Published in final edited form as:
J Hepatol. 2012 August ; 57(2): 384–391. doi:10.1016/j.jhep.2012.03.024.

Modest alcohol consumption is associated with decreased prevalence of steatohepatitis in patients with nonalcoholic fatty liver disease (NAFLD)

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Abstract

Background & Aims—Nonalcoholic fatty liver disease (NAFLD) is a cardiovascular risk factor. Although modest alcohol consumption may reduce the risk for cardiovascular mortality, whether patients with NAFLD should be allowed modest alcohol consumption remains an important unaddressed issue. We aimed to evaluate the association between modest alcohol drinking and nonalcoholic steatohepatitis(NASH), among subjects with NAFLD.

Methods—In a Cross-sectional analysis of adult participants in the NIH NASH Clinical Research Network, only modest or non-drinkers were included: participants identified as 1) drinking > 20gm/day, 2) binge drinkers, or 3) non-drinkers with previous alcohol consumption

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were excluded. The odds of having a histological diagnosis of NASH and other histological features of NAFLD were analyzed using multiple ordinal logistic regression.

Results—The analysis included 251 lifetime non-drinkers and 331 modest drinkers. Modest drinkers compared to nondrinkers had lower odds of having a diagnosis of NASH (Summary odds ratio 0.56, 95%CI 0.39–0.84, p=0.002). The odds of NASH decreased as the frequency of alcohol consumption increased within the range of modest consumption. Modest drinkers also had significantly lower odds for fibrosis (OR 0.56 95%CI 0.41–0.77) and ballooning hepatocellular injury (OR 0.66 95%CI 0.48–0.92) than lifetime non-drinkers.

Conclusions—In a large, well-characterized population with biopsy-proven NAFLD, modest alcohol consumption was associated with lesser degree of severity as determined by lower odds of the key features that comprise a diagnosis of steatohepatitis, as well as fibrosis. These findings demonstrate the need for prospective studies and a coordinated consensus on alcohol consumption recommendations in NAFLD.

Keywords

nonalcoholic fatty liver disease; nonalcoholic steatohepatitis; alcohol; liver biopsy

INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is the most common liver disease in the United States (U.S.) affecting as many as one third of adults[1]. Only a small subset of patients with NAFLD, namely those with a more severe subtype known as steatohepatitis (NASH), which is characterized by inflammatory infiltrates, ballooning hepatocellular injury, and fibrosis in addition to steatosis, are thought to be at risk for cirrhosis related mortality.

The metabolic risk factors for NAFLD are also closely associated with coronary heart disease (CHD) [2]. Patients with NAFLD[3], and especially those with NASH[4], are at risk for coronary heart disease. Patients with NAFLD are approximately two times more likely to die from coronary heart disease than liver disease[5]. Therefore management of CHD risk in patients with NAFLD is imperative. CHD risk can be modified through lifestyle changes, including diet, exercise and smoking cessation. In addition, modest alcohol consumption has been shown to reduce the risk of coronary heart disease mortality and improve metabolic risk factors related to both coronary heart disease and NAFLD [6, 7]. As many as 50% of the adults in the United States regularly consume a modest amount of alcohol[8]. Excessive alcohol, however, can cause alcoholic liver disease[9]. In the general population, the daily threshold [10–12] of alcohol for liver injury is thought to be between 1–3 drinks per day in women and 2–3 in men. In patients with metabolic risk factors for NAFLD, the threshold may be lower [13]. Despite this, cross-sectional studies have suggested that modest alcohol consumption may protect the liver from NASH and NAFLD [14, 15]. The relationship between modest alcohol consumption and NAFLD severity has not been analyzed in detail. Whether patients with NAFLD should abstain from alcohol or be allowed modest alcohol consumption remains an important question. In practice, physicians often recommend abstinence from alcohol for patients with NAFLD, although the data to support this approach are lacking.

To provide counseling on alcohol consumption for patients with NAFLD, it is important to know whether modest alcohol consumption is associated with NAFLD disease severity. We hypothesize that modest alcohol consumption is associated with lower prevalence of NASH in patients with NAFLD. The primary aim of this study was to investigate a potential association between modest alcohol drinking and steatohepatitis in patients with NAFLD.

Secondary aims were to test the association between modest alcohol drinking and the individual histological features of NAFLD including fibrosis.

METHODS

Study Sample

This was a cross-sectional study of the association between modest alcohol consumption and the histological presence and / or severity of recognized lesions in NAFLD. We included baseline data from participants 21 years or older enrolled in two recently published NASH Clinical Research Network (CRN) studies: (1) a cohort study, the NAFLD Database[16]; and (2) a clinical trial, Pioglitazone versus Vitamin E versus Placebo for the Treatment of Non-diabetic Patients with Nonalcoholic Steatohepatitis (PIVENS; Clinical Trial number NCT00063622) [17]. For the NAFLD Database, inclusion required histological diagnosis of NAFLD, imaging suggestive of NAFLD, histological diagnosis of cryptogenic cirrhosis, or clinical evidence of cryptogenic cirrhosis. Patients were excluded if they had other forms of liver diseases, or average alcohol consumption >20gm daily during the 2 years before entry. PIVENS inclusion additionally required patients to have histological evidence of NASH without cirrhosis and the absence of diabetes. Details of the study design can be found elsewhere [16, 17]. The dataset for the current analysis was made up of participants who had central review of pathology completed as of May 2010. The inclusion and exclusion flowchart is illustrated in Figure 1. A minimum age of 21 was chosen for inclusion in this analysis because it is the legal drinking age in the U.S. Participants who had liver biopsy more than 24 months before completing the Alcohol Use Disorders Identification Test (AUDIT) were excluded. Participants consuming on average more than 140g of alcohol per week were already excluded as a part of the enrollment criteria in the NASH CRN. In addition those reporting consumption of more than 2 drinks of alcohol in a typical drinking day and those reporting binge drinking at least once a month were excluded. In order to reduce potential selection bias that subjects may also stop drinking alcohol because of illness related to alcohol, non-drinkers who previously drank alcohol were also excluded. Finally, participants whose biopsy did not have at least 5% steatosis on central reading by the NASH CRN Pathology Committee were not considered to have NAFLD and were excluded from these analyses. Study protocols were approved by all participating center Institutional Review Boards. Each participant provided written informed consent.

Histological Features

The primary outcome for this analysis was the diagnosis of steatohepatitis, reported as none, borderline or definite, by the central review by the Pathology Committee. The assignment of a diagnostic category was based on consensus recognition of the global histological features including those characteristic of steatohepatitis including steatosis and ballooning hepatocellular injury with a zone 3 predominance as well as lobular inflammation.[18]. Secondary outcomes included the following histological variables: fibrosis (stage 0, 1, 2, 3, 4), steatosis (5–33%, 34–66%, >66%), lobular inflammation (<2, 2–4 and >4 under 20X magnification), portal inflammation (none, mild, more than mild), ballooning hepatocellular injury(none, few, many), microvesicular steatosis (absent, present), Mallory-Denk bodies (absent or rare, many), megamitochondria (absent or rare, many), acidophil bodies (absent or rare, many), large lipogranulomas (absent, present).

Alcohol Consumption

The primary exposure was modest alcohol consumption compared to lifetime abstinence from alcohol. Current alcohol consumption was assessed using the AUDIT[19]. Participants were asked “how often do you have a drink containing alcohol?” Those who responded “never” were considered non-drinkers. Participants were subsequently asked “how many

drinks containing alcohol do you have on a typical day when you are drinking?" Those who reported drinking more than 2 drinks on a drinking day were excluded. Participants were asked "how often do you have six or more drinks on one occasion". Those who reported binge drinking once monthly or more were excluded because previous publications have indicated that episodic heavy drinking as little as once a month is associated with fibrosis progression in patients with NAFLD[20].

Prior alcohol consumption was measured using the Lifetime Drinking History questionnaire[21]. Participants were asked "Over the course of your lifetime have you ever had at least one drink of alcohol, beer, liquor, wine, or wine coolers, per month during a 12-month time period, or at least three drinks per day for at least three consecutive days?" Non-drinkers who responded "yes" to this lifetime drinking history question were also excluded as being previous drinkers.

Social, Demographic and Lifestyle Confounders

Social, demographic and lifestyle confounders including age, gender, race, income, education and physical activity level were collected using standardized questionnaires. Race and ethnicity were categorized into Asian, Hispanic, non-Hispanic white, Pacific Islander and others. Annual household income was categorized into <$30,000, $30,000 to $50,000, or more than $50,000. Education was categorized into less than high school graduation, high school graduation, some college, and bachelor's degree or higher. Smoking history was categorized as never, previous and current smoker. Dietary variables include total calories per day, percent calories from carbohydrates and percent calories from fat. Number of METS per week for non-recreational activity and recreational activity was calculated from self-reported physical activity assessed using the Physical Activity Questionnaire[22]. Height and weight were measured in a standardized fashion and body mass index (BMI) was calculated as weight[kg]/height[m]^2.

Patatin-like Phospholipase Domain-containing Protein 3 (PNPLA3) Genotyping

Recently a nonsynonymous sequence variation (rs738409) that substitutes methionine for isoleucine at codon 148 in the gene encoding patatin-like phospholipase domain-containing (PNPLA3) has been shown to be associated with hepatic steatosis [23], as well as the severity of histological injury in both NAFLD[24] and in alcoholic liver disease[25]. Therefore, we included these data to determine whether the relationship between modest alcohol consumption and the odds of having NASH differed by rs738409 genotype (CC, CG, GG). Genotyping for rs738409 was done using the Sequenom MassARRAY iPLEX Gold platform (Sequenom, Inc., San Diego, CA) and recently published by the NASH CRN[24].

Statistical Analysis

Most histological features were ordinally graded. To account for the ordinal nature of histological features, we used multiple ordinal logistic regression to address the association between the histological features and modest alcohol drinking. Under the proportional odds assumption, the odds ratio is the same regardless of how the histological features are dichotomized. The proportional odds assumption was verified using the score test. Multiple regression analyses adjusted for the social, demographic and lifestyle confounders listed above. Income, non-recreational and recreational activity data were missing in 37 participants. Rather than excluding these participants from statistical analysis, multiple imputation was used to replace each missing value with five imputed values in five complete datasets. Multiple regression analysis was performed on each of the five datasets and summary statistics were generated using the SAS procedure MIANALYSE. The dose response association of alcohol frequency and steatohepatitis was tested using Jonckheere-

Terpstra trend test, which is similar to the Cochran-Armitage trend test but allows the response to be ordinal rather that binomial. While there was no evidence of interaction, we performed a gender-based sub-analysis because there is a considerable body of literature describing gender-based differences as well as threshold differences in risk for liver injury between men and women. A PNPLA3 genotyped based sub-analysis was also performed to measure the effect of modest alcohol consumption in the CC, GC and GG genotype. SAS 9.1 (SAS Institute Inc., Cary. NC) was used for statistical analysis.

Sensitivity Analyses

In order to address potential biases, 5 sets of sensitivity analysis were performed. Participants might change their alcohol consumption over time. While the main analysis excluded participants with liver biopsy more than 24 months before completing the alcohol history, sensitivity analysis #1 further excluded participants with liver biopsy more than 12 months old. Current nondrinkers who previously stopped drinking may have different histology than lifetime non-drinkers. The main analysis excluded non-drinkers who previously had higher alcohol consumption. Sensitivity analysis #2 included the non-drinkers who were previous drinkers. The main analysis used multiple imputation to handle missing confounder variables. Sensitivity analysis #3 excluded participants with missing values on confounders rather than using multiple imputation. A person may discontinue drinking after a diagnosis of diabetes. The main analysis did not include the history of diabetes as a confounder because diabetes can potentially be in the causal pathway. Sensitivity analysis #4 additionally controlled for a history of diabetes, while sensitivity analysis #5 excluded participants with a history of diabetes.

RESULTS

Study Sample

The study sample included 252 lifetime non-drinkers and 331 modest drinkers enrolled in the NASH CRN studies with central pathology readings. The social, demographical, lifestyle and metabolic characteristics of the two groups are presented in Table 1. As compared to lifetime non-drinkers, modest drinkers were more likely to be male, have higher income and education, have higher insulin sensitivity and HDL, and less likely to have diabetes.

Prevalence of Histological Features in Lifetime Non-drinkers and Modest Drinkers

The frequency and adjusted odds ratio for each histological feature are presented in Table 2. The primary outcome, steatohepatitis, was present in 69.8% of lifetime non-drinkers and 53.2% of modest drinkers. Compared to nondrinkers, modest drinkers had 0.49 (95% CI 0.33 – 0.72) times the adjusted odds of having steatohepatitis, and 0.64 (95% CI 0.40 – 1.03) times the adjusted odds of having steatohepatitis or borderline steatohepatitis. The proportional odds assumption was verified using the Score test. The adjusted summary odds ratio was 0.52 (95% CI 0.36 – 0.76). Refer to table 3 for the univariable and multivariable summary odds ratios adjusting for each confounder. In addition, the association was stronger in those who drank more frequently. For modest drinkers who drank ≤ once weekly vs. ≥ twice a week, the adjusted summary odds were 0.54 (95% CI 0.38 – 0.79) and 0.24 (95% CI 0.10 – 0.55) respectively. The Jonckheere-Terpstra Test for dose response was highly significant ($p < 0.0001$). In gender based sub-group analysis, male modest drinkers had 0.47 (95% CI 0.24 – 0.91) time the adjusted summary odds of having steatohepatitis, while female had 0.57 (95%CI 0.36 – 0.90) times the odds. For male modest drinkers who drank ≤ once weekly vs. ≥ twice a week, the adjusted summary odds were 0.49 (95% CI 0.25 – 0.97) and 0.24 (95% CI 0.07 – 0.79) respectively. For female modest drinkers, the odds were 0.55 (0.37 – 0.79) and 0.24 (0.1 – 0.55) respectively.

In addition, modest drinkers had significantly lower summary odds ratio than lifetime nondrinkers for fibrosis (OR 0.56 95% CI 0.41 – 0.78), ballooning hepatocellular injury (OR 0.62 95%CI 0.45 – 0.87) and Mallory-Denk bodies (OR 0.65, 95%CI 0.43 – 0.97). In contrast, there were no significant differences in degree of macrovesicular steatosis (p=0.35) or lobular inflammation (p=0.46) and minimal differences in degree of portal inflammation (p=0.052).

In the five sets of sensitivity analysis, the odds ratios for all histological features were similar, except the results for Mallory-Denk bodies were not statistically significant in 3 of the sensitivity analysis (#1, 3 and 5). For example, adjustment for diabetes (sensitivity analysis #4) changed the odds ratio for steatohepatitis slightly from 0.52 (95% CI 0.36 – 0.76) to 0.58 (95% CI 0.41 – 0.84).

Data for the rs738409 SNP were available for 184 lifetime non-drinkers and 252 modest drinkers. The genotype distribution was not different (p = 0.82) based upon classification of drinking habits: lifetime non-drinkers (CC 27.2%, CG 42.4%, GG 30.4%) and modest drinkers (CC 29.0%, CG 43.2%, GG 27.8%). In sub-group analysis based on PNPLA3 genotype, the adjusted summary odds of having steatohepatitis in modest drinker compared to non-drinker was 0.28 (95% CI 0.10 – 0.72) in CC, 0.40 (95% CI 0.19 – 0.86) in GC, and 0.39 (95% CI 0.16 – 0.93) in GG.

DISCUSSION

We studied the association of modest alcohol consumption and steatohepatitis in a sample of well-characterized study participants with biopsy-proven NAFLD from referral centers across the U.S. These data suggest that among subjects with biopsy-proven NAFLD, modest alcohol consumption up to 2 drinks per day was associated with half the odds of steatohepatitis. Modest drinkers also had a lesser severity of fibrosis and ballooning hepatocellular injury. Notably, a dose response was observed; among this overall group of participants who drank ≤ 2 drinks on a drinking day, those who drank more often appeared to have more protection. These associations were persistent for both men and women, as well as for all rs738409 genotypes.

Excessive alcohol consumption is a well known cause of alcoholic liver disease. Data from prospective cohort studies showed that the threshold for alcohol consumption to increase the risk for cirrhosis or cirrhosis- related mortality is 2 – 3 drinks for men and 1 – 3 drinks for women daily[10–12]. In the Copenhagen Heart Study, the threshold for increased cirrhosis-related hospitalization or death was 1 to 2 drinks per day for women and 2 to 3 drinks per day for men[11]. In the Cancer Prevention Study – II, the threshold for increased cirrhosis related death was 2 to 3 drinks per day for both men and women [10]. In the Nurse's health study, the threshold was 3 or more drinks per day in woman [12]. Modest alcohol consumption has been shown to ameliorate metabolic risk factors for NAFLD, possibly through a protective mechanism on insulin resistance[6, 7]. A number of studies have suggested that modest alcohol consumption may be protective against liver disease[15]. A "J" shaped association between alcohol and cirrhosis risk was suggested in the Copenhagen study [11] and the Nurse's Health Study [12]. In NHANES III, modest drinkers (up to 10g of alcohol / day) had a lower prevalence of suspected NAFLD than non-drinkers[14]. Importantly, this association was mainly attributable to wine and not other forms of alcohol.

Patients with NAFLD, and especially NASH, are at risk for CHD[2]. CHD is among the two most common causes of death in patients with NAFLD[3, 4]. Given the risk, clinical care must address CHD risk in addition to cirrhosis risk. Coronary heart disease risk can be modified through lifestyle changes, including diet, exercise and smoking cessation. In

addition, modest alcohol consumption may be beneficial to both cardiovascular risk and liver histology. The survival benefit of modest alcohol consumption has been demonstrated by a meta-analysis of 34 prospective studies including over one million subjects. Modest alcohol consumption, up to 2 drinks a day in women and 3 drinks a day in men, was associated with a relative risk of 0.82 and 0.83 for overall mortality respectively[26]. This benefit was even greater in patients with diabetes[27]. Notably, initiating modest alcohol consumption can modify CHD risk. Data from the Atherosclerosis Risk in Communities study showed that lifetime nondrinkers who began moderate alcohol consumption lowered their risk of cardiovascular event by 38%.[28]

Despite the potential benefit of moderate alcohol consumption in some settings, even a small amount of alcohol can aggravate the risk of certain diseases such as breast cancer [29]. Certain conditions, such as hepatitis C, may not be compatible with even a small amount of alcohol [30]. In patients with NASH cirrhosis, social drinking may be associated with increased incidence of hepatocellular carcinoma [31]. The potential risk for developing alcoholism should also be considered when counseling patients about initiating or maintaining modest alcohol use. It has been estimated that when non-drinkers begin alcohol consumption, 94% would start modest drinking and 6% would start heavier drinking[28]. Over 24 years, only 2% of modest drinkers subsequently developed alcoholism[32]. Whether a person should continue or start modest alcohol consumption must be determined on a case by case basis with careful consideration of the individual's risk profile.

The current study had a number of important methodological characteristics that allowed for accurate assessment of the association between modest alcohol drinking and biopsy diagnosis of steatohepatitis. Histology is the reference standard for NASH. The histology was reviewed together by a committee of 9 pathologists specialized in NAFLD diagnosis to minimize potential misclassification bias. Modest drinkers and nondrinkers often differ in socioeconomic factors. To adjust for confounders, social, demographic and lifestyle covariates were entered into multiple regression analysis. Nondrinkers may have stopped drinking due to health issues related to alcohol. To avoid selection bias, non-drinkers who previously consumed alcohol were excluded. This minimized, although did not completely exclude, the possibility that self-report of modest alcohol drinking was a surrogate marker of other unmeasured lifestyle factors. The main findings were sufficiently robust that the result remained unchanged after 5 sets of sensitivity analysis.

There were, however, a number of limitations in the current study. Alcohol consumption was assessed using two widely used, validated questionnaires [33, 34]. Nevertheless, quantification of alcohol use by self-report may be inaccurate. The inclusion criteria were predicated upon having biopsy-proven NAFLD. The extent to which these data are generalizable to persons with undiagnosed NAFLD is unclear. Finally, the cross-sectional design cannot address temporal relationship or causality between modest alcohol consumption and steatohepatitis.

Conclusion

In a large, well-characterized population with NAFLD, modest alcohol consumption was associated with a significantly lower odds of biopsy-diagnosed NASH. Speculation regarding the role of modest alcohol consumption in prevention or treatment of NASH is tempting but premature. It is likely, however, that most non-cirrhotic patients with NAFLD who already drink modestly are not at risk for aggravating their liver disease. Whether a person with NAFLD should be abstinent or consume alcohol modestly needs to be evaluated individually. The provocative nature of these findings juxtaposed against the well-

established dangers of excessive alcohol consumption presents a need for future prospective studies and a coordinated consensus on alcohol consumption recommendations in NAFLD.

Acknowledgments

Funding: The Nonalcoholic Steatohepatitis Clinical Research Network (NASH CRN) is supported by the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) (grants U01DK061718, U01DK061728, U01DK061731, U01DK061732, U01DK061734, U01DK061737, U01DK061738, U01DK061730, U01DK061713), and the National Institute of Child Health and Human Development (NICHD). Several clinical centers use support from General Clinical Research Centers or Clinical and Translational Science Awards in conduct of NASH CRN Studies (grants UL1RR024989, M01RR000750, M01RR00188, UL1RR02413101, M01RR000827, UL1RR02501401, M01RR000065, M01RR020359). This study is supported in part by the Intramural Research Program of the National Cancer Institute. Other grant support include T32 DK07202 from National Institute of Health Nation Research Service Award, P60 MD00220 for the San Diego EXPORT Center from the National Center of Minority Health and Health Disparities, M01 RR000827 from the National Center for Research Resources for the General Clinical Research Center at UCSD, and DK080506 for the UCSD Digestive Diseases Research Development Center. The National Institute of Diabetes and Digestive and Kidney Diseases provided scientific advice for the design and conduct of the study; the collection, management, analysis, and interpretation of the data; and the preparation, review, and approval of the manuscript and funding for the study. However, the contents of this work are solely the responsibility of the authors and do not necessarily represent the official views of the National Institutes of Health.

Abbreviations

AUDIT	Alcohol Use Disorders Identification Test
CHD	Coronary Heart Disease
CI	confidence interval
NAFLD	nonalcoholic fatty liver disease
NASH	nonalcoholic steatohepatitis
NASH CRN	Nonalcoholic Steatohepatitis Clinical Research Network
OR	odds ratio

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Potential Conflict of Interest: None

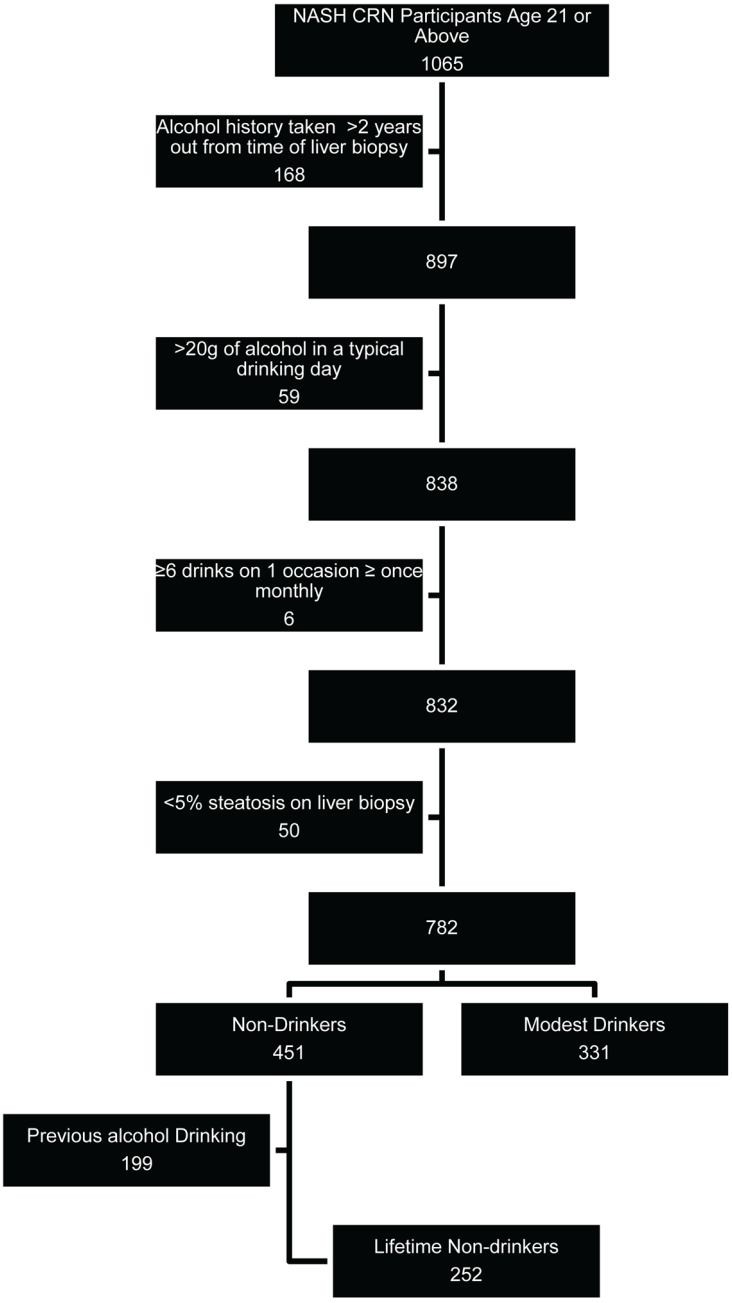

Figure 1.
Inclusion and Exclusion Flow Chart.

Table 1

Social, Demographic, Lifestyle and metabolic characteristics of modest drinkers and lifetime non-drinkers

	Life time non-drinkers (N= 251)	Modest Drinkers (N = 331)	p
Age, mean (SD), year	49.0(12.0)	47.7(11.7)	0.16
Gender, n (%)			0.007
Male	70(27.9)	128(38.7)	
Female	181(72.1)	203(61.3)	
Race, n (%)			0.12
White	179(71.3)	261(78.9)	
Hispanic	36(14.3)	34(10.3)	
Asians or Pacific Islander	15(6.0)	20(6.0)	
Other	21(8.4)	16(4.8)	
Income, n (%)			0.001
<30K	68(27.4)	48(14.8)	
30–50K	58(23.4)	64(19.7)	
>50K	122(49.2)	213(65.5)	
Education, n (%)			<0.001
<High School	39(15.6)	11(3.3)	
High School Graduation	73(29.2)	61(18.4)	
Some College	84(33.6)	117(35.4)	
Bachelor or Higher	54(21.6)	142(42.9)	
Diabetes, n (%)	94(37.5%)	73(22.1%)	<0.001
BMI, mean (SD), kg/m^2	35.1(7.0)	33.8(6.2)	0.04
Waist, mean (SD), cm			0.17
Male	113(15)	110(12)	
female	107(14)	106(14)	
Hip, mean (SD), cm			0.23
Male	113(12)	112(10)	
Female	120(16)	118(15)	
WHR,, mean (SD)			0.77
Male	1.00(0.06)	0.98(0.06)	
female	0.90(0.07)	0.90(0.07)	
SBP, mean (SD), mmHg	133(17)	132(14)	0.55
DBP, mean (SD), mmHg	76(11)	77(10)	0.24
triglyceride, mean (SD), mg/dl	187(155)	178(119)	0.51
HDL, mean (SD), mg/dl			0.007
Male	37(10)	41(13)	
Female	47(11)	48(13)	
QUICKI, mean (SD)	0.31(0.03)	0.31(0.03)	0.03
Work Related Activity, mean (SD), METS /wk	80(43)	83(39)	0.52

	Life time non-drinkers (N= 251)	Modest Drinkers (N = 331)	p
Non-work Related Activity, mean (SD), METS / wk	35(29)	39(39)	0.12
Smoking			0.02
Never	160(63.7)	171(52.1%)	
Past	69(27.5%)	121(36.9%)	
Current	22(8.8%)	36(11.0%)	
Total Calorie	1756(923)	1898(869)	0.57
%Carbohydrate	49.3(9.5)	46.0(8.3)	<0.001
%fat	37.6(7.9)	39.4(7.3)	0.006
%protein	15.1(3.6)	15.9(2.9)	0.004

Table 2

Prevalence and Adjusted* Odds ratio for Histological Features in Lifetime non-drinkers and Modest Drinkers

		Life Time Non Drinker	Modest Drinker	Adjusted Summary Odds Ratio (95% Confidence Interval)	p
Steatohepatitis	None	16.3%	22.7%	0.52(0.36 – 0.76)	0.0006
	Borderline	13.9%	24.2%		
	Definite	69.8%	53.2%		
Macrovesicular Steatosis	5–33%	44.8%	41.7%	1.11(0.79 – 1.55)	0.56
	34–66%	32.9%	35.6%		
	>66%	22.2%	22.7%		
Microvesicular Steatosis	Absent	86.1%	92.7%	0.57(0.31 – 1.06)	0.57
	Present	13.9%	7.3%		
Lobular Inflammation*	<2 foci	52.2%	51.1%	1.09(0.77 – 1.55)	0.63
	2–4 foci	37.3%	36.0%		
	>4 foci	10.7%	13.0%		
Portal Inflammation	None	9.5%	18.7%	0.69(0.48 – 1.00)	0.052
	mild	64.3%	62.8%		
	>mild	26.2%	18.4%		
Fibrosis Stage†	0	19.9%	30.0%	0.56(0.41 – 0.78)	0.0005
	1	24.7%	33.6%		
	2	22.3%	15.6%		
	3	20.3%	14.7%		
	4	12.7%	6.1%		
Ballooning Degeneration	None	25.8%	37.8%	0.62(0.45 – 0.87)	0.006
	Few	26.2%	26.3%		
	Many	48.0%	36.0%		
Mallory-Denk bodies	Rare/absent	65.1%	74.9%	0.65(0.43 – 0.97)	0.04
	Many	34.9%	25.1%		
Acidophil Bodies	Rare/absent	71.0%	68.3%	1.16(0.79 – 1.72)	0.46
	Many	29.0%	31.7%		
Large Lipogranulomas	Absent	57.5%	61.3%	0.73(0.50 – 1.07)	0.11

		Life Time Non Drinker	Modest Drinker	Adjusted Summary Odds Ratio (95% Confidence Interval)	p
	Present	42.5%	38.7%		
Megamitochondria	Rare/absent	81.7%	87.0%	0.71(0.43 – 1.18)	0.19
	Many	18.3%	13.0%		

Multivariate models adjusted for gender, age, race, income, education BMI, recreational and non-recreational physical activity, smoking, total calories per day, percent calories from carbohydrates and percent calories from fat

*Foci per 200x field

†Fibrosis stage 0 = none, stage 1 = perisinusoidal or periportal, stage 2 = perisinusoidal and portal / periportal, stage 3 = bridging fibrosis, stage 4 = cirrhosis

Table 3

Summary Odds Ratio for Steatohepatitis in Modest Drinkers compared to Lifetime non-drinkers, adjusting for each confounders

Model	Summary Odds Ratio
Univariable	0.53(0.45 – 0.61)
Adjusted for	
Gender	0.55(0.47 – 0.64)
Age	0.53(0.46 – 0.62)
Race / Ethnicity	0.53(0.45 – 0.61)
Income	0.54(0.47 – 0.63)
Education	0.54(0.46 – 0.63)
Body Mass Index	0.53(0.45 – 0.61)
Work & non-work related physical activity	0.53(0.46 – 0.62)
Smoking	0.51(0.44 – 0.60)
Total Calories, % carbohydrate, % fat	0.52(0.44 – 0.60)
All of the above confounders	0.52(0.36 – 0.76)