Subjective Response to Alcohol and *ADH* Polymorphisms in a Select Sample of Young Adult Male East Indians and Africans in Trinidad and Tobago

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ABSTRACT. Objective: Level of response to alcohol has been associated with risk of alcohol dependence in a number of ethnic groups. In the present study, subjective and objective responses to alcohol were evaluated in Indo-Trinidadians (Indo-T) and Afro-Trinidadians (Afro-T). Associations of alcohol dehydrogenase polymorphisms with response to alcohol, using the Subjective High Assessment Scale (SHAS), and breath alcohol concentrations (BrAC) were tested. **Method:** Regular male drinkers without alcohol dependence (n = 112) ages 18–25 years participated in alcohol challenge sessions consisting of placebo and two doses of alcohol (target BrAC: 0 g/dl for placebo, .04 g/dl low dose, and .08 g/dl high dose) and genotyped for variants in *ADH1B*3* and *ADH1C*2*. **Results:** Indo-T had significantly higher BrAC, pulse rates, and cortisol levels when compared with Afro-T but did not have significantly higher SHAS values. Higher responses on the SHAS items *muddle/confused*

and *nauseated* were significantly associated with the presence of at least one *ADH1B*3* allele following the high dose of alcohol in Afro-T. Indo-T with at least one *ADH1C*2* allele displayed significantly different Drug × Time interactions for the SHAS item *effects of alcohol* at the low dose and for the SHAS items *clumsy, muddle/confused, effects of alcohol, floating, drunk,* and *total* at the high dose from Indo-T with two *ADH1C*1* alleles. **Conclusions:** This is the first study that has investigated individual sensitivity to alcohol in a Caribbean population and in people of East Indian descent. Indo-T with at least one *ADH1C*2* allele may be at higher risk for heavy drinking by feeling less of the effects of alcohol, including nausea. In Afro-T, having at least one *ADH1B*3* allele appears to exert a protective effect by enhancing the unpleasant effects of alcohol, such as nausea and confusion. (*J. Stud. Alcohol Drugs, 75,* 827–838, 2014)

MEASURES OF SUBJECTIVE RESPONSE to alcohol intoxication are one of the most widely studied markers of alcohol sensitivity and tolerance in human studies. A number of clinical studies have verified that not all individuals respond to alcohol in a similar manner and that this may vary significantly depending on the individual's ethnic origin (Pedersen and McCarthy, 2013). More importantly, a less intense response to alcohol has been associated with an increased risk for the development of alcohol-related problems (Schuckit and Smith, 1996).

A typical method used to assess the subjective response to alcohol intoxication or the lower level of response (LR) to alcohol is the alcohol challenge paradigm. In this paradigm, participants are given doses of alcohol/placebo in a con-

trolled laboratory setting (Schuckit and Gold, 1988; Schuckit and Smith, 1996). The Subjective High Assessment Scale (SHAS) has been shown to be a robust measure of LR to alcohol in alcohol challenge paradigms (Schuckit and Gold, 1988). Other assessment methods to estimate differences between risk groups on their response to alcohol have included measuring levels of cortisol (Schuckit et al., 1987a), prolactin (Schuckit et al., 1987b), and electroencephalographic activity (Ehlers and Schuckit, 1988). The heritability of LR to alcohol has been estimated to be approximately 60% (Heath et al., 1999). Therefore, the investigation of LR to alcohol may be a useful tool in the understanding of how genetics influences drinking behaviors and the development of alcohol dependence in a given population.

Studies using similar alcohol challenge methodologies among groups at lower risk for alcoholism have provided additional support for the idea that individual sensitivity to alcohol might also mediate protection from developing alcoholism. Individuals of Asian heritage who have mutations in the aldehyde dehydrogenase gene (Wall, 2005; Wall et al., 1992) and individuals of Jewish descent (Monteiro et al., 1991), two groups with low rates of alcoholism, were

found to have more intense, although not necessarily more negative, self-reported responses to alcohol than matched control participants of average alcoholism risk. More intense responses to alcohol have also been found in alcohol challenge studies investigating non-Asians with *ADH1B*2* alleles (Duranceaux et al., 2006), whereas, in a high-risk group of Native American Indians, a less intense objective and subjective response to alcohol was seen in an alcohol challenge paradigm (Ehlers et al., 1998, 2001; Garcia-Andrade et al., 1997; Wall et al., 1996). Lastly, in an African American population, individuals with at least one *ADH1B*3* allele, who as a group are at lower risk for alcoholism, displayed an increase in sedation following alcohol challenge (McCarthy et al., 2010).

The present investigation is part of a larger study evaluating the risk and protective factors for alcohol dependence in the two major ethnic groups of the southernmost islands of the Lesser Antilles, Trinidad and Tobago. The population of Trinidad and Tobago is multiethnic but mainly composed of people of East Indian (Indo-T) and African (Afro-T) ancestry (Ministry of Planning and Sustainable Development, 2010). Some researchers have consistently reported that alcoholism is higher among patients of East Indian ancestry when compared with those of African ancestry. However, this is mainly based on surveys as well as alcohol-related hospital admissions (National Alcohol and Drug Abuse Prevention Centre, 2000; Singh and Maharajh, 1991).

Genetically influenced metabolic factors have been strongly implicated in the etiology of alcoholism, although these factors have not been extensively studied in ethnic groups living in the Caribbean. The major enzymes involved in alcohol metabolism, alcohol dehydrogenase (ADH) and mitochondrial aldehyde dehydrogenase (ALDH2), both exist as multiple isoenzymes that differ in their kinetic properties. Because the frequency of these isoenzymes differs across ethnic groups, the genes that code for them (*ADH* on chromosome 4, *ALDH2* on chromosome 12) have been considered candidate genes that could contribute to variation in alcohol metabolism, variability in response to alcohol, and differences in individual vulnerability for developing alcohol dependence and alcohol-related disability (see Bosron et al., 1993; Crabb, 1995; Crabb et al., 2004; Li, 2000; Thomasson et al., 1995; Wall, 2005; Whitfield, 2005). An excess of acetaldehyde produced either by a more active *ADH* (*ADH1B*2* and *ADH1B*3*) or less active *ALDH* is thought to lead to either a more intense response to alcohol or more alcohol-induced side effects. These more intense or more unpleasant effects of alcohol are postulated to contribute to lowered levels of drinking and thus protection from alcohol dependence (Wall, 2005). The *ADH1B*3* genotype has also been associated with fewer health-related effects of alcohol in African Americans (Scott and Taylor, 2007).

Polymorphisms in *ADH1C* (previously termed *ADH3*) have been associated with both alcohol dependence (Grove et al., 1998; Konishi et al., 2004; Monzoni et al., 2001; Mulligan et al., 2003; Osier et al., 2002; Zintzaras et al., 2006) and alcoholic liver disease (Day et al., 1991; Frenzer et al., 2002). It has been suggested that the presence of the *ADH1C*2* allele that codes for a less active enzyme may increase the risk for alcoholism by delaying the formation of acetaldehyde, perhaps leading to a less intense response to alcohol and/or fewer alcohol-induced negative side effects, ultimately resulting in higher levels of drinking (Frenzer et al., 2002).

Evidence of genetic differences in alcohol-metabolizing enzymes that are associated with alcohol-related phenotypes have been uncovered in the population of Trinidad and Tobago. These differences include (a) a significantly higher distribution of the *ADH1C*2* polymorphism in Indo-T, which is associated with alcohol dependence (Montane-Jaime et al., 2006); (b) a protective effect against alcohol consumption levels and alcohol dependence in Afro-T with at least one *ADH1B*3* allele (Ehlers et al., 2007); and (c) an association of the *ALDH1A1*2* genotype with significantly higher levels of current alcohol consumption and alcohol dependence in Indo-T (Moore et al., 2007). An additional study revealed distinct patterns between the two major ethnic groups studied with respect to the clinical course of alcoholism. In that study, alcoholics of Indian descent endorsed more heavy episodic drinking, tolerance, blackouts, and withdrawal than their African-descent counterparts (Montane-Jaime et al., 2008). Also, the pattern of comorbidity with anxiety and affective disorders with alcohol use disorders was found to differ between the two ethnic groups (Shafe et al., 2009).

The overall goal of this research program is to understand the pathophysiology of alcoholism in people of Indo-T and Afro-T ancestry from the southernmost islands of the Lesser Antilles, Trinidad and Tobago. Although our previous studies have demonstrated that Indo-T and Afro-T populations have unique alcohol-related symptom patterns and genetic profiles, there are no studies to date addressing the LR to alcohol in any Caribbean population. Therefore, the purpose of the present study was to investigate the response to alcohol using a laboratory-based alcohol challenge protocol in young men in the two major ethnic groups of Trinidad and Tobago before the development of alcoholism. The specific aims of the study were (a) to test objective and subjective responses (breath alcohol concentrations [BrAC], SHAS, cortisol, pulse rate) to two doses (low and high) of alcohol and placebo beverage in Indo-T and Afro-T young men; (b) to assess whether the two groups differed in response to alcohol, taking into account any differences between the groups in blood alcohol concentrations; (c) to determine if common polymorphisms in *ADH1B* were associated with a more intense LR to alcohol in Afro-T; and (d) to determine if common polymorphisms in *ADH1C* were associated with a less intense LR to alcohol in Indo-T.

Method

Participant selection

This study was approved by the ethics committees of the Faculty of Medical Sciences, The University of the West Indies, San Fernando General Hospital, Ministry of Health for Caura Hospital, Scarborough Hospital, and the institutional review board at the Scripps Research Institute. All participants gave written informed consent before inclusion in the study. This study evaluated 120 university students (56 Indo-T and 64 Afro-T) between ages 18 and 25 years. They were recruited from the Faculty of Medical Sciences and the University of the West Indies St. Augustine campus, Trinidad and Tobago. Participants were asked whether their grandparents were of Indian/African origin and if they had any admixture with any other ethnic groups, including African, White, Chinese, Spanish/Latin, and Arab. Participants qualified for the study if they had at least three grandparents of the same ethnic group, assuring some consistency in definition, and had normal liver and renal function tests. Participants were required to be regular drinkers, defined as having drunk at least one drink a month for the last 6 months.

Screening for alcohol drinking status and psychiatric diagnoses

To confirm that participants were regular drinkers but did not have alcohol use disorders, they were initially screened using the Alcohol Use Disorders Identification Test. If they had a score lower than 15, they were scheduled to undergo an interview using the Semi-Structured Assessment for the Genetics of Alcoholism (SSAGA; Bucholz et al., 1994), which was conducted by a consultant psychiatrist. The SSAGA also served to exclude those who had any major medical illness, a diagnosis of alcohol or other drug dependence, and/or Axis I psychiatric disorders. Based on their SSAGA data, 112 participants met the study requirements.

Beverage

The alcohol used in the study was locally produced 80-proof rum (40% alcohol). It was mixed with a sugar- and caffeine-free drink, also made in the country. The low dose and high dose of alcohol used in the study were calculated for each participant based on the their total body water, height in centimeters, weight in kilograms, age in years, and desired blood alcohol concentration (Watson et al., 1980, 1981). The desired BrAC was zero (0) for placebo, .04 g/dl for the low dose, and .08 g/dl for the high dose. Three milliliters of 80-proof rum was placed on top of the container where the sugar-free, caffeine-free drink was provided to simulate the taste and smell of an alcoholic beverage during the placebo session. The total volume of the beverage for each participant ranged from 445 ml to 790 ml.

Alcohol challenge sessions

Participants were asked to take part in three sessions in which they received either placebo or a low or high alcohol dose. They were instructed to abstain from alcohol use and from taking any medication for 3 days before the study sessions. They were also asked not to eat or drink any fluids other than water the morning of the study session. The sessions were conducted by a psychiatric nurse. The typical session started at 9:00 A.M. and lasted approximately 5 hours. Participants were given a standard breakfast consisting of plain wheat toast and 250 ml of Nestle Orchard orange juice. During the following 30 minutes, height and weight were measured, and then baseline measurements for BrAC and the SHAS were taken. BrAC was measured by an Alco-Sensor breath alcohol–measuring device (Intoximeter, Inc., St. Louis, MO). An intravenous catheter was placed in a forearm vein, and a blood sample was taken for DNA and cortisol analyses. To minimize the likelihood of participants differentiating between the placebo and alcoholic drinks, they were instructed to rinse their mouths with a nonalcoholic mouthwash before drinking the beverages, which they were then asked to consume within 10 minutes. After 5 hours, participants were provided with lunch and were not allowed to leave until their BrAC was less than .02 g/dl or not detectable. No participant was released until the breath alcohol measurement was confirmed a second time, and then the participant was allowed to leave only with a friend or in a taxi.

Subjective and objective responses to alcohol

Response to an acute alcohol intake was assessed with the SHAS, which includes both positive and negative effects of alcohol. Participants were asked to rate, on a 36-point Likert scale, the degree to which they felt the effects of alcohol from baseline (no alcohol) to extreme alcohol effects. These effects were measured at the same time BrAC and pulse rate were obtained, that is, every 30 minutes after the alcoholic or placebo beverage was ingested. Blood samples for cortisol were obtained at 30, 60, and 90 minutes following beverage administration. Whole-blood samples were drawn into 4 ml heparinized tubes and placed on ice. After centrifugation, the serum was transferred to 3 ml tubes and stored at -80 °C. Total plasma cortisol levels were determined using a commercially available enzyme-linked immunoassay kit (ALPCO Immunoassays-Diagnostics, Salem, NH). The optical density of the 96-well plate was read at 450 nm using a Synergy HT plate reader (BioTek Instruments, Inc., Winooski, VT). Results were extrapolated using standard curves. Data are expressed in nanograms per milliliter of plasma, and the lowest limit of detectability was 0.4 μg/dl.

Genotyping

Dried blood samples obtained through venipuncture were collected and sent to the Genomics and Bioinformatics Core of the Indiana Alcohol Research Center for genotyping, where the relevant portions of the *ADH* and *ALDH* loci were amplified using polymerase chain reaction followed by hybridization with allele-specific radiolabeled oligonucleotide probes (Xu et al., 1988), as previously described (Ehlers et al., 2007; Montane-Jaime et al., 2006). DNA samples that were collected after 2005 were genotyped using TaqMan SNP Genotyping Assays (Life Technologies, Grand Island, NY) as previously described (Hendershot et al., 2009).

Statistical analysis

Statistical analyses were designed to test the four specific aims. The first aim was to test objective and subjective responses to alcohol. To test this aim, an analysis of variance (ANOVA) model was used to assess the effects of ethanol on the BrAC, cortisol, pulse rate, and SHAS measures for the two doses (low and high) of alcohol compared with placebo. Because Indo-T had significantly higher BrAC levels than Afro-T, and because BrAC and SHAS are collinear variables, the two ethnic groups were run separately. For the SHAS and pulse rate analyses, a drug (alcohol vs. placebo) × six time points following beverage repeated-measures ANOVA was performed for the low dose and high dose separately for each ethnic group. In the cortisol analyses, only the first three time points following beverage were available for analyses. The second aim was to assess whether the two ethnic groups differed in response to alcohol, taking into account any differences between the groups in blood alcohol concentrations. To test this aim, a two ethnic group (Indo-T vs. Afro-T) × drug (alcohol vs. placebo) × six time points following beverage repeated-measures ANOVA that covaried for BrAC was performed for the low and high doses of alcohol separately. For the third aim, to determine if common polymorphisms in *ADH1B* were associated with a more intense LR to alcohol,

in Afro-T a two genotype group (*ADH1B*1* vs. *ADH1B*3*) × drug (alcohol vs. placebo) × six time points following beverage repeated-measures ANOVA was performed for the low and high dose of ethanol separately. The fourth aim was to determine if common polymorphisms in *ADH1C*2* were associated with a less intense LR to alcohol in Indo-T. To test this aim, in Indo-T a two genotype group (*ADH1C*2* vs. *ADH1C*1*) × drug (alcohol vs. placebo) × six time points following beverage repeated-measures ANOVA was performed for the low and high dose of ethanol separately. *P* values for main effects were set at <.01; *p* values for interactions and post hoc analyses following significant main effects were set at <.05.

Results

Participants

The 112 participants studied had a mean age of 21 years and 15 years of education. They drank on an average of 4 days per month and consumed an average of four drinks per occasion. Complete demographic data for the Indo-T and Afro-T participants are presented in Table 1. Afro-T had a higher body mass index (BMI) than Indo-T; there were no other differences in demographic or drinking variables between Afro-T and Indo-T.

Response to alcohol in Indo-T and Afro-T

The response to high- and low-dose alcohol, as compared with placebo, was determined for SHAS, pulse rate, BrAC, and cortisol in Indo-T and Afro-T young men. A repeated-measures ANOVA revealed that the low dose of alcohol produced a significant overall drug effect for all SHAS items, at the *p* < .01 level, compared with placebo in Afro-T except for the items *great, terrible, muddle or confused, activated,* and *nauseated.* Significant time and Drug × Time effects for this analysis were also seen for all items except for g*reat, terrible, muddle or confused, nauseated,* and *sleepy.* In Indo-T, low-dose alcohol was significantly different from placebo at the *p* < .01 level for overall drug effects on all items except for *clumsy, muddle or confused, slurred speech, floating, dizzy, nauseated, drunk,* and *terrible.* Significant time and Drug × Time effects for this analysis were also seen for all items except *uncomfortable, muddle or confused, slurred speech, dizzy, nauseated, activated, sleepy, great,* and *terrible.* At the high dose of alcohol, a significant drug effect was seen for all SHAS items in both Indo-T and Afro-T. In addition, all time and Drug × Time effects were significant except *sleepy* in the Afro-T. To assess whether the two ethnic groups differed in response to alcohol on the SHAS, a repeated-measures ANOVA that covaried for BrAC was performed at each dose level. No significant overall effects of ethnic group membership (Indo-T

TABLE 1.   Demographic characteristics of participants according to ethnic group

Variable	Afro-Trinidadians M (SD) or % (n) (n = 61)	Indo-Trinidadians M (SD) or % (n) (n = 51)
Age in years	21.07 (1.62)	20.49 (1.82)
Years of school	15.44 (0.81)	15.55 (0.95)
Body mass index, kg/m²	25.16 (4.65)	21.58 (4.26)**
Drinking frequency, times per month	3.61 (3.89)	3.45 (3.83)
Drinking quantity, drinks per occasion	3.72 (3.0)	3.88 (2.24)
Income >$20,000[a]/year	88% (50)[b]	85% (27)[b]

[a]In U.S. dollars; [b]n for whom income data were available
**p < .01.

or Afro-T) or significant interactions between drug (alcohol vs. placebo) and ethnic group membership were found on any of the SHAS items for either dose in these analyses.

Figure 1 shows the mean BrAC as a function of time in Indo-T and Afro-T. Repeated-measures ANOVA revealed that Indo-T had significantly higher BrAC levels than Afro-T at the low dose, $F(1, 103) = 11.02$, $p < .001$, of alcohol; at the high dose, the difference between the two groups did not reach significance ($p < .023$). These findings were unchanged when the analyses were covaried for BMI or current drinking levels. Figure 1 also displays the pulse rate changes as a function of time in Indo-T and Afro-T following low- and high-dose alcohol. Repeated-measures ANOVA revealed that alcohol did not have a significant overall effect on pulse rate but that an overall ethnic group effect was present at both the low, $F(1, 210) = 18.8$, $p < .0001$, and high doses, $F(1, 211) = 22.5$, $p < .0001$, of alcohol. No Alcohol × Ethnic Group × Time interactions were found at either dose. Similarly, as seen in Figure 2, there were significant differences in cortisol values, with Indo-T having higher values at the 60-minute, $F(1, 85) = 4.6$, $p < .035$, and 90-minute, $F(1, 85) = 4.9$, $p < .03$, time points during the placebo condition. However, there were no effects of alcohol on cortisol values and no differences between Indo-T and Afro-T with high- or low-dose alcohol.

ADH1B*3 and response to alcohol in Afro-T

Of the 61 Afro-T participants, 40 were homozygous for the major allele, 20 were heterozygous, and 1 was homozygous for the minor allele (major allele frequency = 0.82, minor = 0.18). A chi-square test demonstrated that there was no significant difference between observed and expected allele frequencies, $\chi^2(1) = 0.726$, $p < .394$. In the Indo-T, only one participant was heterozygous, and the chi-square test was also not significant when observed and expected allele frequencies were tested, $\chi^2(1) = 0.005$, $p < .944$.

The third aim was to determine if common polymorphisms in *ADH1B* were associated with a more intense LR to alcohol in Afro-T. To test this aim, a two genotype group (*ADH1B*1* vs. *ADH1B*3*) × six time points following beverage repeated-measures ANOVA was performed for the two doses of alcohol as compared with placebo in the Afro-T group. There were no significant differences on any of the SHAS items in response to low-dose alcohol between the two genotype groups. At the high dose of alcohol, a genotype group effect was seen for the items *muddle/confused,* $F(1, 55) = 8.2$, $p = .006$, and *nauseated,* $F(1, 55) = 8.9$, $p = .004$, with the presence of at least one *ADH1B*3* allele being associated with higher SHAS responses on those items in Afro-T participants, as seen in Figure 3. In addition, significant Time and Genotype Group × Time effects were seen for the items *uncomfortable, muddle/confused, slurred speech,* and *nauseated.* There were no significant differences

in BrAC between those with at least one *ADH1B*3* allele and those with only *ADH1B*1* alleles.

ADH1C*2 and response to alcohol in Indo-T

Of the 51 Indo-T participants, 24 were homozygous for the major allele, 18 were heterozygous, 7 were homozygous for the minor allele (major allele frequency = 0.67, minor = 0.33), and 2 had missing data. A chi-square test demonstrated that there was no significant difference between observed and expected allele frequencies, $\chi^2(1) = 1.33$, $p < .249$. In the Afro-T, 47 were homozygous for the major allele, 13 were heterozygous, and 1 was homozygous for the minor allele (major allele frequency = 0.88, minor = 0.12), and the chi-square test was also not significant when observed and expected allele frequencies were tested, $\chi^2(1) = 0.009$, $p < .926$.

The fourth aim was to determine if common polymorphisms in *ADH1C* were associated with a more intense LR to alcohol in Indo-T. To test this aim, a two genotype group (*ADH1C*2* vs. *ADH1C*1*) × six time points following beverage repeated-measures ANOVA was performed for the two doses of alcohol as compared with placebo in the Indo-T group. At the low dose of alcohol, no overall genotype group differences were seen; however, a Time and Genotype Group × Time interaction was significant for the SHAS item *effects of alcohol.* At the high dose of alcohol, also, no overall genotype group differences were seen; however, Time and Genotype Group × Time interactions were significant for the SHAS items *clumsy, muddle/confused, effects of alcohol, floating, drunk,* and *total.* These results are presented in Figure 4. There were no significant differences in BrAC between those with at least one *ADH1C*2* allele and those with only *ADH1C*1* alleles.

Discussion

This study determined whether there is a difference in the response to alcohol following an alcohol challenge in young adult men representing the two major ethnic groups of Trinidad and Tobago. We directly compared Indo-T and Afro-T individuals using the same alcohol challenge paradigm, which facilitated the identification of potential differences in subjective response to alcohol. We further investigated if these differences were explained by common variants of the *ADH1B* and *ADHC* genes frequently found in Afro-T and Indo-T, respectively.

Response to alcohol in Indo-T and Afro-T

Indo-Trinidadians. Our previous findings suggested that Indo-Ts are at a higher risk for alcoholism compared with Afro-T. Research has found that Indo-T alcoholics report more tolerance, have experienced more heavy episodic
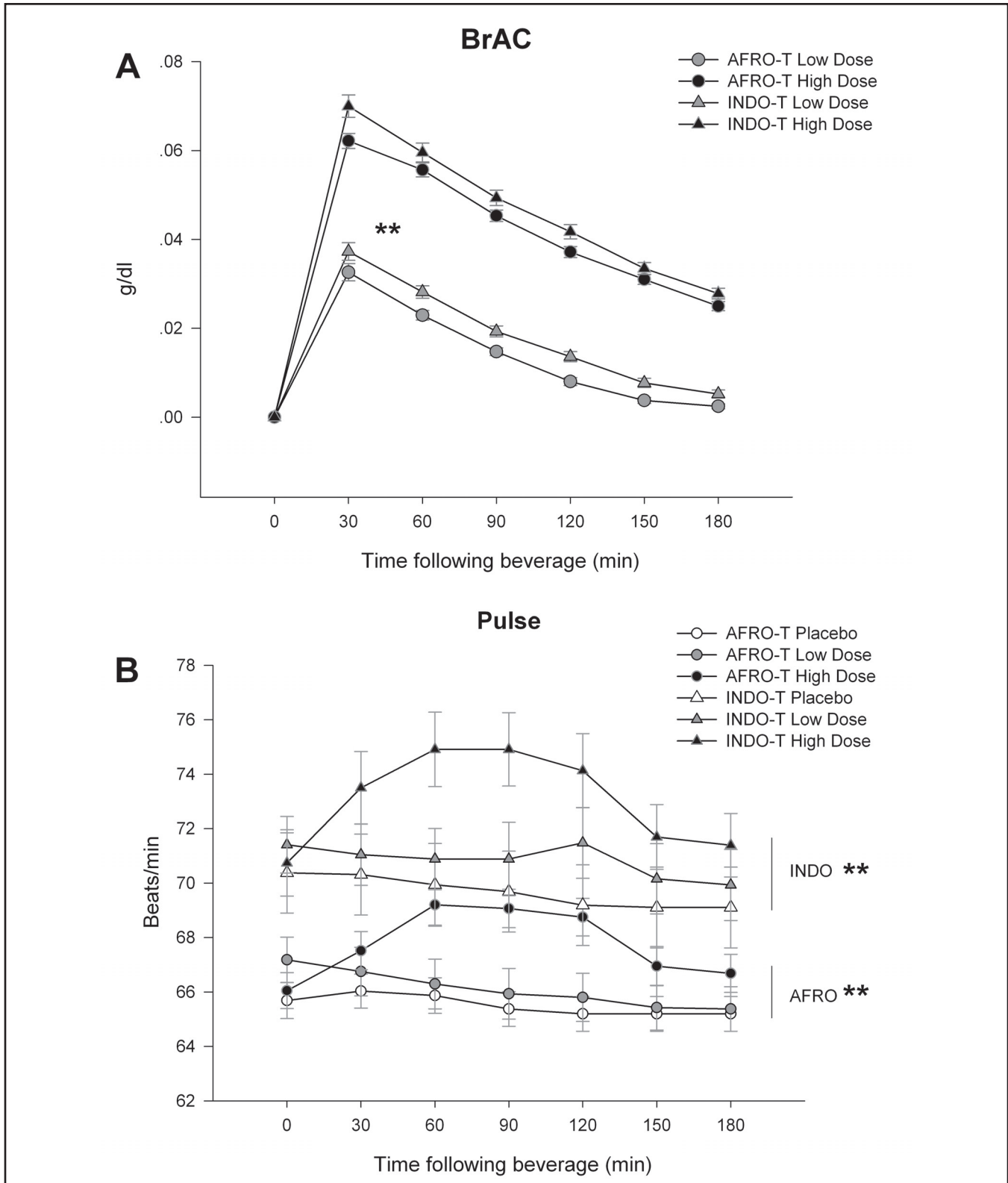
FIGURE 1.   (A) Blood alcohol levels determined by breath alcohol analysis shown for Indo-Trinidadians (Indo-T) and Afro-Trinidadians (Afro-T) at low doses and high doses of alcohol at 30-minute intervals following beverage. Indo-T had significantly higher breath alcohol concentration (BrAC) compared with Afro-T at the low dose. (B) Pulse rates shown for the same 30-minute time points are also significantly higher in Indo-T compared with Afro-T. Min = minutes. Error bars = *SEM*.
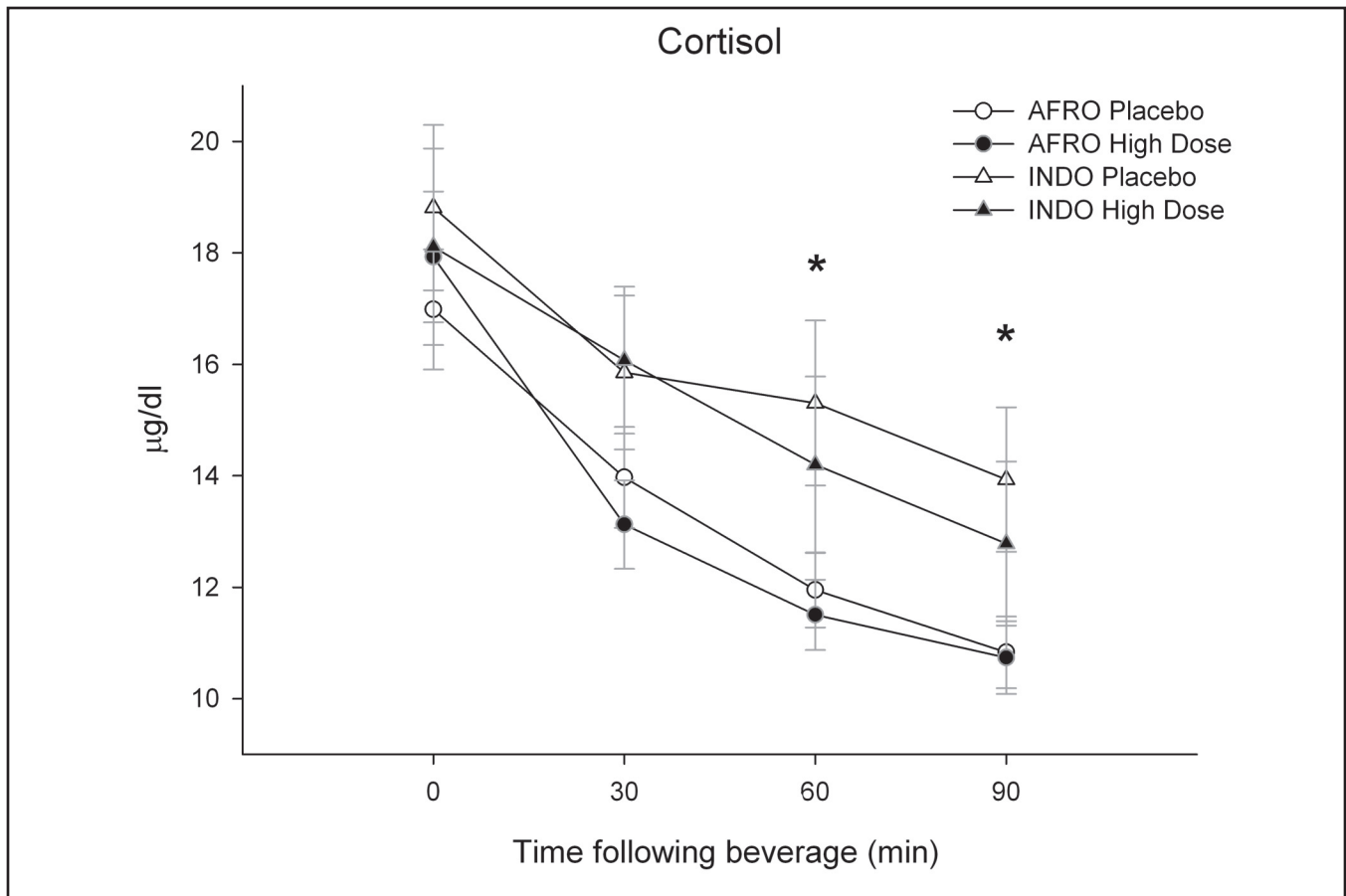**indicates *p* < .01 ethnic group effect.

FIGURE 2.   Blood cortisol levels measured every 30 minutes following beverage in Indo-Trinidadians (Indo-T) and Afro-Trinidadians (Afro-T). Indo-T had significant higher cortisol levels at the 60- and 90-minute time points following the placebo dose. Min = minutes. Error bars = *SEM*. *indicates $p < .05$, post hoc analyses.

drinking (Montane-Jaime et al., 2008), and report drinking more often and more drinks per occasion than Afro-T alcoholics (Montane-Jaime et al., 2006). In the same study, Indo-T also reported experiencing more blackouts, shakes, withdrawal, and health problems than Afro-T (Montane-Jaime et al., 2008).

The present study did not find that young, adult, male, nonalcoholic Indo-T had significantly lower SHAS responses to alcohol than Afro-T using a standard alcohol challenge paradigm. However, Indo-T did have significantly higher levels of BrAC than Afro-T, even when the analyses were covaried for BMI or current drinking history. This may be because of differences in absorption, distribution, or metabolism of alcohol between the two ethnic groups not accounted for in the anthropomorphic equation used to estimate the doses of alcohol used in the study. Indo-T did have significantly higher cortisol levels and pulse rates during the placebo session and higher pulse rates during the alcohol session as compared with Afro-T. This may suggest that Indo-T were more stressed during the laboratory procedures or may potentially represent another inherent risk factor that could also

contribute to more drinking and alcohol dependence in this ethnic group.

Ethnic differences in stress hormone levels at baseline and following stress have been previously reported. For instance, lower cortisol levels upon waking and during the day (after controlling for socioeconomic and psychological factors) have been found in African Americans compared with Whites (Bennett et al., 2004; Cohen et al., 2006). In addition, African Americans reportedly have a lower HPA axis response to psychological stress compared with Whites (Chong et al., 2008). The ethnic variations in adrenocortical response to the stress during laboratory procedures may also be explained by the differences in BMI observed in Indo-T and Afro-T (Afro-T had higher BMI than Indo-T). Higher BMI has been negatively associated with the awakening rise and early decline in cortisol levels (Champaneri et al., 2013).

Afro-Trinidadians. Kerr et al. (2006) have reported that young African Americans needed fewer drinks to feel the effects of alcohol than White Americans. More recently, Pedersen and McCarthy (2013) found that African Americans are more sensitive to some of the effects of alcohol
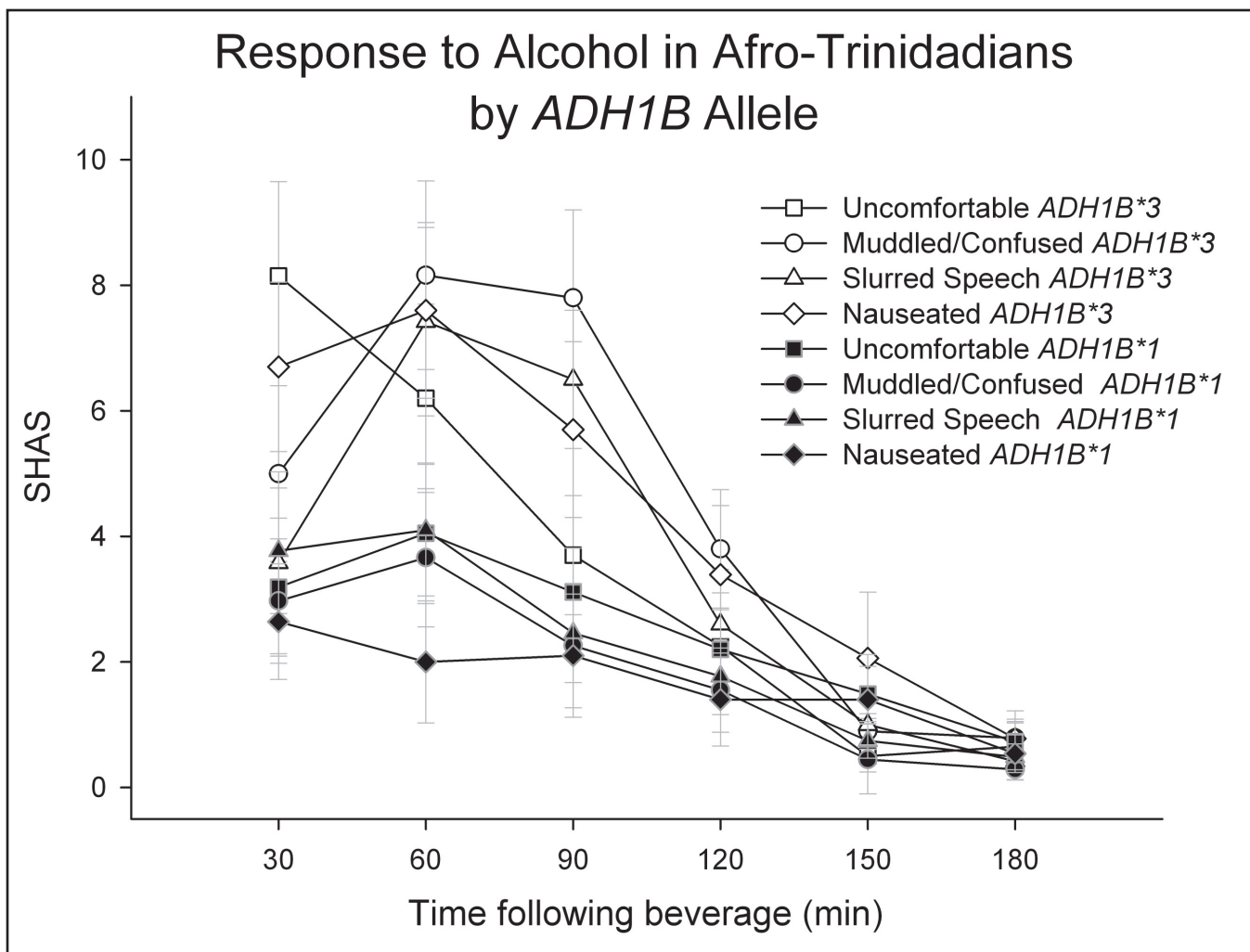
FIGURE 3. *ADH1B* alleles in Afro-Trinidadians (Afro-T) and their responses to the Subjective High Assessment Scale (SHAS) over time following consumption of alcohol. At the high dose of alcohol, a significant ($p < .01$) genotype group effect was seen for SHAS items *nauseated* and *muddled/confused*. In addition, at the high dose of alcohol, a significant ($p < .05$) Genotype × Time effect was seen for the SHAS items *uncomfortable, muddled/confused, slurred speech,* and *nauseated*. Subjects with at least one *ADH1B*3* show an overall more intense response to alcohol. Min = minutes. Error bars = *SEM*.

than European Americans. However, comparison of Afro-T with African Americans with respect to LR to alcohol is hindered because of the limited available data as well as several methodological differences. First, previous studies in African Americans have not included a placebo beverage. Second, study samples were composed of both males and females, whereas our study included only males. In addition, although all participants were of African descent, inclusion criteria required that they have at least one parent of African ancestry, whereas in our study, at least three of their grandparents needed to be of African ancestry.

*ADH1B*3 and response to alcohol in Afro-T*

The third aim of our study was to determine if polymorphisms in *ADH1B*3* were associated with a more intense

LR to alcohol in Afro-T. We found a significant association of *ADH1B*3* with the SHAS symptoms *muddle/confused* and *nauseated* in Afro-T challenged with the high dose of alcohol. We also found additional Genotype × Time interactions for the items *uncomfortable, muddle/confused, slurred speech,* and *nauseated* following the high dose of alcohol. Our results are consistent with previous reports by McCarthy et al. (2010). The authors reported that participants with at least one *ADH1B*3* allele experienced greater sedation following a moderate dose of alcohol. They assessed sedation using the Biphasic Alcohol Effects Scale, which measures self-reports of the stimulant and sedative effects of alcohol as separate and distinct constructs. The sedation subscale of the Biphasic Alcohol Effects Scale includes items such as difficulty concentrating, heavy head, sluggish, and inactive, which may be comparable to the SHAS items of
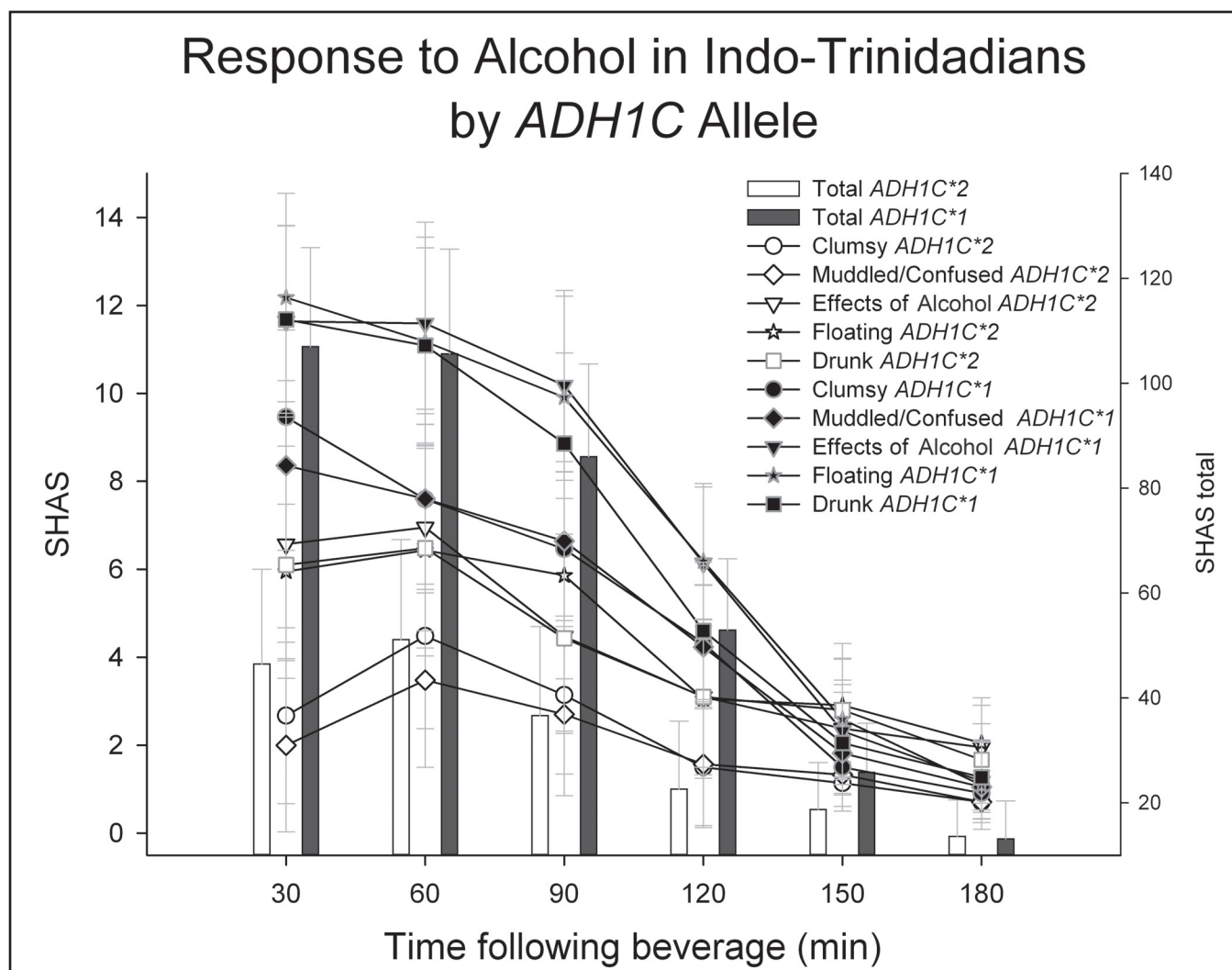
FIGURE 4. *ADH1C* alleles in Indo-Trinidadians (Indo-T) and their responses to the Subjective High Assessment Scale (SHAS) over time following consumption of alcohol. At the high dose of alcohol, a significant time ($p < .01$) and Genotype × Time ($p < .05$) effect was seen for the SHAS items *clumsy, muddled/confused, effects of alcohol, floating, drunk* (left axis), as well as the total of all SHAS items (right axis). Subjects with at least one *ADH1C*2* allele show an overall less intense response to alcohol. Min = minutes. Error bars = *SEM*.

muddle/confused we found significantly associated with the *ADH1B*3.*

We did not find a significant association between the *ADH1B* genotype groups and BrAC in any of the two alcohol challenge sessions. These findings are also in agreement with those obtained in the above-mentioned study in African Americans (McCarthy et al., 2010). In that study, BrAC was not significantly different in participants with the *ADH1B*3* genotype as compared to those with the *ADH1B*1* genotype. Failure to find an association between alcohol-metabolizing enzyme genes and BrAC has been related to the route used to administer alcohol (McCarthy et al., 2010). In a study performed by Neumark et al. (2004), the effect of *ADH1B*2* on alcohol elimination rates in healthy Jewish male participants was demonstrated for the first time. Participants with the *ADH1B*2* genotype had higher alcohol elimination rates compared with those homozygotes for *ADH1B*1,* but they used the alcohol clamp method. Future research may be required to define if there is any association between the presence of *ADH1B*3* and higher concentrations of acetaldehyde in African populations and if this mediates the protecting effect by enhancing alcohol's unpleasant effects. Therefore, the mechanism of *ADH1B*3*-induced unpleasant effects of alcohol such as nausea and confusion in Afro-T and sedation in African Americans remains unclear.

*ADH1C*2 and response to alcohol in Indo-T*

Another aim of our study was to determine if common polymorphisms in *ADH1C* were associated with a less

intense LR to alcohol in Indo-T. We anticipated a positive association because an earlier study that included Indo-T alcoholics and controls revealed that the *ADH1C*2* allele was frequent in Indo-T and that it was related to alcohol dependence (Montane-Jaime et al., 2006). Although these analyses did not show an overall genotype effect in response to alcohol, Time and Genotype × Time interactions were significant for the SHAS item *effects of alcohol* at the low dose and for the SHAS items *clumsy, muddle/confused, effects of alcohol, floating, drunk,* and *total* at the high dose. There were no significant differences in BrAC between those with at least one *ADH1C*2* allele and those with only *ADH1C*1* alleles. The association of this allele with alcohol dependence was previously reported in other populations by several authors (Kortunay et al., 2012; Mulligan et al., 2003). The most recent positive report was in a meta-analysis involving a large sample of alcoholics and controls of Asian, European, African, and Native American origins (Li et al., 2012).

McCarthy et al. (2010) obtained similar results in African Americans. The authors did not find significant differences when African American participants with two *ADH1C*1* alleles were compared with those with at least one *ADH1C*2* allele. Likewise, Duranceaux et al. (2006) found no significant association between the *ADH1C*2* allele and LR to alcohol in a study that included non-Asian populations. These results, however, contradict those from a fairly recent study that provided pharmacokinetic evidence of the role of the *ADH1C*2* in decreasing alcohol metabolic rates in healthy White individuals (Martínez et al., 2010). As with the *ADH1B*3* allele, the association between the *ADH1C*2* allele and LR may need further exploration using methods other than oral administration of alcohol, such as the alcohol clamp method (Neumark et al., 2004).

Conclusions

In summary, this is the first study investigating individual sensitivity to alcohol and its heritability in a Caribbean population and in people of East Indian descent. The finding may be of interest to other East Indian populations because there are no other studies of this kind performed in this ethnic group. We did not find differences in subjective responses to alcohol between the two ethnic groups. However, we did find that within the two ethnic groups, subgroups with allele differences in ADH isozymes did differ in the expected direction in response to alcohol. The *ADH1B*3* allele was associated with response to alcohol and may exert its protecting effect by enhancing some of the unpleasant effects of alcohol in Afro-T, and the *ADH1C*2* allele is a risk factor for Indo-T, as individuals with that allele report fewer effects of alcohol. This study is limited in that results may not be generalized to all Indo-T or Afro-T, as the participants in the present study were young adult male students. The lack of

significance in the present findings may have been affected by the relatively small sample size and lack of statistical power to perform more comprehensive analyses. Information on the environmental effects that may influence response to alcohol in the two ethnic groups was not studied in this investigation. Despite these limitations, these preliminary findings provide further understanding of the risk and protective factors for alcohol dependence in Trinidad and Tobago. More research is needed to validate the hypothesis that LR to alcohol in Indo-T leads to higher alcohol consumption in order to feel the effects of alcohol intoxication, and eventually to alcohol dependence.

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