drinks per day for men and one or less drink per day for women. Moderate-risk drinking is defined as three to four drinks per day for men and two to three drinks per day for women. Hazardous-

risk drinking is defined as five or more drinks per day for men and four or more drinks per day for women. Binge drinking, colloquially referred to as 'alcohol poisoning' in the US, is define as the consumption of five or more drinks per occasion (i.e. in a short period of time) for men and four or more drinks per occasion for women. Application of these criteria to epidemiological studies, however, is complicated by lack of appropriate control for confounding factors including consumption of other recreational drugs such as cigarettes and e-cigarettes, caffeine, marijuana, cocaine, amphetamines and methamphetamines, or therapeutic agents (e.g. acetaminophen, benzodiazepines, etc.) and xenobiotics that are metabolized by the same cytochrome-operated system (Microsomal Ethanol Oxidizing System) that oxidizes alcohol to acetaldehyde in hepatocytes and cardiac myocytes through a process that increases reactive oxygen species formation.[9,10]

The consumption of a low dose of alcohol has consistently been associated with a lower risk of cardiovascular-based mortality when compared with abstainers, whereas the consumption of high doses of alcohol has always been associated with a higher risk of cardiovascular-based mortality. This dose-response association is commonly referred to as the 'J-shaped' or the 'U-shaped' curve. [5, 7, 11, 12] A meta-analysis carried out by di Castelnuovo et al. on 34 studies involving more than a million individuals identified the cardiovascular beneficial effects of alcohol at a dose equivalent to half or 1 drink daily for women, and 1 to 2 drinks daily for men, the consumption of more than 2 drinks per day for women and more than 4 drinks per day for men resulting in higher death-rates, in a dose-dependent manner.[13] When considering the benefits of consuming low doses of alcohol, additional attention has to be paid to the type of alcohol consumed. Red wines contain high levels of polyphenols, and these agents have been proven to possess anti-oxidant and anti-inflammatory effects in addition to anti-platelets aggregation properties, which translate primarily in reduced plasma viscosity, increased high density lipoproteins, decreased platelet aggregation and enhanced endothelial functions among other beneficial effects.[11] Consumption of beer in moderation also appears to manifest several of the same protective effects observed with red wine consumption, although the exact mechanism of protection is not well elucidated. On the other hand, larger consumption of beer has been plagued more than wine by pathological cardiac complications associated to the presence of significant trace amounts of arsenic and cobalt in the beverage as a result of the filtration process and as a foam-stabilizer, respectively.[14, 15] As recently as 2013, arsenic and cobalt have been postulated to act synergistically with alcohol to give rise to severe and widely spread cardiovascular pathologies, including cardiomyopathy and pericarditis in both Europe and North-America.[15] Even when present in limited amounts, arsenic retained in beer as a result of the filtration process can reach up to 10 times the concentration deemed acceptable in drinking water in the US and in Europe, a condition that can easily be exacerbated when beer is consumed in large quantities.[15] The importance of maintaining low-alcohol consumption as a key requisite to retain beneficial cardiovascular effects is underscored by results showing that occasional but recurrent heavy consumption of alcohol, as observed for example during binge-drinking events, increases by almost 50% morbidity and mortality risk due to the onset cardiac dysfunction and myocardial ischemia.[16] Taken together, these observations strongly suggest that the deleterious effects of excessive alcohol drinking on the cardiovascular system and the consequent risk of premature mortality far out-weight the beneficial effects observed at low consumption doses.

**Alcohol Ingestion and Myocardial Metabolism**

Acute consumption of alcohol, especially in large doses has significant effects on cardiac metabolism.[1 and refs. therein] The concentrations of potassium and phosphate increase markedly in the coronary flow, and the extraction of free fatty acids from the serum decreases while that of triglycerides increases, with major implications in terms of intracellular and interfibrillar lipid accumulation. At the mitochondrial level, isocitric and malic dehydrogenases are released at rest and following an increase in cardiac rhythm, indication that the organelle undergoes major functional alterations.[1] Because alcohol is primarily metabolized through cyt-P4502E1 in cardiac cells, the generated acetaldehyde and reactive oxygen species (ROS) have been indicated as the major biochemical mechanisms responsible for cardiac damage as both acetaldehyde and ROS can form adducts with proteins and lipids within the cardiac cells that results in decreased contractility and enzymatic activity.[10] In addition, acetaldehyde has been reported to cause positive chronotropic and inotropic effect through persistent norepinephrine release.[17] On the other hand, the conversion of

alcohol to acetaldehyde by the nicotinamide adenine dinucleotide phosphate (NADP)-dependent cytP4502E1 increases the NADP/NADPH ratio within the sarcoplasmic reticulum further impacting in a negative way lipid and pyruvate metabolism and resulting in increased triglyceride synthesis and lactate production from pyruvate.[18]

**Development of Alcoholic Cardiomyopathy**

Both the European Society of Cardiology and the American Heart Association consider alcoholic cardiomyopathy (ACM) a specific pathology caused by excessive alcohol consumption and characterized by left ventricular dysfunction and dilatation, thereby considered as a form of dilated cardiomyopathy (DCM) without ischemic connotations.[19,20] Usually, the diagnosis of ACM is one of exclusion, supported by a history of heavy alcohol consumption, usually 80 g of alcohol or more per day for at least 5 years.[21-24] It has to be noted, however, that this amount of alcohol consumption can remain an underestimate of the actual consumption in patients, even more so if we take into account that many epidemiological studies not necessary include information about patients' distribution based on gender and/or body mass index (BMI). Hence, controversy or at least uncertainty persists as to whether ACM severity in terms of structural damage within the ventricular cardiac myocytes is proportionally linked to the amount of alcohol consumed per day or a threshold exists above which the damage by excessive alcohol consumption takes place regardless of the duration of consumption, in that the dose of alcohol has overcome any protective mechanisms present within the cell. In this regard, studies from Askanas et al.[25] and Lazarevic et al.[26] appear to lend support to the latter hypothesis in that these studies suggest that once individuals consume ~120 g of alcohol per day or more, the duration of alcohol consumption becomes less of a factor in the development of ACM, as similar clinical signs are observed in patients who have consumed such a large amount of alcohol per day for 5 to 14 years as in those patients who have consumed a similar amount of alcohol for 15 years or more. Consistent with this observation, the study by Kino et al. [27] indicates that the amount of alcohol consumed per day would primarily affect left ventricle wall thickness, as this parameter is proportionally and significantly enhanced in patients consuming ~100 g of alcohol per day as compared to patients who consume 60 g per day.[27] However, as this study did not report the duration of alcohol consumption at either of those doses, no comment can be made as to whether time lapse can be factored in for ACM to develop.

The histopathology of ACM at the macroscopic level has remained well defined and determined since the original observation by Bollinger in 1886,[1,2] and has become enriched at the microscopic level by the observation of intracellular swelling, glycogen and lipid intracellular accumulation and ultra-stuctural damage of mitochondria and sarcoplasmic reticulum, thus helping to explain the ventricle dilation and the signs of left heart failure typically associated with ACM.[15,28] Several pathological mechanisms have been indicated as possible culprits in the development of these microscopic alterations. These pathological mechanisms have been potentially identified as follows: 1) apoptosis; 2) alterations of mitochondria functions and oxidative phosphorylation; 3) functional alterations in the sarcoplasmic reticulum, especially $Ca^{2+}$ cycling; 4) abnormalities of $Ca^{2+}$ excitation/contraction coupling; 5) persistent abnormalities of cytosolic free $Ca^{2+}$ levels with associated changes in $Ca^{2+}$ sensitivity of the contractile myofilaments; 6) abnormal representation of contractile myofilaments; 7) abnormal myosin ATPase activity rate; 8) up-regulation of L-type $Ca^{2+}$ channels; 9) increased oxidative stress; 10) production of acetaldehyde and formation of protein-aldehyde adducts; 11) activation of the renin-angiotensin-aldosterone system; 11) activation of the sympathetic nervous system.[28,29 and refs therein] The review articles by Fenandez-Sola,[28] and Guzzo-Merello et al.[29] have described these mechanisms in detail, and we refer the interested readers to those articles for an in-depth description of the mechanisms and their interplay. Suffice here to mention that because some of these pathological mechanisms become activated during cardiac hypertrophy but also as a compensatory mechanism following loss of cardiac ventricular mass replaced by fibrosis, it is still debated which of these two events (i.e. hypertrophy or cardiac loss) is the initial cause of ACM onset and progression, or whether they coexist to some extent. Additionally, the possibility that genetic predisposition plays a key role in the onset of ACM cannot be excluded altogether in that not all alcoholics develop ACM, as observed already for alcoholic liver disease (ALD) progression.[30] Consequently, because the diagnosis of ACM remains primarily a diagnosis of

exclusion and is based on the co-presence of heavy alcohol consumption and signs of myocardial dysfunction such as decreased ejection fraction (<45%) and increased left ventricular end-diastolic diameters in the absence of other clear etiological causes,[31] the prevalence of ACM may be underestimated. In this regard, autopsy observation of pathological changes in cardiac structures, often in the absence of clear clinical symptoms during the life-time of a patient remains the only viable modality for a posthumous diagnosis.[32]

**Alcoholic Cardiomyopathy and other Clinical Pathologies linked to Alcohol Consumption**

As mentioned previously, alcoholic cardiomyopathy (ACM) is considered a particular form of dilated cardiomyopathy (DCM), characterized by dilation of the left ventricle, and signs and symptoms of left heart failure (LHF). However, as the alcohol-induced damage of the heart can impact specific cardiac structures such as cardiac ventricular myocytes or the conduction system, or cardiac regulatory mechanisms such as the renin-angiotensin-aldosterone system or the sympathetic system, a variety of clinical conditions other than the classical, histological well-described, ACM can be observed in alcohol-drinking patients. Among these conditions hypertension, myocarditis, coronary artery disease, arrhythmias, and stroke, are most commonly observed in alcoholics. Yet, as mentioned previously, it is unclear whether they are consequences of alcohol consumption or co-existing pathologies exacerbated by alcohol consumption.

**a. Hypertension**

Alcohol consumption is known to cause an acute but transient vasodilation as well as more long-term pressor effects. The transient vasodilation is most likely mediated by the release of the atrial natriuretic peptide that counter-regulates the renin-angiotensin system.[33] On the other hand, the long term pressor effects of alcohol appears to be primarily mediated by the renin-angiotensin-aldosterone system and the release of vasopressin, and associated cardiac remodeling.[34,35] Alcohol also exerts central hypertensive effects by interacting with brain stem receptor receptors.[36] Interestingly, this central effect would occur already after the consumption of 3 drinks per day, thereby showing what appears to be a lower alcohol threshold than many other alcohol-related pathological effects, and would interfere with concurrent anti-hypertensive treatments, further complicating blood pressure stabilization.[15,36]

**b. Alcoholic Myocarditis**

Myocarditis in the context of alcohol consumption was first reported by McKenzie in 1902.[37] The histopathological assessment indicated the presence of lymphocytic infiltrates in the context of focal myocyte necrosis and degeneration, with increased expression of both human leukocyte antigen (HLA) and intercellular adhesion molecule (ICAM).[38,39] The increased expression of HLA and ICAM has been related to altered lymphocyte function, inhibited neutrophil chemotaxis and suppressed cytokines production associated with the dysregulated inflammatory response normally observed following prolonged alcohol consumption, both systemically and locally.[40-42]

**c. Coronary Artery Disease**

As stated at the beginning of this review, the relationship between alcohol consumption and coronary artery disease, mainly angina, is a complex one. Low doses of alcohol have long been considered as a direct coronary artery dilator and therefore beneficial – at least temporarily – in patients with angina or coronary artery disease on atherosclerotic bases. Yet, alcohol is not a direct coronary dilator,[43] and the symptomatic relief of angina symptoms following alcohol consumption can be explained by its general anesthetic effects, its peripheral vasodilatory effects, which decrease temporarily the cardiac oxygen demand, its anti-aggregate effect on platelets and coagulation system, and to its effect at increasing high density lipoproteins, especially sub-fractions 2 and 3, while decreasing low density lipoproteins.[44-47] Many of these beneficial effects can act synergistically to ameliorate angina conditions, and they are attributed to the levels of polyphenols and resveratrol found primarily in red wine.[11]

**d. Arrhythmias**

Acute effects of alcohol ingestion often result in disturbances of the cardiac rhythm, and these disturbances tend to disappear with abstinence. A Kaiser Permanente Study reported a double-relative risk of arrhythmias in heavy drinkers consuming more than 6 drinks per day as compared to low-risk drinkers.[48] Several hypotheses have been put forward to explain

the insurgence of arrhythmias including the underlying hypertension and the cardiac remodeling associated with activation of the renin-angiotensin system, a lowering of the resting membrane potential in the conduction system of the heart and/or the cardiac ventricular myocytes, the prolongation of impulse conduction, and the reduced responsiveness of the adenyl-cyclase downstream the cardiac □1adreneric receptor.[49-51]

**Vascular Complications**

The effect of alcohol consumption on the vascular system in general and coronary system in particular is not fully elucidated. Experimental and epidemiological studies largely report that moderate consumption of alcohol is associated with a transient vasodilation followed by a rebounding increase in blood pressure, as mentioned under the Hypertension paragraph. Assessment of 28 alcohol-consuming patients of an age between 28 and 65 undergoing randomized and sequential exposure for 1 week to wine consumption, to beer consumption, to non-alcoholic wine consumption and to abstinence[52] indicated an increase in heart rate more sustained during the evening and at night while blood pressure in the same individuals increased primarily during the day. The reason for these diurnal changes in blood pressure and heart rate were primarily attributed to circadian changes in the sympathetic and vagal nervous systems, although an increased urinary extrusion of endothelin-1 was observed following alcohol consumption, regardless of whether alcohol was administered as wine or beer.[52] This latter observation suggests that alcohol increases endothelin-1 production and ultimately urinary secretion, with increased pressory effect during the day.[52] Because in their study Zilkens and collaborators did not assess the occurrence of circadian changes in endothelin-1 levels both in the absence and in the presence of alcohol consumption, it remains unanswered whether the observed changes in blood pressure and heart rate are the result of alcohol-induced changes in endothelin-1 levels during the day, or alcohol-mediated exacerbation of a physiological condition already in place. In addition, as the authors selected for their study individuals already consuming alcohol prior to their recruitment for the study, it cannot be excluded that the alcohol-induced changes in blood pressure and heart rate were already in place, at least to some extent, prior to the beginning of the study. On the other hand, because the intake of a volume of non-alcoholic wine containing an amount of resveratrol, the most abundant polyphenol in red wine, comparable to that present in an equal volume of wine did not induce any detectable change in blood pressure, heart rate and endothelial secretion, the results of the study by Zilkens et al.[52] strongly suggest that the effects exerted by alcoholic beverages are due to alcohol itself. This observation contrasts what reported by Agerwall et al.[53], and the reason for the discrepancy has been attributed to the different changes in endothelial functions and endothelin-1 release observed in the two studies.

Strictly considering the effects of alcohol on the vasculature, the most important modifications in endothelial functions have been primarily related to three areas: atherosclerotic plaque, nitric oxide (NO), and reactive oxygen species (ROS) formation.

a. **Atherosclerotic Plaque**

Atherosclerosis is a chronic inflammatory condition characterized by thickening of the artery wall due to accumulation of cholesterol, in particular LDL that can be more readily oxidized and macrophages, and proliferation of smooth muscle cells.[54] The thickening of the arterial wall results in a narrowing of the arterial lumen and a reduced blood flow, ultimately leading to coronary artery disease, culminating in angina and myocardial infarction, and cerebrovascular disease and stroke.[54] As LDL become oxidized, free radical production increases and ultimately leads to atherothrombosis, through a process that includes recruitment of circulating monocytes, increased expression of intercellular adhesion molecule-1 (ICAM-1) and platelet derived growth factor (PDGF), and foam cell production, all conditions that promote smooth muscle cells proliferation within the intima of the arterial wall.[54 and refs. therein.] Owing to the effect of alcohol consumption in moderate doses in preventing platelet aggregation, studies by Pahor et al.[55], and Pomp et al.[56] have evidenced a decreased risk of deep vein thrombosis and pulmonary embolism in people of 68 years of age or older. Further, the data by Pomp et al.[56] confirmed a reduced risk of thromboembolism particularly in women, and explained the outcome with a

decreased in fibrinogen levels. Once again, the more pronounced protective effect was observed in individuals consuming red wine, and linked to the high level of resveratrol present therein.[57]

b. **Nitric Oxide (NO) production**

Nitric oxide (NO) is a well-known endothelium-derived relaxing factor.[58] NO contributes to vascular homeostasis by causing vasodilation and through its antioxidant and anti-inflammatory properties, which antagonize atherosclerosis progression.[59] Exposure of rats to moderate alcohol intake for 8 weeks resulted in increased expression of eNOS (endothelial nitric oxide synthase) protein in the vasculature and increased levels of NO in the circulation, and improved systolic and diastolic functions, with some apparent gender-related differences that were attributed to increased levels of iNOS (inducible Nitric Oxide Synthase) in female rats.[60] In contrast, exposure of rats to high levels of alcohol (4 g/kg) for 12 weeks resulted in the development of hypertension, impaired vascular relaxation, reduced eNOS expression and increased vascular oxidative stress.[61] Studies carried out in humans reported similar results. Red wine consumption acutely increased NO production in healthy humans,[62] through a mechanism that involves the rapid activation of mitochondrial aldehyde dehydrogenase 2 (ALDH2) to prevent reactive oxygen species formation, and ultimately eNOS activation.[63] Prolonged exposure of human endothelial cells to high doses of alcohol, on the other hand, markedly reduced NO production.[64] Taken together, these studies – irrespective of the experimental model utilized – confirm the underlying tenant that moderate doses of alcohol primarily exert a vasodilatory effect by increasing NO production whereas high (toxic) doses of alcohol induce vasoconstrictive effects, possibly through increased production of endothelin-1.[65]

c. **Reactive Oxygen Species formation**

An increased production of reactive oxygen species (ROS) is involved in a variety of cardiovascular dysfunctions including atherosclerosis.[54] The production of ROS is due to an imbalance between pro-oxidative enzymes (i.e. NADPH oxidase, xanthine oxidase, and mitochondrial electron chain) and anti-pxidative enzymes (i.e. superoxide dismutase, glutathione peroxidase, thioredoxin peroxidase, catalase, and paraoxonase) that ultimately favors pro-oxidative enzymes. Further, consumption of high doses of alcohol results in the conversion of ethanol to acetaldehyde via the cytP4502E1, a process that is associated with ROS production within the sarcoplasmic reticulum of the cardiac myocytes, i.e. a location where ROS can more readily damage $Ca^{2+}$ storage and cycling.[10] Consistent with this notion, data from Rocha et al.[66] suggest that low and moderate alcohol consumption induce vascular relaxation through ROS generation, in contrast to the hypertensive state associated with abnormal ROS formation as a result of chronic, elevated alcohol consumption.[67] An implication of this observation in need of a more-in-depth assessment would be that the low levels of ROS generated following modest alcohol intake could have transient regulatory effects on specific cellular functions before being scavenged by anti-oxidant mechanisms. In contrast, the massive ROS production generated by chronic and abusive alcohol consumption over-powers the anti-oxidant mechanisms and results in abnormal, persistent activation of these cellular function, ultimately leading to cardiac myocyte damage.

**Is there an unappreciated role of Magnesium in the onset of Alcoholic Cardiomyopathy and other Cardiovascular Complications?**

Magnesium ($Mg^{2+}$) is the second most abundant cation after potassium within mammalian cells, including cardiac myocytes.[68] Being primarily an intracellular cation, $Mg^{2+}$ is highly concentrated within mitochondria, endoplasmic or sarcoplasmic reticulum, and nucleus, where it controls a variety of enzymes including adenyl cyclase, mitochondrial electron chain complexes and adenine nucleotide translocase, and reticular $Ca^{2+}$ cycling.[68] Cellular $Mg^{2+}$ homeostasis is maintained through the operation of a specific entry and exit mechanisms: a cAMP-activated $Na^{+}/Mg^{2+}$ exchanger (or Solute

Carrier 41: SLC41A1) extrudes cellular $Mg^{2+}$ in exchange for extracellular $Na^{+}$, whereas protein kinase C-regulated Transient Receptor Potential cation channel, subfamily M, member **7** (TRPM7 channel) promote $Mg^{2+}$ entry.[69] Additional transporters, which are only partially identified and characterized, contribute to maintain proper $Mg^{2+}$ distribution within cellular organelles.[69] Adrenergic stimulation of □1-adrenergic receptors in cardiac cells results in a marked release of cellular $Mg^{2+}$ into the extracellular space or the circulation within 8-10 min from the administration of the adrenergic stimulus, in both perfused hearts and collagenase-dispersed cardiac ventricular myocytes, and in the whole animal.[68-70] This elicited $Mg^{2+}$ extrusion from cardiac cells appears to regulate mitochondrial energetic production and $Ca^{2+}$ cycling across the sarcoplasmic reticulum membrane as well as the duration of the ventricular action potential.[69 and refs therein] Interestingly, acute alcohol administration to cardiac cells results in a dose- and time-dependent extrusion of $Mg^{2+}$ that parallels the decrease in cardiac ATP levels resulting from the conversion of alcohol to acetaldehyde via cytP4502E1.[71,72] This loss of $Mg^{2+}$ affects primarily the cytosolic pool of $Mg^{2+}$ but also the pools located within mitochondria and sarcoplasmic reticulum.[71] Pharmacological inhibition of alcohol oxidation to acetaldehyde or the use of anti-oxidants, which scavenge reactive oxygen species and prevents their deleterious effects on cellular components, both prevents the loss of cytosolic- and organelle-based $Mg^{2+}$ and ATP, and help cardiac myocytes to maintain functions and ultrastructural morphology.[72]

The role of cellular $Mg^{2+}$ in maintaining proper cardiovascular functions and cardiac cells ultrastructural properties is largely underestimate. However, $Mg^{2+}$ is postulated to act as a natural $Ca^{2+}$ channel blocker and a cellular $Na^{+}$ content regulator, especially in cardiac cells.[69] This notion is indirectly supported by the clinical observation that magnesium boluses are far more effective than anti-arrhythmic drugs in preventing or sedating cardiac arrhythmias, torsade de point in particular.[73] Interestingly, hearts of animals fed magnesium-deficient diets present with the same ultrastructural modifications in terms of altered contractile elements, swollen mitochondria and sarcoplasmic reticulum dilation, and increased intracellular fat deposits that have been observed in the hearts of heavy alcoholics,[1] and exogenous $Mg^{2+}$ supplementation with the diet after alcohol withdrawal renormalizes heart size, isometric force and isotonic shortening of the cardiac fibers.[74] How exactly magnesium supplementation elicits these beneficial effects is not understood, and the topic warrants more attentive research, especially in lieu of the observation that continuous exposure to alcohol prevents restoration of $Mg^{2+}$ homeostasis and nullify its beneficial effects.[69,71] Speculatively, it can be envisioned that these beneficial effects occur via restoration of cellular $Ca^{2+}$ content and cycling, adrenergic receptor-mediated adenyl-cyclase activity, or protein synthesis and mRNA translation, based on the key cellular targets on which magnesium plays a strong regulatory role.

**Prognosis and Treatment**

The prognosis of ACM in patients with low or moderate alcohol consumption is usually more benign that in heavy drinkers, in that the former two groups of individuals are more liable to follow an abstinence regimen and they usually present with modest cardiac damage and symptoms as compared to the latter group.[15] The prognosis of ACM in heavy drinkers, on the other hand, is far more complex as the health status of these individuals is often hindered by other complications including hemorrhagic stroke, as a result of alcohol-supported hypertension, arrhythmias, altered coagulation process, atherosclerosis, and overall a poor compliance to alcohol withdrawal.

Alcohol withdrawal remains the corner stone of any alcohol-based pathology, and ACM is not different in this regard. An indirect benefit of alcohol withdrawal is that also promotes a better responsiveness to cardiovascular oriented therapies, especially anti-hypertensive treatment.[1,15,29] Abstinence supportive therapies can also be implemented, including naltrexone to reduce alcohol-craving, acamprosate, which also reduce craving, disulfiram to inhibit acetaldehyde dehydrogenase and use the negative side-effects of acetaldehyde build-up (i.e. nausea, sweating, and tachycardia) to limit alcohol use, or specific serotonin reuptake inhibitors.[75]

Aside from these support therapies, because the main symptoms of ACM are those of heart failure, the therapeutic regimen follows the usual steps of heart failure treatment, which include angiotensin converting enzyme (ACE) inhibitors or angiotensin-II receptor blockers (ARB), beta-blockers, diuretics including spironolactone, digitalis to support cardiac contraction, and anticoagulants as appropriate, although this last class of drugs carries significant counter-indications (namely poor compliance, risk of post-traumatic bleeding, and over-dosage in case of hepatic dysfunction). Anti-

arrhythmic drugs are usually not included as part of the medical treatment owing to their intrinsic risk of being arrhythmogenic.[15,29] A question remains whether heart transplant can be considered as a viable treatment for patients with ACM.[76] Post-surgery assessment of the procedure indicates that heart transplant can be safely performed in alcoholic patients, with clear and long-term benefits, and a post-surgery mortality rate at 5 and 10 years that is not statistically different from that of non-alcoholics: ~79% at 3 years, and 67% at 5 years, and ~49% at 10 years (https://www.healthguideinfo.com/surgical-heart-procedures/p79774/).[76] The main limitation remains the strict adherence by the patient to 6 months of abstinence prior to undergoing the procedure.

**Conclusion**

In the present review article we have attempted to provide an overview of the onset, development, prognosis and treatment of alcoholic cardiomyopathy, and relate the specifics of this pathology to the dose of alcohol required to generate the conditions. It is clear from the review of the available literature that the more consistent and reliable results and information on ACM have been obtained when large numbers of patients were assessed against multiple variables that included the amount of alcohol consumed per drink, the number of drinks consumed per week, and the duration of alcohol consumption. Yet, some uncertainties persist, including the importance of body mass index and the possible role of co-morbidities in promoting a faster progression towards ACM. As mentioned in the review, several clinical manifestations including hypertension and arrhythmias can be related in a multi-factorial manner to alcohol consumption in a cause-effect manner but can also be considered alcohol-independent risk factors that make the progression of ACM more severe. Further studies with more well-defined selection criteria and more attention to alcohol consumption and other variables are necessary to clarify some of the above uncertainties. For example, it is still unclear whether a threshold exists above which ACM develops and whether such a threshold is different based on gender, body mass index, the type of alcohol ingested (e.g. wine vs. beer vs. hard liquor), or it strictly depends on the amount of alcohol ingested. Because the cytP4502E1 can be induced by extensive alcohol consumption, leading to increased reactive oxygen species production, it needs further clarification whether an increase in cytP4502E1 expression is beneficial or detrimental to the onset and progression of alcoholic cardiomyopathy. On the other hand, data from studies implicating atherosclerosis, nitric oxide production, and reactive oxygen species generation in the vasculature of alcohol-consuming individuals or animal models concur in indicating that low or moderate alcohol consumption is beneficial and induces vasodilation whereas chronic and massive alcohol consumption results in vasoconstriction, increased blood pressure, and abnormal regulation of the sympathovagal systems.

In this review, we have detoured from the canonical aspects of alcohol disease to provide an alternative lens to consider how changes in cellular magnesium that occur following acute and chronic exposure of cardiac cells to alcohol may play a yet to define role in the development of some of the ultrastructural and energetic dysfunctions that occur in cardiac cells observed in alcohol-exposed cardiac cells. Because the role of magnesium in the onset and progression of ACM and its clinical associations has not been investigated in great detail and no proper clinical trial has addressed the issue in all its variables, the picture we have is far from being clear, and several aspects still remain poorly understood. Owing to the limited information and attention paid on the possible beneficial role of magnesium in preventing the development of ACM, it is still largely undetermined whether $Mg^{2+}$ supplementation in conjunction with more classical therapeutic agents can improve the patient's health and minimize the progression of alcoholic cardiomyopathy, and whether the beneficial effect of magnesium supplementation are permanent or transient. Further, if we consider magnesium as a therapeutic agents or co-adjuvant, it remains to be determined the minimal dose at which the effects of magnesium are already maximal, and whether these effects are permanent or transient. This latter question is not trivial in that if the effects are transient, implication is there that magnesium supplementation has to be maintained for extended period of time to guarantee a long-lasting beneficial effect of cardiac myocytes physiology and contractility.

These are just some of the major questions still plaguing the field of ACM and its complications that are in need of experimental and clinical clarification.

**References**


1. Shanoff HM. Alcoholic cardiomyopathy: an introductory review *CMA Journal*; 1972;106(1):55-62.
2. Meister HP. The "Munich Beer Heart"- Revisited Das Münchner Bierherz, 1976;157(1):1-13. https://www.sciencedirect.com/journal/beitrage-zur-pathologie. Accessed May 19, 2022.
3. Sanders MG. Alcoholic cardiomyopathy: a critical review. *Q J Stud Alcohol*; 1970;31(2):324-368.
4. Evans W. Alcoholic myocardiopathy, *Progr Cardiovasc Dis*;1964 Sep;7:151-171.
5. Xi B, Veeranski SP, Zhao M, Ma C, Yan Y, Mi J. Relationship of alcohol consumption to all-cause, cardiovascular, and cancer-related mortality in US adults. *J Am Coll Cardiol*;2017;70(8): 913-922.
6. Global Burden of Disease Study (GBD) 2016 Alcohol Collaborators, Griswold MG, Fullman N, Hawley C, et al. Alcohol use and burden for 195 countries and territories, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016. *Lancet*;2018;392(10152):1015–1035.
7. O'Keefe EL, DiNicolantonio JJ, O'Keefe JH, Lavie CJ. Alcohol and CV health: Jekyll and Hyde J-curves. *Prog Cardiovasc Dis*;2018;61(1):68–75.
8. Kalinowski A, Humphreys K. Governmental standard drink definitions and low-risk alcohol consumption guidelines in 37 countries. *Addiction*;2016;111(7):1293–1298.
9. Lieber CS. The Discovery of the Microsomal Ethanol Oxidizing System and Its Physiologic and Pathologic Role. *Drug Metabolism Reviews*;2004;36(3-4):511-529.
10. Lu D, Ma Y, Zhang W, et al. Knockdown of cytochrome P450 2E1 inhibits oxidative stress and apoptosis in the cTnT(R141W) dilated cardiomyopathy transgenic mice. *Hypertension*;2012; 60(1):81–89.
11. Fernández-Solà J. Cardiovascular risks and benefits of moderate and heavy alcohol consumption. *Nat Rev Cardiol*;2015;12(10):576–587.
12. Goel S, Sharma A, Garg A. Effect of alcohol consumption on cardiovascular health. *Curr Cardiol Rep*;2018;20(4):19. DOI: 10.1007/s11886-018-0962-2 . Accessed May 19, 2022
13. Di Castelnuovo A, Costanzo S, Bagnardi V, Donati MB, Iacoviello L, de Gaetano G. Alcohol dosing and total mortality in men and women: an updated meta-analysis of 34 prospective studies. *Arch Intern Med*;2006;166(22):2437-2445.
14. http://www.npr.org/sections/thesalt/2013/04/08/176587506/arsenic-in-beer-may-come-from-widely-used-filtering-process (accessed 04/28/2022)
15. Maisch B. Alcoholic cardiomyopathy: the result of dosage and individual predisposition. *Herz*; 2016;41(6):484-493.
16. Roerecke M, Rehm J. Irregular heavy drinking occasions and risk of ischemic heart disease: a systematic review and meta-analysis. *Am J Epidemiol*; 2010;171(6):633 –644.
17. James TN, Bear ES. Effects of ethanol and acetaldehyde on the heart. *Am Heart J*;1967;74(2): 243-255.
18. Gould L. Cardiac effect of alcohol. *Am Heart J*;1970;79(3):422-425.
19. Elliott P, Andersson B, Arbustini E, et al. Classification of the cardiomyopathies: a position statement from the European Society Of Cardiology Working Group on Myocardial and Pericardial Diseases. *Eur Heart J*;2008;29(2):270-276.
20. Maron BJ, Towbin JA, Thiene G, et al. Contemporary definitions and classification of the cardiomyopathies: an American Heart Association Scientific Statement from the Council on Clinical Cardiology, Heart Failure and Transplantation Committee; Quality of Care and Outcomes Research and Functional Genomics and Translational Biology Interdisciplinary Working Groups; and Council on Epidemiology and Prevention. *Circulation*;2006;113(14):1807-1816.
21. Haissaguerre M, Fleury B, Gueguen A, et al. [Mortality of dilated myocardiopathies as a function of continuation of alcohol drinking. Multivariate analysis concerning 236 patients]. *Presse Med*; 1989;18(14):711-714.
22. Gavazzi A, De Maria R, Parolini M, Porcu M. Alcohol abuse and dilated cardiomyopathy in men. *Am J Cardiol*;2000;85(9):1114-1118.
23. Fauchier L, Babuty D, Poret P, et al. Comparison of long-term outcome of alcoholic and idiopathic dilated cardiomyopathy. *Eur Heart J*;2000;21(4):306-314.
24. Prazak P, Pfisterer M, Osswald S, Buser P, Burkart F. Differences of disease progression in congestive heart failure due to alcoholic as

compared to idiopathic dilated cardiomyopathy. *Eur Heart J*;1996;17(2):251-257.
25. Askanas A, Udoshi M, Sadjadi SA. The heart in chronic alcoholism: a noninvasive study. *Am Heart J*;1980;99(1):9-16.
26. Lazarević AM, Nakatani S, Nesković AN, et al. Early changes in left ventricular function in chronic asymptomatic alcoholics: relation to the duration of heavy drinking. *J Am Coll Cardiol*; 2000;35(6):1599-1606.
27. Kino M, Imamitchi H, Morigutchi M, Kawamura K, Takatsu T. Cardiovascular status in asymptomatic alcoholics, with reference to the level of ethanol consumption. *Br Heart J*;1981; 46(5):545-551.
28. Fernandez-Sola J. The effects of ethanol on the heart: alcoholic cardiomyopathy. *Nutrients*;2020: 12(2):572. DOI: 10.3390/nu12020572. Accessed May 19, 2022
29. Guzzo-Merello G, Cobo-Marcos M, Gallego-Delgado M, Garcia-Pavia P. Alcoholic cardio-myopathy. *World J Cardiol*;2014;6(8):771-781.
30. Fernández-Solà J, Nicolás JM, Oriola J, et al. Angiotensin-converting enzyme gene polymorphism is associated with vulnerability to alcoholic cardiomyopathy. *Ann Intern Med*; 2002;137(5 Part 1):321-326.
31. Kupari M, Koskinen P, Suokas A. Left ventricular size, mass and function in relation to the duration and quantity of heavy drinking in alcoholics. *Am J Cardiol*;1991;67(4):274–279.
32. Steinberg JD, Hayden MT. Prevalence of clinically occult cardiomyopathy in chronic alcoholism. *Am Heart J*;1981;101(4):461–464.
33. Abe H, Kawano Y, Kojima S, et al. Biphasic effects of repeated alcohol intake on 24-hour blood pressure in hypertensive patients. *Circulation*;1994;89(6):2626–2633.
34. Wigle DA, Pang SC, Sarda IR. Acute ethanol ingestion modifies the circulating plasma levels of atrial natriuretic peptide. *Alcohol*;1993;10(4):275–280.
35. Guillaume P, Jankowski M, Gianoulakis C, Gutkowska J. Effect of chronic ethanol consumption on the atrial natriuretic system of spontaneously hypertensive rats. *Alcohol Clin Exp Res*;1996; 20(9):1653–1661.
36. Rupp H, Brilla CG, Maisch B. Hypertension and alcohol: Central and peripheral mechanisms. *Herz*;1996;21(4):258–264.
37. MacKenzie J. *The Study of the Pulse*. Pentland YJ ed., Edinburgh;1902
38. Wilke A, Kaiser A, Ferency I, Maisch B. Alcohol and myocarditis. *Herz*;1996;21(4):248–257.
39. Vasiljevic JD, Kanjuh V, Seferovic P, Sesto M, Stojsic D, Olse EG. The incidence of myocarditis in endomyocardial biopsy samples from patients with congestive heart failure. *Am Heart J*;1990; 120(6 Pt 1):1370–1377.
40. Bernstein IM, Webster KH, Williams RC, Strickland RG. Reduction in circulating T lymphocytes in alcoholic liver disease. *Lancet*;1974;2(27879):488–490.
41. Arbabi S, Garcia I, Bauer GJ, Maier RV. Alcohol (ethanol) inhibits IL-8 and TNF: role of the p38 pathway. *J Immunol*;1999;162(12):7441–7445.
42. Cook RT. Alcohol abuse, alcoholism, and damage to the immune system–a review. *Alcohol Clin Exp Res*;1998;22(9):1927–1942.
43. Klatsky AL, Friedman GD, Siegelaub AB. Alcohol consumption before myocardial infarction. Results from the Kaiser-Permanente epidemiologic study of myocardial infarction. *Ann Intern Med*;1974;81(3):294–301.
44. Gaziano JM, Buring JE, Breslow JL, et al. Moderate alcohol intake, increased levels of high-density lipoprotein and its subfractions, and decreased risk of myocardial infarction. *N Engl J Med*;1993;329(25):1829–1834.
45. Ridker PM, Vaughan DE, Stampfer MJ, Ridker PM, Vaughan DE, Stampfer MJ. Association of moderate alcohol consumption and plasma concentration of endogenous tissue-type plasminogen activator. *JAMA*;1994;272(12):929–933.
46. Mennen LI, Balkau B, Vol S, Cacès E, Eschwège E. Fibrinogen: A possible link between alcohol consumption and cardiovascular disease? *Arterioscler Thromb Vasc Biol*;1999;19(4):887–892.
47. Renaud SC, Beswick AD, Fehily AM, Sharp DS, Elwood PC. Alcohol and platelet aggregation: The Caerphilly Prospective Heart Disease study. *Am J Clin Nutr*;1992:55(5):1012–1017.
48. Klatsky AL. Alcohol and cardiovascular diseases: A historical review and 2005 update, 2005 http://www.aim-digest.com/gateway/pages/heart/articles/CVD/klatsky1105.htm. Accessed May 2, 2002.
49. Patterson E, Dormer KJ, Scherlag BJ, Kosanke SD, Schaper J, Lazzara R. Long-term intra coronary ethanol administration

electrophysiologic and morphologic effects. *Alcohol*;1987;4(5): 375–384.
50. Greenspon AJ, Schaal SF. The "Holiday Heart": Electrophysiologic studies of alcohol effects in alcoholics. *Ann Intern Med*;1983;98(2):135–139.
51. Strasser RH, Nuchter I, Rauch B, Marquetant R, Seitz H. Changes in cardiac sinal transduction systems in chronic ethanol treatment preceding the development of alcoholic cardiomyopathy. *Herz*;1996;21(4):232–240.
52. Zilkens RR, Burke V, Hodgson JM, Barden A, Beilin LJ, Puddey IB. Red wine and beer elevate blood pressure in normotensive men. *Hypertension*; 2005;45(5):874-879.
53. Agewall S, Wright S, Doughty RN, Whalley GA, Duxbury M, Sharpe N. Does a glass of red wine improve endothelial function? *Eur Heart J*; 2000;21(1):74–78.
54. Libby P, Ridker PM, Hansson GK. Progress and challenges in translating the biology of atherosclerosis. *Nature*; 2011;473(7347):317–325.
55. Pahor M, Guralnik JM, Havlik RJ, et al. Alcohol consumption and risk of deep venous thrombosis and pulmonary embolism in older persons. *J Am Geriatr Soc*;1996;44(9):1030–1037.
56. Pomp ER, Rosendaal FR, Doggen CJ. Alcohol consumption is associated with a decreased risk of venous thrombosis. *Thromb Haemost*;2008;99(1):59–63.
57. Hansen-Krone IJ, Braekkan SK, Enga KF, Wilsgaard T, Hansen JB. Alcohol consumption, types of alcoholic beverages and risk of venous thromboembolism—the Tromso Study. *Thromb Haemost*; 2011;106(2):272–278.
58. Furchgott RF, Vanhoutte PM. Endothelium-derived relaxing and contracting factors. *FASEB J*; 198;3(9):2007–2018.
59. Gkaliagkousi E, Ferro A. Nitric oxide signalling in the regulation of cardiovascular and platelet function. *Front. Biosci*; 2011;16(5):1873–1897.
60. Abou-Agag LH, Khoo NK, Binsack R, et al. Evidence of cardiovascular protection by moderate alcohol: Role of nitric oxide. *Free Radic Biol Med*;2005;39(4):540–548.
61. Husain K, Vazquez-Ortiz M, Lalla J. Down regulation of aortic nitric oxide and antioxidant systems in chronic alcohol-induced hypertension in rats. *Hum Exp Toxicol*; 2007;26(5):427–434.
62. Matsuo S, Nakamura Y, Takahashi M, et al. Effect of red wine and ethanol on production of nitric oxide in healthy subjects. *Am J Cardiol*; 2001;87(8):1029–1031.
63. Xue L, Xu F, Meng L, et al. Acetylation-dependent regulation of mitochondrial ALDH2 activation by SIRT3 mediates acute ethanol-induced eNOS activation. *FEBS Lett*; 2012;586(2): 137–142.
64. Kuhlmann CR, Li F, Ludders DW, et al. Dose-dependent activation of $Ca^{2+}$-activated $K^{+}$ channels by ethanol contributes to improved endothelial cell functions. *Alcohol Clin Exp Res*; 2004:28(7): 1005–1011.
65. Altura BM, Zou LY, Altura BT, Jelicks L, Wittenberg BA, Gupta RK, Beneficial vs. detrimental actions of ethanol on heart and coronary vascular muscle: Roles of $Mg^{2+}$ and $Ca^{2+}$. *Alcohol*; 1996; 13(5):499–513.
66. Rocha JT, Hipolito UV, Callera GE, et al Ethanol induces vascular relaxation via redox-sensitive and nitric oxide-dependent pathways. *Vasc Pharmacol*;2012;56(1-2):74–83.
67. Yogi A, Callera GE, Hipolito UV, Silva CR, Touyz RM, Tirapelli, CR. Ethanol-induced vasoconstriction is mediated via redox-sensitive cyclo-oxygenase-dependent mechanisms. *Clin Sci*;2010;118(11):657–668.
68. Romani A, Scarpa A. Regulation of cell magnesium. *Arch Biochem Biophys*;1992;298(1):1-12.
69. Romani AM. Magnesium in health and disease. *Met Ions Life Sci*;2013;13:49-79.
70. Romani A, Scarpa A. Hormonal regulation of $Mg^{2+}$ in the heart. *Nature*;1990;346(6287):841-844.
71. Romani AMP. Effect of acute and prolonged alcohol administration on $Mg^{2+}$ homeostasis in cardiac cells. *Alcohol*;2015;49(3):265-273.
72. Nguyen H, Romani AMP. Effect of alcohol administration on Mg2+ homeostasis in H9C2 cells. *J Cardiovasc Dis Diagn*;2014;2(6):179. doi: 10.4172/2329-9517.1000179. Accessed May 21, 2022
73. Tzivoni D, Banai S, Schuger C, et al. Treatment of torsade de point with magnesium sulfate. *Circulation*;1988;77(2):392-397.
74. Brown RA, Crawford M, Natavio M, Petrowski P, Ren J. Dietary magnesium supplementation attenuates ethanol-induced myocardial dysfunction. *Alcohol Clin Exp Res*;1998;22(9):2062-2072.
75. Connor JP, Huber PS, Hall WD. Alcohol use disorders. *Lancet*;2016;387(10022):988–998.
76. Parker R, Armstrong MJ, Corbett C, Day EJ, Neuberger JM. Alcohol and substance abuse in

solid-organ transplant recipients. *Transplantation*;2013:96(12):1015-1024.