GENETIC LINKS TO THE REINFORCING EFFECTS OF ALCOHOL IN A SOCIAL CONTEXT

by

Kasey Griffin Creswell

B.S., University of Pittsburgh, 2001

M.S., University of Pittsburgh, 2007

Submitted to the Graduate Faculty of

Dietrich School of Arts and Sciences Department of Psychology in partial fulfillment

of the requirements for the degree of

Doctor of Philosophy

University of Pittsburgh

2012

UNIVERSITY OF PITTSBURGH

DIETRICH SCHOOL

This dissertation was presented

by

Kasey Griffin Creswell

It was defended on

July 9, 2012

and approved by

Stephen B. Manuck, Distinguished University Professor, Psychology

Robert E. Ferrell, Professor, Human Genetics

Shirley Y. Hill, Professor, Psychiatry

FeiFei Ye, Assistant Professor, Psychology in Education

Jeffrey Cohn, Professor, Psychology

Dissertation Director: Michael A. Sayette, Professor, Psychology and Psychiatry

GENETIC LINKS TO THE REINFORCING EFFECTS OF ALCOHOL IN A SOCIAL CONTEXT

Kasey Griffin Creswell, Ph.D.

University of Pittsburgh, 2012

Development of interpersonal relationships is a fundamental human motivation, and behaviors facilitating social bonding are prized. Some individuals experience enhanced reward from alcohol in social contexts and may be at heightened risk for developing and maintaining problematic drinking. There has been little systematic research conducted in group settings, though, and no prior studies have tried to link genetic variation to alcohol's socially reinforcing effects. This research investigated whether the rewarding effects of alcohol in a group setting are associated with genetic variation implicated in the development of alcohol use disorders. Specifically, this study tested the moderating influence of genes encoding the dopamine D2 and D4 receptors, the serotonin transporter, and the alpha receptor for gamma-aminobutryic acid ($GABA_A$) on the effects of alcohol on social bonding. Social drinkers (N=427; males=50.12%) were assembled into three-person unacquainted groups, and given a moderate dose of alcohol, placebo, or a non-alcohol (control) beverage, which they consumed over 36-min. To assess social bonding, participants completed the Perceived Group Reinforcement Scale immediately after the group drinking period. In addition, their social interaction was video-recorded, and the duration of facial behaviors was systematically coded using the Facial Action Coding System. After applying the Bonferroni correction to control for false positives in multiple genotype

comparisons, there was one significant gene x environment interaction. Results showed that carriers of at least one copy of the 7-repeat allele of the *DRD4* VNTR reported higher perceived social bonding in the alcohol, relative to placebo or control conditions, whereas alcohol did not affect ratings of 7-absent allele carriers. Findings indicate that carriers of the 7-repeat allele were especially sensitive to alcohol's effects on social bonding. These data converge with other recent gene-environment interaction findings implicating the *DRD4* polymorphism in the development of alcohol use disorders, and results suggest a specific pathway by which social factors may increase risk for problematic drinking among 7-repeat carriers.

TABLE OF CONTENTS

TABLE OF CONTENTS vi

LIST OF TABLES xi

LIST OF FIGURES xiii

1.0 INTRODUCTION 1

1.1 RELEVANCE OF SOCIAL CONTEXTS TO UNCOVER IMPORTANT GENETIC EFFECTS ON ALCOHOL RESPONSE 3

1.2 CONTROLLED SOCIAL INTERACTIONS IN THE LABORATORY 5

1.3 GENETIC LINKS TO THE REINFORCING EFFECTS OF ALCOHOL 6

1.4 DOPAMINE 6

1.5 SEROTONIN 13

1.6 GAMMA AMINOBUTYRIC ACID (GABA) 17

1.7 ALCOHOL EXPECTANCIES VS. PHARMACOLOGICAL EFFECTS OF ALCOHOL 19

1.8 RATIONALE FOR DRINK CONDITIONS 20

1.9 MEASUREMENT OF ALCOHOL'S REINFORCING EFFECTS 21

1.10 GENDER DIFFERENCES IN GENETIC VULNERABILITY TO DEVELOP ALCOHOLISM 22

1.11 FAMILY HISTORY OF ALCOHOLISM 23

1.12 SUMMARY AND SIGNIFICANCE 23

1.12.1 Specific Aim 24
1.12.2 Specific Hypotheses 24
2.0 METHOD 26
2.1 OVERVIEW 26
2.2 PARTICIPANTS 27
2.3 PROCEDURES 28
2.3.1 Pre-drink Assessment 28
2.3.2 Drink Administration 30
2.3.3 Post-drink Assessment 32
2.4 FACIAL CODING 33
2.4.1 Positive Affect 33
2.4.2 Negative Affect 34
2.5 RELIABILITY OF MEASUREMENT 35
2.6 DATA ANALYSES 36
2.6.1 Data Processing 36
2.6.2 Molecular Genetics Analyses 36
2.6.3 *DRD4* VNTR 37
2.6.4 *DRD2* Taq1A 38
2.6.5 5-HTTLPR 39
2.6.6 $GABA_{A\alpha 6}$ 40
2.7 STATISTICAL MODELING 40
2.7.1 Level-1 Model 44
2.7.2 Level-2 Model 44
2.7.3 Level-3 Model 44

2.7.4 Combined Model 45

2.7.5 Level-1 Model 46

2.7.6 Level-2 Model 46

2.7.7 Combined Model 46

3.0 RESULTS 47

3.1 PARTICIPANT CHARACTERISTICS AND RANDOMIZATION 47

3.2 BEVERAGE MANIPULATION CHECK 48

3.3 MAIN EFFECTS OF BEVERAGE CONDITION ON SOCIO-EMOTIONAL RESPONSES 48

3.3.1 Perceived Social Bonding 48

3.3.2 Alcohol Response/Affect Measures 49

3.3.3 Facial Expressions 50

3.4 GENOTYPE EFFECTS ON PARTICIPANT CHARACTERISTICS AND BASELINE MEASURES 51

3.4.1 *DRD4* VNTR 51

3.4.2 *DRD2* Taq1A 52

3.4.3 5-HTTLPR 52

3.4.4 $GABA_{A\alpha6}$ 53

3.5 TESTS OF SPECIFIC AIM: INTERACTIONS BETWEEN BEVERAGE CONDITION AND GENOTYPE ON SOCIO-EMOTIONAL RESPONSES 53

3.5.1 DRD4 VNTR 53

3.5.1.1 Perceived Social Bonding 53

3.5.1.2 Alcohol Response/Affect Measures 55

3.5.1.3 Facial Expressions 55

3.5.2 *DRD2* Taq1A 56
3.5.3 5-HTTLPR 56
3.5.3.1 Perceived Social Bonding 56
3.5.3.2 Alcohol Response/Affect Measures 56
3.5.3.3 Facial Expressions 57
3.5.3.4 Supplementary Analyses 57
3.5.4 GABAAα6 58
3.5.4.1 Perceived Social Bonding 58
3.5.4.2 Alcohol Response/Affect Measures 58
3.5.4.3 Facial Expressions 59
3.5.5 Family History of Alcoholism 59
4.0 DISCUSSION 60
4.1 DOPAMINE 60
4.1.1 *DRD4* VNTR 60
4.1.2 *DRD2* Taq1A 63
4.2 SEROTONIN 64
4.2.1 5-HTTLPR 64
4.3 GABA 67
4.3.1 $GABAA_{\alpha 6}$ 67
4.4 STRENGTHS AND IMPLICATIONS 69
4.5 LIMITATIONS 72
4.6 FUTURE DIRECTIONS 75
4.7 SUMMARY AND SIGNIFICANCE 78
APPENDIX A: TABLES AND FIGURES 80

BIBLIOGRAPHY **106**

LIST OF TABLES

Table 1. *DRD4* VNTR Allele and Genotype Frequencies 81

Table 2. *DRD4* Genotype Distribution Across Beverage Conditions 82

Table 3. *DRD2* Taq1A Genotype Distribution Across Beverage Conditions 83

Table 4. 5-HTTLPR Genotype Distribution Across Beverage Conditions 84

Table 5. $GABA_{A\alpha 6}$ Genotype Distribution Across Beverage Conditions 85

Table 6. Beverage Response Variables 86

Table 7. Socio-emotional Responses (M ± SE) Across Beverage Conditions 87

Table 8. Facial Expressions (M ± SE) Across Beverage Conditions 88

Table 9. Correlations Between Outcome Measures 89

Table 10. HLM: Model Predicting PGRS from Beverage Condition and *DRD4* Genotype 90

Table 11. PGRS scores (M ± SE) by Beverage Condition and Genotype 91

Table 12. Variance Components and Model Fit 92

Table 13. HGLM: Model Predicting Social Smiles from Beverage Condition and *DRD4* Genotype 93

Table 14. HLM: Model Predicting PGRS from Beverage Condition and 5-HTTLPR Genotype 94

Table 15. HLM: Model Predicting Negative Affect from Beverage Condition and 5-HTTLPR Genotype 95

Table 16. HGLM: Model Predicting Social Smiles from Beverage Condition and 5-HTTLPR Genotype 96

Table 17. HLM: Model Predicting PGRS from Beverage Condition and 5-HTTLPR Genotype using Alternative Genotype Classification 97

Table 18. HLM: Model Predicting Negative Affect from Beverage Condition and 5-HTTLPR Genotype using Alternative Genotype Classification 98

Table 19. HGLM: Model Predicting Negative Affect Composite from Beverage Condition and 5-HTTLPR using Alternative Genotype Classification 99

Table 20. HLM: Model Predicting PGRS from Beverage Condition and $GABA_{A\alpha6}$ Genotype 100

Table 21. HLM: Model Predicting BAES-Stimulation from Beverage Condition and $GABA_{A\alpha6}$ Genotype 101

Table 22. HLM: Model Predicting Positive Affect from Beverage Condition and $GABA_{A\alpha6}$ Genotype 102

Table 23. HLM: Model Predicting Negative Affect from Beverage Condition and $GABA_{A\alpha6}$ Genotype 103

LIST OF FIGURES

Figure 1. *DRD4* Genotype by Alcohol vs. No-alcohol Condition Interaction..... 104

Figure 2. PGRS Scores (Mean, SE) by *DRD4* Genotype and Beverage Condition 105

1.0 INTRODUCTION

Most people who drink alcohol do so in moderation, but a significant subset of people develop severe alcohol problems. In the United States, approximately 1 in every 12 adults (or about 17.6 million people) abuse alcohol or are alcohol dependent (Merikangas & McClair, 2012; U.S. Department of Health and Human Services, 2007). Alcohol use disorders (AUDs) are associated with multiple adverse health consequences and claim the lives of over 100,000 people each year (Stinson, Nephew, Dufour, & Grant, 1996). In fact, excessive alcohol use remains the third leading cause of preventable death in the United States (Mokdad, Marks, Stroup, & Gerberding, 2004). It is a research priority to identify individuals at risk to develop AUDs, as such information would greatly inform prevention and treatment strategies (Fromme, de Wit, Hutchison, Ray, Corbin, Cook et al., 2004).

Because alcohol dependence has been shown to be largely (52 to 64%) heritable (Kendler, 2001), there is much interest in uncovering the genetic bases of AUDs (Goldman, Oroszi, & Ducci, 2005). One approach has been to conduct large-scale family based (linkage) or case-control (association) studies and perform genome-wide scans to identify chromosome regions and genes associated with AUDs. Several samples have been collected for these analyses including the Irish Roscommon study (Prescott, Sullivan, Kuo, Webb, Vittum, Patterson et al., 2006), the Collaborative Study on the Genetics of Alcoholism (COGA; Reich, Edenberg, Goate, Williams, Rice, Van Eerdewegh et al., 1998), and the Pittsburgh multiplex family study (Hill,

Shen, Zezza, Hoffman, Perlin, & Allan, 2004). Results have been promising, and functional loci moderating risk for alcoholism have been identified. Examples include a region on chromosome 11 close to the *DRD4* gene (Ehlers, Gilder, Wall, Phillips, Feiler, & Wilhelmsen, 2004.; George, Cheng, Nguyen, Israel, & O'Dowd, 1993; Hill, Zezza, Wipprecht, Xu, & Neiswanger, 1999; Long, Knowler, Hanson, Robin, Urbanek, Moore et al., 1998; Reich et al., 1998), a region on chromosome 4p near the centromere containing a GABA receptor A ($GABA_A$) gene cluster (Long et al., 1998; Zinn-Justin & Abel, 1999), and a region on chromosome 17 (at marker D17S1857), which contains the serotonin transporter gene (Hill et al., 2004; cf. Edenberg, Reynolds, Koller, Begleiter, Bucholz, Conneally et al., 1998).

These studies have provided valuable information regarding genetic variation that moderates risk for AUDs, but progress has been slow, and replication has been difficult (e.g., Bierut, Agrawal, Bucholz, Doheny, Laurie, Pugh et al., 2010). Indeed, despite intense investigation, few well-replicated genetic markers of alcoholism have been identified (Goldman et al., 2005; Li & Burmeister, 2009). Alcohol abuse and dependence are complex, heterogeneous phenotypes that are likely caused by multiple sources of vulnerability, and attempts to identify specific genes related to such distal phenotypes have proved immensely challenging (Ducci & Goldman, 2008; Volkow & Muenke, 2012).

This state of affairs has prompted researchers to focus increasingly on intermediate alcohol-related processes that presumably lie closer to the biological actions of functional genetic variation than the more complex and distal phenotypes of alcoholism (Goldman et al., 2005; Gottesman & Gould, 2003). Nevertheless, these studies, which often examine effects of specific gene polymorphisms on individuals' sensitivity to the rewarding effects of alcohol or to alcohol-associated cues, have also yielded mixed findings (see Dick & Foroud, 2003). This may be due

to limited statistical power afforded by smaller study samples typically recruited for laboratory research, variation in drinking histories and tolerance levels of participants, or the result of unmeasured or unknown environmental moderators of gene effects (i.e., gene-environment interaction; Ducci & Goldman, 2008; Moffitt, Caspi, & Rutter, 2005). It is becoming increasingly clear that in order to detect gene effects in AUDs, environmental factors must be considered (Enoch, 2006). The examination of alcohol's effects in a social context may be a powerful approach to uncover genetic vulnerability to AUDs (Fromme et al., 2004; Slutske, Hunt-Carter, Nabors-Oberg, Sher, Bucholz, Madden et al., 2004).

1.1 RELEVANCE OF SOCIAL CONTEXTS TO UNCOVER IMPORTANT GENETIC EFFECTS ON ALCOHOL RESPONSE

Social factors play an instrumental role in the development and maintenance of AUDs (Kendler, Gardner & Prescott, 2011; Sher, Grekin & Williams, 2005). Older adolescents and young adults do nearly all of their drinking with others (Bachman, Johnston, O'Malley & Schulenberg, 2006), suggesting that social processes may be particularly important in shaping drinking behavior early on and may play a key role in the development of problematic drinking (McGue & Iacono, 2004). Surveys indicate that people commonly endorse social motives for drinking (Cooper, 1994; Cooper, Frone, Russell & Mudar, 1995; Goldman, Brown & Christiansen, 1987), and expectancies of social facilitation are especially powerful in young adult drinkers (Park, Sher & Krull, 2008; Patrick, Schulenberg, O'Malley, Maggs, Kloska, Johnston et al., 2011). Moreover, the belief that alcohol facilitates social functioning is associated with problematic drinking in cross-sectional studies (Conway, Swendsen & Merikangas, 2003; Engels, Wiers, Lemmers &

Overbeek, 2005; Mann, Chassin & Sher, 1987) and, in prospective studies, predictive of actual alcohol use (Smith, Goldman, Greenbaum & Christiansen, 1995) and AUDs. For instance, Patrick et al. (2011) showed that social/recreational reasons for drinking at age 18 predicted symptoms of AUDs 17 years later, and Beseler, Aharonovich, Keyes, and Hasin (2008) showed that adults with a family history of alcoholism who drank for social facilitation and to reduce negative affect had a greater risk of alcohol dependence 10 years later.

In addition to the general importance of social factors in the etiology of AUDs, there are also likely individual differences in the extent to which alcohol is socially reinforcing. Individuals who experience more reward from alcohol in social settings may be at increased risk to misuse alcohol (Kirchner, Sayette, Cohn, Moreland & Levine, 2006; Sher & Wood, 2005), suggesting that individual differences in the socially reinforcing effects of alcohol may be related to genetic makeup. Social contexts can moderate the impact of genetic risk factors for a wide range of psychopathologies (Moffitt, Caspi & Rutter, 2006) including alcohol-related traits (Dick, Rose, Viken, Kaprio & Koskenvuo, 2001). Indeed, the "contextual triggering" model of Shanahan and Hofer (2005) states that social contexts can trigger a genetic predisposition. The social context in which drinking occurs may be an especially salient environmental factor with potential to modulate genetic influences on alcohol response (Kendler, Gardner & Dick, 2011; Slutske et al., 2004; Volkow & Li, 2004).

Surprisingly, experimental paradigms designed to examine the reinforcing effects of alcohol have largely failed to consider social context. These laboratory studies recruit participants who almost always drink in social settings (Bachman et al., 2006), but nearly all test these social drinkers in isolation (Kirchner et al., 2006). Accordingly, most studies create uncommon conditions to assess the reinforcing effects of alcohol. Without considering social

context, it is unsurprising that investigators have struggled to reliably explain the reinforcing effects of alcohol (see Sayette, 1993) or genetic mechanisms underlying these effects (Fromme et al., 2004).

1.2 CONTROLLED SOCIAL INTERACTIONS IN THE LABORATORY

Group settings offer a unique chance to uncover important reinforcing effects of alcohol that might otherwise go unnoticed when examining participants in isolation (Doty & de Wit, 1995; Kirchner et al., 2006). In fact, many of the subjectively pleasant effects of alcohol that confer increased risk for alcohol misuse (e.g., increased sociability) must be studied in a group setting (de Wit, 2005). There has been little systematic research on the effects of alcohol conducted in group settings, though, and despite the noted importance of contextual variables in the study of genetic effects (Ducci & Goldman, 2008; Goldman et al., 2005), no prior laboratory study has examined the moderating role of genetic variation on alcohol's reinforcing effects in a controlled social setting. Recent advances in small groups research on the one hand (Levine & Moreland, 1990, 1998), and the measurement and analysis of social behavior on the other (Bakeman, 1999; Sayette, Cohn, Wertz, Perrott, & Parrott, 2001a), make the goal of testing alcohol's effects in a social context more attainable than in the past. The present study used the Facial Action Coding System (FACS; Ekman, Friesen, & Hagar, 2002) to reliably and unobtrusively assess participants' facial expressions in real time as they interacted in a controlled social setting (details below).

1.3 GENETIC LINKS TO THE REINFORCING EFFECTS OF ALCOHOL

Uncovering genetic variation associated with individual variability in the reinforcing effects of alcohol has been an active area of research interest over the past several years (Sher & Wood, 2005). Generally, efforts have focused on genetic polymorphisms that have previously been shown to be associated with AUDs in large case-control or family based studies (e.g., Hill, 2010; Hill & Tessner, 2010; Wilhelmsen, Schuckit, Smith, Lee, Segall, Feiler et al., 2003) or on genes that are plausibly involved in the pathophysiology of AUDs (Kwon & Goate, 2000). While several genes have been implicated in AUDs, the present study focuses primarily on genetic variation related to the functioning of the dopamine system, as the brain's reward system is thought to play a major role in both AUDs and social behavior. Given the novelty of the present study's methodology (i.e., a controlled micro-social environment, described in more detail below), genetic variation associated with the functioning of the serotonin and GABA systems was also explored. These systems have also been intensely investigated in an effort to understand the mechanisms of AUDs.

1.4 DOPAMINE

Because both the reinforcing effects of alcohol (Weiss & Koob, 1991) and the rewarding effects of social interactions (Krach, Paulus, Bodden, & Kircher, 2010) are mediated via dopamine-dependent activity of the brain's mesocorticolimbic reward system, polymorphic variations in dopamine-regulating genes offer rational candidates for the genetic study of problematic drinking (Gorwood, Le Strat, Ramoz, Dubertret, Moalic, & Simonneau, 2012; Hill et al., 1999)

and the study of interactions between AUDs and social behaviors (Young, Gobrogge, & Wang, 2011). One particularly prominent polymorphism in psychiatric and behavioral genetics consists of a Variable Number of Tandem Repeats (VNTR) in exon 3 of the gene encoding the dopamine D4 receptor (*DRD4*), represented by common length variants of 2, 4, and 7 repeats in most populations (Van Tol, Wu, Guan, Ohara, Bunzow, Civelli et al., 1992). Activation of the G-protein-linked D4 receptor attenuates intracellular signaling by inhibiting adenylyl cyclase coupling, and this inhibitory effect is blunted by presence of the 7-repeat allele (Asghari, Sanyal, Buchwaldt, Paterson, Jovanovic, & Van Tol, 1995; Oak, Oldenhof & Van Tol, 2000; Ding, Chi, Grady, Morishima, Kidd, Kidd et al., 2002). It is this attenuated response to dopamine produced by the 7-repeat variant that putatively underlies hypothesized associations of this polymorphism with addiction-related phenotypes (McGeary, 2009; Wang, Ding, Flodman, Kidd, Kidd, Grady et al., 2004).

The 7-repeat allele of the *DRD4* polymorphism has been associated with several traits, behaviors, and experiences, such as novelty seeking (Ebstein, Novick, Umansky, Priel, Osher, Blaine et al. 1996), heavy drinking in male adolescents (Laucht, Becker, Blomeyer, & Schmidt, 2007), cigarette smoking (Laucht, Hohm, Esser, Schmidt, & Becker, 2007; Laucht, Becker, El-Faddagh, Hohm, & Schmidt, 2005), cue-elicited craving (Filbey, Ray, Smolen, Claus, Audette, & Hutchinson, 2008; Hutchison, McGeary, Smolen, Bryan & Swift, 2002; Mackillop, Menges, McGeary, & Lisman, 2007; Ray, Miranda, Tidey, McGeary, MacKillop, Gwaltney et al., 2010; cf. van den Wildenberg, Janssen, Hutchison, van Breukelen & Wiers, 2007), pathological gambling (Comings, Gade-Andavolu, Gonzalez, Wu, Muhleman, Chen et al., 2001; Perez de Castro, Ibanez, Torres, Saiz-Ruiz, & Fernandez-Piqueras, 1997), laboratory measures of financial risk taking and inhibitory motor control (e.g., Congdon, Lesch, & Canli, 2008; Dreber,

Apicella, Eisenberg, Garcia, Zamore, Lum et al., 2009; Eisenegger, Knoch, Ebstein, Gianotti, Sándor, & Fehr, 2010; Kuhnen & Chiao, 2009), fairness preference (Zhong, Israel, Shalev, Xue, Ebstein, & Chew, 2010), human assortative mating patterns (Eisenberg, Apicella, Campbell, Dreber, Garcia, & Lum, 2010), and infidelity/sexual promiscuity (Garcia, MacKillop, Aller, Merriwether, Wilson, & Lum, 2010), as well as disorders, such as Attention Deficit/Hyperactivity Disorder (ADHD) (Faraone, Doyle, Mick & Biederman, 2001; Gizer, Ficks, & Waldman, 2009; Li, Sham, Owen & He, 2006). Notably, too, a growing literature shows many developmental effects of this VNTR on early behavioral outcomes (e.g., attachment organization, externalizing disorders, sensation seeking, and prosocial behaviors) to vary as a function of naturally occurring or experimentally manipulated environmental exposures (Bakermans-Kranenburg & van Ijzendoorn, 2011), which in turn marks this polymorphism as a prime candidate for gene-environment interaction (e.g., Sweitzer, Halder, Flory, Craig, Gianaros, Ferrell et al., 2012). Thus, although association studies linking DRD4 genotype to AUDs have largely yielded null findings (e.g., Dick & Foroud, 2003; McGeary, 2009), there is accumulating evidence to suggest that *DRD4* genotype interacts with certain environmental factors to influence alcohol outcomes (Park, Sher, Todorov & Heath, 2011).

Two recent studies underscore the importance of social factors in the link between *DRD4* genotype and alcohol outcomes. Larsen, van der Zwaluw, Overbeek, Granic, Franke, and Engels (2010) reported that individuals carrying the 7-repeat allele drank more in the presence of a heavy-drinking confederate than those of other *DRD4* genotypes, and Park et al. (2011) found college/Greek involvement to be associated with increased risk of alcohol dependence, but only among students with at least one copy of the 7-repeat allele. Taken together, these two studies conducted in different laboratories suggest a gene-environment interaction, such that the *DRD4*

VNTR is associated with problematic drinking only in the presence of certain social-environmental factors (specifically, heavy drinking peers and college/Greek involvement). The pathways by which social factors increase risk for problematic drinking among 7-repeat carriers have yet to be articulated. As noted by Park et al. (2011), "Specific factors in college environments that interact with the *DRD4* gene to increase alcohol dependence in emerging adulthood need to be identified."

One factor of particular relevance to young adults is the formation of social bonds (Baumeister & Leary, 1995). To my knowledge, however, no prior study has examined whether effects of alcohol on social bonding may be moderated by *DRD4* variation (or any other gene polymorphism). Accordingly, the present study sought to extend the findings of Larsen et al. (2010) and Park et al. 2011 to investigate whether experimentally manipulated alcohol consumption would promote social bonding in randomly assigned groups of three unacquainted young adults and would do so differentially among those of differing *DRD4* genotype. It was hypothesized that alcohol would increase perceived social bonding and that individuals carrying the 7-repeat allele would be especially sensitive to alcohol's effects on social bonding.

The current study also examined whether genetic variation in *DRD4* moderates individual differences in the reinforcing effects of alcohol (e.g., enhanced positive affect and reduced negative affect/anxiety). One prior study tested this question and did not find an association between the *DRD4* VNTR polymorphism and self-reported vigor and negative mood after alcohol consumption (Ray et al., 2010). This study used ecological momentary assessment, however, and asked participants to record their responses after having two drinks of alcohol. While this paradigm increased the study's external validity, the key measures (i.e., BAC level and exact time of recorded responses) were estimated and not directly measured. The present

study used an experimental design to manipulate alcohol consumption in the laboratory and used both self-report and behavioral assessments of positive and negative affect (i.e., facial expressions, described below), thus providing a more controlled test of whether individuals carrying the 7-repeat allele experience more affect-related reinforcing effects of alcohol in group settings. It was hypothesized that individuals carrying the 7-repeat allele would be especially sensitive to alcohol's effects on positive and negative affect.

The D2 dopamine receptor (*DRD2*) is also thought to play a critical role in the mechanism of drug reward (e.g., Volkow, Wang, Fowler, Logan, Gatley, Gifford et al., 1999), and the *DRD2* gene on chromosome 11 (q22–q23) is a widely studied candidate gene implicated in AUDs. The most researched polymorphism of the *DRD2* gene is the Taq1A polymorphism (*rs1800497*), a C → T substitution located in a noncoding region of the *DRD2* locus. [More recently, this polymorphism has been described as residing within a neighboring gene (i.e., *ANKK1*; Dubertret, Gouya, Hanoun, Deybach, Adès, Hamon et al., 2004; Neville, Johnstone, & Walton, 2004). The variant will be referred to throughout as the *DRD2* Taq1A polymorphism, though, to reflect the nomenclature used in the majority of published studies to date.] Since the initial report of a link between the *DRD2* Taq1A polymorphism and severe alcoholism (Blum, Noble, Sheridan, Montgomery, Ritchie, & Jagadeeswaran, 1990), about 40 studies have tried to replicate the link between the A1 allele and alcoholism with mixed results (see Smith, Watson, Gates, Ball, & Foxcroft, 2008 for a meta-analysis). More extensive genotyping across *DRD2* and *ANKK1* in the COGA sample suggested that evidence for association was strongest in genetic variants in the *ANKK1* gene and a small number of SNPs in *DRD2* (Dick, Wang, Plunkett, Aliev, Hinrichs, Bertelsen et al., 2007). Nevertheless, a significant excess of the A1 allele of the Taq1A polymorphism compared to the A2 allele was found in alcohol dependent individuals in more

recent meta-analyses (Le Foll, Gallo, Strat, Lu, & Gorwood, 2009; Munafò, Matheson, & Flint, 2007).

Furthermore, although the functional role of the Taq1A polymorphism is unknown, the A1 allele has been associated with low D2 receptor density in postmortem striatal samples from individuals with and without alcohol dependence (Noble, Blum, Ritchie, Montgomery, & Sheridan, 1991), as well as in healthy individuals without psychiatric disorders (Thompson, Thomas, Singleton, Piggott, Lloyd, Perry et al., 1997). In addition, *in vivo* studies have revealed an association between the A1 allele and lower mean metabolic rate in human dopaminergic brain regions (Noble et al., 1997), leading to the hypothesis that the A1 allele links to a general 'reward deficiency syndrome' in humans (Noble, 1998). Due to lower levels of striatal dopamine D2 receptor availability, individuals possessing the A1 allele are thought to have decreased sensitivity to naturally occurring rewards and increased vulnerability to abuse alcohol as a way to compensate for this reward deficiency (Volkow et al., 1999).

Neurobiological data support this claim. Lower striatal D2 receptor availability (associated with the A1 allele) predicted increased reinforcement from drugs of abuse, including alcohol, in non-dependent individuals, suggesting that the A1 allele may be involved in the predisposition to drug abuse (e.g., Volkow, Folwer, Wang, Baler, & Telang, 2009). Also supporting the hypothesis that low levels of D2 receptors may be implicated in the risk of AUDs is the observation that higher than expected D2 receptor availability is found in nonalcoholic members of alcoholic families (Volkow, Wang, Begleiter, Porjesz, Fowler, Telang et al., 2006). The current study sought to examine the role of the Taq1A polymorphism in moderating the rewarding effects of alcohol in a large sample of social drinkers during a controlled social

interaction in the lab. It was hypothesized that individuals carrying at least one copy of the A1 allele would show increased sensitivity to the reinforcing effects of alcohol.

In relation to the TaqIA polymorphism, the C957T SNP of the *DRD2* gene has been associated with more substantial changes in *DRD2* expression *in vitro* (Duan, Wainwright, Comeron, Saitou, Sanders, Gelernter et al., 2003) and *in vivo* in humans (Hirvonen, Laakso, Någren, Rinne, Pohjalainen, & Hietala, 2004). Specifically, the CC genotype, as opposed to CT or TT genotypes, is associated with low striatal *DRD2* availability in healthy volunteers (Hirvonen et al., 2004; Hirvonen, Laakso, Någren, Rinne, Pohjalainen, & Hietala 2005), which preclinical data suggest may modulate reinforcing effects of alcohol (Fadda, Mosca, Colombo, & Gessa, 1989; McBride, Chernet, Dyr, Lumeng, & Li, 1993; Sari, Bell, & Zhou, 2006) and general reward sensitivity (Davis, Levitan, Kaplan, Carter, Reid, Curtis et al., 2008). Furthermore, the C957T polymorphism has been implicated in alcohol dependence. Hill, Hoffman, Zezza, Thalamuthu, Weeks, Matthews et al. (2008) found a twofold increase in likelihood of carrying the T allele among alcohol dependent individuals in a large sample using within family association analyses. [Note: Ponce, Hoenicka, Jiménez-Arriero, Rodríguez-Jiménez, Aragüés, Martín-Suñé et al. (2008) report an association between the C allele and alcohol dependence, but their data are based on a sample of individuals of Spanish descent who likely have allele frequencies that differ from more outbred Caucasian populations, such as the individuals in the Hill et al. (2008) study. Furthermore, Ponce et al. (2008) used a much smaller sample size than is generally recommended for studies using a case/control design.] A more recent study has linked T/T homozygotes with higher levels of self-reported dysfunctional impulsivity and less efficiency with inhibiting prepotent responses (a common, behavioral measure of impulsivity) (Colzato, van den Wildenberg, van Does, & Hommel, 2010), a finding

that is consistent with the notion that the T allele results in a net decrease in DA levels in the synapses of the striatum by decreasing receptor synthesis *in vitro* (Duan et al., 2003) and reducing dopamine tone *in vivo* (Hirvonen, Laakso, Någren, Rinne, Pohjalainen, & Hietala, 2009). The Hill et al. (2008) finding linking the T allele with alcohol dependence is also consistent with these findings.

Studies of the C957T SNP have not been conducted in individuals receiving alcohol. The present research was the first to examine whether *DRD2* C957T variation modifies the rewarding effects of alcohol during a controlled social interaction in the lab. Based on prior literature regarding the role of the C957T variant in alcohol dependence and impulsivity, it was hypothesized that individuals carrying at least one copy of the T allele would show increased sensitivity to the reinforcing effects of alcohol.

1.5 SEROTONIN

The functioning of the serotonin (5HT) system has also been intensely investigated to understand the mechanisms of alcohol use, abuse, and dependence. Research suggests that deficits in serotonergic neurotransmission play a key role in both the etiology and maintenance of alcohol misuse (Beck, Borg, Edman, Fyro, Oxenstierna, & Sedvall, 1984; Heinz, Jones, Gorey, Bennet, Suomi, Weinberger et al., 2003; LeMarquand, Pihl, & Benkelfat, 1994; Mosner, Kuhlman, Roehm, & Vogel, 1997; Nevo & Hamon, 1995), making serotonergic genes good candidates for the study of alcohol-related phenotypes. The serotonin transporter gene (*SLC6A4*) encodes a transmembrane transporter involved in reuptake of serotonin at the synapse (Gelernter, Kranzler, & Cubells, 1997). A functional insertion/deletion polymorphism in the 5' regulatory

region of the serotonin transporter gene (5-HTTLPR) has been described, which results in different transcriptional efficiencies (Heils, Teufel, Petri, Stöber, Riederer, Bengalet et al., 1996). The deletion (or short (S) allele) reduces transcriptional efficiency of the transporter gene by several-fold, resulting in reduced serotonin re-uptake, relative to the alternate long (L) allele (Collier, Stöber, Li, Heils, Catalano, Di Bella et al., 1996). The S allele has been associated with increased trait negative affect (Munafò, Clark, Moore, Payne, Walton, & Flint, 2003; Schinka, Busch, & Robichaux-Keene, 2004) and has predicted diverse psychopathologies, most notably by interacting with environmental factors (e.g., stressful life events) to produce depression (see Karg, Burmeister, Shedden, & Sen, 2011 and Monroe & Reid, 2008). A recent meta-analysis also provides support for the effects of 5-HTTLPR variation on amygdala activation in response to experimentally manipulated exposures to emotional stimuli, suggesting that this locus may account for up to10% of phenotypic variance in emotional reactivity (Munafò, Brown, & Hariri, 2008).

Several studies have examined the relationship between 5-HTTLPR variation and alcohol dependence, but the findings remain equivocal. For instance, some studies have reported no association between 5-HTTLPR and alcohol dependence (Gelernter et al., 1997; Hill, Stoltenberg, Bullard, Li, Zucker, & Burmeister, 2002; Edenberg et al., 1998; Jorm, Henderson, Jacomb, Christensen, Korten, Rodgers et al., 1998; Gorwood, Batel, Ades, Hamon, & Boni, 2000; Kranzler, Lappalainen, Gelernter, & Nellissery, 2002), a link between the S allele and the diagnosis of alcohol dependence (Feinn, Nellissery, & Kranzler, 2005; Hammoumi, Payen, Favre, Balmes, Bernard, Hussonet al., 1999; Lichtermann, Hranilovic, Trixler, Franke, Jernej, Delmoet al., 2000; M. Thompson, Gonzalez, Nguyen, Comings, George, & O'Dowd , 2000), and an association between individuals homozygous for the L allele and greater risk for alcohol

dependence (Schuckit, Mazzanti, Smith, Ahmed, Radel, Iwata et al., 1999). A growing literature has also investigated the role of 5-HTTLPR variation in intermediate phenotypes (e.g., alcohol sensitivity, alcohol consumption) in individuals who are not alcohol dependent (i.e., social drinking populations). Again, these studies have reported inconsistent results, with one study finding no association between 5-HTTLPR and subjective responses to alcohol (intoxication and high) on each limb of the blood alcohol curve (Corbin, Fromme, & Bergeson, 2006), some finding an association between the S allele and lower alcohol sensitivity (Fromme et al., 2004; Türker, Sodmann, Goebel, Jatzke, Knapp, Lesch et al., 1998) and increased alcohol consumption (Herman, Philbeck, Vasilopoulos, & Depetrillo, 2003; Munafò, Lingford-Hughes, Johnstone, & Walton, 2005), and some showing a link between one or two copies of the L allele and lower alcohol sensitivity (Hinckers, Laucht, Schmidt, Mann, Schumann, Schuckit et al., 2006; Hu, Oroszi, Chun, Smith, Goldman, & Schuckit, 2005; Schuckit et al., 1999).

There are likely several reasons for these discrepant results. Only one of the studies (Hu et al., 2005) genotyped individuals for the triallelic 5-HTTLPR genotype (S, L_A, L_G) (Nakamura, Ueno, Sano, & Tanabe, 2000). The three alleles appear to act codominantly (e.g., Zalsman, Huang, Oquendo, Burke, Hu, Brent et al., 2006), with the L_G allele being equivalent in expression to the S allele (Hu et al., 2005), which potentially could explain some of the discrepant findings. Inconsistent findings may also be a result of limited statistical power (due to small sample sizes), inadequate behavioral response paradigms, and the lack of consideration of social context. Since associations of 5-HTTLPR genotype with alcohol response could be highly context dependent, some have recommended testing this polymorphism in larger samples and with "a more refined response paradigm that would allow for a better characterization of high and low responders" (Fromme et al., 2004).

The current study examined 5-HTTLPR in one of the largest alcohol administration studies ever conducted, which is roughly double the size of the largest laboratory study evaluating 5-HTTLPR conducted thus far (Corbin et al., 2006). In addition, it allowed for a more comprehensive, multi-modal assessment of alcohol's reinforcing effects during a controlled social interaction, something which may be particularly relevant for uncovering associations between the 5-HTTLPR genotype and alcohol response (Corbin et al., 2006; Morzorati, Ramchandani, Flury, Li, & O'Conner, 2002; Newlin & Thomson, 1990). Specifically, the present study aimed to extend the literature by assessing not only level of response to alcohol (i.e., intoxication level; Schuckit & Smith, 1996), which was the main outcome measure in most prior studies in this area, but also employing behavioral measures to assess the rewarding effects of alcohol on the ascending limb of alcohol absorption on a moment-to-moment basis (i.e., facial expressions linked to positive and negative affect, described below). In summary, the current study set out to clarify the inconsistent findings regarding 5-HTTLPR and alcohol response by (1) providing a more ecologically valid social drinking paradigm than in prior studies, (2) allowing for precise measurement of alcohol's reinforcing effects, as well as individuals' intoxication level (3) offering a sample size that is at least twice the size of previous studies, and (4) providing the opportunity to examine the triallelic 5-HTTLPR genotype.

Since the S allele has been associated with increased trait negative affect (Munafò et al., 2003; Schinka et al., 2004), greater psychological sensitivity to stress (Gotlib, Joormann, Minor, & Hallmayer, 2008) and, in particular, greater cortisol reactivity to social stress (Way & Taylor, 2010), it was hypothesized that individuals carrying the S allele would be especially sensitive to alcohol's anxiolytic properties during the controlled social interaction than individuals without the S allele. Specifically, these individuals were hypothesized to show less negative affect-

related facial expressions during alcohol consumption and to report less anxiety after consuming alcohol in the unstructured social interaction.

1.6 GAMMA AMINOBUTYRIC ACID (GABA)

GABA is the main inhibitory CNS neurotransmitter, and the negatively reinforcing effects of alcohol are putatively mediated by alcohol's effects on $GABA_A$ receptors (see Fromme & D'Amico, 1999; Korpi, Uusi-Oukari, Wegelius, Casanova, Zito, & Kleinman, 1993). $GABA_A$ agonists have cross tolerance with alcohol (Schuckit & Klein, 1991), and those with alcohol dependence have lower levels of $GABA_A$ functioning (e.g., Weiner, Brozowski, Harris, & Dunwiddie, 1997). After initial reports of a link between a cluster of genes encoding $GABA_A$ subunits and alcohol phenotypes were published (Long et al., 1998; Reich et al., 1998), nine studies including COGA samples, replicated this link wholly or in part (Bierut et al., 2010; see Mathews, Hoffman, Zezza, Stiffler, & Hill, 2007 and Enoch, 2008 for reviews). There is accumulating evidence that genetic variation in genes encoding $GABA_A$ receptor subtypes is implicated in AUDs.

In comparison to the large number of case-control and family based studies described above, few studies have tested the impact of the $GABA_A$ system on response to alcohol. Indeed, no prior studies have examined the *GABRA2* variant, which is the most well-replicated marker associated with AUDS, on alcohol sensitivity. Research indicates, though, that those with hypersensitive GABA systems may be more likely to experience the sedative-like effects of alcohol on the descending limb of alcohol absorption (Holdstock & de Wit, 1999; 2001), implicating variations in this system with differential response to alcohol and to risk of alcohol

dependence (Newlin & Renton, 2010). Among the various $GABA_A$ subunit genes, the $_{\alpha}Y_6$ subunit alleles may affect the overall intensity of response to alcohol (Korpi et al., 1993; Loh, Higuchi, Matsushita, Murray, Chen, & Ball, 2000). A common amino acid (Pro385Ser) substitution of the $GABA_{A\alpha6}$ genotype was described (Radel, Iwata, & Goldman, 1998) and shown to relate to a low response to alcohol in men (Schuckit et al., 1999; Hu et al., 2005). Specifically, there was a trend for Pro/Ser heterozygous males to be more likely than Pro/Pro homozygotes to have lower sensitivity to alcohol, indicating that Ser385 alleles may contribute to a low response to alcohol and subsequent AUDs. These studies, however, offered only preliminary evidence of this relationship, as the sample sizes were small. In fact, the authors warned that any conclusions from these data should be considered tentative, and they encouraged others to test the role of this polymorphism in predicting alcohol response in larger samples that include women (Hu et al., 2005; Schuckit et al., 1999).

The present research did not include measurements taken on the descending limb of alcohol absorption, when GABA's role in influencing alcohol's sedative-like effects is most prominent (e.g., Holdstock & de Wit, 2001). However, given the substantial evidence linking genetic variation in genes encoding $GABA_A$ receptor subtypes to risk for AUDs, and the association of the $GABA_{A\alpha6}$ polymorphism with low response to alcohol in a series of alcohol challenge studies (described above), the present study aimed to extend the current literature by testing associations between the $GABA_{A\alpha6}$ polymorphism and alcohol response in a relatively large sample that included women. Although the GABA system plays a significant role in the regulation of anxiety, it is unclear how genetic variation in the α6 subunit of the $GABA_A$ receptor will relate to socio-emotional responses measured in a group setting. Thus, hypotheses are not offered for this polymorphism and analyses will be exploratory in nature.

1.7 ALCOHOL EXPECTANCIES VS. PHARMACOLOGICAL EFFECTS OF ALCOHOL

Individuals have expectancies about the acute effects of alcohol (Goldman, Del Boca, & Darkes, 1999), and these beliefs have related to alcohol use in both cross-sectional (Palfai & Wood, 2001) and prospective (e.g., Christiansen, Smith, Roehling, & Goldman, 1989) designs. These findings underscore the importance of controlling for alcohol expectancies when examining individual differences in response to alcohol (Fillmore, Vogel-Sprott, & Gavrilescu, 1999; Goldman et al., 1999). Importantly, lab studies that examined the impact of genetic variation on alcohol response have not tended to include placebo conditions (e.g., Corbin et al., 2006) or did not analyze available placebo data (e.g., Schuckit et al., 1999). To my knowledge, no prior study on this topic has included a no-alcohol expectancy control condition. Thus, it is unknown whether documented links between particular genotypes and alcohol response (e.g., Hinckers et al., 2006) reflect alcohol-specific associations, associations attributable to expectancies regarding alcohol's effects, or direct links between gene polymorphisms and response measures. The current study sought to clarify the nature of relationships between specific genotypes and alcohol response measures.

The bulk of behavior studies examining the impact of alcohol on mood and social behavior indicate that the pharmacological effects of alcohol are more important than expectancies about drink content produced by a placebo beverage (see Bushman & Cooper, 1990; Hull & Bond, 1986; Sayette, 1993). Accordingly, the primary hypothesis of the current study is that links between genetic variation and responding will be more prominent in the alcohol condition compared to the placebo and control conditions. By including three drink conditions (i.e., alcohol, placebo, and no-alcohol control, described in the next paragraph), the

present study was uniquely suited to determine whether differences in alcohol response across genotypes represent true pharmacodynamic response variation.

1.8 RATIONALE FOR DRINK CONDITIONS

The four-condition balanced placebo design (Marlatt & Rohsenow, 1980), which purports to assess the separate effects of alcohol and the beliefs that alcohol has been consumed, was not used in the current study, as a wealth of data now exists showing that orthogonal manipulation of drink instruction and actual beverage content at doses exceeding .5g/kg is extremely problematic (Hull & Bond, 1986; Martin & Sayette, 1993; Sher, Wood, Richardson, & Jackson, 2005). This is especially true for the antiplacebo condition in which participants receive alcohol, but are told their drink contains no alcohol at all (e.g., Levenson, Sher, Grossman, Newman, & Newlin, 1980; Lyvers & Maltzman, 1991; Sayette, Smith, Breiner, & Wilson, 1992; Sayette, Breslin, Wilson, & Rosenblum, 1994; Sher et al., 2005; Yankofsky, Wilson, Adler, Hay, & Vrana, 1986). The present study relied on the remaining three conditions of the balanced placebo design (Fillmore et al, 1999; Giancola, 2002; Sayette et al., 1993), as this design provides the necessary controls to test the proposed hypotheses (Testa, Fillmore, Norris, Abbey, Curtin, Leonard et al., 2006). Differences between the alcohol and placebo conditions allow the effect of pharmacology to be estimated, while differences between placebo and control conditions provide an estimate of the effects of believing that one has consumed alcohol (see Martin & Sayette, 1993).

1.9 MEASUREMENT OF ALCOHOL'S REINFORCING EFFECTS

FACIAL EXPRESSIONS. Most prior studies that have attempted to assess differential behavioral responsivity to alcohol have asked participants to report on their subjective feelings of intoxication and/or the negative/positive reinforcing effects of alcohol (see Newlin & Thompson, 1990 and Sayette, Martin, Perrott, Wertz, & Hufford, 2001b for reviews). While self-report measures are essential for assessing the subjective effects of alcohol, they also have important limitations, which may be exacerbated following alcohol consumption (Sher, 1987). For instance, self-reports can be subject to various distortions and biases (Nisbett & Wilson, 1977; Schwarz, 1999), and they likely are distal to the neurobiological mechanisms of interest (Kalivas & Volkow, 2005). The ability to find genetic associations with alcohol's effects may require methods that include more direct assessments of underlying mechanisms (Filbey et al., 2008). These concerns with self-report suggest the utility of a multidimensional assessment strategy.

One complementary approach to self-report is to assess the effects of alcohol unobtrusively, using observational methods. Systematic observational methods using predefined operational behavior codes provide measurement of responses as they unfold over time (Bakeman, 1999). The current study used the most comprehensive, anatomically-based coding system [the Facial Action Coding System (FACS; Ekman et al., 2002)] to identify expressions thought to be related to emotions (Schmidt & Cohn, 2001). FACS allows all possible facial displays, referred to as action units (AUs), to be coded, and this system has been used to examine the effects of alcohol on emotion in individuals (e.g., Kushner, Massie, Gaskel, Mackenzie, Fiszdon, & Anderson, 1997; Levenson, 1987; Sayette et al., 2001a) and in groups (Kirchner et al., 2006). FACS is particularly well-suited to capture socially-relevant emotions, such as social anxiety (Keltner, 1995), felt happiness (Frank, Ekman, & Friesen, 1993; Ruch, 1993), and

negative emotions (Ekman & Friesen, 1982). Furthermore, the use of FACS permitted the accurate assessment of moment-to-moment fluctuations in emotions during the group interaction, rather than asking participants to provide an aggregated summary score of their emotional experience during or after the interaction. As such, the present study provided an optimal test of alcohol's socially reinforcing effects (e.g., de Wit, 2005) by using a group design and systematic measurement of emotional responding, in addition to more traditional measures of the subjective intoxication effects of alcohol (e.g., Biphasic Alcohol Effects Scale, described below).

1.10 GENDER DIFFERENCES IN GENETIC VULNERABILITY TO DEVELOP ALCOHOLISM

Genes contribute to vulnerability to alcohol dependence about equally in men and women (Bierut et al., 2010; Heath, Bucholz, Madden, Dinwiddie, Slutske, Bierut et al., 1997), but it is unclear whether the phenotypic heterogeneity evident across gender represents distinct genetic liabilities or a common genetic vulnerability with differential expression among men and women (see Hill & Smith, 1991). Historically, most studies attempting to uncover risk for AUDs have focused exclusively on male samples (Madrid, MacMurray, Lee, Anderson, & Comings, 2001; Schuckit & Smith, 1996), and far fewer studies have examined genetic links to alcohol response variation in females than in males (Han & Evans, 2005). Although the present study was not powered to examine whether gender moderated the link between genetic variation and response to alcohol, the 246 female participants in this study provided an opportunity to determine whether documented associations between specific polymorphisms and alcohol response measures were also evident in women.

1.11 FAMILY HISTORY OF ALCOHOLISM

Children of alcoholics are at increased risk to develop alcoholism, with risk estimates ranging from 3- to 10-fold compared to children of non-alcohol dependent parents (Cloninger, Sigvardsson, Gilligan, von Knorring, Reich, & Bohman 1988; McGue, 1995; Sher, 1991). Many studies have linked a positive family history with increased sensitivity to alcohol's positive and negative reinforcing effects (Croissant, Rist, Demmel, & Olbrich, 2006; Gabbay, 2005; Sayette, Martin, Perrott, & Wertz, 2001c; see Newlin & Renton, 2010 for review), but no prior study has examined this association in individuals drinking alcohol in a social setting. While the current study was not powered to examine the potential moderating role of family history of alcoholism in the relationship between genotype and alcohol response, it provided a large number of participants with a positive family history of AUDs (n=139 family history positive, and n=235 family history negative). [Note: Data are missing for 53 participants because this questionnaire was added after the start of the study.] A series of correlations was calculated to determine the extent to which a positive family history of alcoholism related to genotype classification, and main effects of family history of alcoholism as well as interactions between family history and alcohol consumption on response variables were explored.

1.12 SUMMARY AND SIGNIFICANCE

Individuals who experience more reward from alcohol in social contexts are at increased risk for the development and maintenance of problematic drinking. There has been little systematic research conducted in group settings, though, and no prior studies have tried to link genetic

variation to alcohol's socially reinforcing effects. Drawing on theory and methods derived from two areas that rarely are integrated in alcohol research (genotyping and the observational measurement of socio-emotional responses in a group setting), this research investigated whether the rewarding effects of alcohol in a group setting are associated with genetic variation implicated in the development of AUDs. By combining human genotyping with a comprehensive multidimensional assessment of alcohol's reinforcing effects in a social context, the mechanisms by which genetic factors influence drinking outcomes may be elucidated.

1.12.1 Specific Aim

The aim of the current study was to:

(1) Determine whether common polymorphic variation in genes associated with increased risk for AUDs [i.e., genes encoding the dopamine D2 and D4 receptors, the serotonin transporter, and the alpha receptor for gamma-aminobutryic acid ($GABA_A$)] are related to socio-emotional responses during a controlled group interaction, and whether these relations reflect alcohol specific associations, associations attributable to beliefs that one has been drinking, or direct links between the polymorphisms and socio-emotional responses.

1.12.2 Specific Hypotheses

(a) Individuals carrying the 7-repeat allele of the *DRD4* VNTR will be especially sensitive to alcohol's socially reinforcing effects (i.e., they will experience enhanced social bonding/positive affect and decreased social anxiety/negative affect assessed across multiple response systems) compared to 7-absent individuals.

(b) Individuals carrying at least one copy of the A1 allele of the *DRD2* Taq1A polymorphism will be especially sensitive to alcohol's socially reinforcing effects compared to individuals homozygous for the A2 allele.

(c) Individuals carrying at least one copy of the T allele of the *DRD2* C957T polymorphism will be especially sensitive to alcohol's socially reinforcing effects compared to individuals homozygous for the C allele.

(d) Individuals carrying the S allele of the 5-HTTLPR polymorphism will be especially sensitive to alcohol's anxiolytic effects (i.e., they will experience decreased social anxiety/negative affect assessed across multiple response systems) compared to individuals homozygous for the L allele.

(e) Collapsing across the two non-alcohol conditions (i.e., the placebo and control conditions), the relationship between genetic variation associated with increased anxiety-related traits (i.e., 5-HTTLPR) and the socio-emotional responses will be opposite in direction to that observed in the alcohol condition. That is, when alcohol is not consumed, persons with genetic variation associated with increased risk to experience anxiety-related traits will show increased negative affect/social anxiety during/after the social interaction.

2.0 METHOD

2.1 OVERVIEW

The present study used data collected in the Alcohol and Smoking Research Laboratory for a prior NIAAA-funded R01 examining the reinforcing effects of alcohol (see Sayette, Creswell, Dimoff, Fairbairn, Cohn, Heckman, et al., in press). The parent dataset included 720 (half female) social drinkers, age 21-28, who were assembled into 240 groups of three unacquainted persons. Each group was randomly assigned to drink a moderate dose of alcohol (males: 0.82 g/kg; females: 0.74 g/kg), a placebo, or a nonalcoholic control drink, over a 36 minute time interval. This group drinking period was digitally recorded, and participants' facial expressions were coded by FACS-certified coders. The parent dataset also included a broad array of questionnaire assessments relating to social bonding, personality, and the subjective effects of alcohol. The present study focused on a subset of Caucasian participants (N=427) that submitted saliva samples for DNA isolation and genotyping. [Note: Seventy percent of the parent sample (n=506) contributed saliva samples for DNA analysis (84.5% European-American, 9.5% African-American, 1 % Hispanic, 2.5% Asian, and 2.5% other). Because allele frequencies for the polymorphisms included in this study are known to vary across different ethnic populations, analyses focus on the 427 Caucasian participants]. The study was approved by the University of

Pittsburgh Institutional Review Board, and informed written consent was obtained from all study participants.

2.2 PARTICIPANTS

Healthy social drinkers were recruited from community and city-wide newspapers. Participants who successfully completed a brief telephone screening were invited to the lab for an additional screening session. Participants were included if they reported social drinking practices (i.e., drinking a mean of at least two drinks on at least one occasion per two weeks, or at least four drinks on at least one occasion per month, over the past year). Participants were excluded based on the following criteria: self-report of an adverse reaction to the type or amount of beverage used in the study; a medical condition that ethically contraindicated alcohol use; a diagnosis of current or past alcohol abuse or dependence based on the *Diagnostic and Statistical Manual of Mental Disorders* (American Psychiatric Association, 1994) criteria; a weight not within 15% of ideal weight for their height, as indexed by the 1983 Metropolitan Life tables (Harrison, 1985); and illiteracy. Women who were pregnant or trying to conceive were also excluded. Sessions for women were conducted between days 3 and 11 of their menstrual cycle, which is a time that is associated with stable responses to stress (Kaplan, Whitsett, & Robinson, 1990) and to alcohol (Sutker, Goist, & King, 1987).

2.3 PROCEDURES

Eligible participants were invited to the experimental session. They were asked to avoid using alcohol or drugs within 24 hr, to avoid consuming caffeine within 4 hr, and to avoid smoking for 1 hr prior to arrival. Participants were told that compliance with these instructions would be confirmed using breath measurement instruments. Participants assigned to the alcohol and placebo conditions were told that they could not drive themselves home from the study; those who needed transportation were given money for a taxi or bus.

2.3.1 Pre-drink Assessment

Upon arrival, participants were given a brief description of the study and signed a consent form. All women took a pregnancy test (ONE-STEP hCG Dipstick Test, SA Scientific, San Antonio, TX). Participants were then given a light snack to prevent hypoglycemic reactions and to slow down the absorption rate of alcohol, thereby increasing the amount of time that they were on the rising limb of the blood alcohol curve (BAC). A subset of participants (n=506) provided a saliva sample for DNA extraction and genotyping. Because genotyping began about 18 months after recruitment started, participants recruited during the first 18 months were invited to return to the lab to provide consent for DNA collection. Of the 214 participants that did not contribute DNA, 201 could not be reached because their phone number was disconnected or changed, 2 moved out of the country, and 11 declined consent for various reasons (e.g., lack of interest, failure to return the saliva kit).

Before group formation, participants completed a baseline assessment battery. Drinking patterns were assessed by asking participants to indicate the number of days per week that they

consume alcohol and the number of drinks consumed on each occasion. Participants completed the NEO Five-factor Inventory (NEO-FFI; Costa & McCrae, 1992), which reliably assesses five domains of adult personality (neuroticism, extraversion, openness to experience, agreeableness, and conscientiousness), and the Biphasic Alcohol Effects Scale (BAES; Martin, Earleywine, Musty, Perrine & Swift, 1993), which discriminates sedating and stimulating effects of alcohol in alcohol administration studies and includes seven items that assess feelings of stimulation (e.g., energized, excited), and seven that assess feelings of sedation (e.g., heavy head, difficulty concentrating).

The baseline assessment also included the Alcohol Expectancy Questionnaire (AEQ) and the Social Interaction Anxiety Scale (SIAS). The AEQ is a widely used measure of the beliefs that people have regarding the general and specific effects of alcohol on their affect and behavior. It is comprised of six scales; Global Positive Change, Enhanced Sexual, Social Pleasure, Social Assertiveness, Relaxation and Tension Reduction, and Arousal and Power. The AEQ has been shown to be a reliable and valid measure, and has predicted future drinking (Brown, Christiansen, & Goldman, 1987). The SIAS contains 19-items designed to assess an individual's general fears and avoidance behaviors concerning social interactions (e.g., distress while initiating and maintaining conversations, anticipatory anxiety of interpersonal situations). Respondents used a five-point Likert-type scale ranging from "*Not at all*" to "*Extremely*." The SIAS has demonstrated strong psychometric properties (Heimberg, Mueller, Holt, Hope, & Schneier, 1992.)

To assess emotional state prior to drinking, participants completed the Positive and Negative Affect Schedule (PANAS; Watson, Clark, & Tellegen, 1988). The PANAS is a reliable and valid measure that comprises two independent affect scales assessing current experiences of

positive (5 items) and negative (5 items) affect. Participants also completed the state version of the State-Trait Anxiety Inventory (STAI-B) immediately before drinking. The STAI-B is a brief (6-item) version of the Spielberger, Gorsuch, and Lushene (1970) STAI-B, which was used to reduce response burden. Participants indicated the extent to which they felt upset, worried, frightened, calm, secure, and self-confident (the latter three items were reverse scored) on a 7-point Likert-type scale ranging from "*Not at all*" to "*Very much so*." Prior research with this brief scale has revealed good psychometric properties and has detected effects of alcohol on anxiety. To assess family history of alcoholism, participants completed the Children of Alcoholics Screening Test (CAST-6; Hodgins, Maticka-Tyndale, El-Guebaly, & West, 1993). This multi-item screening instrument was developed to identify individuals with alcoholic parents and has strong psychometric properties. Finally, an initial BAC reading was obtained using a DataMaster breath alcohol instrument (National Patent Analytical Systems, Mansfield, Ohio), and participants rated their intoxication level using a subjective intoxication scale (SIS) on which 0 = *not at all intoxicated* and 100 = *the most intoxicated I have ever been*.

2.3.2 Drink Administration

Several steps were taken to ensure that the groups included three participants who were unacquainted with one another. First, four people were always scheduled to come in to the lab for the experimental session. They were told that there was a small chance that they might be asked to return on another day, in which case they would be given an additional $20. Second, participants were introduced to each other, one at a time, while two experimenters (one of whom was FACS-certified) checked for any signs of recognition. Finally, participants were told that groups of acquainted and unacquainted participants were needed for the study, which likely

decreased any motivation to falsely deny knowing another participant. These measures were designed to ensure that all of the groups consisted of three strangers.

Group members were told that they would drink their beverages together before completing tasks related to memory and cognitive performance to begin about 40-min later (the ostensible study aim). Participants were told that the group drinking format made it easier to monitor their beverage consumption. They were then seated equidistant from each other around a circular (75 cm diameter) table in the experimental room. Separate wall-mounted cameras faced each person, and a common microphone was placed at the center of the table. Participants were told that if they had any questions during the drink period, they should speak loudly to converse with an experimenter in the adjacent room. Participants were also told that the cameras were focused on their drinks (the cameras appeared to be facing down at the table rather than on the participants' faces), and would be used to monitor their consumption rate from an adjoining room. With the exception of mentioning their level of intoxication, there were no restrictions on what was discussed. A custom-designed video control system permitted synchronized video output for each subject, as well as an overhead image of the group.

The alcoholic beverage was 1 part vodka and 3.5 parts cranberry juice cocktail (Ocean Spray). For those drinking alcohol, the vodka bottle contained 100-proof vodka (Smirnoff); for those drinking a placebo, the vodka bottle contained flattened tonic water (Schweppes). Control participants were told they did not receive alcohol and were given cranberry juice in equal volume. Drinks were mixed in front of participants to increase credibility in the placebo condition, and total beverage was isovolemic in the alcohol, placebo, and control conditions. Prior work shows that this approach offers a successful execution of the placebo manipulation (Martin & Sayette, 1993; Rohsenow & Marlatt, 1981). Beginning at time 0, which ranged from

12:30 to 1:30 p.m., participants received one third of their beverage [alcohol participants were given one third of a moderate dose of alcohol (0.82 g/kg males/0.74 g/kg females)] and were asked to consume it evenly over 12-min. The experimenter re-entered the room just before the end of each 12-min drinking block (at 12- and 24-min) to give participants the middle and final thirds of the drink. During each pour, participants were asked to consume the beverage evenly over 12-min intervals. Other than briefly entering the room to fill participants' glasses, the experimenter was not present during the group drink period. The entire period was recorded.

2.3.3 Post-drink Assessment

After drinking the final third (36-min), participants were separated and BAC and SIS ratings were recorded. The DataMaster calibrates infrared measurement systems prior to each test with an accuracy of +/-0.003% at BAC of .1%. This model provided the rapid assessments needed in this group design, and it was custom-designed for false BAC display in the placebo condition. To help control for dosage set, placebo participants received a BAC reading ranging from .041% to .043% (randomly assigned), which is about the highest credible reading for deceived participants (see Martin & Sayette, 1993). This false reading aids in placebo deception (Rohsenow & Marlatt, 1981) (actual BAC readings were also recorded). While separated, participants completed the Perceived Group Reinforcement Scale (PGRS) to assess the perception of social bonding (described below), the 8-item Mood Measure (8-MM) to assess eight different affective states after drinking (i.e., cheerful, annoyed, upbeat, sad, irritated, happy, bored, content), the STAI-B to assess symptoms of anxiety, and the BAES to assess felt stimulation/sedation after drinking.

The PGRS included 12 items, such as "*I like this group"* and *"The members of this group are interested in what I have to say,*" which were summarized as a composite score (Cronbach's

α=.90). Items were adapted from the Group Attitude Scale (Evans & Jarvis, 1986) and the Perceived Cohesion Scale (Bollen & Hoyle, 1990). The PGRS has good face validity and it has proven sensitive to the effects of alcohol on social bonding in our prior research. Importantly, the PGRS demonstrates good convergent validity as well, as it correlates with other non-verbal measures of social bonding (see Kirchner et al., 2006). Following completion of additional cognitive and decision tasks unrelated to the present study (see Sayette, Dimoff, Levine, Moreland, & Votruba-Drzal, in press), BAC and SIS were again obtained (40-min postdrink). Placebo participants were presented with a false BAC reading between .039% and .037% and, along with control participants, were debriefed, paid $60, and allowed to leave. Alcohol participants recorded their BACs, and ate lunch and relaxed. When their BACs dropped below .025%, they were debriefed, paid $60, and allowed to leave (they were not permitted to drive).

2.4 FACIAL CODING

During drinking, facial expressions were scored separately for each participant by a certified FACS-coder blind to drink content and to the behavior of other group members. Observer Video-Pro software for computer-assisted coding of time-locked video was used (Noldus Information Technology, 2003).

2.4.1 Positive Affect

Combined movement of the zygomaticus major muscles, which pull the lip corners up (AU 12), and the obicularis oculi muscles, which cause the cheeks to lift and produce wrinkles around the

eyes (AU 6), has been shown to reflect positive affective states such as happiness, pleasure, or enjoyment (Frank et al., 1993). Ekman (1989) labeled this smile of enjoyment as the Duchenne smile. Smiles that do not include AU 6 are considered to be social smiles. Social smiling is not an index of positive affect per se, but it is thought to reflect self-presentational concerns and cooperative intention and has been suggested to be an adaptive social signal (DePaulo, 1992). While it is possible to deliberately pose a Duchenne smile (Krumhuber & Manstead, 2009), Duchenne smiles require more effort and are harder to fake than social smiles (Schmidt & Cohn, 2001), and they have been more effective in eliciting facial responses from others (Gonzaga, Keltner, Londahl, & Smith, 2001; Keltner & Bonanno, 1997; Surakka & Hietanen, 1998). Duchenne and social smiles were coded according to FACS (Ekman et al., 2002).

2.4.2 Negative Affect

Specific AUs and AU combinations were classified as negative affect-related AUs on the basis of a review of FACS literature. Negative affect was defined by the presence of any of the following AUs: 9 (nose wrinkle), 14 (dimpler), 15 (lip corner depress), or 20 (lip stretch) which, appear during the expression of negative emotion (Ekman & Friesen, 1982, 1986; Sayette & Parrott, 1999). In line with our previous research (Sayette, Cresswell et al., in press), these AUs were combined to create a composite negative affect code. [Note: Action Units included in the negative affect composite occurred in the absence of a smile, as they may not represent a negative affect state when paired with a smile.] Smile controls (Reed, Sayette, & Cohn, 2007) were coded as an additional indicator of negative affect. Smile controls involve facial actions that potentially counteract the upward pull of the smile and/or obscure the smile (Keltner, 1995). These facial expressions have been linked to socially relevant negative affective states, such as

social anxiety/embarrassment (Keltner & Buswell, 1997). Smile controls were defined by the presence of AU 12 and at least one of the following AUs: 14 (dimpler), 15 (lip corner depress), 23 (lip tightener), or 24 (lips presser).

2.5 RELIABILITY OF MEASUREMENT

Certification in FACS requires the coder to complete a standardized exam and attain an agreement ratio of at least .70. This coding reliability has been shown to generalize to the coding of spontaneous emotions in research settings (see Sayette et al., 2001a). All coders in the proposed study were FACS certified. In addition, reliability coding was assessed on a random sample of 10% of all participants (n=72 participants or 1,944 minutes of FACS coding). Coders were considered in agreement if both coded the same behavior during the same 1-second sampling interval (Kirchner et al. 2006). Cohen's kappa statistic was calculated to assess interrater agreement corrected for chance. Kappa values indicated that the coders generally achieved good agreement (for AU 6, κ = .88; for AU 9, κ =.86; for AU 12, κ = .84; for AU 20, κ = .71). Coders were not able to reliably differentiate between AUs 14 and 15 and, within the context of a smile (i.e., concurrent with AU 12), between AUs 23 (or 24) vs. AUs 14 (or 15) (all κ's < .47 for these AUs). Accordingly, AUs 14 and 15 were merged into one behavior (and merged with AUs 23/24 in the context of smile (κ = .65). Subsequent analyses focus only on these merged AUs.

2.6 DATA ANALYSES

2.6.1 Data Processing

Facial expressions were coded on a frame-by-frame basis, with thirty frames per second. This was accomplished using binary coding (0 or 1) to indicate the presence or absence of each AU during each frame of video. Behavioral counts were then computed to indicate the number of frames per one minute bin (i.e., duration of time) each participant displayed behaviors of interest across the interaction period. To ensure that groups in the three drink conditions did not differ at the beginning of the interaction (i.e., before much alcohol was absorbed), the first 3 minutes of the drinking period were coded and analyzed for all groups. (No differences emerged during this baseline.) This baseline period was entered as a covariate in all models examining behavioral outcomes (see detailed explanation below). Coding was done continuously during consumption of the second and third portions of the drink (i.e., minutes 13 through 36 of the interaction, with the exception of a brief interval during which the investigator entered the room to refill drinks). Just over 23.6 million video frames of behavioral data were coded.

2.6.2 Molecular Genetics Analyses

DNA isolation and genotyping were performed in the Human Genetics Laboratory at University of Pittsburgh under the direction of Dr. Ferrell following standard procedures. DNA was collected from saliva using Oragene kits (DNA Genotek, Kanata, Ontario, Canada), which allowed for long-term preservation and storage of DNA at room temperature. All DNA samples were labeled with an anonymous code designed to protect the privacy of participants. Genomic

DNA was isolated following the manufacturer's protocol. First, DNA samples were heat-treated to maximize DNA yield and to ensure that nucleases were permanently inactivated. Next, DNA samples were mixed with Oragene DNA Purifier to remove impurities and then ice-incubated. After this, each sample was centrifuged at room temperature and the clear supernatant was transferred to a new tube. 100% ethanol was added to precipitate the DNA, and then the tubes were centrifuged again at room temperature. DNA samples were then washed with ethanol and rehydrated.

Candidate polymorphisms were assayed by standard procedures in the Human Genetics Laboratory (see below). Specifically, samples were assayed by DNA amplification of the sequence of interest by PCR using unique sequence flanking primers and the fluorescence polarization method of Chen, Levine, and Kwok (1999) for SNPs and electrophoresis in polyacrylamide gels (for length polymorphisms). Each genotype was scored by two observers by comparison to sequence-verified controls of known genotype assayed in parallel. Significant departures from Hardy-Weinberg equilibrium were tested.

2.6.3 *DRD4* VNTR

The 48 bp VNTR in Exon 3 of *DRD4* was genotyped by the method of Lichter, Barr, Kennedy, Van Tol, and Kidd (1993). Allele and genotype frequencies are presented in Table 1 (genotyping was unsuccessful for 1.2% of the sample). The genotype frequencies did not deviate significantly from Hardy-Weinberg equilibrium (p = .56). Due to the low frequency of individuals homozygous for the 7-repeat allele (2.6%) and in accordance with prior convention (e.g., Larsen et al., 2010), participants were classified as 7-present (i.e., homozygous or heterozygous for the 7-repeat allele) or 7-absent (i.e., neither allele is 7-repeat). [Note: Most studies examining an

association between the *DRD4* VNTR and a multitude of disorders and traits, including alcohol-related phenotypes, have assumed that a linear association exists between repeat length and functionality. We rely, however, on data indicating that this is unlikely, with 10 repeats functionally resembling 2 repeats more so than 7 repeats (Asghari et al., 1995; Jovanovic, Guan, & Van Tol, 1999; Jovanovic et al., 1999; Oak et al., 2000). Regardless, there were only 13 individuals (3%) with > 7 repeats in our sample, and results were unchanged when using the long/short classification of alleles (i.e., including individuals with repeats > 7 in the 7-present classification presented here).] As shown in Table 2, *DRD4* genotypes were evenly distributed across beverage conditions, $\chi^2(df=2, N = 422) = 3.25$, $p = .20$.

2.6.4 *DRD2* Taq1A

The *DRD2* Taq1A (*rs18004970*) polymorphism was genotyped by amplification by the polymerase chain reaction using unique sequence flanking primers, followed by digestion with Taq1 restriction endonuclease according to the method of Dubertret, Gouya, Hanoun, Deyback, Ades, Hamon et al. (2004). Genotyping was unsuccessful for 6.32% of the sample. The distribution of participant genotypes was 18 (4.5%) A1/A1, 128 (32%) A1/A2, and 254 (63.5%) A2/A2. The genotype frequencies did not deviate significantly from Hardy-Weinberg equilibrium ($p = .76$). As in prior studies (see Munafò et al., 2007 for a review), participants were classified as A1 allele carriers (i.e., homozygous or heterozygous for the A1 allele; n=146) or non-carriers (i.e., homozygous for the A2 allele; n=254). As shown in Table 3, *DRD2* genotypes were evenly distributed across beverage conditions, $\chi^2(df=2, N = 400) = .43$, $p = .81$.

[NOTE: The C957T SNP (*rs6277*) of the *DRD2* gene was genotyped by fluorescence polarization by the method of Chen et al. (1999). Genotyping was unsuccessful for the majority of participants, and this polymorphism was omitted from analyses.]

2.6.5 5-HTTLPR

The 5-HTTLPR length polymorphism and SNP *rs25531* in the serotonin transporter gene (*SLC6A4*) were genotyped by the multiplex PCR protocol followed by double restriction endonuclease digestion as described by Wendland, Martin, Kruse, Lesch, and Murphy (2006). Genotyping was unsuccessful for 1.64% of the sample. The distribution of participant genotypes was 111 (26.4%) L_A/L_A, 26 (6.2%) L_A/L_G, 2 (.5%) L_G/L_G, 184 (43.8%) S_A/L_A, 18 (4.3%) S_A/L_G, 78 (18.6%) S_A/S_A, and 1 (.2%) S_A/S_G. Given their functional equivalence (Hu et al., 2006), S_A/S_G and L_G alleles were combined (jointly labeled S) to compare with the L_A allele (labeled L), which produced the following genotype frequencies: 111 (26.4%) L/L, 210 (50%) S/L, and 99 (23.6%) S/S. Based on prior literature and evidence that the S allele might act in a dominant fashion (Heils et al., 1996), participants were then classified by the presence (i.e., S/S and S/L; n=111) or absence (i.e., L/L; n=309) of the S allele (see Munafò et al., 2008 for a review). Frequencies did not deviate significantly from Hardy-Weinberg equilibrium (p = .78). As shown in Table 4, 5-HTTLPR genotypes were evenly distributed across beverage conditions, $\chi^2(df=2, N = 420) = .02$, $p = .99$.

2.6.6 $GABA_{A\alpha6}$

The Pro>Ser substitution (*rs34907804*) in the $GABA_{A\alpha6}$ subunit gene was genotyped by amplification using primers rs34907804-F: 5'-CTGGCCGCAAGCTATTCA-3' and rs34907804-R: 5'-GATCACTTCCTCTGTCTTTG-3' followed by digestion with restriction endonuclease Fok1, and resolution of the fragments on 2% agarose gel. Genotyping was unsuccessful for 3.3% of the sample. The distribution of participant genotypes was 2 (.5%) Ser/Ser, 66 (16%) Pro/Ser, and 345 (83.5%) Pro/Pro. The genotype frequencies did not deviate significantly from Hardy-Weinberg equilibrium (p = .30). Participants were classified as Ser allele carriers (i.e., homozygous or heterozygous for the Ser allele; n=68) or non-carriers (i.e., homozygous for the Pro allele; n=345) (Iwata, Cowley, Deborah, Radel, Roy-Byrne, & Goldman, 1999). As shown in Table 5, $GABA_{A\alpha6}$ genotypes were evenly distributed across beverage conditions, $\chi^2(df=2, N = 413) = .78$, p = .68. [Note: Due to the low frequency of the rare Ser allele (.09), there were very few individuals (i.e., *N*'s of 22-24) in three of the experimental cells, resulting in low power to test the hypothesized associations. Nonetheless, previous studies had smaller sample sizes (Hu et al., 2005; Iwata et al., 1999; Schuckit et al., 1999) and found significant links between the $GABA_{A\alpha6}$ polymorphism and alcohol response and benzodiazepine sensitivity.]

2.7 STATISTICAL MODELING

The primary aim of the analyses was to test whether polymorphic variation in candidate genes implicated in AUDs is related to socio-emotional responses during a controlled social

interaction, and whether these relations reflect alcohol-specific associations, associations attributable to beliefs that one has been drinking, or direct links between the polymorphisms and socio-emotional responses. Although emotion is generally considered to be comprised of loosely coupled response systems (P. Lang, 1968), previous alcohol research has found little relationship among different response modalities (A. Lang, Patrick, & Stritzke, 1999; Sayette, Contrada, & Wilson, 1990). To protect against type-2 error, and to identify aspects of emotion that are most sensitive to alcohol's effects, the self-report and facial-expression measures were analyzed individually (Levenson et al., 1987; Sayette & Wilson, 1991; Sher & Walitzer, 1986). This analytic technique is customary in the alcohol field (e.g., Hu et al., 2005; Ray et al., 2010; Schuckit et al., 1999). To be conservative, genotype effects were only analyzed when significant overall alcohol effects emerged (see Sher & Levenson, 1982). Thus, a set of preliminary analyses were conducted to test the effects of alcohol on socio-emotional responses during group formation. As outlined in the parent study, we hypothesized that alcohol would enhance self-reported bonding and displays of positive affect and reduce displays of negative affect (see Sayette, Creswell et al., in press). The Bonferroni correction was applied in interpreting study results to control for false positives in multiple genotype comparisons. Specifically, results for each dependent variable were compared to a p-value of .01, and results were only discussed when $p < .01$.

Given the nested structure of the data (i.e., behavioral observations are nested within participant, and individuals are nested within groups), hierarchical linear modeling was used to account for interdependence of within-individual and between-participant responses (Kashy & Kenny, 2000). The intraclass correlation coefficient (ICC), which assesses the degree of clustering or non-independence of measures, was calculated to be .23 for PGRS scores (i.e.,

within groups) and to exceed .20 for many AUs (i.e., within person). These ICC values indicated substantial clustering of PGRS scores within groups and behavioral observations within participants (Singer, 1998), which violates a key assumption of the statistical model used by ANOVA (i.e., independence of observations). As such, hierarchical linear modeling, a well-established system for analyzing hierarchically nested data, was employed as the primary data analytic approach (Raudenbush & Bryk, 2002). [Note: Because genotyping began after the start of the study, there was complete (i.e., all three group members) data for less than half (i.e., 48%) of the full sample. Although every participant in the current dataset drank his/her beverage in a group with two other members (which is a crucial element of the study), only 71 "groups" included all three members, 80 "groups" included data for just two members, and 67 "groups" contributed only one member's data to the analyses. A notable strength of hierarchical linear modeling, the primary data analytic technique used in the current study, is that it has been shown to be robust to missing or incomplete data (Raudenbush & Bryk, 2002).]

Facial data (i.e., frame counts) were not normally distributed, with high kurtosis and skewness values. Histograms revealed a large proportion of zero values across the interaction period, with higher values becoming increasingly less frequent. Distributions such as these are characteristic of count variables, and the Poisson distribution has been shown to be a good fit for these data characteristics (Atkins & Gallop, 2007; King, 1988). The Poisson distribution makes very stringent assumptions about the dispersion of data, however, such that the variance is assumed to be equal to the mean. This assumption rarely holds with real data, though (Coxe, West, & Aiken, 2009), and an overdispersion parameter is required. Therefore, hierarchical generalized linear modeling specifying a log link function and Poisson-distributed errors was used to examine behavioral outcomes (Agresti, Booth, Hobert, & Caffo, 2000; Littell, Milliken,

Stroup, & Wolfinger, 1996), and overdispersion of level-1 variance was measured and accounted for in the analyses (Atkins & Gallop, 2007). Results from models with robust standard errors are reported to protect against potential violations of model assumptions. A complete, orthogonal set of contrast codes was used that compared (a) alcohol to both no alcohol groups and (b) placebo to control. Significant findings were followed up by independently comparing placebo and control conditions to the alcohol condition.

All individual-level behavioral responses were examined in models that included three levels of analysis, accounting for within-individual observations across time at level one, individual-level variables (i.e., genotype classification) at level two, and group-level variables (i.e., beverage condition) at level three (see equations below). Consistent with many prior studies (e.g., Sayette & Hufford, 1995), women were more expressive than men (all p's $<.001$), and thus gender was entered as a control variable into all behavioral models. Covariates (i.e., individual-level baseline behavior and gender) were entered at level-2, in accordance with the standard means-as-outcomes procedure outlined by Raudenbush and Bryk (2002). Due to the small size of the groups in this study (three members) models examining cross-level interactions between individual and group-level variables estimated level-2 slopes as fixed, modeling the interdependence of groups in the random variation of the intercepts (Kashy & Kenny, 2000; Kenny, Mannetti, Pierro, Livi, & Kashy, 2002).

Below is a description of the model used to test for a main effect of *DRD4* genotype as well as a *DRD4* genotype by beverage condition interaction on behavioral outcome variables. Discrete analyses were run to examine each of the following variables individually: Duchenne smiles, social smiles, negative affect composite, and smile controls (descriptions above). The term "facial affect" will be used broadly to represent all facial behavioral dependent variables, as

the same overall model structure was used in each case. Tests of the other three genotypes replicated this model building strategy.

2.7.1 Level-1 Model

$$\ln(\text{FacialAffect}_{ijk}) = \pi_{0jk} + e_{ijk}$$

At Level-1, the natural logarithm of within-individual facial affect of individual "j" at time "i" in group "k" was modeled as a function of average individual-level differences (π_{0jk}) and a random variance component (e_{ijk}). Facial affect was measured as frame counts per one minute bin (i.e., duration of time) that each participant displayed the behavior of interest.

2.7.2 Level-2 Model

$$\pi_{0jk} = \beta_{00k} + \beta_{01k}*(\text{BaselineAffect}_{jk}) + \beta_{02k}*(\text{DRD4}_{jk}) + \beta_{03k}*(\text{GENDER}_{jk}) + r_{0jk}$$

At Level-2, between-individual facial affect is modeled as a function of average group-level intercept (β_{00k}), individual-level predictors, and a random variance component (r_{0jk}), the latter of which models unexplained between-individual variance at level-2. Covariates (i.e., individual-level baseline behavior and gender) as well as predictors (i.e., *DRD4* genotype) were entered at this level. These variables were centered or contrast coded.

2.7.3 Level-3 Model

$$\beta_{00k} = \gamma_{000} + \gamma_{001}(\text{AlcoholVs.NoAlcohol}_{k}) + \gamma_{002}(\text{PlaceboVs.Control}_{k}) + u_{00k}$$

$$\beta_{01k} = \gamma_{010}$$

$\beta_{02k} = \gamma_{020} + \gamma_{021}(\text{AlcoholVs.NoAlcohol}_k) + \gamma_{022}(\text{PlaceboVs.Control}_k)$

$\beta_{03k} = \gamma_{030}$

At Level-3, group-level differences were modeled as a function of the grand mean (γ_{000}), group-level predictors, and a random variance component (u_{00k}), the latter of which models unexplained between-group variance at level-3. Here, group characteristics (i.e., beverage condition) were used to predict average facial affect in groups. A complete, orthogonal set of contrast codes that compared (a) alcohol to both no alcohol groups and (b) placebo to control were entered at this level to test the interaction between *DRD4* genotype and beverage condition (see full mixed model below). The significance of the *DRD4*AlcoholVs.NoAlcohol* slope coefficient in the mixed model (γ_{021}) tested whether *DRD4* genotype moderated facial affect displays of individuals consuming alcohol.

2.7.4 Combined Model

$$\ln(\text{FacialAffect}_{ijk}) = \gamma_{000} + \gamma_{001}(\text{AlcoholVs.NoAlcohol}_k) + \gamma_{002}(\text{PlaceboVs.Control}_k) + \gamma_{010}*(\text{BaselineAffect}_{jk}) + \gamma_{020}*(\text{DRD4}_{jk}) + \gamma_{021}*(\text{DRD4}_{jk})*(\text{AlcoholVs.NoAlcohol}_k) + \gamma_{022}*(\text{DRD4}_{jk})*(\text{PlaceboVs.Control}_k) + \gamma_{030}*(\text{GENDER}_{jk}) + r0jk + u00k$$

Individual-level self-report responses were examined in two-level models, with individual-level variables (i.e., genotype classification) entered at level one and group-level variables (i.e., beverage condition) entered at level two. Below is a description of the model used to test for a main effect of *DRD4* genotype as well as a *DRD4* genotype by beverage condition interaction on self-report responses. Discrete analyses were run to examine each of the self-report response variables (e.g., PGRS, BAES—descriptions above) individually. The term "self-report" will be used broadly to represent all individual-level self-report responses, as the same

overall model structure was used in each case. Tests of the other three genotypes replicated this model building strategy.

2.7.5 Level-1 Model

$$\text{Self-Report}_{ij} = \beta_{0j} + \beta_{1j}*(\text{DRD4}_{ij}) + r_{ij}$$

2.7.6 Level-2 Model

$$\beta_{0j} = \gamma_{00} + \gamma_{01}*(\text{ALCCNTRA}_j) + \gamma_{02}*(\text{PLACCNTR}_j) + u_{0j}$$

$$\beta_{1j} = \gamma_{10} + \gamma_{11}*(\text{ALCCNTRA}_j) + \gamma_{12}*(\text{PLACCNTR}_j)$$

2.7.7 Combined Model

$$\text{Self-Report}_{ij} = \gamma_{00} + \gamma_{01}*\text{ALCCNTRA}_j + \gamma_{02}*\text{PLACCNTR}_j + \gamma_{10}*\text{DRD4}_{ij} + \gamma_{11}*\text{ALCCNTRA}_j*\text{DRD4}_{ij} + \gamma_{12}*\text{PLACCNTR}_j*\text{DRD4}_{ij} + u_{0j} + r_{ij}$$

3.0 RESULTS

3.1 PARTICIPANT CHARACTERISTICS AND RANDOMIZATION

Participants (N=427; males=50.12%) had a mean age of 22.3 years (SD=1.8). Participants drank on average slightly more than twice a week [M= 3.79 (SD = 0.90) using a 7-point scale with "3" = 1-2 occasions/week and "4" = 2-3 occasions/week] and consumed an average of 4.30 (*SD* = 1.91) drinks per occasion. Randomization was effective in creating similar experimental groups. Hierarchical linear modeling was used to determine that the following participant characteristics and baseline variables were equivalent across the three drink conditions: gender, age, marital status, income, family history status (i.e., positive/negative for presence of alcoholism in biological parent), felt stimulation/sedation prior to drinking (as assessed by the BAES), positive/negative affect (as assessed by the PANAS), prior drinking patterns (i.e., drinking amount and frequency), expectancies regarding alcohol's effects (as assessed by the AEQ), social interaction anxiety level (as assessed by the SIAS), the 5 personality dimensions assessed by the NEO-FFI, and smoking status (all *p*'s > .20). There were also no significant differences in observational data collected during the first 3 minutes of the drinking period across the three drink conditions (i.e., Duchenne smiles, social smiles, negative affect composite, and smile controls) (all *p*'s > .46). Significant differences emerged across drink conditions on the STAI-B at baseline. Specifically, participants assigned to consume placebo reported significantly more

anxiety at baseline than participants assigned to consume alcohol (b = 0.18, t (215) = 2.18, p = .03) or the no-alcohol control beverage, (b = 0.27, t (215) = 3.18, p = .002). As such, baseline STAI-B was entered as a covariate in subsequent analyses.

3.2 BEVERAGE MANIPULATION CHECK

BACs and SIS scores across drink conditions appear in Table 6. Alcohol participants were on the ascending limb of the BAC curve with a BAC about .06% following the interaction period. Consistent with our prior studies (Sayette et al., 2001b; 2001c), placebo participants reported experiencing some level of intoxication, more than control participants and less than alcohol participants.

3.3 MAIN EFFECTS OF BEVERAGE CONDITION ON SOCIO-EMOTIONAL RESPONSES

Scores for self-reported socio-emotional responses ($M \pm SE$) assessed after the interaction period across beverage conditions are shown in Table 7. Table 8 depicts average durations for facial expressions ($M \pm SE$) evinced across time during the interaction.

3.3.1 Perceived Social Bonding

Hierarchical linear modeling analyses examining main effects of beverage condition on social bonding revealed that participants drinking alcohol reported significantly higher perceived social

bonding on the PGRS than participants not consuming alcohol, (b = 0.42, t (215) = 2.25, p = .026). Follow-up contrasts showed that alcohol participants reported higher PGRS scores than placebo participants (p = .002), but similar scores to control participants (p=.65). In addition, placebo participants reported significantly lower PGRS scores than control participants (b = -0.49, t (215) = -2.65, p = .01).

3.3.2 Alcohol Response/Affect Measures

After beverage consumption, participants drinking alcohol reported significantly higher levels of stimulation on the BAES than participants not consuming alcohol, (b = 1.35, t (215) = 5.33, p< 0.001). Follow-up contrasts revealed that alcohol participants reported higher stimulation compared to both placebo (p < .001) and control participants (p < .001). Placebo and control participants did not differ on their reported level of stimulation after beverage consumption (p = .65). Participants drinking alcohol also reported significantly higher levels of sedation on the BAES than participants not consuming alcohol, (b = 0.43, t (215) = 2.40, p = .017). Follow-up contrasts showed that alcohol participants reported higher levels of sedation than control participants (p < .001), but similar scores to placebo participants (p=.98). In addition, placebo participants reported significantly higher levels of sedation than control participants (b = 0.63, t (215) = 4.43, p < .001).

Participants drinking alcohol reported higher positive affect scores on the 8-item mood measure after the drinking period than participants not consuming alcohol, (b = 0.38, t (215) = 3.49, p< 0.001). Follow-up contrasts showed that alcohol participants reported higher positive affect compared to both placebo participants (p < .001) and control participants (p = .019). Placebo and control participants did not differ on their reported positive affect (p = .22).

Participants drinking alcohol also reported lower negative affect scores on the 8-item mood measure than participants not consuming alcohol, (b = -0.49, t (215) = -6.67, p = < 0.001). Follow-up contrasts revealed that alcohol participants reported lower negative affect compared to both placebo (p < .001) and control participants (p < .001). Placebo and control participants did not differ on their reported negative affect (p = .23). Participants did not differ on their reported level of anxiety as assessed by the STAI-B after drinking (all p's >.11).

3.3.3 Facial Expressions

During the interaction, participants drinking alcohol displayed Duchenne smiles for significantly more frames (i.e., significantly more time) than those drinking nonalcoholic beverages, (b = 0.79, t (215) = 8.52, p = < 0.001). Follow-up contrasts revealed that alcohol participants expressed Duchenne smiles for significantly more time compared to both placebo (p < .001) and control participants (p < .001). There were no differences between placebo and control conditions in the time spent displaying Duchenne smiles (p = .13). Participants drinking alcohol also displayed Social smiles for significantly more time than those drinking nonalcoholic beverages, (b = 0.22, t (215) = 2.67, p = 0.008). Follow-up contrasts showed that alcohol participants expressed Social smiles for significantly more time compared to placebo participants (p = .002) but similar time to control participants (p = .23). In addition, placebo participants spent significantly less time displaying Social smiles than control participants (b = -0.18, t (215) = -2.28, p = .024).

Participants drinking alcohol displayed negative affect (as assessed by the negative affect composite index) for significantly less time than those drinking nonalcoholic beverages, (b = -0.52, t (215) = -4.11, p = < 0.001). Follow-up contrasts revealed that alcohol participants

expressed negative affect for significantly less time compared to both placebo ($p < .001$) and control participants ($p < .001$). There were no differences between placebo and control conditions in the time spent displaying negative affect ($p = .46$). Finally, participants drinking alcohol displayed smile controls for significantly less time than those drinking nonalcoholic beverages, ($b = -0.66$, t (215) = -5.11, $p = < 0.001$). [Note: Since smile controls were coded only in the presence of a smile, a variable reflecting the duration of smiling was entered into the model as a control variable at level-1.] Follow-up contrasts revealed that alcohol participants displayed smile controls for significantly less time compared to both placebo ($p < .001$) and control participants ($p < .001$). There were no differences between placebo and control conditions in the time spent displaying smile controls ($p = .75$).

Correlations between outcome measures are presented in Table 9.

3.4 GENOTYPE EFFECTS ON PARTICIPANT CHARACTERISTICS AND BASELINE MEASURES

3.4.1 *DRD4* VNTR

Gender, age, marital status, income, family history status, felt stimulation/sedation prior to drinking (as assessed by the BAES), negative affect (as assessed by the PANAS at baseline), prior drinking patterns, expectancies regarding alcohol's effects (as assessed by the AEQ at baseline), social interaction anxiety level (as assessed by the SIAS at baseline), anxiety (as assessed by the STAI-B), the 5 personality dimensions assessed by the NEO-FFI, and smoking status were equivalent across *DRD4* genotypes. Carriers of the 7-repeat allele reported higher

positive affect scores on the PANAS at baseline (M=26.3, SD=6.7) than 7-absent participants (M=24.8, SD=6.4), F (1, 421) = 4.91, p = .027. [Note: Analyses regarding genotype effects on baseline assessment measures were carried out using ANOVA rather than hierarchical linear modeling, as participants were not yet assigned to groups. Results were unchanged, however, when using hierarchical linear modeling for baseline analyses. Unless otherwise specified, there were no differences on baseline measures for the remaining genotypes described below.]

3.4.2 *DRD2* Taq1A

There were *DRD2* genotype effects on three of the subscales of the AEQ at baseline (there were no differences on the AEQ Total score across *DRD2* genotypes). Specifically, compared to A2/A2 individuals, participants carrying the A1 allele reported (1) lower expectations regarding alcohol's ability to enhance sexual satisfaction (M=2.2, SD=2.0) than did A2/A2 individuals (M=1.6, SD=1.8), F (1, 397) = 7.2, p = .008, (2) a trend toward lower expectations regarding alcohol's ability to provide social pleasure (M=7.5, SD=1.3) than did A2/A2 individuals (M=7.2, SD=1.4), F (1, 397) = 3.75, p = .053, and (3) a trend toward lower expectations regarding alcohol's ability to reduce tension/provide relaxation (M=5.5, SD=2.2) than did A2/A2 individuals (M=6.0, SD=2.1), F (1, 397) = 3.24, p = .072. All other baseline measures were equivalent across *DRD2* genotypes.

3.4.3 5-HTTLPR

There was a non-significant trend for carriers of the S allele to report higher sedation levels on the BAES at baseline (M=1.86, SD=1.4) than L/L participants (M=1.58, SE=1.4), F (1, 414) =

2.96, p = .086. There was also a non-significant trend for carriers of the S allele to report higher extraversion scores on the NEO-FFI (M=33.3, SD=6.5) than L/L participants (M=32.08, SD=6.6), F (1, 417) = 2.81, p = .095. Otherwise, baseline measures were not significantly different across 5-HTTLPR genotypes.

3.4.4 $GABA_{A\alpha6}$

Carriers of the Ser allele had lower agreeableness scores on the NEO-FFI (M=31.23, SD=6.5) than Pro/Pro participants (M=32.9, SD=6.0), F (1, 411) = 4.01, p = .045. In addition, there was a non-significant trend for carriers of the Ser allele to be more likely to be male, χ^2(df=2, N = 413) = 3.36, p = .066. Finally, there was a non-significant trend for carriers of the Ser allele to report higher drinking amounts per drinking occasion (M=4.21, SD=1.8) than Pro/Pro participants (M=4.62, SD=2.0), F (1, 411) = 2.57, p = .109. All other baseline measures were equivalent across $GABA_{A\alpha6}$ genotypes.

3.5 TESTS OF SPECIFIC AIM: INTERACTIONS BETWEEN BEVERAGE CONDITION AND GENOTYPE ON SOCIO-EMOTIONAL RESPONSES

3.5.1 DRD4 VNTR

3.5.1.1 Perceived Social Bonding

As can be seen in Table 10, there was no main effect of *DRD4* genotype on PGRS scores (p >.2). As predicted, there was a significant *DRD4* genotype by alcohol consumption interaction, (p =

.008). As depicted in Figure 1, follow-up contrasts revealed that 7-present individuals reported higher PGRS ratings in the alcohol-consuming condition (M=7.4, SE=.17) than in the no-alcohol consuming condition (M=6.6, SE=.14; p =.0006), whereas alcohol consumption did not significantly affect ratings of 7-absent carriers (alcohol; M=7.0, SE=.15: no-alcohol; M=7.0, SE=.11; p =.82). To further probe this significant interaction, contrasts across genotypes and all three beverage conditions were examined (see Table 11 for PGRS scores ($M \pm SE$) across the six experimental cells). As Shown in Figure 2, 7-repeat carriers reported higher PGRS ratings in the alcohol condition than in either the placebo (t = 2.08, p<.0001) or control conditions (t = 3.94, p<.04), whereas alcohol did not significantly affect ratings of 7-absent carriers (p's > .17). Results were unchanged when gender and baseline positive affect (as assessed by the PANAS) were entered as covariates in the model. [Note: Several steps were taken to ensure that the effects were not due to possible outliers. First, skewness (-0.8) and kurtosis tests (0.5) suggested that the data were fairly evenly distributed. Visual inspection of the PGRS data distribution, along with the acceptable skewness value, suggested that the distribution could be considered symmetrical. To be certain of this, however, individuals whose PGRS score fell outside 3 standard deviations of the mean (n=3) were removed and the analyses were re-run in this smaller dataset. The results did not change. Thus, results do not appear to be driven by a few individuals with extreme scores.]

The specified model explained 3% of level-1 variance and 13% of level-2 variance (represented by proportional reductions in the variance-component residual of each additional model in comparison with the empty model without explanatory variables; Singer & Willett, 2003). Table 12 shows the variance components and goodness of fit characteristics associated with each model estimated. As can be seen, the model with the interaction term included

provided a significantly better fit to the data than both the empty model and the model including only main effect estimates.

3.5.1.2 Alcohol Response/Affect Measures

Among those drinking alcohol, there were no differences between 7-present and 7-absent genotypes on BACs, on ratings of subjective intoxication (as assessed by the SIS), on felt stimulation and sedation (as assessed by the BAES), on positive and negative affect (as assessed by the 8-item mood measure), and on anxiety (as assessed by the STAI-B) after alcohol consumption (all *p*'s >.20). These results suggest that the PGRS findings are unlikely due to systematic differences between the two genotype groups on the above mentioned variables.

3.5.1.3 Facial Expressions

There were no significant main effects of *DRD4* genotype on any of the behavioral outcomes (i.e., Duchenne smiles, social smiles, negative composite, and smile controls), but there was a trend for 7-present individuals to display Duchenne smiles for more time than 7-absent individuals (b = 0.09, t (200) = -1.78, p = 0.078). In addition, *DRD4* genotype moderated displays of social smiles between individuals consuming placebo and control beverages, such that among 7-absent individuals, control participants displayed social smiles for significantly longer than did placebo participants (see Table 13). Otherwise, there were no significant *DRD4* genotype by drink condition interactions on behavioral outcomes (all other *p*'s > 0.24).

3.5.2 *DRD2* Taq1A

There were no significant main effects of *DRD2* genotype and no significant *DRD2* genotype by drink condition interactions on PGRS or any other self-report measure after alcohol consumption (all *p*'s > 0.45). There were also no significant main effects of *DRD2* genotype and no significant *DRD2* genotype by drink condition interactions on behavioral outcomes (all *p*'s > 0.32). Among those drinking alcohol, there were no differences between *DRD2* genotypes on BACs.

3.5.3 5-HTTLPR

3.5.3.1 Perceived Social Bonding

As depicted in Table 14, there was a main effect of 5-HTTLPR genotype on PGRS such that individuals carrying one or two copies of the S allele reported significantly less perceived social bonding on the PGRS (M=6.93, SE=.08) than did L/L individuals (M=7.22, SE=.12). 5-HTTLPR genotype did not significantly interact with beverage condition to affect PGRS scores (*p*'s > .56).

3.5.3.2 Alcohol Response/Affect Measures

As shown in Table 15, there was a main effect of 5-HTTLPR genotype on negative affect after beverage consumption such that individuals carrying one or two copies of the S allele reported significantly more negative affect on the 8-item mood measure (M=.62, SE=.06) than did L/L individuals (M=.52, SE=.03). As can be seen, 5-HTTLPR genotype did not significantly interact with drink condition to predict negative affect scores (*p* > .96). There were no significant 5-HTTLPR genotype by drink condition interactions on any self-report measures after alcohol

consumption (all *p*'s > .38). Among those drinking alcohol, there were no differences between 5-HTTLPR genotypes on BACs (*p*'s >.42).

3.5.3.3 Facial Expressions

There were no significant main effects of 5-HTTLPR genotype on behavioral outcomes. Table 16 shows that 5-HTTLPR genotype moderated displays of social smiles between individuals consuming placebo and control beverages. Follow-up contrasts revealed that, among individuals carrying the S allele, placebo participants displayed social smiles for significantly longer than did control participants (p = 0.01). Otherwise, there were no significant 5-HTTLPR genotype by drink condition interactions on behavioral outcomes (all other *p*'s > 0.24).

3.5.3.4 Supplementary Analyses

To explore possible associations between the S/S genotype of 5-HTTLPR and outcome measures, in a series of supplementary analyses, individuals were grouped as being either homozygous for the S allele (i.e., S/S; n=99) or heterozygous or homozygous for the L allele (i.e., S/L and L/L; n=321) (Uher & McGuffin, 2007). The main effect of 5-HTTLPR genotype on PGRS became more pronounced with this classification (see Table 17). Individuals homozygous for S allele reported significantly less perceived social bonding on the PGRS (M=6.67, SE=.14) than did S/L and L/L individuals (M=7.11, SE=.07). Again, 5-HTTLPR genotype did not significantly interact with beverage condition to affect PGRS scores (*p*'s > .34). As depicted in Table 18, there was an interaction between 5-HTTLPR genotype and alcohol consumption on self-reported negative affect after the interaction period. Follow-up contrasts revealed that individuals carrying one or two copies of the L allele who drank alcohol reported significantly less negative affect after the interaction period (M=.266, SE=.05) than individuals carrying one

or two copies of the L allele who drank non-alcohol beverages (M=.682, SE=.04; p <.001). Contrary to predictions, alcohol did not significantly reduce negative affect in S/S individuals (alcohol M=.477, SE=.09; no-alcohol M=.689, SE=.07; p = .13).

Finally, there was a main effect of 5-HTTLPR genotype on the amount of time displaying negative affect during the interaction period (see Table 19), such that S/S individuals spent more time displaying negative affect than did S/L and L/L individuals. 5-HTTLPR genotype did not interact with beverage condition to influence negative facial expressions (p's >.74). There were no other significant effects using this classification of 5-HTTLPR genotypes.

3.5.4 GABAAα6

3.5.4.1 Perceived Social Bonding

As can be seen in Table 20, there was a main effect of $GABA_{A\alpha6}$ genotype on PGRS scores such that individuals carrying one or two copies of the Ser allele reported significantly less perceived social bonding on the PGRS (M=6.46, SE=.18) than did Pro/Pro individuals (M=7.09, SE=.07). $GABA_{A\alpha6}$ genotype did not interact with beverage condition to influence perceptions of social bonding (p's >.59).

3.5.4.2 Alcohol Response/Affect Measures

Table 21 shows that there was a main effect of $GABA_{A\alpha6}$ genotype on stimulation scores after the interaction period such that individuals carrying one or two copies of the Ser allele reported significantly lower stimulation scores on the BAES after the interaction period (M=4.0, SE=.23) than did Pro/Pro individuals (M=4.8, SE=.10). $GABA_{A\alpha6}$ genotype did not interact with beverage condition to influence stimulation scores (p's >.49). As shown in Table 22, there was also a main

effect of $GABA_{A\alpha6}$ genotype on positive affect scores such that individuals carrying one or two copies of the Ser allele reported significantly lower positive affect on the 8-item Mood Measure after the interaction period (M=3.12, SE=.11) than Pro/Pro individuals (M=3.43, SE=.04). $GABA_{A\alpha6}$ genotype did not interact with beverage condition to influence positive affect (*p*'s >.48). A main effect of $GABA_{A\alpha6}$ genotype on negative affect scores (see Table 23) revealed that individuals carrying one or two copies of the Ser allele reported significantly higher negative affect after the interaction (M=.74, SE=.08) than did Pro/Pro individuals (M=.51, SE=.03). $GABA_{A\alpha6}$ genotype did not interact with beverage condition to influence negative affect (*p*'s >.24). Since there was a trend ($p = .06$) for males to be more likely than females to carry one or more copies of the Ser allele, gender was entered as a covariate in the above analyses focusing on self-report data. Results were unchanged with the addition of this covariate to the models. Finally, among those drinking alcohol, there were no differences between $GABA_{A\alpha6}$ genotypes on BACs (*p*'s > .47).

3.5.4.3 Facial Expressions

There were no significant main effects of $GABA_{A\alpha6}$ genotype and no significant $GABA_{A\alpha6}$ genotype by drink condition interactions on behavioral outcomes (all *p*'s > 0.21).

3.5.5 Family History of Alcoholism

Family history status did not correlate with any of the genotypes (all *p*'s >.32). Family history status exerted no main effects nor did it interact with alcohol consumption to affect any of the self-report or behavioral outcome measures (all *p*'s >.11).

4.0 DISCUSSION

The current study is the first to examine the impact of genetic variation, alcohol consumption, and alcohol reward (including enhanced perceptions of social bonding) among previously unacquainted individuals in a controlled group setting. The specific aim was to determine whether common polymorphic variation in genes associated with increased risk for AUDs [i.e., genes encoding the dopamine D2 and D4 receptors, the serotonin transporter, and the alpha receptor for gamma-aminobutryic acid ($GABA_A$)] were related to socio-emotional responses during a controlled group interaction, and whether these relations reflected alcohol specific associations, associations attributable to beliefs that one has been drinking, or direct links between the polymorphisms and socio-emotional responses.

4.1 DOPAMINE

4.1.1 *DRD4* VNTR

It was hypothesized that individuals carrying the 7-repeat allele of the *DRD4* VNTR would be especially sensitive to alcohol's socially reinforcing effects compared to 7-absent individuals. This study provides initial evidence for a moderating effect of the *DRD4* polymorphism on the relationship between alcohol consumption and perceived social bonding. Specifically, 7-present

individuals reported increased social bonding in an unstructured group setting after drinking alcohol, compared to placebo and non-alcohol control beverages. In contrast, alcohol did not affect perceived social bonding of 7-absent individuals. These results converge with and extend those of Larsen et al. (2010) and of Park et al. (2011) suggesting that *DRD4* may be linked to the development of problematic drinking partly through the formation of social relationships. Developing interpersonal relationships is a fundamental human motivation (Baumeister & Leary, 1995), and behaviors that support the formation of social bonds are highly rewarding (e.g., Shore & Heerey, 2011). Current results suggest that one possible pathway by which alcohol may become more reinforcing for 7-repeat carriers is by the facilitation of perceived social bonding.

Social reward and the reinforcing effects of drugs of abuse, including alcohol, are mediated in part through the mesocorticolimbic dopamine system (Krach et al., 2010), and recent studies have focused on the role of dopamine in regulating interactions between alcohol and social factors (e.g., Young et al., 2011). Because 7-repeat carriers may be more sensitive to the dopamine response triggered by priming doses of alcohol and alcohol-related cues (Hutchison et al., 2002; Larsen et al., 2010; Ray et al., 2010), they may perceive enhanced social bonding while drinking due to an augmented dopamine response in the brain's reward circuitry. This explanation is generally consistent with prior reports showing that 7-present individuals respond to alcohol consumption with increased craving (Hutchison et al., 2002) and respond to positive-feedback with increased reward-related reactivity in the ventral striatum (Forbes, Brown, Kimak, Ferrell, Manuck et al., 2009) compared to 7-absent individuals.

Consistent with prior studies (Hutchison et al., 2002; Ray et al., 2010), 7-present individuals reported feeling neither more intoxicated nor more stimulated (e.g., elated, energetic, excited) after alcohol consumption, indicating that 7-present individuals generally did not appear

to be feeling more of the euphoric effects of alcohol than 7-absent individuals. Rather, the results suggest a separate pathway by which alcohol becomes more rewarding for 7-present individuals by increasing their perceived ability to bond with their peers. Future work should examine the relationship between increased stimulation/euphoria and enhanced perception of social bonding more fully, though, as these results are based only on the BAES. Furthermore, a statistical trend was found for a difference in perceived social bonding between 7-repeat carriers and non-carriers within the alcohol condition at this alcohol dose (p = .10) such that, as expected, carriers of the 7-repeat reported increased perceived social bonding compared to 7-absent individuals. Further research is indicated that varies alcohol dose, as higher doses might generate more pronounced effects.

Contrary to hypotheses, 7-present individuals did not evince more positive and less negative affect-related facial expressions during the interaction period compared to 7-absent individuals. Although speculative, 7-present individuals may be more sensitive to socially-relevant rewarding cues (e.g., smiling) under conditions of alcohol than 7-absent individuals, something that may not be captured by examining differences in intercepts (i.e., means) of facial behavior between individuals (which was tested in the current study). This hypothesis is consistent with recent findings showing that individuals carrying the 7-repeat appear to be more sensitive than noncarriers to other people's drinking behavior when randomly assigned to a heavy alcohol drinking condition (Larsen et al., 2010). More sophisticated data analyses than used here (e.g., time-lagged correlations; actor-partner analyses) could be used to test this prediction, showing for example, that 7-present individuals are more responsive to the smiling of others when intoxicated (with the putative cause-effect relationship reflected in lag-times between group members' smiles) than 7-absent individuals (Cook & Kenny, 2005). Analyses

such as these would allow for the examination of interactional processes between group members, which may be more likely to detect the influence of *DRD4* genotype on alcohol's socially reinforcing effects than analyses focusing on individual-level facial affect.

It remains unclear whether 7-repeat carriers possess an actual increased ability to bond with others or if they only perceive their ability to be enhanced. Regardless of this distinction, though, it may be that their perception of increased social bonding is what leads to problematic drinking. Future work is indicated, however, that examines whether 7-repeat carriers are rated as being more sociable by their peers under conditions of alcohol. In the current data set, future analyses can explore whether 7-present individuals are better at eliciting smiles from other group members when intoxicated than are 7-absent individuals. It will also be essential for future studies to test whether carriers of the 7-repeat allele choose to drink more alcohol in social contexts as a result of their perception of enhanced social bonding.

4.1.2 *DRD2* Taq1A

It was also hypothesized that individuals carrying at least one copy of the A1 allele of the *DRD2* Taq1A polymorphism would be especially sensitive to alcohol's socially reinforcing effects compared to individuals homozygous for the A2 allele. This hypothesis was unsupported. There were no significant *DRD2* genotype by drink condition interactions on perceived social bonding or any other self-report/behavioral measure of alcohol reinforcement after alcohol consumption. Despite the oft-cited hypothesis that individuals with the A1 allele experience less reinforcement from natural rewards and more reinforcement from drugs of abuse, including alcohol (e.g., Noble et al., 1998; Volkow et al., 1999; 2009), no prior study has tested the role of this polymorphism on individual differences in acute alcohol sensitivity in the laboratory. The present study had

adequate power to detect small to medium sized interaction effects between genetic variation and drink condition. The current null findings suggest that this polymorphism does little to influence level of alcohol reward in social drinkers, at least as assessed by the self-report and observational measures used in this study.

It should be emphasized that the Taq1A variant is a polymorphism with unknown functional significance that is thought to actually reside in *ANKK1*, which has not yet been detected in the brain (Neville et al., 2004). In contrast to this variant, there is strong evidence suggesting that the C957T SNP of the *DRD2* gene is related to substantial changes in dopaminergic functioning *in vitro* and *in vivo* (e.g., Hirvonen et al., 2004; 2009). A major limitation of this study was the inability to genotype participants at this locus. Future work is planned, however, to re-analyze the data using an alternate method of genotyping (i.e., pyrosequencing), which has proven successful for this particular SNP in other samples. Results considering the role of the C957T variant in determining alcohol reward may shed light on the exact relationship between *DRD2* and the reinforcing effects of alcohol in social drinkers.

4.2 SEROTONIN

4.2.1 5-HTTLPR

It was hypothesized that individuals carrying the S allele of the 5-HTTLPR polymorphism would be especially sensitive to alcohol's anxiolytic effects (i.e., they would experience decreased social anxiety/negative affect assessed across multiple response systems) compared to individuals homozygous for the L allele. In addition, it was expected that among participants not

consuming alcohol (i.e., placebo and control participants), the relationship would be reversed, such that persons with genetic variation associated with increased risk to experience anxiety-related traits (those carrying the S allele) will show increased negative affect/social anxiety during/after the social interaction.

Contrary to predictions, when comparing individuals carrying the S allele to those homozygous for the L allele, there were no significant 5-HTTLPR genotype by drink condition interactions on perceived social bonding or any other measure of alcohol reinforcement after alcohol consumption. [Note: These null findings remained non-significant when using the alternate classification system, in which S/S individuals were compared to S/L and L/L individuals].

Prior studies, which have focused exclusively on the role of 5-HTTLPR on individual differences in level of response to alcohol (i.e., assessment of subjective intoxication level; Schuckit & Smith, 1996), have reported mixed results. The current null findings are consistent with results reported by Corbin et al. (2006) who found no association between 5-HTTLPR and subjective responses to alcohol (intoxication and high) on each limb of the blood alcohol curve (Corbin et al., 2006). Results are inconsistent, however, with studies finding an association between the S allele and lower alcohol sensitivity (Fromme et al., 2004; Türker et al., 1998), as well as those showing a link between one or two copies of the L allele and lower alcohol sensitivity (Hinkers et al., 2006; Hu, Oroszi, Chun, Smith, Goldman, & Schuckit, 2005; Schuckit et al., 1999).

The current study sought to clarify inconsistent findings regarding 5-HTTLPR and alcohol response by testing the role of the triallelic 5-HTTLPR genotype in a much larger sample of young adult drinkers than any prior study in the literature. In addition to intoxication level, the

present study assessed behavioral measures of alcohol's reinforcing effects and did so in an ecologically valid social drinking paradigm. Null findings suggest that this polymorphism may not influence alcohol's anxiolytic effects (or level of alcohol reward) in social drinkers consuming alcohol in a social setting, at least as assessed by the measures used in the current study.

In contrast to null findings regarding an interaction between 5-HTTLPR genotype and drink condition, there was evidence that 5-HTTLPR genotype exerted main effects on some outcome measures. For instance, there was a main effect of 5-HTTLPR genotype on negative affect after beverage consumption such that individuals carrying one or two copies of the S allele reported significantly more negative affect on the 8-item mood measure than did L/L individuals. In addition, individuals homozygous for S allele reported lower perceived social bonding after the interaction compared to S/L and L/L individuals. Finally, there was a main effect of 5-HTTLPR genotype on the amount of time displaying negative affect during the interaction period, such that S/S individuals spent more time displaying negative affect than did S/L and L/L individuals. This effect just nearly missed significance, however, after the Bonferroni correction was applied.

Taken together, these results suggest that individuals carrying the S allele (and S/S individuals in particular) may have experienced the structured interaction period negatively, regardless of the drink condition to which they were randomly assigned. These participants reported feeling more negative affect, they displayed more negative affect-related facial expressions, and they reported lower perceived social bonding with their group members than L allele individuals. These results are consistent with prior studies that have linked the S allele to increased trait negative affect (Munafo et al., 2003; Schinka et al., 2004) and greater

psychological sensitivity to stress (Gotlib, Joormann, Minor, & Hallmayer, 2007). Notably, the findings contribute to an emerging literature aiming to understand the specific mechanisms by which 5-HTTLPR genotype contributes to risk for psychological impairment. To this end, prior studies have linked the S allele to increased amygdala reactivity in response to experimentally manipulated exposures to emotional stimuli (Munafò et al., 2008) and to greater cortisol reactivity to experimentally manipulated psychosocial stress (Way & Taylor, 2010). Given the importance of social bonding to overall psychological well-being, as well as the adverse psychological and physical health consequences associated with social rejection (Slavicha, Way, Eisenberger & Taylor, 2010) and loneliness (e.g., Hawkley & Cacioppo, 2010), the current findings suggest a potential intermediate phenotype that might predispose S allele carriers to increased risk of psychological dysfunction. Specifically, these individuals appear to both feel and evince (facially) more negative affect when interacting with unknown peers and to report less perceived bonding after an unstructured social interaction than similar individuals who do not carry a copy of the S allele. These results should be considered to be preliminary, though, and future studies should attempt to replicate this finding.

4.3 GABA

4.3.1 $GABA_{A\alpha6}$

The current study aimed to extend the literature by testing associations between the $GABA_{A\alpha6}$ polymorphism and alcohol response in a much larger sample of male and female social drinkers than previous studies. In contrast to prior studies focusing on the link between this variant and

alcohol sensitivity, current results revealed several main effects of $GABA_{A\alpha6}$ genotype (i.e., independent of alcohol exposure) on outcome measures. Specifically, individuals carrying one or two copies of the Ser allele reported significantly less perceived social bonding, lower stimulation, lower positive affect, and higher negative affect after the interaction period. In contrast, $GABA_{A\alpha6}$ genotype did not interact with beverage condition to influence any of the outcome measures.

A series of prior studies (Hu et al., 2005; Schuckit et al., 1999) linked the Ser allele with a low response to a laboratory alcohol challenge in a smaller number of male participants. Importantly, though, neither of these studies analyzed available placebo data when testing the association between Ser and alcohol response. The current study challenges the previous assumption that the link between $GABA_{A\alpha6}$ genotype and alcohol response is alcohol-specific, and rather provides evidence (in a larger sample of individuals) that the $GABA_{A\alpha6}$ genotype exerts main effects on response measures, independent of alcohol consumption. Thus, the current study, which is unique in that it includes a placebo as well as a non-alcohol control condition, potentially clarifies the nature of the relationship between $GABA_{A\alpha6}$ genotype and alcohol response. Although the function of the Pro385Ser amino acid substitution polymorphism of the $GABA_{A\alpha6}$ gene is unknown, current findings suggest that it may *not* contribute to AUDs by differentially affecting alcohol response. Results should be viewed as preliminary at best, though, given the low frequency of the Ser allele and the small number of participants carrying this variant that consumed alcohol in the present study. Perhaps most notably, findings highlight the importance of including placebo and no-alcohol control conditions in studies aiming to test the effects of specific genotypes on alcohol response measures.

4.4 STRENGTHS AND IMPLICATIONS

The current study had several strengths. Among these is the fact that the study was sufficiently powered to comprehensively examine the moderating roles of *DRD4, DRD2,* and 5-HTTLPR genetic variation on the reinforcing effects of alcohol in groups. Because the typical effect size for genetic variation acting on behavioral phenotypes is small (Chakravarti, 1999; Ducci & Goldman, 2008; Lander, 1996; Reich & Lander, 2001; Shanahan & Hofer, 2005), large samples are necessary to draw firm conclusions about how certain polymorphisms modulate the experience of alcohol. In one of the largest alcohol administration studies yet conducted, we were able to detect small though potentially meaningful *DRD4* effects on perceived social bonding. The size of this effect is similar to effect size values reported in other studies examining genetic links to complex human traits (e.g., Frazer, Murray, Schork, & Topol, 2009).

The current study also included three drink conditions (i.e., alcohol, placebo, and control), which made it possible to determine whether differences in alcohol response across genotypes represented true pharmacodynamic response variation. Importantly, no prior study on this topic has included a placebo and a no-alcohol control condition. Indeed, current findings challenge the notion that variation in $GABA_{A\alpha6}$ and 5-HTTLPR differentially affect alcohol response and, in contrast, show that these polymorphisms exert main effects on response measures independent of drink condition. It will be important for future studies to consider whether observed links between particular genotypes and alcohol response reflect alcohol-specific associations, associations attributable to expectancies regarding alcohol's effects, or direct links between gene polymorphisms and response measures.

Several methodological advances were employed to better understand the influence of genetic variation on the reinforcing effects of alcohol in a group setting. This study used

sophisticated observational measures of facial responding to study alcohol's effects on emotion. By attending to the limb of the BAC curve and controlling for familiarity among group members, the current study sought to clarify the role of specific genetic polymorphisms in understanding the reinforcing effects of alcohol. Furthermore, a large number of participants received alcohol, and drinking patterns were equivalent across groups. Personality traits thought to relate to social bonding also did not vary across groups. Use of an ecologically valid social drinking context, in which unacquainted young adults consumed alcohol together, increases the generalizability of our results to the natural environment. More generally, this design, which manipulated the environment through random assignment, uniquely allowed causal inferences to be drawn regarding the *DRD4* by drink condition (i.e., gene-environment) interaction (Rutter, Pickles, Murray & Eaves, 2001).

The present gene-environment interaction findings are preliminary and will need to be replicated. While some argue that genotype-dependent interactions should be the primary focus of alcohol research (e.g., Heath & Nelson, 2002), there is also growing skepticism about the utility of examining gene-environment interactions in the context of addiction and psychopathology. This is mainly due to the fact that some initial, positive gene-environment interaction findings failed to replicate in other samples (Munafo, Durrant, Lewis & Flint, 2009; Risch, Herrell, Lehner, Liang, Eaves, Hoh et al., 2009) but see (Karg, Burmeister, Shedden & Sen, 2011; Monroe & Reid, 2008). In general, many of the notable replication difficulties in the literature relate to studies of distal behavioral phenotypes (e.g., depression) and molar environmental moderators (e.g., life events), where differences in methodologies across studies may yield unstable findings (Monroe & Reid, 2008). It is worth noting that, in the case of the 5-HTTLPR literature, gene-environment interactions in experimental studies (e.g., effects of

transporter variation on amygdala response to experimentally manipulated exposures to emotional stimuli) have fared well in terms of replication (see Munafo et al., 2008). Still, although the present study utilizes an experimental design and builds upon an emerging literature highlighting the importance of social factors in the association of *DRD4* genotype and drinking outcomes (Larsen et al., 2010; Park et al., 2011), replication is crucial.

In contrast to gene-environment studies focusing on naturally occurring variation in putative environmental moderators and down-stream behavioral phenotypes, studies of genetic influences moderated by experimentally manipulated environmental exposures (as is the case in the present study) have at least two advantages. First, these designs allow for observations to be made under controlled and uniform stimulus conditions, and second, these paradigms better permit causal inferences, because the environmental effect is not subject to contamination by gene-environment correlation (Moffitt, Caspi & Rutter, 2006). The present study examined a gene-environment interaction in the context of an experimentally manipulated environmental factor, which offers a more powerful tool for identifying gene-environment interactions than do population based studies (Moffitt et al., 2006; Rutter et al., 2001). Together with other recent findings targeting *DRD4,* current results suggest that interventions may benefit from focusing on social reward as an important underlying mechanism for the development of problematic drinking in a subset of young adults. More generally, these findings highlight the potential utility of employing transdisciplinary methods that integrate genetic methodologies, social psychology, and addiction theory to improve theories of alcohol use/abuse and to help predict who may be at risk of developing drinking problems.

4.5 LIMITATIONS

Despite notable strengths, the present study had several limitations. Although the alcohol participants reported higher perceived social bonding on the PGRS than did the placebo participants, they did not differ significantly from the control (no alcohol) participants. This may indicate that a higher dose of alcohol might have been more useful to test. Higher doses of alcohol, however, can lead to subjects becoming ill or not being able to participate. Research also would be useful to further probe the role of dosage-set, as the present data reveal that placebo participants reported lower PGRS scores than did control participants. This seemingly counterintuitive pattern has been observed for cognitive processes where compensatory mechanisms are implicated (Vogul-Sprott & Fillmore, 1999), but it is unclear how this would apply to the social interaction.

In addition, although every participant in the current dataset drank his/her beverage in a group with two other members, there was complete group data for less than half of the full sample. As mentioned above, a notable strength of HLM (the primary data analytic technique used in the current study) is that it has been shown to be robust to missing or incomplete data (Raudenbush & Bryk, 2002). Nonetheless, future work should attempt to replicate the findings in a sample with complete group data. Also, while the distribution of group gender compositions was evenly distributed across the six cells of the experiment (for each polymorphism tested), this variable was not controlled for and the study was not sufficiently powered to examine its influence on the results. Future studies with even larger samples would permit the examination of potentially interesting three-way interactions including gender and group gender composition as variables. Though such studies raise ethical considerations, it potentially would also be valuable to extend these findings in individuals who meet criteria for alcohol use disorders.

As is true in all prior alcohol administration studies, the sample size of the current study precluded performing a genome-wide scan, an approach that typically requires many more individuals in order to be informative. Compared to genome-wide scans, though, candidate gene studies may be better suited to detect genes underlying common and more complex disorders, such as AUDs (Kwon & Goate, 2000; McCarroll & Altshuler, 2007; McCarthy, Goncalo, Abecasis, Cardon, Goldstein, Little et al., 2008; cf. Hill, 2010). Thus, the present research explored the relationship between the reinforcing effects of alcohol and four polymorphisms selected a priori on the basis of the literature (i.e., polymorphisms that have previously been linked to increased risk for AUDs).

Because the parent study was not designed explicitly to investigate family history of alcoholism, family history assessment was based on participant reports. Although some studies have found that classification derived from such data corresponds fairly well to that derived from structured clinical interviews with both participants and a collateral parent (e.g., Crews & Sher, 1992; Cuijpers & Smit, 2001), family history classification based on participant reports is clearly not the preferred assessment method (see Rice, Reich, Bucholz, Neurnan, Fishman, Rochberg et al., 1995). The current study allowed for at least a preliminary examination of the extent to which a positive family history of alcoholism, as assessed by participant reports, related to alcohol response and genotype classification. Nevertheless, it remains unclear whether the current null findings resulted from measurement error in the assessment of family history.

A limitation of alcohol administration research in general is the variability in blood alcohol concentrations that results from oral consumption. This variability was mitigated to some degree in the current study by several steps taken prior to and during consumption (e.g., including only using participants who were within 15% of normal body weight using the

Metropolitan Life charts, adjusting for gender in the dosage charts, and pouring one third of the beverage into participants' glasses every 12-mintues to help ensure even rate of consumption). Notably, too, the current laboratory study allowed for precise measurement of BACs. The only prior study that attempted to examine the relationship between genotype and alcohol response in a social setting used field data and thus was forced to estimate BACs for each participant (see Ray et al., 2010).

Another limitation of the current study is that it offers no information about how genetic variation impacts alcohol response on the descending limb of alcohol absorption, as key measures were assessed on the ascending limb only. It is also important to note that several other candidate genes that have been implicated in AUDs were not included in this study (e.g., *NPY*). The current study, however, focused on polymorphisms that may be particularly relevant for the study design (i.e., genes that have been implicated in both AUDs and that may also have relevance for social/emotional phenotypes). For instance, as noted above, previous studies have highlighted the importance of social factors in the link between *DRD4* genotype and alcohol outcomes (e.g., Larsen et al., 2010, Laucht et al., 2007; Park et al., 2011). In addition, 5-HTTLPR variation has been linked not only to AUDs but also to depression and anxiety-related traits (Lesch et al., 1996), which in turn are associated with problematic alcohol use. Future studies examining other possible candidate genes are indicated (see future directions section below). Finally, as noted previously, a major limitation of this study was that genotyping for the C957T SNP of *DRD2* was unsuccessful for most participants. Future work is planned to re-analyze the data using an alternate method of genotyping (i.e., pyrosequencing), which has proven successful for this particular SNP in other samples.

4.6 FUTURE DIRECTIONS

There is a substantial genetic component for drinking behavior and AUDs, but few well-replicated genetic markers of alcoholism have been identified. Because of the complexity associated with behavioral phenotypes of AUDS, researchers are focusing more and more on intermediate phenotypes (e.g., alcohol sensitivity), which are thought to mediate genetic effects on clinical phenotypes. Prior to the current project, these studies have focused exclusively on testing individuals' responses to alcohol in isolation. Adolescents and young adults do nearly all of their drinking with others, though, suggesting that social processes may be particularly important in shaping drinking behavior early on and may play a key role in the development of problematic drinking. Future studies should continue to explore how genetic factors interact with social factors to confer increased risk for AUDs. This research should be conducted in the context of experimentally manipulated social-environmental factors which, in contrast to studies examining naturally occurring (and therefore non-random) variation in environmental moderators, will allow investigators to rule out gene-environment correlations and draw causal inferences.

Other candidate genes should be examined in relation to the socio-emotional outcome variables assessed in the current study. For example, the SNP A118G (*rs1799971*) of *OPRM1*, which exerts functional effects on μ-opioid receptors (Bond, LaForge, Tian, Melia, Zhang & Borg et al., 1998; Zhang, Wang, Johnson, Papp, & Sadee, 2005), has been shown to influence subjective reports of the reinforcing effects of drugs of abuse, including alcohol (Ray, 2011). *DRD4* genotype appears to exert its influence on alcohol-related phenotypes by increasing 7-repeat carriers' sensitivity to cues under conditions of alcohol [e.g., increased craving in response to alcohol cues after a priming dose of alcohol (Hutchison et al., 2002), increased sensitivity to

others' drinking behavior under a heavy alcohol drinking condition (Larsen et al., 2010), and, as the current study found, increased perceptions of bonding when consuming alcohol]. In contrast, variation in genes involved in the opioid system may be more likely to affect hedonic responses to alcohol on the ascending limb of alcohol absorption (Ray et al., 2010). Specifically, in controlled alcohol administration studies, individuals with the Asp40 allele of *OPRM1* reported higher feelings of intoxication, stimulation, and positive feelings and lower levels of negative mood across rising levels of BAC compared to individuals carrying the Asn40 allele (Ray, 2011; Ray & Hutchison, 2004, 2007; Ray et al., 2010). Although examination of this SNP was beyond the scope of the original project, this variant has been genotyped in the current sample. Future work is planned to determine whether variation in *OPRM1* affects alcohol's socially reinforcing effects in a controlled group setting.

If future work were to replicate the current *DRD4* findings (as well as those reported by Larsen et al., 2010 and Park at al., 2011), it may then be useful to consider implementing more process-oriented designs in order to delineate possible mechanisms underlying the link between *DRD4*, social factors, and alcohol outcomes. For example, using a previously developed paradigm to assess the relative ability of social (genuine smiles) and nonsocial feedback (monetary rewards) to shape choice behavior (Shore & Heerey, 2011), it could be determined whether 7-present individuals deem social signals to have more reward value than 7-absent individuals and whether this effect is potentiated under conditions of alcohol. The specificity of *DRD4* effects should also be tested. At this point, it remains unknown whether individuals carrying the 7-repeat allele are also more sensitive to negative social cues under conditions of alcohol compared to 7-absent individuals. To this end, it would be interesting to determine whether 7-present individuals are more sensitive to social rejection paradigms (e.g., Williams,

Cheung & Choi, 2000) than 7-absent individuals and whether this effect is moderated by alcohol consumption. In addition, it would be illuminating to determine whether 7-present individuals are generally more myopic when intoxicated than 7-absent individuals, perhaps by drawing on study designs from the alcohol myopia literature (Steele & Josephs, 1990). Future work could also examine whether 7-present individuals are more impulsive under conditions of alcohol, by using known behavioral paradigms to assess impulsivity (e.g., Connors, 2000) or delay discounting (e.g., Bickel & Marsch, 2001). Studies such as these will help to elucidate specific mechanisms of increased risk underlying links between *DRD4* genotype and alcohol outcomes.

Finally, in addition to the examination of other polymorphisms in relation to alcohol's socially reinforcing effects (e.g., *DRD2* C957T, *OPRM1*), this uniquely large data set will permit an investigation of more diverse genetic effects on processes underlying social interactions, independent of alcohol exposure. For example, I intend to examine whether variation in genes that regulate the social neuropeptides (oxytocin, arginine vasopressin) is associated with social behavior during this controlled social interaction. Such investigations, which have not yet been conducted, will benefit from several aspects of the current study's design. First, the dataset includes a multidimensional, behavioral assessment of social processes occurring in real time during a controlled social interaction, making it especially well-suited to the examination of social behavior. Second, the study focused on the initial period of group formation (amongst unacquainted young adults), which is a phase of social integration characterized by self-awareness, self-presentational concerns, and social anxiety (Leary & Kowalski, 1995), as well as enjoyment (Kirchner et al., 2006). Finally, the use of observational face and speech measures permitted unobtrusive capture of moment-to-moment fluctuations in emotional responses, which is crucial when studying dynamic, coordinated social interaction. As such, the current project

may well shed light on important genetic mechanisms underlying social behavior, regardless of drink condition assignment.

4.7 SUMMARY AND SIGNIFICANCE

The current study tested the moderating role of four polymorphisms (i.e., *DRD4* VNTR, *DRD2* Taq1A, 5-HTTLPR, and $GABA_{A\alpha6}$) on alcohol's socially reinforcing effects among previously unacquainted individuals in a controlled group setting. Of the many models that were tested, only one genotype by drink condition interaction remained significant after controlling for Type 1 error with multiple genotype comparisons. Importantly, this lone (highly) significant interaction involves the polymorphism that has received the strongest prior support linking social factors and drinking (Larsen et al., 2010; Park et al., 2011). As hypothesized, individuals carrying the 7-repeat allele of the *DRD4* VNTR were especially sensitive to alcohol's socially reinforcing effects compared to 7-absent individuals. Specifically, 7-present individuals reported increased social bonding in an unstructured group setting after drinking alcohol, compared to placebo and non-alcohol control beverages. In contrast, alcohol did not affect perceived social bonding of 7-absent individuals. None of the other genotype by drink condition interactions was significant.

This single finding raises several questions and concerns. In particular, one might ask whether this result was found by chance alone, given the number of different genotypes and hypotheses tested in the current study. Although the Bonferroni correction was used to control for Type 1 error, there remains a legitimate concern over whether this is indeed a real effect. On the other hand, the size of the observed *DRD4* genotype by drink condition interaction finding in the current study is in line with what would be expected for genetic variation acting on a

behavioral phenotype (Chakravarti, 1999; Ducci & Goldman, 2008; Lander, 1996; Reich & Lander, 2001; Shanahan & Hofer, 2005), providing some evidence for the veracity of the effect. Thus, one might make the case that the finding, while small in size, is indeed a real effect. Furthermore, because none of the other genotypes showed any relationship at all to alcohol's effects, one could argue for the discriminant validity of *DRD4* genotype in moderating alcohol's reinforcing effects. [Note: Inspection of mean values for key dependent variables across the experimental cells revealed very little differences across these values]. Clearly, replication will be crucial if we are to truly understand the significance of the present results. Regardless, the present study suggests that alcohol administration studies that involve genotyping relatively large samples should include a social context in order to understand the interaction of genes and alcohol in social drinkers.

APPENDIX A

TABLES AND FIGURES

Table 1. *DRD4* VNTR Allele and Genotype Frequencies

Allele	*n*	%
Allele		
2	70	8.30
3	31	3.67
4	547	64.81
5	11	1.30
7	172	20.38
8	13	1.54
Total	844	100
Genotype		
2/2	2	0.47
2/3	1	0.24
2/4	47	11.14
2/7	17	4.03
2/8	1	0.24
3/3	1	0.24
3/4	17	4.03
3/7	10	2.37
3/8	1	0.24
4/4	175	41.47
4/5	7	1.66
4/7	117	27.73
4/8	9	2.13
5/7	4	0.94
7/7	11	2.60
7/8	2	0.47
Total	422	100
Genotype Classification		
7-present	161	38.15
7-absent	261	61.85
Total	422	100

Table 2. *DRD4* Genotype Distribution Across Beverage Conditions

	Alcohol		Placebo		Control		Total	
	n	%	*n*	%	*n*	%	*n*	%
7-present	68	43.31	42	33.07	51	36.96	161	38.15
7-absent	89	56.69	85	66.93	87	63.04	261	61.85
Total	157	100	127	100	138	100	422	100

Table 3. *DRD2* Taq1A Genotype Distribution Across Beverage Conditions

	Alcohol		Placebo		Control		Total	
	n	%	*n*	%	*n*	%	*n*	%
A1 carriers	54	36.99	47	38.21	45	34.35	146	36.50
A2/A2	92	63.01	76	61.79	86	65.65	254	63.50
Total	146	100	123	100	131	100	400	100

Table 4. 5-HTTLPR Genotype Distribution Across Beverage Conditions

	Alcohol		Placebo		Control		Total	
	n	%	*n*	%	*n*	%	*n*	%
S carriers	114	73.55	93	73.23	102	73.91	309	73.57
L/L	41	26.45	34	26.77	36	26.09	111	26.43
Total	155	100	127	100	138	100	420	100

Table 5. $GABA_{A\alpha 6}$ Genotype Distribution Across Beverage Conditions

	Alcohol		Placebo		Control		Total	
	n	%	*n*	%	*n*	%	*n*	%
Ser carriers	22	14.38	22	17.46	24	17.91	68	16.46
Pro/Pro	131	85.62	104	82.54	110	82.09	345	83.54
Total	153	100	126	100	134	100	413	100

Table 6. Beverage Response Variables

	Alcohol		Placebo		Control		
Characteristic	*Mean*	*SD*	*Mean*	*SD*	*Mean*	*SD*	*F*
BAC post-interaction	0.054^{a}	0.012	0.001^{b}	0.001	0.001^{b}	0.001	2649.51^{**}
BAC 40-min post-drink†	0.062^{a}	0.011	0.001^{b}	0.001	-----	-----	3896.09^{**}
SIS post-interaction	38.39^{a}	16.89	15.26^{b}	10.31	0.09^{c}	0.73	396.44^{**}
SIS 40-min post- drink†	34.75^{a}	16.53	9.85^{b}	11.34	-----	-----	208.63^{**}

* $p = < .05$ ** $p = < .001$

† analyses did not include control participants as they were not asked to provide these data

Note. BAC = blood alcohol concentration. SIS = subjective intoxication scale (values ranging from 0 to 100). Groups with non-overlapping superscripts differed significantly ($p < .05$).

Table 7. Socio-emotional Responses (M ± SE) Across Beverage Conditions

Drink Condition	PGRS	Positive Affect	Negative Affect	Stimulation	Sedation	Anxiety
Alcohol	7.2 (.09)	3.6 (.07)	.32 (.03)	5.3 (.17)	1.8 (.11)	1.9 (.05)
Placebo	6.6 (.13)	3.2 (.06)	.74 (.05)	4.4 (.16)	1.8 (.11)	2.1 (.06)
Control	7.1 (.10)	3.3 (.06)	.64 (.05)	4.3 (.14)	1.2 (.09)	1.9 (.05)

Note. PGRS=Perceived Group Reinforcement Scale (individual items on scale ranged from 1 to 9). Positive and negative affect were assessed by the 8-item Mood Measure (values ranging from 0 to 5). Stimulation and sedation were assessed by the Biphasic Alcohol Effects Scale (values ranging from 0 to 10). Anxiety was assessed by the state version of the State-Trait Anxiety Inventory (values ranging from 1 to 7).

Table 8. Facial Expressions (M ± SE) Across Beverage Conditions

Drink Condition	Duchenne smiles	Social smiles	Negative Composite	Smile controls
Alcohol	270.34 (12.84)	337.74 (14.07)	24.62 (2.43)	21.56 (1.94)
Placebo	157.35 (7.62)	266.18 (13.03)	29.95 (2.16)	27.97 (2.48)
Control	173.90 (8.42)	309.95 (13.72)	29.88 (2.42	30.95 (2.82)

Note. Values represent frames per minute. Participants, on average, showed a 1% decrease per minute in the number of frames they spent displaying negative affect (as assessed by the negative composite; t=-2.32, df=215, p =.02) and the number of frames they spent displaying social smiles (t=-5.92, df=215, $p < .001$). The average amount of time participants spent displaying Duchenne smiles and smile controls did not change significantly over time during the interaction.

Table 9. Correlations Between Outcome Measures

	Duchenne Smile	Social Smile	Smile Control	Negative Comp	PGRS	Positive Affect	Negative Affect	Stimulation	Sedation	Anxiety
Duchenne Smile	1									
Social Smile	.240***	1								
Smile Control	.117*	.273***	1							
Negative Composite	-.162***	-.254***	.320***	1						
PGRS	.200***	.059	.013	.000	1					
Positive Affect	.239***	.004	.067	.021	.427***	1				
Negative Affect	-.199***	-.071	-.015	-.002	-.445***	-.329***	1			
Stimulation	.180***	-.003	-.029	-.040	.353***	.724***	-.266***	1		
Sedation	-.047	.010	-.087	-.056	-.236**	-.147**	.395***	-.098*	1	
Anxiety	.021	-.067	-.057	.053	-.263**	-.336***	.288***	-.280***	.302***	1

*p<.05, **p<.01, ***p<.001

Table 10. HLM: Model Predicting PGRS from Beverage Condition and *DRD4* Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean PRGS, β_1				
Intercept, γ_{00}	7.015	.089	78.10	<.001
Alcohol vs No-Alc, γ_{01}	0.058	.242	.239	.811
Placebo vs Control, γ_{02}	-0.490	.229	-2.14	.034
Model for DRD4 Slopes, β_1				
Intercept, γ_{10}	-0.142	.135	-1.05	.293
Alcohol vs No-Alc, γ_{11}	0.952	.354	2.69	.008
Placebo vs Control, γ_{12}	-0.041	.356	-0.114	.909

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.421	0.66	<.001
Level-1 effect, *r*	1.281	1.13	

Table 11. PGRS scores (M ± SE) by Beverage Condition and Genotype

	Alcohol	Placebo	Control	*Genotype Mean*
7-present	7.37 (.17)	6.35 (.20)	6.86 (.18)	6.86 (.10)
7-absent	7.04 (.15)	6.75 (.15)	7.24 (.15)	7.01 (.09)
Beverage Mean	7.21 (.11)	6.55 (.13)	7.05 (.13)	

Note. Possible range = 1 – 9. Contrasts examining carriers and non-carriers within each beverage condition failed to reach significance.

Table 12. Variance Components and Model Fit

	Empty Model *Coef (SE)*	*DRD4* *Coef (SE)*	*DRD4* and Drink *Coef (SE)*	*DRD4* x Drink *Coef (SE)*
Variance Components				
Variance in group intercepts	0.46(.68)	0.46(.68)	0.39(.63)	0.40(.63)
Variance within groups	1.30(1.14)	1.30(1.14)	1.30(1.14)	1.26(1.13)
Goodness of Fit				
No. of Parameters	3	4	6	8
Deviance (FIML)	1419.93	1419.38	1407.46	1399.82
Chi-square statistic		0.55	11.93	7.64
Degrees of freedom		1	2	2
P-value		>0.50	0.003	0.021

Table 13. HGLM: Model Predicting Social Smiles from Beverage Condition and *DRD4* Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Social Smiles, β1				
Intercept, γ000	5.488	.053	101.74	<.001
Alcohol vs No-Alc, γ001	0.207	.104	1.99	.047
Placebo vs Control, γ002	-0.294	.093	-3.15	.002
Model for Gender, β01				
Intercept, γ010	-0.033	.056	-0.587	.558
Model for DRD4, β02				
Intercept, γ020	0.029	.049	.596	.552
Alcohol vs No-Alc, γ021	0.069	.138	.505	.614
Placebo vs Control, γ022	0.299	.124	2.42	.016
Model for Baseline Social Smiles, β1				
Intercept, γ030	0.000	.000	20.69	<.001

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, r0	.421	0.18	<.001
Level-1 effect, e	131.7	11.48	
r0 / e, u00	0.076	0.277	<.001

Table 14. HLM: Model Predicting PGRS from Beverage Condition and 5-HTTLPR Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean PRGS, β_1				
Intercept, γ_{00}	7.18	.12	59.89	<.001
Alcohol vs No-Alc, γ_{01}	0.311	.317	.982	.327
Placebo vs Control, γ_{02}	-0.341	.311	-1.09	.274
Model for 5-HTTLPR Slopes, β_1				
Intercept, γ_{10}	-0.275	.142	-1.94	.054
Alcohol vs No-Alc, γ_{11}	0.148	.371	.399	.691
Placebo vs Control, γ_{12}	-0.202	.371	-0.544	.587

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.375	0.61	<.001
Level-1 effect, *r*	1.32	1.15	

Table 15. HLM: Model Predicting Negative Affect from Beverage Condition and 5-HTTLPR Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean Negative Affect, β_1				
Intercept, γ_{00}	0.456	.043	10.41	<.001
Alcohol vs No-Alc, γ_{01}	-0.498	.100	-4.96	<.001
Placebo vs Control, γ_{02}	0.291	.124	2.34	.020
Model for 5-HTTLPR Slopes, β_1				
Intercept, γ_{10}	0.142	.053	2.65	.009
Alcohol vs No-Alc, γ_{11}	0.006	.128	.048	.962
Placebo vs Control, γ_{12}	-0.268	.149	-1.79	.074

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.043	.208	<.001
Level-1 effect, *r*	.255	.505	

Table 16. HGLM: Model Predicting Social Smiles from Beverage Condition and 5-HTTLPR Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Social Smiles, β_1				
Intercept, γ_{000}	5.50	.058	94.34	<.001
Alcohol vs No-Alc, γ_{001}	0.436	.136	3.18	.002
Placebo vs Control, γ_{002}	0.050	.121	.418	.676
Model for Gender, β_{01}				
Intercept, γ_{010}	-0.038	.058	-0.665	.507
Model for 5-HTTLPR, β_{02}				
Intercept, γ_{020}	-0.000	.056	-.009	.993
Alcohol vs No-Alc, γ_{021}	-0.291	.157	-1.85	.065
Placebo vs Control, γ_{022}	-0.287	.139	-2.05	.041
Model for Baseline Social Smiles, β_1				
Intercept, γ_{030}	0.000	.000	21.08	<.001

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, r_0	0.169	0.41	<.001
Level-1 effect, *e*	131.44	11.47	
r_0 / e, u_{00}	0.089	0.298	<.001

Table 17. HLM: Model Predicting PGRS from Beverage Condition and 5-HTTLPR Genotype using Alternative Genotype Classification

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean PRGS, β_1				
Intercept, γ_{00}	7.07	.075	93.98	<.001
Alcohol vs No-Alc, γ_{01}	0.349	.208	1.677	.095
Placebo vs Control, γ_{02}	-0.372	.188	-1.97	.049
Model for 5-HTTLPR Slopes, β_1				
Intercept, γ_{10}	-0.369	.139	-2.64	.009
Alcohol vs No-Alc, γ_{11}	0.217	.381	.570	.569
Placebo vs Control, γ_{12}	-0.335	.354	-.946	.345

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.356	0.59	<.001
Level-1 effect, *r*	1.32	1.14	

Table 18. HLM: Model Predicting Negative Affect from Beverage Condition and 5-HTTLPR Genotype using Alternative Genotype Classification

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean Negative Affect, β_1				
Intercept, γ_{00}	0.549	.032	17.15	<.001
Alcohol vs No-Alc, γ_{01}	-0.564	.076	-7.35	<.001
Placebo vs Control, γ_{02}	0.038	.088	.428	.669
Model for 5-HTTLPR Slopes, β_1				
Intercept, γ_{10}	0.050	.060	.827	.409
Alcohol vs No-Alc, γ_{11}	0.335	.157	2.13	.034
Placebo vs Control, γ_{12}	0.217	.160	1.35	.177

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.047	.216	<.001
Level-1 effect, *r*	.254	.504	

Table 19. HGLM: Model Predicting Negative Affect Composite from Beverage Condition and 5-HTTLPR using Alternative Genotype Classification

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Negative Comp, β_1				
Intercept, γ_{000}	2.671	.065	40.98	<.001
Alcohol vs No-Alc, γ_{001}	-0.472	.131	-3.58	<.001
Placebo vs Control, γ_{002}	0.033	.120	.275	.783
Model for Gender, β_{01}				
Intercept, γ_{010}	0.466	.082	5.66	<.001
Model for 5-HTTLPR, β_{02}				
Intercept, γ_{020}	0.212	.089	2.36	.012
Alcohol vs No-Alc, γ_{021}	-0.081	.255	-.318	.731
Placebo vs Control, γ_{022}	-0.062	.218	-.286	.752
Model for Baseline Negative Comp, β_1				
Intercept, γ_{030}	0.004	.000	13.70	<.001

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, r_0	.401	0.63	<.001
Level-1 effect, *e*	41.58	6.44	
r_0 / e, u_{00}	0.099	0.315	<.001

Table 20. HLM: Model Predicting PGRS from Beverage Condition and $GABA_{A\alpha 6}$ Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean PRGS, β_1				
Intercept, γ_{00}	7.07	.075	94.02	<.001
Alcohol vs No-Alc, γ_{01}	0.461	.199	2.31	.022
Placebo vs Control, γ_{02}	-0.447	.194	-2.29	.023
Model for $GABA_{A\alpha 6}$ Slopes, β_1				
Intercept, γ_{10}	-0.666	.172	-3.86	<.001
Alcohol vs No-Alc, γ_{11}	0.013	.470	.029	.977
Placebo vs Control, γ_{12}	-0.227	.436	-.523	.602

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.453	0.67	<.001
Level-1 effect, *r*	1.24	1.11	

Table 21. HLM: Model Predicting BAES-Stimulation from Beverage Condition and $GABA_{A\alpha 6}$ Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean BAES, β_1				
Intercept, γ_{00}	4.79	.097	49.07	<.001
Alcohol vs No-Alc, γ_{01}	1.32	.281	4.71	<.001
Placebo vs Control, γ_{02}	0.186	.235	.791	.430
Model for $GABA_{A\alpha 6}$ Slopes, β_1				
Intercept, γ_{10}	-0.703	.236	-2.96	<.001
Alcohol vs No-Alc, γ_{11}	0.276	.721	.384	.702
Placebo vs Control, γ_{12}	-0.351	.531	-.661	.509

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.002	0.04	<.001
Level-1 effect, *r*	3.51	1.87	

Table 22. HLM: Model Predicting Positive Affect from Beverage Condition and $GABA_{A\alpha 6}$ Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean Positive Affect, β_1				
Intercept, γ_{00}	3.41	.042	81.04	<.001
Alcohol vs No-Alc, γ_{01}	0.408	.116	3.49	<.001
Placebo vs Control, γ_{02}	-0.082	.105	-.785	.434
Model for $GABA_{A\alpha 6}$ Slopes, β_1				
Intercept, γ_{10}	-0.302	.121	-2.49	<.001
Alcohol vs No-Alc, γ_{11}	-0.131	.370	-.356	.722
Placebo vs Control, γ_{12}	-0.187	.271	-.691	.490

Random Effects	*Variance Component*	*SD*	*p-value*
Intercept, u_0	.019	0.13	<.001
Level-1 effect, *r*	.615	.78	

Table 23. HLM: Model Predicting Negative Affect from Beverage Condition and $GABA_{A\alpha 6}$ Genotype

Fixed Effects	*Coefficient*	*SE*	*t-ratio*	*P-value*
Model For Mean Negative Affect, β_1				
Intercept, γ_{00}	0.530	.030	17.32	<.001
Alcohol vs No-Alc, γ_{01}	-0.498	.073	-6.78	<.001
Placebo vs Control, γ_{02}	0.067	.084	.798	.426
Model for $GABA_{A\alpha 6}$ Slopes, β_1				
Intercept, γ_{10}	0.204	.074	2.763	<.001
Alcohol vs No-Alc, γ_{11}	0.006	.193	.032	.974
Placebo vs Control, γ_{12}	0.223	.194	1.14	.252

Random Effects	*Variance Component*	*SD*	p-*value*
Intercept, u_0	.036	0.19	<.001
Level-1 effect, *r*	.259	.50	

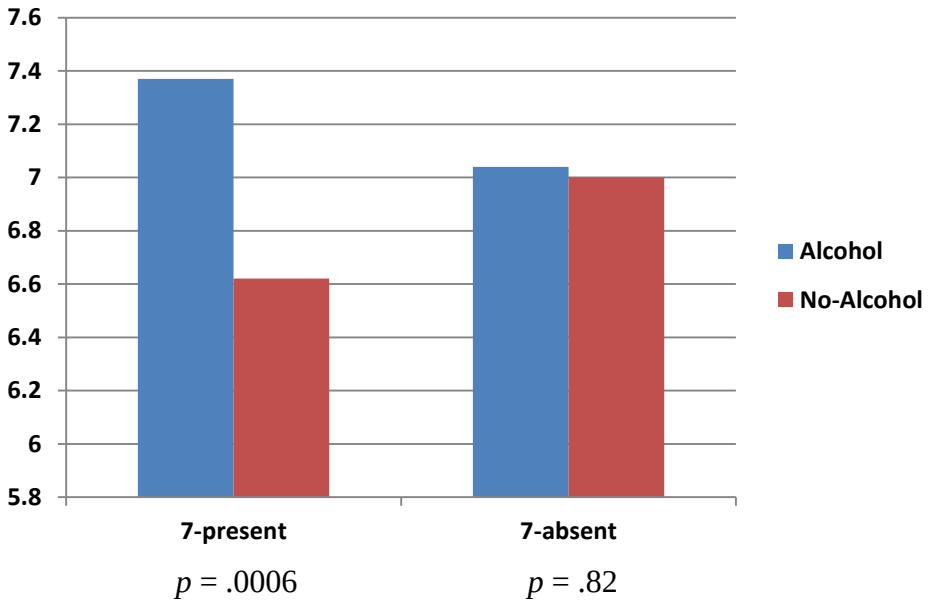
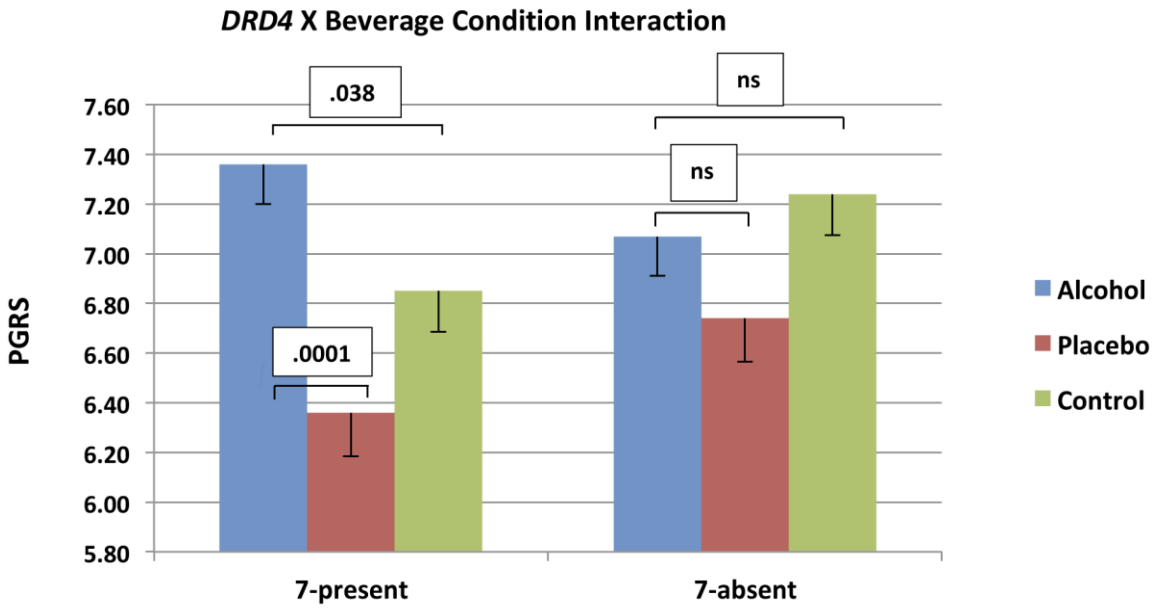


Figure 1. ***DRD4*** **Genotype by Alcohol vs. No-alcohol Condition Interaction**



Note: P-values are displayed in boxes.

Figure 2. PGRS Scores (Mean, SE) by *DRD4* Genotype and Beverage Condition

BIBLIOGRAPHY

Agresti, A., Booth, J. G., Hobert, J. P., & Caffo, B. (2000). Random-effects modeling of categorical response data. *Sociological Methodology, 30*(1), 27–80.

American Psychiatric Association. (1994). Diagnostic and statistical manual of mental disorders (4th ed.). Washington, DC: American Psychiatric Association.

Asghari, V., Sanyal, S., Buchwaldt, S., Paterson, A., Jovanovic, V., & Van Tol, H. H. (1995). Modulation of intracellular cyclic AMP levels by different human dopamine D4 receptor variants. *Journal of Neurochemistry, 65*, 1157–1165.

Atkins, D. C., & Gallop, R. J. (2007). Rethinking how family researchers model infrequent outcomes: A tutorial on count regression and zero-inflated models. *Journal of Family Psychology, 21*(4), 726–735.

Bachman, J. G., Johnston, L. D., O'Malley, P. M., & Schulenberg, J. E. (2006). *The Monitoring the Future project after thirty-two years: Design and procedures.* Ann Arbor, MI: Institute for Social Research.

Bakeman, R. (1999). Behavioral observation and coding. In H. T. Reis & C. M. Judd (Eds.), *Handbook of research methods in social and personality psychology* (pp. 138–159). Cambridge, U.K. New York: Cambridge University Press.

Bakermans-Kranenburg, M. J., & van Ijzendoorn, M. H. (2011). Differential susceptibility to rearing environment depending on dopamine-related genes: New evidence and a meta-analysis. *Development and Psychopathology, 23,* 39–52. doi:10.1017/S0954579410000635

Baumeister, R. F., & Leary, M. R. (1995). The need to belong: Desire for interpersonal attachments as a fundamental human motivation. *Psychological Bulletin, 117,* 497–529.

Beck, O., Borg, S., Edman, G., Fyro, B., Oxenstierna, G., & Sedvall, G. (1984). 5-hydroxytryptophol in human cerebrospinal fluid: Conjugation, concentration gradient, relationship to 5-hydroxyindoleactic acid, and influence of hereditary factors. *Journal of Neurochemistry, 43,* 58–61.

Beseler, C. L., Aharonovich, E., Keyes, K. M., & Hasin, D. S. (2008). Adult transition from at-risk drinking to alcohol dependence: The relationship of family history and drinking motives. *Alcoholism: Clinical and Experimental Research, 32*(4), 607–616.

Bickel, W. K., & Marsch, L. A. (2001). Toward a behavioral economic understanding of drug dependence: Delay discounting processes. *Addiction, 96,* 73–86.

Bierut, L. J., Agrawal, A., Bucholz, K. K., Doheny, K. F., Laurie, C., Pugh, E., et al. (2010). A genome-wide association study of alcohol dependence. *Proceedings of the National Academy of Sciences of the United States of America, 107*(11), 5082–5087.

Blum, K., Noble, E. P., Sheridan, P. J., Montgomery, A., Ritchie, T., & Jagadeeswaran, P. (1990). Allelic association of human dopamine D2 receptor gene in alcoholism. *Journal of the American Medical Association, 263,* 2055–2060.

Bollen, K. A., & Hoyle, R. H. (1990). Perceived cohesion: A conceptual and empirical examination. *Social Forces, 69,* 479–504.

Brown, S. A., Christiansen, B., & Goldman, M. S. (1987). The Alcohol Expectancy Questionnaire: An instrument for the assessment of adolescent and adult alcohol expectancies. *Journal of Studies on Alcohol, 48*, 483–491.

Bushman, B. J., & Cooper, H. M. (1990). Effects of alcohol on human aggression: An integrative research review. *Psychological Bulletin, 107,* 31–354.

Chen, X., Levine, L., & Kwok, P. Y. (1999). Fluorescence polarization in homogeneous nucleic acid analysis. *Genome Research, 9*(5), 492–498.

Christiansen, B. A., Smith, G. T., Roehling, P. V., & Goldman, M. S. (1989). Using alcohol expectancies to predict adolescent drinking behavior after one year. *Journal of Consulting and Clinical Psychology, 57,* 93–99.

Cloninger, C. R., Sigvardsson, S., Gilligan, S. B., von Knorring, A. L., Reich, T., & Bohman, M. (1988). Genetic heterogeneity and the classification of alcoholism. *Advances in Alcohol & Substance Abuse, 7,* 3–16.

Cohen, J., Cohen, P., West, S., & Aiken, L. (2003). *Applied multiple regression/correlation analysis for the behavioral sciences* (3rd ed.). Erlbaum: Mahwah, NJ.

Collier, D. A., Stöber, G., Li, T., Heils, A., Catalano, M., Di Bella, D., et al. (1996). A novel functional polymorphism within the promoter of the serotonin transporter gene: Possible role in susceptibility to affective disorders. *Molecular Psychiatry, 1,* 453–460.

Colzato, L. S., van den Wildenberg, W.P.M., van der Does, W., & Hommel, B. (2010). Genetic markers of striatal dopamine predict individual differences in dysfunctional, but not functional impulsivity. *Neuroscience, 170,* 782–788.

Comings, D. E., Gade-Andavolu, R., Gonzalez, N., Wu, S., Muhleman, D., Chen, C., et al. (2001). The additive effect of neurotransmitter genes in pathological gambling. *Clinical Genetics, 60*(2), 107–116.

Congdon, E., Lesch, K. P., & Canli, T. (2008). Analysis of DRD4 and DAT polymorphisms and behavioral inhibition in healthy adults: Implications for impulsivity. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics, 147B,* 27–32.

Conners, C. K. (2000). Conners' Continuous Performance Test (CPT II) Computer Program for Windows Technical Guide and Software Manual. North Tonawanda, NY: Multi-Health Systems, Inc;

Conway, K. P., Swendsen, J. D., & Merikangas, K. R. (2003). Alcohol expectancies, alcohol consumption, and problem drinking: The moderating role of family history. *Addictive Behaviors, 28,* 823–836.

Cook, W. L., & Kenny, D. A. (2005). The actor–partner interdependence model: A model of bidirectional effects in developmental studies. *International Journal of Behavioral Development, 29*(2), 101–109.

Cooper, M. L, Frone, M. R., Russell, M., & Mudar, P. (1995). Drinking to regulate positive and negative emotions: A motivational model of alcohol use. *Journal of Personality and Social Psychology, 69,* 990–1005.

Cooper, M. L. (1994). Motivations for alcohol use among adolescents: Development and validation of a four-factor-model. *Psychological Assessment, 6*(2), 117–128.

Corbin, W. R., Fromme, K., & Bergeson, S. E. (2006). Preliminary data on the association among the serotonin transporter polymorphism, subjective alcohol experiences, and drinking behavior. *Journal of Studies on Alcohol and Drugs, 67,* 5–13.

Costa, P. T., Jr., & McCrae, R. R. (1992). Revised NEO personality inventory (NEO PI-R) and NEO five-factor inventory (NEO-FFI) professional manual. Odessa, FL: Psychological Assessment Resources, Inc.

Coxe, S., West, S. G., & Aiken, L. S. (2009). The analysis of count data: A gentle introduction to poisson regression and its alternatives. *Journal of Personality Assessment, 91*(2), 121–136.

Crews, T. M., & Sher, K. J. (1992). Using adapted short MASTs for assessing parental alcoholism: Reliability and validity. *Alcoholism: Clinical and Experimental Research, 16,* 576-584.

Croissant, B., Rist, F., Demmel, R., & Olbrich, R. (2006). Alcohol-induced heart rate response dampening and rewarding stress paradigms in subjects at risk for alcoholism. *International Journal of Psychophysiology, 61,* 253–261.

Cuijpers, P., & Smit, F. (2001). Assessing parental alcoholism: A comparison of the Family History Research Diagnostic Criteria versus a single-question method. *Addictive Behaviors, 26,* 741-748.

Davis, C., Levitan, R. D., Kaplan, A. S., Carter, J., Reid, C., Curtis, C., et al. (2008). Reward sensitivity and the D2 dopamine receptor gene: A case-control study of binge eating disorder. *Progress in Neuro-Psychopharmacology & Biological Psychiatry, 32*(3), 620–628.

de Wit, H. (2005). Relationships between personality and acute subjective responses to stimulant drugs. In M. Earleywine (Ed.), *Mind-altering drugs: The science of subjective experience.* NY: Oxford University Press.

DePaulo, B. M. (1992). Nonverbal behavior and self-presentation. *Psychological Bulletin, 111,* 203–243.

Dick, D. M., & Foroud, T. (2003). Candidate genes for alcohol dependence: A review of genetic evidence from human studies. *Alcoholism: Clinical and Experimental Research, 7,* 868–879.

Dick, D. M., Rose, R. J., Viken, R. J., Kaprio, J., & Koskenvuo, M. (2001). Exploring gene-environment interactions: Socioregional moderation of alcohol use. *Journal of Abnormal Psychology, 110*(4), 625–632.

Dick, D. M., Wang, J. C., Plunkett, J., Aliev, F., Hinrichs, A., Bertelsen, S., et al. (2007). Family-based association analyses of alcohol dependence phenotypes across DRD2 and neighboring gene ANKK1. *Alcoholism: Clinical and Experimental Research, 31*(10), 1645–1653.

Ding, Y. C., Chi, H. C., Grady, D. L., Morishima, A., Kidd, J. R., Kidd, K. K., et al. (2002). Evidence of positive selection acting at the human dopamine receptor D4 gene locus. *Proceedings of the National Academy of Sciences of the United States of America, 99,* 309–314.

Doty, P., & de Wit, H. (1995). Effects of setting on the reinforcing and subjective effects of ethonol in social drinkers. *Psychopharmacology, 118,* 19–27.

Dreber, A., Apicella, C., Eisenberg, D., Garcia, J., Zamore, R., Lum, J. K., et al. (2009) The 7R polymorphism in the dopamine receptor D4 gene (DRD4) is associated with financial risk-taking in men. *Evolution and Human Behavior, 30,* 85–92.

Duan, J., Wainwright, M. S., Comeron, J. M., Saitou, N., Sanders, A. R., Gelernter, J., et al. (2003). Synonymous mutations in the human dopamine receptor D2 (DRD2) affect mRNA stability and synthesis of the receptor. *Human Molecular Genetics, 12,* 205–216.

Dubertret, C., Gouya, L., Hanoun, N., Deybach, J. C., Adès, J., Hamon, M., et al. (2004). The 3' region of the DRD2 gene is involved in genetic susceptibility to schizophrenia. *Schizophrenia Research, 67,* 75–85.

Dubertret, C., Gouya, L., Hanoun, N., Deybach, J. C., Ades, J., Hamon, M., et al. (2004). The 3′ region of the DRD2 gene is involved in genetic susceptibility to schizophrenia. *Schizophrenia Research, 67,* 75–85.

Ducci, F., & Goldman, D. (2008). Genetic approaches to addiction: Genes and alcohol. *Addiction, 103,* 1414–1428.

Ebstein R. P., Novick O., Umansky R., Priel B., Osher Y., Blaine, D., et al. (1996). Dopamine D4 receptor (D4DR) exon III polymorphism associated with the human personality trait of novelty seeking. *Nat Genet, 12,* 78–80.

Edenberg, H. J., Reynolds, J., Koller, D. L., Begleiter, H., Bucholz, K. K., Conneally, P. M., et al. (1998). A family-based analysis of whether the functional promoter alleles of the serotonin transporter gene HTT affect the risk for alcohol dependence. *Alcoholism: Clinical and Experimental Research, 22,* 1080–1085.

Ehlers, C. L., Gilder, D. A., Wall, T. L., Phillips, E., Feiler, H., & Wilhelmsen, K. C. (2004). Genomic screen for loci associated with alcohol dependence in Mission Indians. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics, 129,* 110–115. doi:1527405110 .1002/ajmg.b.30057.

Eisenberg, D. T. A., Apicella, C. L., Campbell, B. C., Dreber, A., Garcia, J. R., & Lum, J. K. (2010) Assortative human pair-bonding for partner ancestry and allelic variation of the dopamine receptor D4 (DRD4) gene. *Social Cognitive and Affective Neuroscience, 5,* 94–202.

Eisenegger, C., Knoch, D., Ebstein, R. P., Gianotti, L. R., Sándor, P. S., & Fehr, E. (2010). Dopamine receptor D4 polymorphism predicts the effect of L-DOPA on gambling behavior. *Biological Psychiatry, 67*(8), 702–706.

Ekman, P. (1989). The argument and evidence about universals in facial expressions of emotion. In H. Wagner & A. Manstead (Eds.), *Handbook of Social Psychophysiology* (pp. 143–164). Chichester: John Wiley Ltd.

Ekman, P., & Friesen, W. V. (1982). Rationale and reliability for EMFACS coders. Unpublished manuscript.

Ekman, P., & Friesen, W. V. (1986). A new pan-cultural facial expression of emotion. *Motivation & Emotion, 10,* 159–168.

Ekman, P., Friesen, W. V., & Hagar, J. C. (2002). Facial Action Coding System on CD-Rom [Computer software]. Salt Lake City, UT: Network Information Research.

Engels, R. C., Wiers, R., Lemmers, L., Overbeek, G. J. (2005). Drinking motives, alcohol expectancies, self-efficacy, and drinking patterns. *Journal of Drug Education, 35*(2), 147–166.

Enoch, M. A. (2006). Genetic and environmental influences on the development of alcoholism: Resilience vs. risk. *Annals of the New York Academy of Sciences, 1094,* 193–201.

Enoch, M. A. (2008). The role of GABA(A) receptors in the development of alcoholism. *Pharmacology Biochemistry and Behavior, 90,* 95–104.

Evans, N. J., & Jarvis, P. A. (1986). The group attitude scale: A measure of attraction to group. *Small Group Research, 17,* 203–216.

Fadda, F., Mosca, E., Colombo, G., & Gessa, G. L. (1989). Effect of spontaneous ingestion of ethanol on brain dopamine metabolism. *Life Sciences, 44,* 281–287.

Faraone, S. V., Doyle, A. E., Mick, E., & Biederman, J. (2001) Meta-analysis of the association between the dopamine D4 gene 7-repeat allele and attention deficit-hyperactivity disorder. *American Journal of Psychiatry, 158,* 1052–1057.

Feinn, R., Nellissery, M., & Kranzler, H. R. (2005). Meta-analysis of the association of a functional serotonin transporter promoter polymorphism with alcohol dependence. *American Journal of Medical Genetics: Part B, Neuropsychiatric Genetics, 133,* 79–84.

Filbey, F. M., Ray, L., Smolen, A., Claus, E. D., Audette, A., & Hutchison, K. E. (2008). Differential neural response to alcohol priming and alcohol taste cues is associated with DRD4 VNTR and OPRM1 genotypes. *Alcoholism: Clinical and Experimental Research, 32*(7), 1113–1123.

Fillmore, M. T., Vogel-Sprott, M., & Gavrilescu, D. (1999). Alcohol effects on intentional behavior: Dissociating controlled and automatic influences. *Experimental and Clinical Psychopharmacology, 7,* 372–378.

Frank, M., Ekman, P., & Friesen, W. (1993). Behavioral markers and recognizability of the smile of enjoyment. *Journal of Personality and Social Psychology, 64,* 83–93.

Fromme, K., & D'Amico, E. (1999). Neurobiological bases of alcohol's psychological effects. In K.E. Leonard & H.T. Blane (Eds.) *Psychological theories of drinking and alcoholism* (2nd ed., pp. 422–455). New York: Guilford Press.

Fromme, K., de Wit, H., Hutchison, K. E., Ray, L., Corbin, W. R., Cook, T. A. R., Wall, T. L., & Goldman, D. (2004). Biological and behavioral markers of alcohol sensitivity. *Alcoholism: Clinical and Experimental Research, 28*(2), 247–256.

Gabbay, F.H. (2005). Family history of alcoholism and response to amphetamine: Sex differences in the effect of risk. *Alcoholism: Clinical and Experimental Research, 29,* 773–780.

Garcia, J. R., MacKillop, J., Aller, E. L., Merriwether, A. M., Wilson, D.S, & Lum, J. K. (2010). Associations between dopamine D4 receptor gene variation with both infidelity and sexual promiscuity. *PLoS ONE, 5*(11), e14162. doi:10.1371/journal.pone.0014162.

Gelernter, J., Kranzler, H., & Cubells, J. F. (1997). Serotonin transporter protein (SLC6A4) allele and haplotype frequencies and linkage disequilibria in African and European American and Japanese populations and in alcohol dependent subjects. *Human Genetics, 101,* 243–246.

George, S. R., Cheng, R., Nguyen, T., Israel, Y., & O'Dowd, B. F. (1993). Polymorphisms of the D4 dopamine receptor alleles in chronic alcoholism. *Biochemical and Biophysical Research Communications, 196,* 107–114.

Giancola, P.R. (2002). Alcohol-related aggression during the college years: Theories, risk factors and policy implications. *Journal of Studies on Alcohol, 14*(Suppl.), 129–139.

Gizer, I. R., Ficks, C., & Waldman, I.D. (2009). Candidate gene studies of ADHD: A meta-analytic review. *Human Genetics, 126,* 51–90.

Goldman, D., Oroszi, G., & Ducci, F. (2005). The genetics of addictions: Uncovering the genes. *Nature Reviews: Genetics, 6,* 521–532.

Goldman, M. S., Brown, S. A., & Christiansen, B. A. (1987). Expectancy theory: Thinking and drinking. In Blane, H. T., & Leonard, K. E. (Eds.), *Psychological theories of drinking and alcoholism* (pp. 181–226). New York: Guilford Press.

Goldman, M. S., Del Boca, F. K., & Darkes, J. (1999). Alcohol expectancy theory: The application of cognitive neuroscience. In K.E. Leonard & H.T. Blane (Eds.), *Psychological theories of drinking and alcoholism* (2nd ed., pp. 203–246). New York: Guilford Press.

Gonzaga, G. C., Keltner, D., Londahl, E. A., & Smith, M. D. (2001). Love and the commitment problem in romantic relations and friendship. *Journal of Personality and Social Psychology, 81,* 247–262.

Gorwood, P., Batel, P., Ades, J., Hamon, M., & Boni, C. (2000). Serotonin transporter gene polymorphisms, alcoholism, and suicidal behavior. *Biological Psychiatry, 48,* 259–264.

Gorwood, P., Le Strat, Y., Ramoz, N., Dubertret, C., Moalic, J. M., & Simonneau, M. (2012). Genetics of dopamine receptors and drug addiction. *Human Genetics, 131*(6), 803–822. doi:10.1007/s00439-012-1145-7.

Gotlib, I. H., Joormann, J., Minor, K. L., & Hallmayer, J. (2008). HPA axis reactivity: A mechanism underlying the associations among 5-HTTLPR, stress, and depression. *Biological Psychiatry, 63,* 847–851.

Gottesman, I. I., & Gould, T. D. (2003). The endophenotype concept I psychiatry: Etymology and strategic intentions. *American Journal of Psychiatry, 160,* 636–645.

Hammoumi, S., Payen, A., Favre, J., Balmes, J., Bernard, J., Husson, M., et al. (1999). Does the short variant of the serotonin transporter linked polymorphic region constitute a marker of alcohol dependence? *Alcohol, 17,* 107–112.

Han, S. C., & Evans, S. M. (2005). Sex and drugs. In M. Earleywine (Ed.), *Mind-altering drugs: The science of subjective experience.* NY: Oxford University Press.

Harrison, G. G. (1985). Height-weight tables. *Annals of Internal Medicine, 103,* 498–494.

Hawkley, L. C., & Cacioppo, J. T. (2010). Loneliness matters: A theoretical and empirical review of consequences and mechanisms. *Annals of Behavioral Medicine, 40,* 218–227.

Heath, A. C., Bucholz, K. K., Madden, P. A., Dinwiddie, S. H., Slutske, W. S., Bierut, L. J., et al. (1997). Genetic and environmental contributions to alcohol dependence risk in a national twin sample: Consistency of findings in women and men. *Psychological Medicine, 27,* 1381–1396.

Heath, A. C., & Nelson, E. C. (2002). Effects of the interaction between genotype and environment: research into genetic epidemiology of alcohol dependence. *Alcohol Health Res World, 26,* 193-201.

Heils, A., Teufel, A., Petri, S., Stöber, G., Riederer, P., Bengel, D., et al. (1996). Allelic variation of human serotonin transporter gene expression. *Journal of Neurochemistry, 66,* 2621–2624.

Heimberg, R. G., Mueller, G. P., Holt, C. S., Hope, D. A., & Schneier, E. R. (1992). Assessment of anxiety in social interaction and being observed by others: The Social Interaction Anxiety Scale and the Social Phobia Scale. *Behavior Therapy, 23,* 53–73.

Heinz, A., Jones, D. W., Gorey, J. G., Bennet, A., Suomi, S .J., Weinberger, D. R., et al. (2003). Serotonin transporter availability correlates with alcohol intake in non-human primates. *Molecular Psychiatry, 8,* 231–234.

Herman, A. I., Philbeck, J. W., Vasilopoulos, N. L., & Depetrillo, P. B. (2003). Serotonin transporter promoter polymorphism and difference in alcohol consumption behavior in a college student population. *Alcohol, 38,* 446–449.

Hill, E. M., Stoltenberg, S. F., Bullard, K. H., Li, S., Zucker, R. A., & Burmeister, M. (2002). Antisocial alcoholism and serotonin-related polymorphisms: Association tests. *Psychiatric Genetics, 12,* 143–153.

Hill, S. Y. (2010). Neural plasticity , human genetics, and risk for alcohol dependence. In M. T. Reilly & D. M. Lovinger (Eds.), *Functional plasticity and genetic variation: Insights into the neurobiology of alcoholism* (p. 53). Academic Press.

Hill, S. Y., & Smith, T. R. (1991). Evidence for genetic mediation of alcoholism in women. *Journal of Substance Abuse, 3,* 159–174.

Hill, S. Y., Hoffman, E. K., Zezza, N., Thalamuthu, A., Weeks, D. E., Matthews, A. G., et al. (2008). Dopaminergic mutations: Within-family association and linkage in multiplex alcohol dependence families. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics,147B*(4), 517–526

Hill, S. Y., Shen, S., Zezza, N., Hoffman, E. K., Perlin, M., & Allen, W. A. (2004). A genome wide search for alcoholism susceptibility genes. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics, 128,* 102–113.

Hill, S. Y., & Tessner, K. (2010). Neural circuitry associated with risk for alcohol dependence. *Neuropsychology Reviews, 20,* 1-20.

Hill, S. Y., Zezza, N., Wipprecht, G., Xu, J., & Neiswanger, K. (1999). Linkage studies of D2 and D4 receptor genes and alcoholism. *American Journal of Medical Genetics,* 88, 676–685.

Hinckers, A. S., Laucht, M., Schmidt, M. H., Mann, K. F., Schumann, G., Schuckit, M. A., et al. (2006). Low level of response to alcohol as associated with serotonin transporter genotype and high alcohol intake in adolescents. *Biological Psychiatry, 60,* 282–287.

Hirvonen, M., Laakso, A., Någren, K., Rinne, J. O., Pohjalainen, T., & Hietala, J. (2004). C957T polymorphism of the dopamine D2 receptor (DRD2) affects striatal DRD2 availability in vivo. *Molecular Psychiatry, 9,* 1060–1061.

Hirvonen, M., Laakso, A., Någren, K., Rinne, J. O., Pohjalainen, T., & Hietala, J. (2005). C957T polymorphism of the dopamine D2 receptor (DRD2) affects striatal DRD2 availability in vivo [Erratum]. *Molecular Psychiatry, 10,* 889.

Hirvonen, M., Laakso, A., Någren, K., Rinne, J. O., Pohjalainen, T., & Hietala, J. (2009). C957T polymorphism of dopamine D2 receptor gene affects striatal DRD2 in vivo availability by changing the receptor affinity. *Synapse, 63,* 907–912.

Hodgins, D. C., Maticka-Tyndale, E., El-Guebaly, N., & West, M. (1993). The CAST-6: Development of a short-form of the Children of Alcoholics Screening Test. *Addictive Behaviors, 18*(3), 337–345.

Holdstock, L., & de Wit, H. (1999). Individual differences in response to ethanol and triazolam. *Behavioural Pharmacology, 10,* 283–295.

Holdstock, L., & de Wit, H. (2001). Individual differences in responses to ethanol and d-amphetamine in humans. *Alcoholism: Clinical Experiments and Research, 25,* 540–548.

Hu, X., Oroszi, G., Chun, J., Smith, T. L., Goldman, D., & Schuckit, M. A. (2005). An expanded evaluation of the relationship of four alleles to the level of response to alcohol and the alcoholism risk. *Alcoholism: Clinical and Experimental Research, 29*(1), 8–16.

Hull, J. G., & Bond, C. F. (1986). Social and behavioral consequences of alcohol consumption and expectancy: A meta-analysis. *Psychological Bulletin, 99,* 347–360.

Hutchison, K. E., McGeary, J., Smolen, A., Bryan, A., & Swift, R. M. (2002). The DRD4 VNTR polymorphism moderates craving after alcohol consumption. *Health Psychology, 21,* 139–146.

Iwata, N., Cowley, D. S., Radel, M., Roy-Byrne, P. P., & Goldman, D. (1999). Relationship between a GABAAα6 Pro385Ser substitution and benzodiazepine sensitivity. *The American Journal of Psychiatry, 156*(9), 1447–1449.

Jorm, A. F., Henderson, A. S., Jacomb, P. A., Christensen, H., Korten, A. E., Rodgers, B., et al. (1998). An association study of a functional polymorphism of the serotonin transporter gene with personality and psychiatric symptoms. *Molecular Psychiatry, 3*, 449–451.

Jovanovic, V., Guan, H.-C., & Van Tol, H H. M. (1999). Comparative pharmacological and functional analysis of the human dopamine D4.2 and D4.10 receptor variants. *Pharmacogenetics, 9*, 561–568.

Kalivas, P. W., & Volkow, N. D. (2005). The neural basis of addiction: A pathology of motivation and choice. *American Journal of Psychiatry, 162*(8), 1403–1413.

Kaplan, B., Whitsett, S., & Robinson, J. (1990). Menstrual cycle phase is a potential confound in psychophysiology research. *Psychophysiology, 27*, 445–450.

Karg, K., Burmeister, M., Shedden, K., & Sen, S. (2011). The serotonin transporter promoter variant (5-HTTLPR), stress, and depression meta-analysis revisited: Evidence of genetic moderation. *Archives of General Psychiatry, 68*(5), 444–454.

Kashy, D. A., & Kenny, D. A. (2000). The analysis of data from dyads and groups. In, Reis, H. T., & Judd, C. M. (Eds.), *Handbook of research methods in social and personality psychology* (pp. 451–477). Cambridge, U.K.; New York: Cambridge University Press.

Keltner, D. (1995). Signs of appeasement: Evidence for the distinct displays of embarrassment, amusement, and shame. *Journal of Personality and Social Psychology, 68*, 441–454.

Keltner, D., & Bonanno, G. A. (1997). A study of laughter and dissociation: The distinct correlates of laughter and smiling during bereavement. *Journal of Personality and Social Psychology, 73*, 687–702.

Keltner, D., & Buswell, B. N. (1997). Embarrassment: Its distinct form and appeasement functions. *Psychological Bulletin, 122*, 250–270.

Kendler, K. S. (2001). Twin studies of psychiatric illness: an update. *Archives of General Psychiatry, 58*, 1005–1014.

Kendler, K. S., Gardner, C. O., & Prescott, C. A. (2011). Toward a comprehensive developmental model for alcohol use disorders in men. *Twin Research and Human Genetics, 14*, 1–15.

Kendler, K. S., Gardner, C., & Dick, D. M. (2011). Predicting alcohol consumption in adolescence from alcohol-specific and general externalizing genetic risk factors, key environmental exposures and their interaction. *Psychological Medicine, 41*, 1507–1516. doi:10.1017/S003329171000190X.

Kenny, D. A., Mannetti, L., Pierro, A., Livi, S., & Kashy, D. A. (2002). The statistical analysis of data from small groups. *Journal of Personality and Social Psychology, 83,* 126–137.

King, G. (1988). Statistical models for political science event counts: Bias in conventional procedures and evidence for the exponential Poisson regression model. *American Journal of Political Science, 32,* 838–863.

Kirchner, T., Sayette, M., Cohn, J., Moreland, R., & Levine, J. (2006). Effects of alcohol on group formation among male social drinkers. *Journal of Studies on Alcohol, 67,* 785–793.

Korpi, E. R., Uusi-Oukari, M., Wegelius, K., Casanova, M., Zito, M., & Kleinman, J. E. (1993). Cerebellar and frontal cortical benzodiazepine receptors in human alcoholics and chronically alcohol-drinking rats. *Biological Psychiatry, 31,* 774–786.

Krach, S., Paulus, F. M., Bodden, M., & Kircher, T. (2010). The rewarding nature of social interactions. *Frontiers in Behavioural Neuroscience, 4,* 22.

Kranzler, H. R., Lappalainen, J., Gelernter, J., & Nellissery, M. (2002). Association study of alcoholism subtypes with a functional promoter polymorphism in the serotonin transporter protein gene. *Alcoholism: Clinical and Experimental Research, 9,* 1330–1335.

Krumhuber, E. G., & Manstead, A. S .R. (2009). Can Duchenne smiles be feigned? New evidence on felt and false smiles. *Emotion, 9*(6), 807–820.

Kuhnen, C. M., & Chiao, J. Y. (2009) Genetic determinants of financial risk taking. *PLoS ONE, 4*(2), e4362. doi:10.1371/journal.pone.0004362.

Kushner, M. G., Massie, E. D., Gaskel, S., Mackenzie, T., Fiszdon, J., & Anderson, N. (1997). Alcohol effects on the facial expressions of anxiety patients undergoing panic provocation. *Addictive Behaviors, 22,* 275–280.

Kwon, J. M., & Goate, A. M. (2000). The candidate gene approach. *Alcohol Research and Health, 24*(3), 164–168.

Lang, A. R., Patrick, C. J., & Stritzke, W. G., K. (1999). Alcohol and emotional response: A multidimensional-multilevel analysis. In Leonard, K. E., & Blane, H. T. (Eds.), *Psychological theories of drinking and alcoholism* (pp. 328–371). New York: Guilford Press.

Lang, P. (1968). Fear reduction and fear behavior: Problems in treating a construct. In E. Shlein (Ed.), *Research in psychotherapy*. Washington, DC: American Psychological Association.

Larsen, H., van der Zwaluw, C. S., Overbeek, G., Granic, I., Franke, B., & Engels, R. C. M. E. (2010). A variable-number-of-tandem-repeats polymorphism in the dopamine D4 receptor gene affects social adaptation of a gene-environment interaction. *Psychological Science, 21,* 1064–1068.

Laucht, M., Becker, K., Blomeyer, D., & Schmidt, M. H. (2007). Novelty seeking involved in mediating the association between dopamine D4 receptor gene exon III polymorphism and heavy drinking in male adolescents: Results from a high-risk community sample. *Biological Psychiatry, 61*, 87–92.

Laucht, M., Becker, K., El-Faddagh, M., Hohm, E., & Schmidt, M. H. (2005). Association of the DRD4 exon III polymorphism with smoking in fifteen-year-olds: A mediating role for novelty seeking? *Journal of the American Academy of Child and Adolescent Psychiatry, 44*(5), 477–484.

Laucht, M., Hohm, E., Esser, G., Schmidt, M. H., & Becker, K. (2007). Association between ADHD and smoking in adolescence: Shared genetic, environmental and psychopathological factors. *Journal of Neural Transmission, 114*(8), 1097–1104.

Leary, M., R., & Kowalski, R., M. (1995). *Social anxiety*. New York: Guilford Press.

Le Foll, B., Gallo, A., Strat, Y. L., Lu, L., & Gorwood, P. (2009). Genetics of dopamine receptors and drug addiction: a comprehensive review. *Behavioural Pharmacology, 20,* 1–17.

LeMarquand, D., Pihl, R. O., & Benkelfat, C. (1994). Serotonin and alcohol intake, abuse, and dependence: Clinical evidence. *Biological Psychiatry, 36,* 326–337.

Levenson, R. W. (1987). Alcohol, affect, and physiology: Positive effects in the early stages of drinking. In: Gottheil, E., Drulev, K. A., Pashko, S., & Weinstein, S. P. (Eds.), *Stress and addiction* (pp. 173–196). New York: Brunner-Mazel, Inc.

Levenson, R. W., Sher, K. J., Grossman, L. M., Newman, J., & Newlin, D. B. (1980). Alcohol and stress response dampening: Pharmacological effects, expectancy, and tension reduction. *Journal of Abnormal Psychology, 89,* 528–538.

Levine, J. M., & Moreland, R. L. (1990). Progress in small groups research. *Annual Review of Psychology, 41,* 585–634.

Levine, J. M., & Moreland, R. L. (1998). Small Groups. In D. T. Gilbert, S. T. Fiske & G. Lindzey (Eds.), *The handbook of social psychology* (4th ed., Vol. 2, pp. 415–469). Boston: McGraw-Hill.

Li, D., Sham, P. C., Owen, M. J., & He, L. (2006). Meta-analysis shows significant association between dopamine system genes and attention deficit hyperactivity disorder (ADHD). *Human Molecular Genetics, 15*(14), 2276–2284.

Li, M. D., & Burmeister, M. (2009). New insights into the genetics of addiction. *Nature Reviews: Genetics, 10,* 225–231.

Lichter, J. B., Barr, C. L., Kennedy, J. L., Van Tol, H. H., Kidd, K. K., &, Livak, K. J. (1993). A hypervariable segment in the human dopamine receptor D4 (DRD4) gene. *Human Molecular Genetics, 2,* 767–773.

Lichtermann, D., Hranilovic, D., Trixler, M., Franke, P., Jernej, B., Delmo, C. D, et al. (2000). Support for allelic association of a polymorphic site in the promoter region of the serotonin transporter gene with risk for alcohol dependence. *American Journal of Psychiatry, 157,* 2045–2047.

Littell, R. C., Milliken, G. A., Stroup, W. W., & Wolfinger, R. D. (1996). SAS system for mixed models [Computer software]. Cary, NC: SAS Institute.

Loh, E. W., Higuchi, S., Matsushita, S., Murray, R., Chen, C. K., & Ball, D. (2000). Association analysis of the GABA-A receptor subunit genes cluster on 5q33-34 and alcohol dependence in a Japanese population. *Molecular Psychiatry, 5,* 301–307.

Long, J. C., Knowler, W. C., Hanson, R. L., Robin, R. W., Urbanek, M., Moore, E., et al. (1998). Evidence for genetic linkage to alcohol dependence on chromosomes 4 and 11 from an autosome-wide scan in an American Indian population. *American Journal of Medical Genetics, 81,* 216–221.

Lyvers, M. F., & Maltzman, I. (1991). The balanced placebo design: Effects of alcohol and beverage instructions cannot be independently assessed. *The International Journal of the Addictions, 26,* 963–972.

MacKillop, J., Menges, D. P., McGeary, J. E., & Lisman, S. A. (2007). Effects of craving and DRD4 VNTR genotype on the relative value of alcohol: An initial human laboratory study. *Behavioral and Brain Functions, 3,* 1–12.

Madrid, G. A., MacMurray, J., Lee, J. W., Anderson, B. A., & Comings, D. E. (2001). Stress as a mediating factor I the association between the DRD2 TaqI polymorphism and alcoholism. *Alcohol, 23,* 117–122.

Mann, L. M., Chassin, L., & Sher, K. J. (1987). Alcohol expectancies and the risk for alcoholism. *Journal of Consulting and Clinical Psychology, 55,* 411–417.

Marlatt, G.A., & Rohsenow, D.J. (1980). Cognitive processes in alcohol use: Expectancy and the balanced placebo design. In N.K. Mello, (Ed.), *Advances in substance abuse: Behavioral and biological research* (Vol. 1, pp. 159–199). Greenwich, Connecticut: JAI Press.

Martin, C., & Sayette, M. (1993). Experimental design in alcohol administration research: Limitations and alternatives in the manipulation of dosage-set. *Journal of Studies on Alcohol, 54,* 750–761.

Martin, C., Earleywine, M., Musty, R., Perrine, M., & Swift, R. (1993). Development and validation of the biphasic alcohol effects scale. *Alcoholism: Clinical and Experimental Research, 17,* 140–146.

Mathews, A. G., Hoffman, E. K., Zezza, N., Stiffler, S., & Hill, S. Y. (2007). The role of the GABRA2 polymorphism in multiplex alcohol dependence families with minimal comorbidity: Within-family association and linkage analyses. *Journal of Studies on Alcohol and Drugs, 68,* 625–633.

McBride, W. J., Chernet, E., Dyr, W., Lumeng, L., & Li, T.-K. (1993). Densities of dopamine D2 receptors are reduced in CNS regions of alcohol-preferring P rats. *Alcohol, 10,* 387–390.

McCarroll, S. A., & Altshuler, D. M. (2007). Copy-number variation and association studies of human disease. *Nature Genetics, 39, S*37–*S*42.

McCarthy, M. I., Abecasis, G. R., Cardon, L. R., Goldstein, D. B., Little, J., Ioannidis, J. P. A., & Hirschhorn, J. N. (2008). Genome-wide association studies for complex traits: Consensus, uncertainty and challenges. *Nature Reviews Genetics 9,* 356-369.

McGeary, J. (2009). The DRD4 exon 3 VNTR polymorphism and addiction-realted phenotypes: A review. *Pharmacology, Biochemistry, and Behavior, 93,* 222–229.

McGue, M. (1995). Mediators and moderators of alcoholism inheritance. In: Turner, J. R., Cardon, L. R., & Hewitt, J. K. (Eds.), *Behavioral genetic approaches in behavioral medicine* (pp. 17–44). New York: Plenum Press.

McGue, M., & Iacono, W.G. (2004). The initiation of substance use in adolescence: A behavioral genetics perspective. In D. F. Lisabeth (Ed.), *Behavior genetics principles: Perspectives in development, personality, and psychopathology* (pp. 41–57). Washington, DC: American Psychological Association.

Merikangas, K. R., & McClair, V. L. (2012). Epidemiology of substance use disorders. *Human Genetics, 131*(6), 779–789. doi:1007/s00439-012-1168-0.

Moffitt, T. E., Caspi, A., & Rutter, M. (2005). Strategy for investigating interactions between measured genes and measured environments. *Archives of General Psychiatry, 62,* 473–481.

Moffitt, T. E., Caspi, A., & Rutter, M. (2006). Measured gene-environment interactions in psychopathology: Concepts, research strategies, and implications for research, intervention, and public understanding of genetics. *Perspectives on Psychological Science, 1*, 5–27.

Mokdad, A. H., Marks, J. S., Stroup, D. F., & Gerberding, J. L. (2004). Actual causes of death in the United States, 2000. *Journal of the American Medical Association, 291,* 1238–1245.

Monroe, S. M., & Reid, M. W. (2008). Gene-environment interactions in depression: Genetic polymorphisms and life stress polyprocedures. *Psychological Science, 19*(10), 947–956.

Morzorati, S. L., Ramchandani, V. A., Flury, L., Li, T.-K., & O'Conner, S. (2002). Self-reported subjective perception of intoxication reflects family history of alcoholism when breath alcohol levels are constant. *Alcoholism: Clinical and Experimental Research, 26,* 1299–1306.

Mosner, A., Kuhlman, G., Roehm, C., & Vogel, W. H. (1997). Serotonergic receptors modify the voluntary intake of alcohol and morphine but not of cocaine and nicotine by rats. *Pharmacology, 54,* 186–192.

Munafò, M. R., Brown, S. M., & Hariri, A. R. (2008). Serotonin transporter (5-HTTLPR) genotype and amygdala activation: A meta-analysis. *Biological Psychiatry, 63,* 852–857. doi:10.1016/j.biopsych.2007.08.016.

Munafò, M. R., Lingford-Hughes, A. R., Johnstone, E. C., & Walton, R. T. (2005). Association between the serotonin transporter gene and alcohol consumption in social drinkers. *American Journal of Medical Genetics Part B: Neuropsychiatric Genetics, 135,* 10–14.

Munafò, M. R., Matheson, I. J., & Flint, J. (2007). Association of the DRD2 gene Taq1A polymorphism and alcoholism: A meta-analysis of case–control studies and evidence of publication bias. *Molecular Psychiatry, 12,* 454–461.

Munafò, M., T. Clark, L. Moore, E. Payne & J. Flint. (2003). Genetic polymorphisms and personality in health adults: A systematic review and meta-analysis. *Molecular Psychiatry, 8,* 471–484.

Nakamura, M., Ueno, S., Sano, A., & Tanabe, H. (2000). The human serotonin transporter gene linked polymorphism (SERT) shows 10 novel allelic variants. *Molecular Psychiatry, 5,* 32–38.

Neville, M. J., Johnstone, E. C., & Walton, R. T. (2004). Identification and characterization of ANKK1: A novel kinase gene closely linked to DRD2 on chromosome band 11q23.1. *Human Mutation, 23,* 540–545.

Nevo, I., & Hamon, M. (1995). Neurotransmitter and nueromodulatory mechanisms involved in alcohol abuse and alcoholism. *Neurochemistry International, 26,* 305–336.

Newlin, D. B., & Renton, R. M. (2010). High risk groups often have higher levels of alcohol than low risk: The other side of the coin. *Alcoholism: Clinical and Experimental Research, 34*(2), 199–202.

Newlin, D. B., & Thomson, J. B. (1990). Alcohol challenge in sons of alcoholics: A critical review and analysis. *Psychological Bulletin, 108,* 383–402.

Nisbett, R. E., & Wilson, T. D. (1977). Telling more than we know: Verbal reports on mental processes. *Psychological Review, 84,* 231–259.

Noble, E. P. (1998). DRD2 gene and alcoholism. *Science, 281,* 1287–1288.

Noble, E. P., Blum, K., Ritchie, T., Montgomery, A., & Sheridan, P. J. (1991). Allelic association of the D2 dopamine receptor gene with receptor-binding characteristics in alcoholism. *Archives of General Psychiatry, 48,* 648–654.

Noldus Information Technology (2003). The Observer: Professional system for collection, analysis, and presentation of observational data (Version 5.0) [Computer software]. Wageningen, The Netherlands.

Oak, J. N., Oldenhof, J., & Van Tol, H. H. (2000). The dopamine D(4) receptor: One decade of research. *European Journal of Pharmacology, 405,* 303–327.

Palfai, T., & Wood, M. D. (2001). Positive alcohol expectancies and drinking behavior: The influence of expectancy strength and memory accessibility. *Psychology of Addictive Behaviors, 15,* 60–67.

Park, A., Sher, K. J., & Krull, J. L. (2008). Risky drinking in college changes as fraternity/sorority affiliation changes: A person-environment perspective. *Psychology of Addictive Behaviors, 22,* 219–229.

Park, A., Sher, K. J., Todorov, A. A., & Heath, A. C. (2011). Interaction between the DRD4 VNTR polymorphism and proximal and distal environments in alcohol dependence during emerging and young adulthood. Journal of *Abnormal Psychology, 120*(3), 585–595.

Patrick, M. E., Schulenberg, J. E., O'Malley, P. M., Maggs, J. L., Kloska, D. D, Johnston, L. D., et al. (2011). Age-related changes in reasons for using alcohol and marijuana from ages 18 to 30 in a national sample. *Psychology of Addictive Behaviors, 25*(2), 330–339. doi:10.1037/a0022445.

Perez de Castro, I., Ibanez, A., Torres, P., Saiz-Ruiz, J., & Fernandez-Piqueras, J. (1997). Genetic association study between pathological gambling and a functional DNA polymorphism at the D4 receptor gene. *Pharmacogenetics, 7,* 345–348.

Ponce, G., Hoenicka, J., Jiménez-Arriero, M. A., Rodríguez-Jiménez, R., Aragüés, M., Martín-Suñé, N., et al. (2008). DRD2 and ANKK1 genotype in alcohol-dependent patients with psychopathic traits: Association and interaction study. *British Journal of Psychiatry, 193,* 121–125. doi:10.1192/bjp.bp.107.041582.

Prescott, C. A., Sullivan, P. F., Kuo, P.-H., Webb, B. T., Vittum, J., Patterson, D. G., et al. (2006). Genomewide linkage study in the Irish affected sib pair study of alcohol dependence: Evidence for a susceptibility region for symptoms of alcohol dependence on chromosome 4. *Molecular Psychiatry, 11,* 603–611.

Radel, M., Iwata, N., & Goldman, D. (1998). Detection of sequence variants in GABAA receptor subunit genes by DHPLC analysis. *Alcoholism: Clinical and Experimental Research, 22*(Suppl.), 98A.

Raudenbush, S. W., & Bryk, A. S. (2002). *Hierarchical linear models: Applications and data analysis methods* (2nd ed.). Newbury Park, CA: Sage.

Ray, L. A. (2011). Stress-induced and cue-induced craving for alcohol in heavy drinkers: Preliminary evidence of genetic moderation by the OPRM1 and CRH-BP genes. *Alcoholism: Clinical and Experimental Research, 35*(1), 166-174.

Ray, L. A., Miranda, R., Tidey, J. W., McGeary, J. E., MacKillop, J., Gwaltney, C. J., et al. (2010). Polymorphisms of the μ-opioid receptor and dopamine D4 receptor genes and subjective responses to alcohol in the natural environment. *Journal of Abnormal Psychology, 119*(1), 115–125.

Reed, L. I., Sayette, M. A., & Cohn, J. F. (2007). Impact of depression on response to comedy: A dynamic facial coding analysis. *Journal of Abnormal Psychology, 116*(4), 804–809.

Reich, T., Edenberg, H. J., Goate, A., Williams, J. T., Rice, J. P., Van Eerdewegh, P., et al. (1998). Genome-wide search for genes affecting the risk for alcohol dependence. American Journal of *Medical Genetics, 81,* 207–215.

Rice, J. P., Reich, T., Bucholz, K. K., Neurnan, R. J., Fishman, R., Rochberg, N., et al. (1995). Comparison of direct interview and family history diagnoses of alcohol dependence. *Alcoholism: Clinical and Experimental Research, 19,* 1018-1023.

Rohsenow, D., & Marlatt, G. A. (1981). The balanced placebo design: Methodological considerations. *Addictive Behaviors, 6*, 107–122.

Ruch, W. (1993). Extraversion, alcohol, and enjoyment. *Personality and Individual Differences, 16*, 89–102.

Sari, Y., Bell, R. L., & Zhou, F. C. (2006). Effects of chronic alcohol and repeated deprivations on dopamine D1 and D2 receptor levels in the extended amygdala of inbred alcohol-preferring rats. *Alcoholism: Clinical and Experimental Research, 30,* 46–56.

Sayette, M. A., & Hufford, M. R. (1995). Urge and affect: A facial coding analysis of smokers. *Experimental and Clinical Psychopharmacology, 3,* 417–423.

Sayette, M. A., & Parrott, D. J. (1999). Effects of olfactory stimuli on urge reduction in smokers. *Experimental and Clinical Psychopharmacology, 7*(2), 151–159.

Sayette, M. A., Breslin, F. C., Wilson, G. T., & Rosenblum, G. (1994). An investigation of the balanced placebo design in alcohol administration research. *Addictive Behaviors, 19,* 333–342.

Sayette, M. A., Cohn, J. F., Wertz, J. M., Perrott, M. A., & Parrott, D. J. (2001a). A psychometric evaluation of the Facial Action Coding System for assessing spontaneous expression. *Journal of Nonverbal Behavior, 25,* 167–186.

Sayette, M. A., Contrada, R. C., & Wilson, G.T. (1990). Alcohol and correspondence between self-report and physiological measures of anxiety. *Behaviour Research and Therapy, 28,* 351–354.

Sayette, M. A., Creswell, K. G., Dimoff, J. D., Fairbairn, C. E., Cohn, J. F., Heckman, B. W, et al. (in press). Alcohol and group formation: A multimodal investigation of the effects of alcohol on emotion and social bonding. *Psychological Science.*

Sayette, M. A., Dimoff, J. D., Levine, J. M, Moreland, R. L., & Votruba-Drzal, E. (in press). The effects of alcohol and dosage-set on risk-seeking behavior in groups and individuals. *Psychology of Addictive Behaviors.* doi:10.1037/a0023903.

Sayette, M. A., Martin, C. S., Perrott, M. A., & Wertz, J. M. (2001c). Parental alcoholism and the effects of alcohol on semantic priming. *Experimental and Clinical Psychopharmacology, 9,* 409–417.

Sayette, M. A., Martin, C. S., Perrott, M. A., Wertz, J. M., & Hufford, M. R. (2001b). A test of the appraisal-disruption model of alcohol on stress. *Journal of Studies on Alcohol, 62,* 247–256.

Sayette, M. A., Smith, D. W., Breiner, M. J., & Wilson, G. T. (1992). The effect of alcohol on emotional response to a social stressor. *Journal of Studies on Alcohol, 53,* 541–545.

Sayette, M., A. (1993). An appraisal-disruption model of alcohol's effects on stress responses in social drinkers. *Psychological Bulletin, 114,* 459–476.

Sayette, M. A., & Wilson, G.T. (1991). Intoxication and exposure to stress: The effects of temporal patterning. *Journal of Abnormal Psychology, 100,* 56–62.

Schinka, J. A., Busch, R. M., & Robichaux-Keene, N. (2004). A meta-analysis of the association between the serotonin transporter gene polymorphism (5-HTTLPR) and trait anxiety. *Molecular Psychiatry, 9*(2), 197–202.

Schmidt, K. L., & Cohn, J., F. (2001). Human facial expressions as adaptations: Evolutionary questions in facial expression research. *Yearbook of Physical Anthropology, 44,* 3–24.

Schuckit, M. A., & Klein, J. L. (1991). Correlations between drinking intensity and reactions to ethanol and diazepam in healthy young men. *Neuropsychopharmacology, 4,* 157–163.

Schuckit, M. A., & Smith, T. L. (1996). An 8-year follow-up of 450 sons of alcoholic and control participants. *Archives of General Psychiatry, 53,* 202–210.

Schuckit, M. A., Mazzanti, C., Smith, T. L., Ahmed, U., Radel, M., Iwata, N., et al. (1999). Selective genotyping for the role of 5-HT2A, 5-HT2C, and GABAAα6 receptors and the serotonin transporter in the level of response to alcohol: A pilot study. *Biological Psychiatry, 45,* 647–651.

Schwarz, N. (1999). Self-reports: How the questions shape the answers. *American Psychologist, 5*, 93–105.

Shanahan, M. J., & Hofer, S. M. (2005). Social context in gene-environment interactions: retrospect and prospect. *Journal of Gerontology, 60B,* 65–76.

Sher, K. J. (1987). Stress response dampening. In H. T. Blane & K. Leonard (Eds.), Psychological theories of drinking and alcoholism. New York: Guilford Press.

Sher, K. J. (1991). Psychological characteristics of children of alcoholics. In D. Gallant and E. Gottheil (Eds.), *Recent developments in alcoholism: Vol. 9. Children of alcoholics* (pp. 301–326). New York: Plenum Press.

Sher, K. J., & Levenson, R. W. (1982). Risk for alcoholism and individual differences in the stress-response dampening effect of alcohol. *Journal of Abnormal Psychology, 91,* 350–367.

Sher, K. J., & Walitzer, K. S. (1986). Individual differences in the stress-response-dampening effect of alcohol: A dose-response study. *Journal of Abnormal Psychology, 95*(2), 159–167.

Sher, K. J., Grekin, E. R., & Williams, N. A. (2005). The development of alcohol use disorders. *Annual Review of Clinical Psychology, 1*, 493–523.

Sher, K., & Wood, M. (2005). Subjective effects of alcohol II: Individual differences. In M. Earleywine (Ed.), *Mind-altering drugs: The science of subjective experience.* NY: Oxford University Press.

Sher, K., Wood, M., Richardson, A. E., & Jackson, K. M. (2005). Subjective effects of alcohol I: Individual differences. In M. Earleywine (Ed.), *Mind-altering drugs: The science of subjective experience.* NY: Oxford University Press.

Singer, J. D. (1998). Using SAS PROC MIXED to fit multilevel models, hierarchical models, and individual growth models. *Journal of Educational and Behavioral Statistics, 24*(4), 323–355.

Singer, J. D., & Willett, J. B. (2003). Survival Analysis. In J. A. Schinka & W. F. Velicer (Eds.), *Handbook of psychology: Vol. 2. Research methods in psychology* (pp. 555–580). New York: John Wiley & Sons.

Slavicha, G. M., Way, B. M., Eisenberger, N. I., & Taylor, S. E. (2010). Neural sensitivity to social rejection is associated with inflammatory responses to social stress. *PNAS, 107*(33), 14817–14822.

Slutske, W. S., Hunt-Carter, E. E., Nabors-Oberg, R. E., Sher, K. J., Bucholz, K. K., Madden, P. A. F., et al. (2004). Do college students drink more than their non-college-attending peers? Evidence from a population-based longitudinal female twin study. *Journal of Abnormal Psychology, 113,* 530–540.

Smith, G. T., Goldman, M. S., Greenbaum, P. E., & Christiansen, B. A. (1995). Expectancy for social facilitation from drinking: The divergent paths of high-expectancy and low-expectancy adolescents. *Journal of Abnormal Psychology, 104*, 32–40.

Smith, L., Watson, M., Gates, S., Ball, D., & Foxcroft, D. (2008). Meta-analysis of the association of the Taq1A polymorphism with the risk of alcohol dependency: A HuGE gene-disease association review. *American Journal of Epidemiology, 167,* 125–138.

Spielberger, C. D., Gorsuch, R. L., & Lushene, R. E. (1970). *The state-trait anxiety inventory*. Palo Alto, CA: Consulting Psychologists Press.

Steele, C. M., & Josephs, R. A. (1990). Alcohol myopia: Its prized and dangerous effects. *American Psychologist, 45*(8), 921-933.

Stinson, F. S., Nephew, T. M., Dufour, M. C., & Grant, B. F. (1996). State trends in alcohol-related mortality, 1979-92. *US alcohol epidemiologic data reference manual: Vol. 5* (1st ed.). Bethesda, MD: National Institute on Alcohol Abuse and Alcoholism.

Surakka, V., & Hietanen, J., K. (1998). Facial and emotional reactions to Duchenne and non-Duchenne smiles. *International Journal of Psychophysiology, 29,* 23–33.

Sutker, P. B., Goist, K. C., & King, A. R. (1987). Acute alcohol intoxication in women: relationship to dose and menstrual cycle phase. *Alcoholism: Clinical and Experimental Research, 11,* 74–79.

Sweitzer, M. M., Halder, I., Flory, J. D., Craig, A. E., Gianaros, P. J., Ferrell, R. E., & Manuck, S. B. (2012). Polymorphic variation in the dopamine D4 receptor predicts delay discounting as a function of childhood socioeconomic status: Evidence for differential susceptibility. *Social Cognitive and Affective Neuroscience.* doi:10.1093/scan/nss020.

Testa, M., Fillmore, M. T., Norris, J., Abbey, A., Curtin, J. J., Leonard, K. E., et al. (2006). Understanding alcohol expectancy effects: Revisiting the placebo condition. *Alcohohlism: Clinical and Experimental Research, 30*(2), 339–348.

Thompson, J., Thomas, N., Singleton, A., Piggott, M., Lloyd, S., Perry, E. K., et al. (1997). D2 dopamine receptor gene (DRD2) Taq1 A polymorphism: reduced dopamine D2 receptor binding in the human striatum associated with the A1 allele. *Pharmacogenetics, 7*(6), 479–484.

Thompson, M. D., Gonzalez, N., Nguyen, T., Comings, D. E., George, S. R., & O'Dowd, B. F. (2000). Serotonin transporter gene polymorphisms in alcohol dependence. *Alcohol, 22,* 61– 67.

Türker, T., Sodmann, R., Goebel, U., Jatzke, S., Knapp, M., Lesch, K. P., et al. (1998). High ethanol tolerance in young adults is associated with the low-activity variant of the promoter of the human serotonin transporter gene. *Neuroscience Letters, 248,* 147–150.

U.S. Department of Health and Human Services (2007). *Behavioral risk factor surveillance system survey data.* Retrieved from http://www.cdc.gov/brfss/technical_infodata /surveydata/2007.htm.

Uher, R., & McGuffin, P. (2007). The moderation by the serotonin transporter gene of environmental adversity in the aetiology of mental illness: Review and methodological analysis. *Molecular Psychiatry, 13,* 131–146.

van den Wildenberg, E., Janssen, R. G., Hutchison, K. E., van Breukelen, G. J., & Wiers, R. W. (2007). Polymorphisms of the dopamine D4 receptor gene (DRD4 VNTR) and cannabinoid CB1 receptor gene (CNR1) are not strongly related to cue-reactivity after alcohol exposure. *Addiction Biology, 12,* 210–220.

Van Tol, H. H. M., Wu, C. M., Guan, H. C., Ohara, K., Bunzow, J. R., Civelli, O., et al. (1992). Multiple dopamine D4 receptor variants in the human population. *Nature, 358,* 149–152.

Volkow, N. D., & Li, T.-K. (2004). Drug addiction: The neurobiology of behaviour gone awry. *Nature Reviews: Neuroscience, 5*(12), 963–970.

Volkow, N. D., & Muenke, M. (2012). The genetics of addiction. *Human Genetics, 131*(6), 773–777. doi:10.1007/s00439-012-1173-3.

Volkow, N. D., Fowler, J. S., Wang, G. J., Baler, R., & Telang F. (2009). Imaging dopamine's role in drug abuse and addiction. *Neuropharmacology, 56*(Suppl.), 3–8.

Volkow, N. D., Wang, G. J., Begleiter, H., Porjesz, B., Fowler, J. S., Telang, F., et al. (2006). High levels of dopamine D2 receptors in unaVected members of alcoholic families: Possible protective factors. *Archives of General Psychiatry, 63*(9), 999–1008.

Volkow, N. D., Wang, G. J., Fowler, J. S., Logan, J., Gatley, S. J., Gifford, A., et al. (1999). Prediction of reinforcing responses to psychostimulants in humans by brain dopamine D2 receptor levels. *American Journal of Psychiatry, 156,* 1440–1443.

Wang, E., Ding, Y.-C., Flodman, P., Kidd, J. R., Kidd, K. K., Grady, D. L., et al. (2004). The genetic architecture of selection at the human dopamine receptor D4 (DRD4) gene locus. *American Journal of Human Genetics, 74,* 931–944.

Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: The PANAS scales. *Journal of Personality and Social Psychology, 54,* 1063–1070.

Way, B. M., & Taylor, S. E. (2010). The serotonin transporter promoter polymorphism (5-HTTLPR) is associated with cortisol response to psychosocial stress. *Biological Psychiatry, 67,* 487–492.

Weiner, J. L., Brozowski, S. J., Harris, R. A., & Dunwiddie, T. V. (1997). Whole cell patch clamp analysis of ethanol actions on human recombinant GABAA receptors. *Alcoholism: Clinical and Experimental Research, 21,* 75A.2.

Weiss, F., & Koob, G. F. (1991). The neuropharmacology of ethanol self-administration. In R.E. Meyer, G. F. Koob, M. J. Lewis & S. M. Paul (Eds.), *Neuropharmacology of ethanol* (pp. 125–162). Boston: Birkhauser.

Wendland, J. R., Martin, B. J., Kruse, M. R., Lesch, J.-P., & Murphy, D. L. (2006) Simultaneous genotyping of four functional loci of human SLC6A4, with a reappraisal of 5-HTTLRP and rs25531. *Molecular Psychiatry, 11,* 224–226.

Wilhelmsen, K. C., Schuckit, M., Smith, T. L., Lee, J. V., Segall, S. K., Feiler, et al. (2003). The search for genes related to a low-level response to alcohol determined by alcohol challenges. *Alcoholism: Clinical and Experimental Research, 27*(7), 1041–1047.

Yankofsky, L., Wilson, G. T., Adler, J. L., Hay, W. M., & Vrana, S. (1986). The effect of alcohol on self-evaluation and perception of negative interpersonal feedback. *Journal of Studies on Alcohol, 47,* 26–33.

Young, K. A., Gobrogge, K. L., & Wang, Z. (2011). The role of mesocorticolimbic dopamine in regulating interactions between drugs of abuse and social behavior. *Neuroscience and Biobehavioral Reviews, 35,* 498–515.

Zalsman, G., Huang, Y.H., Oquendo, M.A., Burke, A.K., Hu, X., Brent, D.A., et al. (2006). Association of a triallelic serotonin transporter gene promoter region (SERT) polymorphism with stressful life events and severity of depression. *American Journal of Psychiatry, 163,* 1588–1593.

Zhong, S., Israel, S., Shalev, I., Xue, H., Ebstein, R. P., & Chew, S. H. (2010). Dopamine D4 receptor gene associated with fairness preference in ultimatum game. *PLoS ONE, 5,* e13765. doi:10.1371/journal.pone.0013765.

Zinn-Justin, A., & Abel, L. (1999). Genome search for alcohol dependence using the weighted pairwise correlation linkage method: Interesting findings on chromosome 4. *Genetic Epidemiology, 17*(Suppl.), 421–426.