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Effects of Aspirin on Gastroduodenal Permeability in Alcoholics and Controls

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Abstract

Alcohol and non-steroidal anti-inflammatory drugs (NSAIDs) are noxious agents that can disrupt the integrity of the gastroduodenal mucosal and damage the epithelial barrier, and lead to increased gastroduodenal permeability. Moreover, it is not uncommon that patients are exposed to these two barrier stressors at the same time. It is thus important to know how simultaneous exposure affects the gastroduodenal barrier, and acquiring that knowledge was the goal of this study. We used a

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method that has been widely used for the assessment of injury to the gastroduodenal barrier induced by these noxious agents – measurement of gastroduodenal permeability as indicated by urinary excretion of ingested sucrose. We used gas chromatography to measure the amount of sucrose excreted in the urine over the 5–12 h following ingestion of a bolus of sucrose. The 148 participants in the study included 92 alcoholics and 56 healthy controls. All study subjects had a baseline permeability test. To determine whether addition of a second noxious agent, in addition to chronic alcohol, further decreases gastroduodenal barrier integrity, a subset of 118 study subjects participated in another permeability test in which they were exposed to aspirin. For this test, participants ingested 1300 mg aspirin twice, 12 hours and 1 hour before the final permeability test. The baseline permeability test showed that alcoholics have significantly higher gastroduodenal permeability than controls. Aspirin caused a significant within group absolute increase in gastroduodenal permeability in both alcoholics and controls (+7.72%, $p=0.003$ and +2.25%, $p = 0.011$, respectively) but the magnitude of these increases were not significantly different from each other. Baseline permeability did vary by gender, self-reported illegal drug use, and employment type. The extent of the permeability increase after aspirin ingestion varied with illegal drug use and recruitment site (a surrogate marker of socioeconomic status). Our data show that alcoholics have greater gastroduodenal permeability than healthy controls. This difference was independent of the duration of any preceding period of sobriety, gender, smoking history, or illicit drug abuse. The injurious effects of alcohol on the gastroduodenal epithelial barrier are long lasting, persisting even after 7 days of sobriety. Although, acute aspirin and chronic alcohol each increase intestinal permeability in alcoholics, their effects appear to be additive rather than synergistic.

Keywords

ethanol; intestinal permeability; sucrose; gastroduodenal permeability; aspirin-induced gut leakiness; intestinal injury

Introduction

Quantifying changes in gastroduodenal permeability became more practical after Meddings et al introduced the sucrose permeability test in 1993 as a non-invasive measure for assessing the extent of gastroduodenal damage induced by non-steroidal anti-inflammatory agents (NSAIDs)(Meddings et al., 1993). Subsequently, several studies showed that abnormal gastroduodenal permeability to sucrose is a reasonable marker for the presence of gastroduodenal injury in NSAID users (DeMeo, 1995; Erlacher et al., 1998; Meddings et al., 1995; Smecuol et al., 2001; Sutherland et al., 1994). Other researchers used sucrose permeability tests to assess damage to the gastroduodenal mucosa induced by oral corticosteroids (Kiziltas et al., 1998), intense exercise (Lambert et al., 2007b; Pals et al., 1997; Smetanka et al., 1999), *Helicobacter pylori* infection (Borch et al., 1998; Goodgame et al., 1997), atrophic gastritis (Sjostedt Zsigmond et al., 2005), Crohn's disease (Wyatt et al., 1997), celiac disease (Smecuol et al., 1997; Vogelsang et al., 1996), coffee (Cibickova et al., 2004), smoking (Gotteland et al., 2002) or a combination of these damaging factors (Gotteland et al., 2002; Lambert et al., 2007a; Lambert et al., 2001; Rabassa et al., 1996; Ryan et al., 1996; Santucci et al., 1995).

Alcohol is another agent that disturbs the integrity of the gastrointestinal barrier. A few studies have shown that acute alcohol consumption increases gastroduodenal permeability (Gotteland et al., 2002; Keshavarzian et al., 1994; Robinson et al., 1981). In the study of Robinson et al, the effect of alcohol was reversible and the increase in gastrointestinal permeability resolved after 2–3 days (Robinson et al., 1981). The effects of chronic exposure to alcohol on gastroduodenal permeability are less well established. We previously showed that chronic alcohol consumption increases gastroduodenal permeability to sucrose (Keshavarzian et al.,

1999). However, another study did not find any difference in PEG 400 absorption among alcoholics compared to healthy controls (Parlesak et al., 2000).

These conflicting data could be due to the fact that other potential confounding factors have not been considered in either study. For example, the effects of gender or duration of pre-study sobriety on alcohol-induced gastroduodenal hyperpermeability tests are not known. Furthermore, alcoholics commonly consume other potentially injurious chemicals, such as illicit drugs and over the counter medications such as NSAIDs and aspirin products, which can also affect gastroduodenal permeability. Accordingly, in the present study, we investigated how exposure to both ethanol and NSAIDs affects gastroduodenal permeability. In addition, we investigated the effects of gender, age, body mass index (BMI), race, education, history of smoking, use of illicit drugs, and the duration of sobriety on gastroduodenal permeability.

Materials and Methods

The study was carried out after obtaining written, informed consent from each participant. The study was reviewed and approved by the Rush University Medical Center IRB.

Subjects

Subjects included 92 alcoholics and 56 healthy controls who had been recruited into a larger study on the impact of chronic alcohol consumption on intestinal barrier function and who had at least one permeability test. Subjects in the larger study were recruited from the Gastroenterology and Hepatology outpatient clinics of Rush University Medical Center, Cook County Hospital outpatient clinics, Chicago area homeless shelters or halfway houses, and Craig's List. A total of 130 alcoholics and 70 healthy controls were initially enrolled. Of the 200, 148 (92 alcoholics, 56 healthy controls) were eligible, chose to participate, and had a baseline GI permeability test. The reasons for exclusion included unwillingness to collect urine for the permeability test and the presence of exclusion criteria (see below). There were no differences in demographic data or alcoholism status between those who participated and those who were excluded. There was also no difference in the rate of participation between the control and alcoholic groups. Of the 148, 105 agreed to a second urine collection (2nd baseline test) that took place within one week, and again there was no difference in participation between alcoholic and control groups. Of the 148 subjects who had the first baseline permeability test, 118 were suitable and agreed to an aspirin challenge test. The overall non-participation rate for the aspirin challenge test was 20%. The reasons for non-participation in the aspirin challenge test included aspirin intolerance, low platelets, and/or a bleeding tendency, particularly in those with liver disease. Based on self-reports, all alcoholic subjects were adhering to complete alcohol abstinence for at least 7 days prior to the post-aspirin permeability test.

We recorded age, gender, BMI, race, years of education (as an indirect measure of socioeconomic status), smoking history, and self-reported use of illicit drugs. All subjects had blood drawn at the time of recruitment to assess biochemical markers of liver injury including serum aspartate amino transferase (AST), amino alanine transferase (ALT), alkaline phosphatase (AP), and total bilirubin (TB). To test intestinal permeability, all subjects were provided with a sugar mixture and a urine container for collection of urine.

Inclusion criteria and definitions

All alcoholic subjects fulfilled NIAAA (O'Connor and Schottenfeld, 1998) & DSM-IV criteria (Ball et al., 1997) for alcoholism. This included a recent drinking history (regular & heavy alcohol consumption for a minimum of 3 months prior to enrollment). A history of alcohol consumption was assessed by a validated NIAAA-endorsed assessment instrument – The

Lifetime Drinking History (Skinner and Sheu, 1982; Sobell et al., 1988). Sobriety was confirmed by the history and by a negative blood alcohol level. Liver disease (LD; regardless of etiology) was defined as the presence of elevated ALT or AST levels that were >1.5 times normal, or clinical or radiological (CT or ultrasound) evidence of LD. Although liver histology is ideal for establishing the presence of liver disease and the degree of liver injury and fibrosis, for ethical reasons, liver biopsy could not be done in alcoholics without biochemical or clinical evidence of liver disease or in those alcoholics with evidence of liver disease when histological diagnosis was not clinically indicated. Control subjects were otherwise healthy individuals with no known liver disease, who did not fulfill any of the exclusion criteria, and who were willing to participate in the study.

Exclusion criteria

Subjects recruited were excluded if they reported gastrointestinal diseases (except for hiatal hernia or hemorrhoids), clinically detectable ascites or severe edema, evidence of ongoing infection, major depression or anxiety requiring therapy, clinically significant lung or heart disease, or recidivism (in alcoholics) during the study or regular use of NSAIDs or metoclopramide.

Test of intestinal permeability

All subjects ingested a mixture of sugars after at least an 8 hour fast. The mixture contained 4 sugars. This included 2 g of mannitol, 7.5 grams of lactulose, 1 g of sucralose, and 40 g of sucrose. The mixture was dissolved in 150 mL of tap water prior to ingestion. Each subject collected his/her own urine in containers that were pretreated with a sodium fluoride preservative to prevent sugar degradation by bacteria. For the aspirin challenge test, 4 tablets each containing 325 mg of aspirin were given 12 hours before ingestion of the sugar mixture and another 4 tables 1 hour before taking sugar drink

Standardization of intestinal permeability data from the first collection

At study onset, a 5-hour urine collection period was used for measuring gastroduodenal permeability. Of the 50 participants recruited up to that point, only 60% provided urine samples. In order to increase participation and to make urine collection easier for subjects, we changed our protocol to a 12-hour overnight urine collection. Participation increased to 80% through the remainder of the study. In order to analyze 5 hour and 12 hour data together, we calculated time-standardized permeability scores. We standardize the 5-h samples using the mean and standard deviation of the 5-h healthy control samples. The standardized scores were calculated by subtracting the measured value from the mean sucrose excretion for healthy controls and dividing by the healthy control standard deviation of sucrose excretion. Likewise, for individuals with 12-h urinary collections, the mean and standard deviations from 12-h healthy controls were used.

Sucrose excretion from the second collection and aspirin challenge

Of the 148 participants, a subset of 105 agreed to a second baseline permeability test one week after the first baseline study to determine reproducibility of the permeability test. Also, of the 148 participants, a subset of 118 subjects participated in the aspirin challenge test. Thus, of the 148 participants, 89 had complete data (two baseline permeability tests and an aspirin challenge permeability test). The second baseline permeability test and the aspirin challenge were done using the same collection period that each individual had used in the first baseline permeability test – 5 hours or 12 hours. This allows direct comparison of the pre-aspirin and post-aspirin urinary sucrose determinations from any individual subject.

Materials and Equipment

Materials, chemicals and chromatographic conditions were as previously described (Farhadi et al., 2006; Farhadi et al., 2003). We calculated sucrose concentrations in urine samples based on the slope and intercept of the line created by the readout of the sugar concentration in the sample before and after being spiked with a known amount of sucrose. In this method, each urine sample serves as its own blank and this decreases the variability in our readout (Farhadi et al., 2006). Using this method, the sugar concentration in each sample was equal to the intercept/coefficient ratio. The total amount of a given sugar in the 5-h and 12-h urine samples was expressed as a percentage of the amount of that sugar that was ingested in the oral dose.

Data Analysis

Data are presented either as counts and percents, mean $\pm$ standard deviation (SD) or medians (25th, 75th) percentiles (box plot, Fig.1). Differences in demographic data between the study groups were compared using chi-square tests or independent t-tests. As the time-standardized scores and raw percent of sucrose excretion were significantly skewed, data were analyzed univariately using Wilcoxon Rank Sum (WRS) tests, Kruskal-Wallis Tests (KW), or Spearman's correlation. To examine differences in groups adjusting for demographic factors, we first categorized sucrose excretion as high (defining patients as "leaky") based on the upper quartile value of healthy controls or normal and then we fit multivariate logistic regression models. Using the same criteria for defining "abnormal" permeability and based on the average value of the two baseline permeability data (when there were two sets of baseline permeability data) and percent changes in sucrose excretion after aspirin challenge, we labeled those with high sucrose excretion at baseline as "leaky" and those with more than a 50% increase after aspirin ingestion as "susceptible to leakiness," and ran multivariate logistic regression models to determine what might predict leakiness and/or susceptibility to leakiness. The Wald p-values from those models are presented along with Odds Ratios (OR) and 95% Confidence intervals (CI). We compared associations between percent change in sucrose excretion and demographics similarly. We also examined within-group changes using Wilcoxon Signed Rank (WSR) tests. All tests were run using a significance level of 0.05, and all analyses were run in SASv. 9.1 (Cary, NC).

Results

Demographics

The healthy controls were comparable to the alcoholics with respect to age. However, alcoholics were more likely to be male, have a higher BMI, and were less likely to be Caucasian or to have a college degree. They were more likely to be smokers or report ever using illegal drugs. Most healthy controls were employed as office or professional workers and were more likely to be recruited from Rush University Medical Center's outpatient clinics, whereas alcoholics were more likely to be unemployed or retired and were recruited from homeless shelters or halfway houses. Demographic information for the entire sample with permeability data and for the subset of data with complete permeability data is presented in Table 1.

Gastroduodenal Permeability

Baseline data—At baseline, the variable most strongly associated with permeability scores, regardless of study subject groups was employment status (KW test, $p = 0.016$); unemployed/retired or office workers/professionals exhibited more leakiness than did service or labor workers. The only other demographic variables that appeared to contribute to higher sucrose excretion were gender (males) and self-reported use of illegal drugs, although these showed borderline significance ($p = 0.048$ for both WRS tests). We then fit a multiple logistic regression model to examine if rates of leakiness varied after adjustment for these potential confounders

(gender, illegal drug use, and employment). We found that employment category was the only statistically significant predictor of high sucrose excretion in the multivariate model (Wald $p = 0.048$). Similar to the univariate models, "Unemployed or Retired" and "Office/Professional" workers both had almost 5 times the odds of having gut leakiness as did those in Service or Labor professions (OR and 95% CI: 4.92 (1.33, 18.13), 5.21 (1.26, 21.48), respectively). Although the majority of unemployed/retired subjects were alcoholics, the impact of employment status on permeability did not appear to be due to alcoholism alone because the effect of employment status remained significant even when the presence of alcohol use was included in the model.

The median baseline sucrose excretion from Week 1 was higher in alcoholics than in healthy controls (Fig. 1; WRS $p = 0.004$), as was the second sucrose excretion, which had been collected one week later (Fig. 2; WRS $p = 0.017$). Table 2 shows median sucrose excretion by demographic factors.

Aspirin Challenge Test—The aspirin challenge resulted in a significant increase in the median permeability of sucrose in both alcoholics and controls (Figure 1). Figure 1 depicts urinary sucrose data as "calculated standardized values" in order to correct for differences in duration of urine collection and demonstrates that gastroduodenal permeability is significantly increased in chronic alcoholics and after acute intake of aspirin. Figure 2 depicts % of an oral sucrose dose excreted in the 5 or 12 hour urine and demonstrates that the median raw sucrose excretion increased 2.25% for healthy controls and 7.72% for alcoholics. These within-groups absolute differences (not percent increases) were significant (WSR $p = 0.011$ and 0.003 , for controls and alcoholics, respectively) indicating that acute aspirin intake disrupts gastroduodenal barrier integrity in both healthy subjects and alcoholics. Although the median for aspirin-induced increases in urinary sucrose was higher in alcoholics (> 3-fold difference) than controls, the overall between-groups difference was not statistically significant (WRS $p = 0.393$). This suggests that the increased gastroduodenal permeability induced by aspirin challenge is at least additive but may not be synergistic with the effect of alcohol.

The change in sucrose excretion did not differ significantly with respect to any demographic characteristics with the exception of location of recruitment (KW $p = 0.017$). Post hoc analyses of location of recruitment show that there was significant increases in absolute sucrose excretion among those recruited from Rush or County Outpatient, and from Shelters, but not among those recruited from Craig's List. Table 3 shows change in sucrose excretion by all demographic factors.

We examined multivariate logistic regression models to more fully examine demographic relationships to sucrose excretion. First we labeled those who had high baseline sucrose excretion (highest quartile) as "leaky". Table 2 includes univariate associations with baseline "leakiness." We then, defined another group as "susceptible to leakiness" based on their response to aspirin challenge. This group experienced an increase in sucrose excretion of 50% or more after aspirin regardless of their baseline values. In order to avoid any ceiling effect (those who were leaky at baseline may not be able to increase leakiness by 50%), we fit logistic regression models to the outcomes "leaky or susceptible to leakiness" as well as "susceptible to leakiness". Table 3 includes univariate results from those models. Consistent with analyses of median sucrose excretion, employment was the strongest predictor of baseline leakiness. Self-reports of ever using illegal drugs, as well as location of recruitment predicted leakiness or susceptibility to leakiness, while illegal drug use was the lone predictor of susceptibility to leakiness among those who were not considered leaky at baseline. After adjusting for these factors, alcoholism was not predictive of leakiness after aspirin challenge (Wald $p = 0.753$), nor was it predictive of susceptibility to leakiness among those who were initially not leaky

(Wald $p = 0.998$). These data again suggest that the deleterious effects of chronic alcohol use and acute aspirin intake are more likely to be additive rather than synergistic.

Discussion

Our study is one of the few that shows the deleterious effects of chronic alcohol consumption and acute ingestion of aspirin on gastroduodenal mucosal barrier integrity. Our data also suggest that the effects of alcohol and aspirin were more likely to be additive rather than being synergistic. This could be particularly important for alcoholics who usually use aspirin and other NSAIDs for management of hangovers or headaches. However, the clinical implication of this potential additivity is not clear at this time. In particular, our study does not exclude the possibility that aspirin and other NSAIDs have synergistic deleterious effects with alcohol in actively drinking alcoholics, individuals who typically take these medications to treat hang over or symptoms of withdrawal. We could not answer this important clinical question because our Institutional Research Review Board required us to recruit only those alcoholics who did not drink during the 3 days prior to the aspirin challenge test in order to ensure their full understanding of the research protocol and that they were without signs and symptoms of alcohol withdrawal. Furthermore, we found no difference in response to aspirin challenge between alcoholics with or without liver disease. However, due to the small number of subjects with alcoholic liver disease, we still can not exclude the possibility that those alcoholics with liver disease may be more susceptible to the injurious effects of aspirin on intestinal barrier function. Indeed, we previously showed that patients with non-alcoholic steatohepatitis [NASH] are more susceptible to aspirin-induced intestinal hyperpermeability (Farhadi, 2008).

One important question is whether all alcoholics are prone to the deleterious effects of alcohol on the GI tract. Indeed, several studies, including our own (Keshavarzian 1999) have demonstrated that not all alcoholics develop tissue injury and organ dysfunction such as leaky gut. Thus there is a wide range of susceptibility among alcoholics to the injurious effects of chronic alcohol exposure (Maddrey, 2000). For example, it is now well established that gender affects susceptibility to alcohol injury. Females are more susceptible to the injurious effects of alcohol on the liver and to the development of cirrhosis – in females these occur after a shorter period and a smaller amount of drinking than in males (Maddrey, 2000). In contrast, we found that male subjects were more prone to having abnormally high gastroduodenal permeability. The mechanism of how gender affects susceptibility to the injurious effects of chronic alcohol consumption is not clear. Several studies suggest that there are modulatory effects of sex hormones on injury pathways (Sumida et al., 2007; Yokoyama et al., 2005). Further studies are needed to determine the mechanism of alcohol-induced disruption of gastroduodenal barrier integrity and the influence of gender on this process.

Other potential factors that could affect alcohol-induced tissue injury and organ dysfunction are socioeconomic status and the magnitude and pattern of drinking (Tao et al., 2003). We found that socioeconomic status of the subjects affected their susceptibility to gastroduodenal leakiness; those who were unemployed had more gastroduodenal leakiness. However, differences in permeability findings across employment categories may not be related only to socioeconomic status. In fact, differences in permeability could be associated with other factors such as stress, which can by itself increase intestinal permeability (Werther, 2000). Our findings regarding location of recruitment support this hypothesis. Those recruited from hospital outpatient clinics, as well as those from shelters may have more health or life-related stress than those recruited from Craig's list. We did not find any correlation between the severity of gastroduodenal hyperpermeability and pattern of drinking behavior (binge vs. daily drinking), duration of sobriety, or the duration or amount of daily drinking. For example, there was no significant difference in baseline gastroduodenal permeability values between those who quit drinking just before the study and those who had quit more than a week before the study. We

also did not observe any differences in gastroduodenal permeability for race, smoking, age or BMI.

Alcohol-induced gastroduodenal leakiness was persistent for at least 7 days. Repeating the gastroduodenal permeability test 7 days after the 1st baseline permeability test showed that the median sucrose permeability remained the same (WSR $p > 0.05$). In fact, alcoholics had a higher median sucrose excretion on the second permeability test after 1 week of reported abstinence. This finding is in contrast to the findings of a prior study that reported a short lived, 2–3 day long, gastrointestinal hyperpermeability in alcoholics (Robinson et al., 1981). Several mechanisms might explain this observation. Unreported recidivism can be considered as a possible cause in some of our subjects considering that most of our alcoholics were recruited from alcohol rehabilitation centers or clinics, even though, all of these patients claimed that they had stopped drinking alcohol before or at the time of recruitment. In our study, we measured serum alcohol at the time of recruitment and used a negative blood alcohol finding as an inclusion criterion. We did not confirm self-reported sobriety objectively throughout the study period. Another possibility is sucrose malabsorption due to a disaccharidase deficiency (i.e. sucrase) that has been observed with chronic alcohol use (Huang et al., 2005; Palacios et al., 1989; Perlow et al., 1977) and that can last for several weeks even when the injurious agents such as alcohol and bacteria are no longer present. Sucrase deficiency can hamper the ability of the body to digest and absorb sucrose in the proximal small bowel and thus, provide more sucrose for permeation over a larger area of the gastrointestinal tract i.e. the distal part of the small bowel and colon. This would lead to higher levels of sucrose in the blood and urine and would result in an overestimation of gastroduodenal permeability in chronic alcoholics. Lastly, the effects of alcohol on the intestine could be long lasting and affect even new epithelial cells and paracellular spaces that were not directly exposed to alcohol. Although, to the best of our knowledge, there are no reports on the effects of alcohol on intestinal stem cells, several studies have demonstrated that alcohol affects neural stem cells and affects neuronal development and regeneration (Morris et al., 2009). Furthermore, the prolonged effects of alcohol could be due to epigenomic effects. For example, we recently showed that microRNA 212, which suppresses production of the key tight junction protein ZO-1 (post-transcriptional modulation) is increased in the colonic mucosa of sober alcoholics (Tang et al., 2008). Further studies are needed to determine whether chronic alcohol consumption affects other microRNAs that control synthesis of proteins involved in regulation of gastroduodenal barrier function.

The mechanism of alcohol-induced gastroduodenal leakiness was not examined in our study. A previous report proposed that mucosal vascular injuries and ischemia are the main mechanisms underlying gastroduodenal mucosal injury due to high concentrations of acute alcohol (Szabo, 1987). In contrast, the mechanism underlying chronic alcohol-induced gastroduodenal hyperpermeability has not been fully elucidated. We showed that iNOS-mediated oxidative stress is the key mechanism for chronic alcohol-induced hyperpermeability of the small and large bowel (Tang et al., 2009) but it remains to be determined whether the same mechanism is also responsible for alcohol-induced hyperpermeability of the gastroduodenal mucosa.

In contrast to studies on the effects of alcohol on the gastroduodenal mucosal layer, many investigators have studied the mechanism of aspirin/NSAID-induced disruption of the gastroduodenal mucosal layer (Allen et al., 1986; Leung et al., 2009). Changes in prostaglandin homeostasis, mucosal blood flow, and mucus and oxidative stress are among the mechanisms that have been shown to be involved in NSAID-induced disruption of the gastroduodenal mucosal layer. It remains to be determined whether alcohol-induced disruption of gastroduodenal barrier integrity is also mediated through changes in prostaglandin homeostasis, mucosal blood flow, and mucus. Our observation that the injurious effects of chronic alcohol and acute aspirin on the gastroduodenal mucosal layer are more additive than

synergistic suggests that the mechanisms of the deleterious effects of these two injurious agents on gastroduodenal mucosa may share some components. This possibility should be investigated in future studies.

Our study has limitations. One of the major shortcomings is the possibility of recidivism in subjects who claimed to be abstaining from alcohol across multiple measurements. The other major limitation is the presence of two types of urine collection procedures in the study. To manage this issue we calculated time-standardized permeability scores. We also needed to consider and adjust for several variables since our alcoholics and controls were from different populations. At the same time, our study may not have had enough power to detect a meaningful difference in all of these variables among our study groups. Additionally, our study question on use of illegal drugs, did not address the length or frequency of use of these drugs. Also, because we were not able to study alcoholics within 3 days of drinking, we can not conclude with certainty that there is no synergy for the deleterious effects of aspirin and alcohol on gastroduodenal mucosal integrity in actively drinking alcoholics.

In conclusion, chronic alcohol consumption and acute aspirin ingestion increase gastroduodenal permeability. The effects of these two barrier stressors appear to be additive. These induced increases in permeability are not affected by age, gender, BMI, race or smoking, but are potentially affected by the socioeconomic status or stress levels of the subjects. The injurious effects of alcohol on gastroduodenal epithelial permeability appear to persist even after weeks of sobriety.

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Figure 1. The effect of chronic alcohol consumption and acute aspirin intake on gastroduodenal permeability

Gastroduodenal permeability (urinary sucrose excretion) for all study subjects (n=49 healthy controls and n=69 alcoholics) at baseline and after aspirin challenge are presented. Urinary sucrose excretion was determined by GC analysis after an oral sugar bolus as described in Methods. Data are presented as standardized scores (see Methods) of medians of percent of oral sucrose dose excreted in the urine (5 or 12 hours). Note that: 1) Alcoholics have significantly higher sucrose excretion (leaky gastroduodenal barrier) than Controls (*, $p=.004$); and 2) Aspirin induces a significant increase in gastroduodenal permeability in both Controls (**, $p=.011$) and Alcoholics (***, $p=.003$).

Figure 2. Stability of gastroduodenal permeability in alcoholic and healthy control subjects

Figure 2 depicts gastroduodenal permeability (urinary sucrose) for those subjects for whom two baseline measurements were taken one week apart (Weeks 1 & 2) and after aspirin challenge ($n=35$ healthy controls and $n=54$ alcoholics). Gastroduodenal permeability was determined as described in Fig 1. Data are presented as means of percent of oral sucrose dose excreted in the urine (5 or 12 hours). Data shown are non-standardized as the second baseline test (Week 2) was only available for those with 12 hour collections. Note that: 1) There were no statistically significant within-group differences between baseline time points (Weeks 1 & 2). However, there is more baseline variability in the Alcoholic group, although no correlation was found between permeability and length of sobriety. 2) Alcoholics had greater gastroduodenal permeability to sucrose at both baseline tests compared to Controls. 3) Overall, aspirin increased gastroduodenal permeability to sucrose in both Controls and Alcoholics, especially in those who had lower baseline values.

Table 1

Descriptive statistics of our study sample.

Characteristic	Total sample		Complete data (n=89)			
	Healthy Controls	Alcoholics	Significance	Healthy Controls	Alcoholics	Significance
Total n	56	92	---	35	54	---
2 nd Permeability test	37 (66%)	68 (74%)	0.308			
Aspirin Challenge	49 (88%)	69 (75%)	0.067			
Gender (Male)	22 (39%)	64 (70%)	<0.001	17 (49%)	44 (81%)	0.001
Age (Mean ± SD)	39.8 ± 12.8	41.9 ± 11.4	0.310	38.9 ± 12.0	41.0 ± 11.1	0.385
BMI (mean ± SD)	25.0 ± 3.5	28.2 ± 6.0	<0.001	25.4 ± 3.6	28.3 ± 6.5	0.007
Race (Caucasian)	31 (54%)	33 (36%)	0.023	16 (46%)	12 (22%)	0.020
Education level (college or higher)	31 (58%)	21 (23%)	<0.001	19 (54%)	8 (15%)	<0.001
Current Smokers	12 (21%)	60 (65%)	<0.001	10 (29%)	35 (65%)	<0.001
Current self-report drinkers	38 (68%)	53 (58%)	0.214	22 (63%)	32 (59%)	0.734
Self-report Illicit drug use (ever)	15 (27%)	75 (82%)	<0.001	13 (37%)	49 (91%)	<0.001
Employment						
Service/Labor	6 (11%)	18 (20%)		4 (11%)	11 (20%)	
Office/Professional	31 (59%)	16 (17%)		18 (52%)	8 (15%)	
Unemployed/retired	16 (30%)	57 (63%)	<0.001	13 (37%)	35 (65%)	0.001
Location of Recruitment						
Rush Outpatient	30 (54%)	14 (15%)		15 (43%)	5 (9%)	
County Outpatient	6 (11%)	14 (15%)		1 (3%)	4 (7%)	
Shelter	11 (19%)	56 (61%)		10 (28%)	37 (69%)	
Craig's List	9 (16%)	8 (9%)	<0.001	9 (26%)	8 (15%)	<0.001

Table 2

Univariate associations between demographics and sucrose excretion.

Characteristic	High Sucrose Permeability	p-value	Median Sucrose	P-value
Group				
Healthy Control	16 (29%)		−0.26 (−0.35, 0.01)	
Alcoholic	36 (39%)	0.192	0.08 (−0.34, 1.62)	0.004
Gender				
Male	29 (34%)		0.02 (−0.33, 1.29)	
Female	23 (37%)	0.671	−0.26 (−0.46, 0.71)	0.050
Age (Mean ± SD)	40.6 ± 11.4	0.697	Spearman's R = 0.049	0.551
BMI (mean ± SD)	27.1 ± 5.7	0.939	Spearman's R = 0.103	0.264
Race				
Caucasian	22 (34%)		−0.16 (−0.33, 0.58)	
Non-Caucasian	30 (36%)	0.824	0.01 (−0.43, 1.53)	0.362
Education level				
High School or less	33 (36%)		0.01 (−0.43, 1.36)	
College or higher	17 (33%)	0.701	−0.16 (−0.33, 0.45)	0.507
Non- smokers	24 (32%)		−0.20 (−0.34, 0.32)	
Current Smokers	28 (39%)	0.352	0.03 (−0.46, 1.60)	0.145
Current self-report drinkers	32 (35%)		−0.16 (−0.35, 0.87)	
Non-drinkers	20 (35%)	0.992	0.03 (−0.34, 1.11)	0.358
Self-report Illicit drug use				
Ever	34 (38%)		0.03 (−0.35, 1.56)	
Never	17 (30%)	0.361	−0.24 (−0.35, 0.26)	0.050
Employment				
Service/Labor	3 (13%)		−0.27 (−0.43, 0.30)	
Office/Professional	17 (36%)		−0.16 (−0.34, 0.08)	
Unemployed/retired	30 (41%)	0.037	0.12 (−0.35, 1.65)	0.016
Location of Recruitment				
Rush Outpatient	13 (30%)		−0.16 (−0.33, 0.20)	
County Outpatient	5 (25%)		−0.13 (−0.66, 0.72)	
Shelter	28 (42%)		0.18 (−0.46, 1.56)	
Craig's List	6 (35%)	0.422	−0.26 (−0.34, −0.12)	0.380

Table 3

Univariate associations between demographics and aspirin-induced sucrose excretion.

Characteristic	High or Susceptible to high Sucrose	p-value	Susceptible to high Sucrose	p-value	Median percent increase of sucrose excretion after aspirin	p-value
n	73/118		36/81			
Group						
Healthy Control	27 (55%)		14 (39%)		2.25 (-63.9, 156.0)	
Alcoholic	46 (67%)	0.203	22 (49%)	0.368	7.72 (-35.4, 120.6)	0.393
Gender						
Male	50 (67%)		25 (50%)		9.00 (-47.8, 150.8)	
Female	23 (53%)	0.156	11 (35%)	0.201	2.56 (-25.0, 128.4)	0.562
Age (Mean $\pm$ SD)	40.6 $\pm$ 11.4	0.697	39.6 $\pm$ 11.6	0.136	Spearman's R = 0.035	0.708
BMI (mean $\pm$ SD)	27.1 $\pm$ 5.7	0.939	27.8 $\pm$ 6.0	0.348	Spearman's R = 0.042	0.692
Race						
Caucasian	27 (53%)		13 (35%)		3.52 (-6.3, 68.6)	
Non-Caucasian	46 (70%)	0.064	23 (53%)	0.100	15.3 (-51.2, 231.0)	0.334
Education level						
High School or less	46 (63%)		23 (46%)		4.90 (-47.8, 152.5)	
College or higher	26 (60%)	0.785	13 (43%)	0.817	4.16 (-10.0, 128.4)	0.959
Non-smokers	35 (56%)		17 (38%)		2.25 (-61.5, 88.6)	
Current Smokers	38 (69%)	0.131	19 (53%)	0.177	17.96 (-35.4, 228.3)	0.093
Current drinkers	46 (61%)		20 (41%)		2.25 (-61.5, 96.1)	
Non-drinkers	27 (63%)	0.875	16 (50%)	0.416	20.00 (-10.0, 238.0)	0.064
Self-report Illicit drug use						
Ever	49 (71%)		24 (55%)		17.96 (-43.8, 150.8)	
Never	24 (49%)	0.015	12 (32%)	0.046	2.56 (-10.0, 85.7)	0.675
Employment						
Service/Labor	10 (48%)		7 (39%)		3.52 (-22.3, 68.9)	

Characteristic	High or Susceptible to high Sucrose	p-value	Susceptible to high Sucrose	p-value	Median percent increase of sucrose excretion after aspirin	p-value
Office/Professional	24 (58%)		10 (37%)		1.16 (-66.5, 88.6)	
Unemployed/retired	38 (70%)	0.160	19 (54%)	0.336	21.43 (-43.0, 228.3)	0.322
Location of Recruitment						
Rush Outpatient	17 (50%)		8 (32%)		2.41 (-6.3, 68.6)	
County Outpatient	6 (40%)		3 (25%)		9.00 (1.81, 231.0)	
Shelter	39 (75%)		20 (61%)		36.83 (-39.2, 226.2)	
Craig's List	11 (65%)	0.030	5 (45%)	0.075	-66.94 (-91.6, 88.6)	0.017