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Genetic Predictors of Cue- and Stress-Induced Cigarette Craving: An Exploratory Study

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Abstract

Cigarette cravings in response to environmental cues and stressors are widely recognized as important predictors of smoking cessation outcomes. Accumulating evidence suggests that genetics plays a role in these craving responses, as well as in smoking cessation more generally. Previous studies of genetic polymorphisms have been limited by examination of single candidate genes and the use of broadly defined phenotypes (e.g., smoking history). In addition, research examining the similarities and differences between cue- and stress-induced cravings has been limited, although some evidence has suggested that they may have common genetic underpinnings. In the current study, we examined associations between a panel of 1,350 candidate genetic polymorphisms and craving responses to laboratory smoking cues and stressors. We hypothesized that common genetic polymorphisms would be predictive of both cue- and stress-induced craving. Nicotine-dependent smokers ($n = 210$) donated a blood sample, were exposed to neutral, smoking-related, and stress-related stimuli, and completed craving questionnaires immediately prior to and following each stimulus. Findings indicated that craving responses to smoking cues and stressors were moderately correlated ($r = .44$). However, genetic analysis revealed that cue and stress-induced cigarette craving were predicted by different polymorphisms, such that variants in the glycine and dopamine pathways were predictive of cue-induced craving, whereas variants in the stress-corticotropin pathway predicted stress-induced craving. Conclusions: Study results provide no support for the hypothesis that cue- and stress-induced craving have the same genetic predictors.

Keywords

smoking; stress; cue; craving; genetic factors

Despite the clear negative health consequences of smoking and repeated attempts by most smokers to quit (American Cancer Society, 2009; Office of the Surgeon General, 2004), smoking cessation success remains elusive for many (Centers for Disease Control & Prevention, 2009). A long recognized reason that quitting smoking is so difficult is the

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ubiquitous presence of environmental cues that trigger strong, difficult-to-resist, cravings to smoke (Ferguson & Shiffman, 2008; Tiffany, 1990). Studies have also suggested that stress can trigger strong craving reactions that may in turn relate to poorer cessation outcomes (Shiffman & Waters, 2004). A large body of literature has demonstrated that such cue- and stress-induced cravings can be reliably modeled in the laboratory. Experimental exposure to smoking cues (e.g., handling a cigarette without smoking it) as well as laboratory stressors has been found to give rise to strong self-reported cigarette craving responses in smokers (Carter & Tiffany, 1999; Erblich, Boyarsky, Spring, Niaura, & Bovbjerg, 2003; Ferguson & Shiffman, 2009; Kassel, Stroud, & Paronis, 2003). Whether or not cue- and stress-induced cigarette cravings have similar mechanisms to one another or represent distinct motivational processes has yet to be determined (Stewart, 2008). This is an important clinical question, because interventions may need to be developed that appropriately, either independently or in combination, address cue- and stress-induced cravings.

Initial research now suggests that genetic factors may contribute to variability in cue- and stress-induced cigarette craving responses. For example, Hutchison et al. (Hutchison, LaChance, Niaura, Bryan, & Smolen, 2002) found that carriers of a genetic polymorphism at the gene that codes for the dopamine D4 receptor exhibited stronger craving reactions to smoking cues than noncarriers. Erblich and colleagues found that polymorphisms at the dopamine transporter and D2 receptor genes were predictive of Diaz, & Bovbjerg, 2004; Erblich, Lerman, Self, Diaz, & Bovbjerg, 2005), suggesting potential commonalities between the two processes.

Until recently, research on genetic factors in addictions generally, as well as smoking in particular, has relied heavily on association studies of either one gene, or a highly limited set of genes, and drug use outcomes. With the advent of high-throughput microarray technologies, contemporaneous analysis of large sets of single nucleotide polymorphisms (SNPs) has become more feasible and affordable. This methodological advance has led to numerous genome-wide association studies (GWAS), in which hundreds of thousands of marker SNPs across the entire genome are analyzed in relation to smoking-related phenotypes (Furberg et al., 2010). Although GWAS have the advantage of surveying the effects of the entire human genome on an outcome, they are fraught with numerous challenges, chief among which are the large number of false positives due to multiple testing, and, along similar lines, the difficulty involved in making sense of putative genetic effects that are not clearly in the causal pathway of interest (Wray, Goddard, & Visscher, 2007; Wray, Goddard, & Visscher, 2008; Ziegler, Konig, & Thompson, 2008).

An intermediate approach has been made possible by the recent development of a large panel of SNPs covering a broad selection of 130 genes associated with addictions and related psychiatric disorders (Hodgkinson et al., 2008). In this panel, 1,350 SNPs across 130 genes were identified (a full list is available in Hodgkinson et al., 2008) representing 13 systems that are relevant to the addictions: dopamine (10 genes), NMDA (six genes), GABA (19 genes), serotonin (12 genes), acetylcholine (seven genes), norepinephrine (eight genes), opioid (eight genes), drug metabolism (11 genes), HPA-related stress (nine genes), glycine (four genes), cannabinoid (two genes), signal transduction (22 genes), and other (12 genes) systems. For this panel, SNPs were selected to cover major haplotype blocks (with high levels of linkage disequilibrium within each block) across a given gene, based on data from the International HapMap Project (www.hapmap.org). Using this panel of SNPs represents an improvement over the single-gene association study approach, while remaining substantially more manageable than a genome-wide approach with regard to concerns about false positives and Type I error. In the present study, we evaluated the relationship between the 1,350 SNPs on the “addiction array” described above and cue- and stress-induced cigarette craving reactions.

Method and Materials

Participants

Adult smokers ($n = 210$) were recruited to a study of smoking behavior by advertisements (for a smoking study) placed in and around an urban medical center in New York City. All participants qualified for a current *DSM-IV* diagnosis of nicotine dependence (American Psychiatric Association, 1994) ascertained during initial telephone contact and reascertained in-person upon arrival. To qualify as a smoker, only participants who reported smoking at least 10 cigarettes per day for at least 5 years were included in the study. Participants were excluded if they reported: (a) fewer than 10 cigarettes per day, on average; (b) smoking history of less than 5 years; (c) current treatment for smoking cessation; (d) a history of other substance abuse; or (d) a history of hospitalization for mental illness.

Measures

Demographic and smoking questionnaire—Participants completed questionnaires assessing demographic information (e.g., age, gender, education, income, ethnicity) and personal smoking history (e.g., cigarettes per day, years smoked).

Fagerstrom Test of Nicotine Dependence (FTND)—The 6-item FTND was used as a measure of the strength of participants' dependence. The instrument has documented reliability and validity (Heatherton, Kozlowski, Frecker, & Fagerstrom, 1991) and has been used extensively in the literature.

Cigarette craving questionnaire—Improving on the use of single-item craving assessments, and following the recommendations of Kozlowski and colleagues (Kozlowski, Mann, Wilkinson, & Poulos, 1989; Kozlowski & Wilkinson, 1987) that craving be assessed using multiple descriptors (e.g., crave, urge, desire), this brief, 5-item, 0–100 instrument is designed specifically to make rapid assessments of craving during experimental manipulations. The instrument has been used as an outcome measure in previous reports by us (Erblich et al., 2003) and others (Hutchison et al., 2002). The craving questionnaire was administered immediately before and after each stimulus. The instrument evidenced high levels of internal consistency (Cronbach's $\alpha = .96$ – 0.97) at all administrations.

Procedures

Potential participants who responded to advertisements were prescreened via telephone to determine eligibility. The study was described over the phone, and if eligible, participants were scheduled for the study. Ninety percent of eligible respondents were enrolled in the study.

Participants were scheduled for an experimental session during which they were seated comfortably in a quiet controlled environment for exposure to three types of stimuli (neutral, smoking-related, stress-related). To avoid ceiling effects in cigarette craving, participants smoked one cigarette immediately before beginning the study procedures. Participants then donated a 6 ml sample of blood for genotyping, completed demographic, smoking history and cessation questionnaires, and the FTND, and were exposed to the experimental stimuli using a classic laboratory imaginal exposure paradigm (Maude-Griffin & Tiffany, 1996). Briefly, participants listened to 60-s scripts and were instructed to imagine the scene in each as vividly as possible; each of the scripts was followed by a 30-s silent period, during which they were to continue imagining the scene. Participants viewed a nature video for 3 minutes between each script presentation. To avoid potential confounding by carryover effects of strong cigarette urges induced by smoking cues observed in prior experimental studies (Heish-man, Lee, Taylor, & Singleton, 2010; Sayette, Griffin, & Sayers, 2010), the neutral

cue was presented first, followed by the stress and smoking cues. Before beginning the exposures, participants were read a practice script (a trip to the grocery store) to familiarize them with the task. The imaginal cues employed have been described elsewhere (Maude-Griffin & Tiffany, 1996); the neutral imagery involved changing a light bulb, the stress imagery involved waiting for dental work, and the smoking imagery involved lighting up a cigarette after a meal. Cigarette cravings were measured immediately before and after exposure to each cue. Upon completion of the study, participants were thanked for their participation, offered referrals for smoking cessation interventions, and offered an honorarium for their time.

Genetic Analyses

DNA was extracted from blood samples using the PureGene DNA isolation kit (Gentra Systems, Minneapolis, MN). Genotyping was performed on a GoldenGate SNP custom panel (Illumina Inc., San Diego, CA) containing 1,350 SNPs across 130 candidate genes for addiction, described elsewhere (Hodgkinson et al., 2008). In addition, the panel contained 186 ancestry informative markers to control for population stratification (for a total of 1,536 SNPs) as described by Hodgkinson et al. (Hodgkinson et al., 2008; see therein for a complete list of all SNPs). SNP analyses were conducted at the Rockefeller University Genomics Resource Center (C. Zhao, Director). Ten percent of randomly selected samples were replicated for quality assurance of genotyping results.

Statistical Analyses

In a first set of analyses, we aimed to characterize the relationship between smoking cue- and stress-induced cigarette cravings. First, we calculated raw change scores (postexposure minus preexposure) for each of the three imaginal cues. We then calculated partial Pearson correlation coefficients for the smoking cue and stress scores, controlling for neutral scores and preexposure craving levels. Next, we performed a repeated-measures ANOVA to compare cue- and stress-induced craving scores, again controlling for neutral scores and preexposure craving levels. Demographic and smoking-related variables were considered as covariates. These analyses were conducted in SAS 9.1 (SAS, Inc., Cary, NC).

To analyze the genetic data, SNP information was imported into HelixTree SVS 6.4.2 software (Golden Helix, Inc., Bozeman, MT). In preliminary linear regressions, raw change scores for cue- and stress-induced cigarette craving were regressed on neutral change scores and preexposure craving levels. Residualized change scores were generated using this method (Draper & Smith, 1998) and served as the dependent variables in the genetic analyses. Next, to control for population stratification, principal components analysis (PCA) of the markers was performed, with six factors specified, as suggested by the validation data in Hodgkinson et al. (Hodgkinson et al., 2008). *p* values in the subsequent association analyses reflect the data corrected for any population stratification. Association analyses were then conducted assuming a dominant model. That is, carrier status of each minor allele was compared against homozygous noncarrier genotype. Consistent with the exploratory objective of this study and the desire to minimize Type II error, results were not Bonferroni corrected (Williams & Haines, 2011). For each SNP, genotype and allele frequencies were calculated and Hardy-Weinberg equilibrium was assessed.

Results

Background Variables

Forty-eight percent of participants ($n = 100$) were men and 52% ($n = 110$) were women. The mean age of the sample was 38.2 years ($SD = 10.3$). Forty-five percent of participants reported African American ethnicity, 22% reported Caucasian ethnicity, 26% reported

Hispanic ethnicity, and 7% reported other ethnic backgrounds. Forty percent of participants had a high school education or lower, and 45% of the sample reported annual incomes of \$20,000 or less. Participants smoked a mean of 18.4 cigarettes ($SD = 9.8$) per day for an average of 19.1 year ($SD = 10.2$). Mean Fagerstrom score of the sample was 5.7 ($SD = 2.2$). Preliminary analyses revealed that demographic variables were not related to cue- or stress-induced craving.

Magnitude of Cue-Induced and Stress-Induced Cravings

Consistent with our previous reports (Erblich et al., 2004; 2005) both stress imagery and smoking imagery elicited robust elevations in cigarette craving. Following exposure to the stress imagery, participants' craving scores increased by a mean of 14.3 (± 2.6) points on the 0–100 craving scale; $t(209) = 9.4$; $p < .0001$. Following exposure to the smoking imagery, participants' craving scores increased by a mean of 26.2 (± 2.4) points on the 0–100 craving scale; $t(209) = 19.0$; $p < .0001$. A head-to-head comparison of stress imagery- and smoking imagery-induced cravings revealed that the smoking imagery elicited significantly higher cravings than the stress imagery; $F(1, 208) = 30.6$; $p < .0001$.

Relationships Between Cue-induced and Stress-induced Cravings

Pearson partial correlation coefficients were performed to evaluate the relationship between smoking cue- and stress-induced cigarette cravings, controlling for neutral cue-induced cravings and prestimulus levels of craving. Findings revealed a significant moderate positive correlation; $r = .44$, $p < .0001$.

Genetic Analyses of SNP Panel

As indicated above, the SNP panel consisted of 1,350 addiction-related markers at 130 genes, as well as 186 ancestry informative markers as controls for population admixture. Of these 1,350 markers, the 93 SNPs associated with alcohol-specific drug metabolism (e.g., SNPs at the *ADH* and *ALDH* receptor loci) were dropped. The remaining 1,443 SNPs were analyzed. Of these, 32 SNPs were excluded because there was no allelic variability in the sample (see Table 1 for a list of the excluded SNPs). The average call rate for the remaining 1,411 SNPs was 99.7% with a replication rate of 99.8. Minor allele frequencies ranged from 0.002 to 0.497, with a mean minor allele frequency of 0.27 (± 0.15).

Relationships Between SNPs and Cue-Induced Cigarette Cravings

Genetic analyses identified 15 SNPs that were correlated with cue-induced cigarette cravings at the $p < .01$ level (see Table 2). Specifically, the significant SNPs were found at loci in the dopamine, glycine, NMDA, GABA, serotonin, and signal transduction systems. Each individual SNP accounted for 3.2 to 5.4% of the variance in craving responses. Only four of the SNPs, rs2053736 (at *SLC6A13*), rs10877970 (at *AVPR1A*), and rs5934186 and rs6630716 (at *GLRA2*), had significant departures from Hardy-Weinberg equilibrium ($p < .01$).

Relationships Between SNPs and Stress-Induced Cigarette Cravings

Genetic analyses identified 13 SNPs that were correlated with stress-induced cigarette cravings at the $p < .01$ level (see Table 3). None of the SNPs predictive of cue-induced cravings were predictive of stress-induced cravings. SNPs at loci in the HPA-stress, NMDA, GABA, serotonin, and signal transduction systems were found to be predictive of stress-induced cravings. Each individual SNP accounted for 3.2 to 5.8% of the variance in craving responses. Only two of the SNPs, rs455892 (at *GRIK1*), and rs9497515 (at *GRM1*), had significant departures from Hardy-Weinberg equilibrium ($p < .01$). Inclusion of race (African American vs. other) and gender as factors did not affect any of the results.

Discussion

The objective of the present study was to test the hypothesis that stress- and cue-induced cigarette cravings would be associated with similar genetic predictors. Consistent with previous research, both exposure to smoking imagery and mild stress imagery led to significant increases in cigarette craving. The two effects were significantly correlated suggesting that these phenomena may share some common mechanisms. Results revealed that none of the individual specific SNPs predicted both cue- and stress-induced cravings. Within the families of SNPs related to specific signaling pathways, SNPs in the glycine and dopamine systems were found to predict cue-induced craving, but not stress-induced craving. On the other hand, stress-induced craving but not cue-induced craving was predicted by a SNP in the HPA stress system. The GABA, NMDA, serotonin and signal transduction families were not selective for cue- versus stress-induced craving.

Findings of this exploratory study suggest that while there appear to be some general underlying commonalities between cue- and stress-induced cravings, they do not have identical predictors at the genetic level. Research on genetic predictors of cue- and stress-induced craving is sparse. The few studies that have been conducted have highlighted the role of dopamine-related genes in smoking cue-induced craving (Franklin et al., 2009; Erblich et al., 2005; Hutchison et al., 2002), but genes in other neurotransmitter systems have not been evaluated. Generally consistent with this literature, the present results suggest a role for dopamine-related genes in cue-induced craving, but also raise the possibility that other systems are involved as well. It is interesting that SNPs in the glycine receptor system were predictive of cue-induced craving. Although research on relations between glycine activity and nicotine dependence are lacking, there are several studies indicating that glycine receptors are located in the dopamine-rich ventral tegmental area (Molander & Soderpalm, 2005), and their activation can influence dopamine activity via stimulation of nicotinic acetylcholine receptors (Soderpalm, Lof, & Ericson, 2009).

Perhaps not surprisingly, a SNP in the HPA stress system (at *CRHBP*, which codes for corticotropin-releasing hormone binding protein) was predictive of stress-induced craving. Consistent with this finding, the CRHBP protein has been shown to be related to stress-induced cocaine-seeking in animals (Wang, You, Rice, & Wise, 2007). In contrast to Erblich et al. (Erblich et al., 2004), dopamine-related genes in the addiction array were not predictive of stress-induced cigarette cravings, although it should be noted that the specific variants in our previous study (rs1800497 at *DRD2* and the 40bp VNTR at *SLC6A3*) were not analyzed in the present study. Interestingly, CRHBP has been found to potentiate activity at both NMDA and dopamine receptors (Ungless et al., 2003), leaving open the possibility that dopamine is involved downstream in the stress-induced craving response.

Consistent with the larger literature on the effects of polymorphisms on complex behavioral endophenotypes, the effect sizes observed in this study were small, with each SNP accounting for only 3–6% of the variability in cigarette craving. It should be noted that in this study, nominal p values at the 0.01 level, were reported; SNPs were not significantly predictive following a more conservative Bonferroni correction. Thus, the results should be interpreted as preliminary. Indeed, given the number of tests performed, it would be anticipated that 14 chance findings each for cue- and stress-induced cravings would occur at the $p = .01$ level, approximately that which was observed. On the other hand, conservative corrections often lead to substantial increases in Type II error, and recent work has pointed to the potential exploratory importance of uncorrected p values (Williams & Haines, 2011). Thus the present results can be viewed as interesting preliminary data requiring follow-up confirmation. Another limitation is that the genetic panel did not include SNPs at the nicotinic acetylcholine receptor or at genes in the cytochrome P450 system responsible for

nicotine metabolism that have recently been found to be associated with smoking-related phenotypes (Ray, Tyndale, & Lerman, 2009).

In addition, the neutral, stress, and smoking stimuli were presented in a fixed order, thus raising the possibility of order effects. However, Sayette and colleagues (Sayette et al., 2010) have underscored important risks of counterbalancing in this work, and as such, we decided to fix the order of presentation, with smoking imagery, thought to be the most likely to elicit strong cravings, administered last. We should also note that we only employed one imaginal stressor and one imaginal smoking cue, and thus cannot rule out the possibility that effects were somehow specific to these cues. Additionally, we did not collect data on menstrual phase or oral contraceptive use among the women in the study, which in one recent study was found to have an impact on cue- and stress-induced cravings (Gray et al., 2010). Finally, although we identified candidate polymorphisms associated with cue- and stress-induced cigarette craving responses among nicotine dependent smokers, it is not clear how these effects relate to the key clinical outcome, smoking cessation. Future research is warranted to further elucidate the pathway from genetics to craving to cessation failures.

The genetic dissociation between cue- and stress-induced cigarette cravings may have implications for the development of more successful cessation treatments. These genetic results are consistent with a growing experimental neuroscience literature that suggests that while these two phenomena are related, they may have distinct neurobiological underpinnings (Stewart, 2008). Complementary pharmacological and behavioral interventions targeted to the specific management of cue- and stress-induced cigarette cravings may be appropriate to address these potentially distinct processes.

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References

- American Cancer Society. Cancer Facts and Figures 2009. Atlanta, GA: American Cancer Society; 2009.
- American Psychiatric Association. Diagnostic and statistical manual of mental disorders, IV (DSM-IV). Washington, DC: Author; 1994.
- Carter BL, Tiffany ST. Meta-analysis of cue-reactivity in addiction research. *Addiction*. 1999; 94:327–340. [PubMed: 10605857]
- Centers for Disease Control and Prevention. Cigarette smoking among adults and trends in smoking cessation—United States, 2008. *Morbidity and Mortality Weekly Report*. 2009; 58:1227–1232. [PubMed: 19910909]
- Draper, N.; Smith, H. *Applied Regression Analysis* (Wiley Series in Probability and Statistics: Texts and References Section). New York, NY: Wiley-Interscience; 1998.
- Erblich J, Boyarsky Y, Spring B, Niaura R, Bovbjerg DH. A family history of smoking predicts heightened levels of stress-induced cigarette craving. *Addiction*. 2003; 98:657–664. [PubMed: 12751983]
- Erblich J, Lerman C, Self DW, Diaz GA, Bovbjerg DH. Stress-induced cigarette craving: Effects of the DRD2 TaqI RFLP and SLC6A3 VNTR polymorphisms. *The Pharmacogenomics Journal*. 2004; 4:102–109. [PubMed: 14732864]
- Erblich J, Lerman C, Self DW, Diaz GA, Bovbjerg DH. Effects of dopamine D2 receptor (DRD2) and transporter (SLC6A3) polymorphisms on smoking cue-induced cigarette craving among African-American smokers. *Molecular Psychiatry*. 2005; 10:407–414. [PubMed: 15381926]

- Ferguson SG, Shiffman S. The relevance and treatment of cue-induced cravings in tobacco dependence. *Journal of Substance Abuse Treatment*. 2009; 36:235–243. [PubMed: 18715743]
- Franklin TR, Lohoff FW, Wang Z, Sciortino N, Harper D, Li Y, Childress AR. DAT genotype modulates brain and behavioral responses elicited by cigarette cues. *Neuropsychopharmacology*. 2009; 34:717–728. [PubMed: 18704100]
- Furberg H, Kim Y, Dackor J, Boerwinkle E, Franceschini N, Ardissino D, Sullivan PF. Genome-wide meta-analyses identify multiple loci associated with smoking behavior. *Nature Genetics*. 2010; 42:441–447. [PubMed: 20418890]
- Gray KM, DeSantis SM, Carpenter MJ, Saladin ME, LaRowe SD, Upadhyaya HP. Menstrual cycle and cue-reactivity in women smokers. *Nicotine and Tobacco Research*. 2010; 12:174–178. [PubMed: 19996146]
- Heatherton TF, Kozlowski LT, Frecker RC, Fagerstrom KO. The Fagerstrom Test for Nicotine Dependence: A revision of the Fagerstrom Tolerance Questionnaire. *British Journal of Addiction*. 1991; 86:1119–1127. [PubMed: 1932883]
- Heishman SJ, Lee DC, Taylor RC, Singleton EG. Prolonged duration of craving, mood, and autonomic responses elicited by cues and imagery in smokers: Effects of tobacco deprivation and sex. *Experimental and Clinical Psychopharmacology*. 2010; 18:245–256. [PubMed: 20545389]
- Hodgkinson CA, Yuan Q, Xu K, Shen PH, Heinz E, Lobos EA, Goldman D. Addictions biology: Haplotype-based analysis for 130 candidate genes on a single array. *Alcohol and Alcoholism*. 2008; 43:505–515. [PubMed: 18477577]
- Hutchison KE, LaChance H, Niaura R, Bryan A, Smolen A. The DRD4 VNTR polymorphism influences reactivity to smoking cues. *Journal of Abnormal Psychology*. 2002; 111:134–143. [PubMed: 11866166]
- Kassel JD, Stroud LR, Paronis CA. Smoking, stress, and negative affect: Correlation, causation, and context across stages of smoking. *Psychological Bulletin*. 2003; 129:270–304. [PubMed: 12696841]
- Kozlowski LT, Mann RE, Wilkinson DA, Poulos CX. “Cravings” are ambiguous: Ask about urges or desires. *Addictive Behaviors*. 1989; 14:443–445. [PubMed: 2782125]
- Kozlowski LT, Wilkinson DA. Use and misuse of the concept of craving by alcohol, tobacco, and drug researchers. *British Journal of Addiction*. 1987; 82:31–36. [PubMed: 3470042]
- Maude-Griffin P, Tiffany ST. Production of smoking urges through imagery: The impact of affect and smoking abstinence. *Experimental and Clinical Psychopharmacology*. 1996; 4:198–208.
- Molander A, Soderpalm B. Glycine receptors regulate dopamine release in the rat nucleus accumbens. *Alcoholism: Clinical and Experimental Research*. 2005; 29:17–26.
- Office of the Surgeon General. The health consequences of smoking: A report of the Surgeon General. Office on smoking and health. Atlanta, GA: Centers for Disease Control and Prevention; 2004.
- Ray R, Tyndale RF, Lerman C. Nicotine dependence pharmacogenetics: Role of genetic variation in nicotine-metabolizing enzymes. *Journal of Neurogenetics*. 2009; 23:252–261. [PubMed: 19169923]
- Sayette MA, Griffin KM, Sayers WM. Counterbalancing in smoking cue research: A critical analysis. *Nicotine and Tobacco Research*. 2010; 12:1068–1079. [PubMed: 20884695]
- Shiffman S, Waters AJ. Negative affect and smoking lapses: A prospective analysis. *Journal of Consulting and Clinical Psychology*. 2004; 72:192–201. [PubMed: 15065954]
- Soderpalm B, Lof E, Ericson M. Mechanistic studies of ethanol’s interaction with the mesolimbic dopamine reward system. *Pharmacopsychiatry*. 2009; 42(Suppl 1):S87–S94. [PubMed: 19434560]
- Stewart J. Review. Psychological and neural mechanisms of relapse. *Philosophical Transactions of the Royal Society of London, B: Biological Sciences*. 2008; 363:3147–3158.
- Tiffany ST. A cognitive model of drug urges and drug-use behavior—Role of automatic and nonautomatic processes. *Psychological Review*. 1990; 97:147–168. [PubMed: 2186423]
- Ungless MA, Singh V, Crowder TL, Yaka R, Ron D, Bonci A. Corticotropin-releasing factor requires CRF binding protein to potentiate NMDA receptors via CRF receptor 2 in dopamine neurons. *Neuron*. 2003; 39:401–407. [PubMed: 12895416]

- Wang B, You ZB, Rice KC, Wise RA. Stress-induced relapse to cocaine seeking: Roles for the CRF(2) receptor and CRF-binding protein in the ventral tegmental area of the rat. *Psychopharmacology*. 2007; 193:283–294. [PubMed: 17437087]
- Williams SM, Haines JL. Correcting away the hidden heritability. *Annals of Human Genetics*. 2011; 75:348–350. [PubMed: 21488852]
- Wray NR, Goddard ME, Visscher PM. Prediction of individual genetic risk to disease from genome-wide association studies. *Genome Research*. 2007; 17:1520–1528. [PubMed: 17785532]
- Wray NR, Goddard ME, Visscher PM. Prediction of individual genetic risk of complex disease. *Current Opinion in Genetics and Development*. 2008; 18:257–263. [PubMed: 18682292]
- Ziegler A, König IR, Thompson JR. Biostatistical aspects of genome-wide association studies. *Biometrical Journal*. 2008; 50:8–28. [PubMed: 18217698]

Appendix

Genes Included in the Analyses (see Hodgkinson et al., 2008, for a complete list of SNPs)

<u>Adrenergic</u>	<u>GABA</u>	<u>Opioid</u>	<u>Signal Transduction (cont'd)</u>
<i>ADRA1A</i>	<i>GABRA2</i>	<i>OPRM1</i>	<i>JUN</i>
<i>ADRA2A</i>	<i>GABRA3</i>	<i>OPRD1</i>	<i>MAPK1</i>
<i>ADRA2B</i>	<i>GABRA4</i>	<i>OPRK1</i>	<i>MAPK3</i>
<i>ADRA2C</i>	<i>GABRA6</i>	<i>OPRL1</i>	<i>MAPK14</i>
<i>ADRB2</i>	<i>GABRB1</i>	<i>PDYN</i>	<i>MPDZ</i>
<i>ARRB2</i>	<i>GABRB2</i>	<i>PENK</i>	<i>NGFB</i>
<i>SLC6A2</i>	<i>GABRB3</i>	<i>PNOC</i>	<i>NTRK2</i>
<i>DBH</i>	<i>GABRD</i>	<i>POMC</i>	<i>NTSR1</i>
	<i>GABRE</i>		<i>NTSR2</i>
<u>Cannabinoid</u>	<i>GABRG2</i>	<u>Serotonin</u>	<i>PPP1R1B</i>
	<i>GABRG3</i>		<i>PRKCE</i>
<i>CNR1</i>	<i>GABRQ</i>	<i>HTR1A</i>	
<i>FAAH</i>	<i>SLC6A7</i>	<i>HTR1B</i>	<u>Stress</u>
	<i>SLC6A11</i>	<i>HTR2A</i>	
<u>Cholinergic</u>	<i>SLC6A13</i>	<i>HTR2B</i>	<i>CRH</i>
	<i>SLC32A1</i>	<i>HTR2C</i>	<i>CRHBP</i>
<i>CHRM1</i>	<i>GAD1</i>	<i>HTR3A</i>	<i>CRHR1</i>
<i>CHRM2</i>	<i>GAD2</i>	<i>HTR3B</i>	<i>CRHR2</i>
<i>CHRM3</i>	<i>DBI</i>	<i>MAOA</i>	<i>GAL</i>
<i>CHRM4</i>		<i>MAOB</i>	<i>NPY</i>
<i>CHRM5</i>	<u>Glycine</u>	<i>SLC6A4</i>	<i>NPY1R</i>
<i>CHRNA4</i>		<i>TPH1</i>	<i>NPY2R</i>
<i>CHRNA2</i>	<i>GLRA1</i>	<i>TPH2</i>	<i>NPY5R</i>
	<i>GLRA2</i>		
<u>Dopamine</u>	<i>GLRB</i>	<u>Signal Transduction</u>	<u>Other</u>
	<i>GPHN</i>		
<i>COMT</i>		<i>ADCY7</i>	<i>BDNF</i>
<i>DDC</i>	<u>NMDA</u>	<i>AVPR1A</i>	<i>CART</i>
<i>DRD1</i>		<i>AVPR1B</i>	<i>CCK</i>
<i>DRD2</i>	<i>GRIK1</i>	<i>CDK5R1</i>	<i>CCKAR</i>

<i>DRD3</i>	<i>GRIN1</i>	<i>CREB1</i>	<i>CCKBR</i>
<i>DRD4</i>	<i>GRIN2A</i>	<i>CSNK1E</i>	<i>CLOCK</i>
<i>DRD5</i>	<i>GRIN2B</i>	<i>FEV</i>	<i>HCRT</i>
<i>SLC18A2</i>	<i>GRIN2C</i>	<i>FOS</i>	<i>LEP</i>
<i>SLC6A3</i>	<i>GRM1</i>	<i>FOSL1</i>	<i>OXT</i>
<i>TH</i>		<i>FOSL2</i>	<i>NR3C1</i>
		<i>GSK3B</i>	<i>SLC29A1</i>
			<i>TAC</i>

Table 1

Thirty-Two SNPs Excluded From Analyses Due to Lack of Allelic Variation

SNP	Gene	SNP	Gene	SNP	Gene	SNP	Gene
rs10266026	<i>CHRM2</i>	rs1800443	<i>DRD4</i>	rs3218737	<i>COMT</i>	rs3891068	<i>AVPR1B</i>
rs1058576	<i>HTR2A</i>	rs1805066	<i>SLC6A2</i>	rs3732791	<i>DRD3</i>	rs5319	<i>DBH</i>
rs11575377	<i>DDC</i>	rs2071644	<i>GRIN1</i>	rs3748814	<i>JUN</i>	rs5327	<i>DBH</i>
rs11575856	<i>OPRM1</i>	rs2179008	<i>GABRA3</i>	rs3762836	<i>CLOCK</i>	rs5566	<i>SLC6A2</i>
rs12658894	<i>GABRA6</i>	rs2227852	<i>DRD5</i>	rs3774395	<i>CCK</i>	rs6352	<i>SLC6A4</i>
rs1333505	<i>SLC6A2</i>	rs2228669	<i>HTR2C</i>	rs3891059	<i>AVPR1B</i>	rs8192492	<i>CRHR2</i>
rs1554817	Control *	rs22267717	<i>CRHR2</i>	rs3891062	<i>AVPR1B</i>	rs852982	<i>NR3C1</i>
rs17110563	<i>TPH2</i>	rs3025417	<i>DBH</i>	rs3891064	<i>AVPR1B</i>	rs9333311	<i>CDK5R1</i>

* ancestry informative marker.

Table 2
SNPs Predicting Elevated Cue-Induced Craving at the $p < .01$ Level (D = Minor allele; HWE = Hardy-Weinberg Equilibrium)

SNP	GENE	GENOTYPE			MINOR ALLELE (D) FREQ.	CORRELATION/ P-VALUE	HWE
		DD	Dd	dd			
rs220573	<i>GRIN2B</i> ^a	50	87	73	0.445	−0.233/0.0008	YES
rs2964608	<i>GLRA1</i> ^b	41	107	62	0.450	0.229/0.001	YES
rs2053736	<i>SLC6A13</i> ^c	5	22	183	0.076	0.216/0.002	NO
rs11055612	<i>GRIN2B</i> ^a	52	105	53	0.498	−0.208/0.003	YES
rs2220159	<i>TPH2</i> ^d	30	83	97	0.340	−0.203/0.004	YES
rs10877970	<i>AVPR1A</i> ^e	39	78	93	0.371	0.203/0.004	NO
rs6630716	<i>GLRA2</i> ^b	23	41	146	0.207	−0.201/0.004	NO
rs2854030	<i>CCKAR</i> ^f	36	96	78	0.400	0.191/0.007	YES
rs860573	<i>FEV</i> ^e	30	78	101	0.330	−0.190/0.007	YES
rs595565	<i>CSNK1E</i> ^e	1	28	180	0.072	0.189/0.007	YES
rs5993883	<i>COMT</i> ^g	34	109	67	0.421	0.184/0.009	YES
rs10857299	<i>GLRB</i> ^b	12	66	132	0.214	0.181/0.01	YES
rs4645869	<i>FOS</i> ^e	0	19	190	0.045	0.181/0.01	YES
rs11930311	<i>GLRB</i> ^b	13	78	119	0.248	−0.181/0.01	YES
rs5934186	<i>GLRA2</i> ^b	24	28	158	0.181	0.180/0.01	NO

^aNMDA.

^bGlycine.

^cGABA.

^dSerotonin.

^eSignal Transduction.

^fCholecystokinin.

^gDopamine.

Table 3

SNPs Predicting Elevated Stress-Induced Cigarette Craving at the $p < .01$ Level (D = Minor allele; HWE = Hardy-Weinberg Equilibrium)

SNP	GENE	GENOTYPE				MINOR ALLELE (D) FREQ.	CORRELATION/ P-VALUE	HWE
		DD	Dd	dd	dd			
rs6497676	<i>GRIN2A</i> ^a	53	101	56		0.493	0.241/0.0006	YES
rs9333303	<i>CDK5R</i> ^b	0	3	207		0.007	-0.237/0.0007	YES
rs9497515	<i>GRM1</i> ^a	40	69	101		0.355	0.236/0.0007	NO
rs4412952	<i>GABRB3</i> ^c	24	90	96		0.329	-0.217/0.002	YES
rs9895782	<i>ARRB2</i> ^b	1	13	196		0.036	-0.207/0.002	YES
rs4763361	<i>GRIN2B</i> ^a	6	46	156		0.139	0.201/0.004	YES
rs7202950	<i>GRIN2A</i> ^a	23	89	98		0.321	-0.192/0.006	YES
rs455892	<i>GRIK1</i> ^a	33	73	104		0.331	0.191/0.006	NO
rs10839535	<i>CKKB</i> ^d	11	75	123		0.232	-0.182/0.01	YES
rs3792738	<i>CRHR1</i> ^e	3	39	168		0.107	0.181/0.01	YES
rs2066713	<i>SLC6A4</i> ^f	21	87	103		0.302	0.181/0.01	YES
rs1386488	<i>TPH2</i> ^f	10	89	106		0.266	0.183/0.01	YES
rs8028997	<i>GABRB3</i> ^c	18	93	99		0.307	-0.180/0.01	YES

^aNMDA.

^bSignal Transduction.

^cGABA.

^dCholecystokinin.

^eStress.

^fSerotonin.