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The Effect of Varenicline on the Neural Processing of Fearful Faces and the Subjective Effects of Alcohol in Heavy Drinkers

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Abstract

Background: Pharmacotherapies for alcohol use disorder have been shown to reduce hazardous drinking and improve overall health. The effect sizes for the effectiveness of these medications, however, are small, underscoring the need to expand the range of therapeutics and develop personalized treatment approaches. Recent studies have suggested that varenicline, an $\alpha 4\beta 2$ -nicotinic partial agonist widely used for smoking cessation, can help alcoholics reduce drinking, but the neurocognitive underpinnings of its effectiveness remain largely unexplored.

Methods: In this double-blind study, 32 heavy drinkers were randomized to receive varenicline (2 mg/d) or placebo. After 2 weeks of dosing, participants underwent functional MRI scans, during which they viewed images of faces with either neutral or fearful expressions at baseline and following an intravenous alcohol infusion to a target breath alcohol concentration of 80 mg%. Blood oxygen level-dependent (BOLD) response was analyzed with Analysis of Functional Neuroimaging software. Linear mixed-effects models were used to examine the effects of facial expression (fearful vs. neutral) and medication (placebo vs. varenicline) on BOLD response. The effect of medication on measures of subjective response to alcohol was also examined.

Results: Results indicated a significant facial expression-by-medication interaction in the left amygdala. The groups showed equivalent activation to neutral faces, but, whereas the placebo group showed increased activation to fearful faces, the varenicline group showed no change in activation. Amygdala activation to fearful faces correlated with number of drinks in the previous 90 days and Obsessive Compulsive Drinking Scale scores. There was no effect of varenicline on subjective response to alcohol.

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SUPPORTING INFORMATION

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Conclusions: Our results indicate that varenicline may disrupt amygdala response to fearful faces in heavy drinkers. Further, amygdala activation correlated with alcohol consumption, suggesting that the effects of varenicline may be related to aspects of drinking behavior. These results suggest that amygdala response to fearful faces may be developed as a biomarker of the effectiveness of medications being developed for the treatment of alcohol use disorder.

Keywords

Alcohol; Intravenous Infusion; Heavy Drinkers; Faces Task; Amygdala

MORE THAN 1 in 5 Americans will develop an alcohol use disorder over the course of their lifetime (Grant et al., 2015; Hasin et al., 2007), underscoring the continued need for effective treatment. Comprehensive analyses of treatment studies have shown that pharmacotherapies for alcohol dependence can reduce drinking and relapse rates, but the effect sizes of existing medications are small (Jonas et al., 2014; Rosner et al., 2010). This may be due to the multiple etiologic pathways that can lead to alcoholism and suggests that personalized treatment approaches are warranted, along with the development of new medications (Heilig et al., 2011). One potential medication is varenicline, a partial agonist at the $\alpha_4\beta_2$ nicotinic acetylcholine receptor (nAChR) that is widely prescribed to help people quit smoking (Coe et al., 2005; Kuehn, 2006; Tonstad et al., 2006). Laboratory studies in humans and animals have shown that varenicline can reduce alcohol consumption (McKee et al., 2009; Steensland et al., 2007). Further, several clinical trials have tested whether varenicline can reduce drinking, providing preliminary evidence that it may be an effective treatment (Litten et al., 2013; although see Erwin and Slaton, 2014; Plebani et al., 2013). Better understanding of varenicline's effects in heavy drinkers may help determine how to maximize its benefit as a treatment.

Varenicline putatively reduces smoking through reducing rewarding effects of cigarettes and relieving adverse symptoms such as withdrawal (Jorenby et al., 2006); these effects are thought to arise from varenicline's partial agonism at the nAChR. Given the high comorbidity between smoking and alcohol use disorders (Dani and Harris, 2005), these drugs presumably affect common neural circuitry. For example, evidence shows that alcohol stimulates the nAChR and repeated alcohol use leads to both tolerance at the nAChR and cross-tolerance to nicotine (Davis and de Fiebre, 2006). Thus, the same mechanisms involved in smoking cessation may also contribute to reduction in drinking. A few studies have shown evidence that varenicline reduces neural correlates of reward in response to smoking cues (Franklin et al., 2011; Ray et al., 2015). In heavy drinkers, varenicline increased self-reported control over alcohol-related thoughts and reduced orbitofrontal cortical response to alcohol cues (Schacht et al., 2014). In smokers, some evidence suggests that varenicline relative to placebo improves participants' speed at recognizing emotions in faces and reduces amygdala activity when viewing images of angry or fearful faces (Loughhead et al., 2013), although there is heterogeneity in this effect (Sutherland et al., 2013). This suggests that varenicline affects neural correlates associated with processing negative emotionality in smokers, which corroborates evidence that directly stimulating nAChRs can reduce anxiety in both animals and humans (Elliott et al., 2004; Kassel and Shiffman, 1997; Turner et al., 2011). This may be particularly relevant for heavy drinkers given the high

comorbidity between anxiety and alcohol use disorders (Kushner et al., 1990). To date, however, no study had tested whether varenicline alters neural processing of negative emotional cues in heavy drinkers.

This study assessed the neural correlates of emotional face processing and examined subjective differences in response to alcohol administration between heavy drinkers treated with either placebo or varenicline. Numerous studies have shown that images of fearful faces increase amygdalar activity (Hariri et al., 2002; Yang et al., 2002). This may be important for heavy drinkers, as evidence suggests that greater attention to threat is associated with more alcohol consumption among healthy individuals (Keough and O'Connor, 2014). Further, amygdala activity in response to emotional faces was recently shown to discriminate which depressed patients were most likely to respond to treatment with medication (Williams et al., 2015), suggesting that amygdala response might signal clinically meaningful information. We hypothesized that varenicline, compared to placebo, would attenuate amygdalar response to threatening cues and would reduce the subjective experience of liking and wanting alcohol.

MATERIALS AND METHODS

Participant Characteristics

Heavy drinkers were recruited for this randomized, double-blind, placebo-controlled experimental medicine study, which occurred over the course of 3 weeks. Part of this study had been reported in a previous manuscript, including use of the same subjects (Vatsalya et al., 2015). The study was conducted at the NIH Clinical Center in Bethesda, MD, and all participants gave informed consent prior to participating in the study. Candidates completed a screening visit that included both a clinical and psychiatric evaluation (Structured Clinical Interview for the DSM-IV; First et al., 2002). We used the Fagerström Test for Nicotine Dependence (Fagerstrom, 1978) and 90-day Timeline Followback (TLFB; Sobell and Sobell, 1992) to assess smoking and drinking history, respectively. Participants also completed the Alcohol Use Disorders Identification Test (AUDIT; Babor et al., 2001) and the Self-Rating of the Effects of Alcohol (SRE; Schuckit et al., 1997). The AUDIT assesses recent drinking by measuring consumption, dependence symptoms, and harmful drinking. The SRE quantifies level of response to alcohol. Participants were included if they (i) consumed an average of >20 drinks per week for men and 15 drinks per week for women and (ii) were not seeking help for alcohol-related problems.

Participants were excluded if they met any of the following criteria: (i) lifetime history of Axis I mood, anxiety, or substance use disorders (other than alcohol or nicotine use disorders); (ii) recent or regular use of illicit or nonprescribed psychoactive substances; (iii) history of clinically significant alcohol withdrawal; (iv) lifetime history of violence, suicide attempts, or self-injurious behavior; (v) current or chronic medical conditions, including cardiovascular conditions, requiring inpatient treatment or frequent medical visits; (vi) use of medications contraindicated with varenicline in the past 90 days, or those that may affect the hemodynamic response (e.g., antihypertensives) within the past 30 days, or those that may interact with alcohol within 2 weeks prior to the study; or (vii) metal in body, left-

handedness, or claustrophobia (MRI exclusion criteria). The complete list of inclusion/exclusion criteria is available at clinicaltrials.gov (NCT00695500).

Forty-nine participants were randomized, but 3 failed to return. Nine participants dropped out or failed to comply with study procedures (3 varenicline participants, 6 placebo participants); 5 participants were removed from the final sample due to excessive motion artifacts during imaging (i.e., >20% of imaging repetition times were censored due to motion; 3 varenicline participants, 2 placebo participants). Thirty-two participants were included in the final dataset (varenicline: N = 17, placebo: N = 15). Participants were given medication in pill bottles, and adherence was monitored by counting the pills remaining at each visit.

Study Procedures

Following enrollment and a baseline visit, participants began medication with varenicline or placebo for 3 weeks. The varenicline dose was titrated during the first week (0.5 mg/d for the first 3 days, 1 mg/d for the next 4 days) to a final regimen of 2 mg/d for the remainder of the study (Vatsalya et al., 2015). After 2 weeks of taking medication, participants completed an fMRI session where they underwent scans prior to and following an IV alcohol infusion. Participants were instructed not to drink alcohol in the 48 hours prior to the study session or smoke after entering the scanning facility.

Self-Report Questionnaires

To examine relationships between brain activation and subjective response, several self-report measures were included.

The Drug Effects Questionnaire (DEQ) is one measure of early response to drug effects, which has been used in studies involving oral alcohol (King et al., 2011). The DEQ prompts participants to answer the following questions on a scale of 0 (not at all) to 100 (extremely): (i) Do you feel any drug effects? (ii) Do you feel high? (iii) Do you like the effects you are feeling now? (iv) Would you like more of what you received, right now? (v) Do you feel intoxicated?

The Obsessive Compulsive Drinking Scale (OCDS) is a 14-item questionnaire of the obsessive and compulsive dimensions of alcohol dependence (Anton et al., 1995). Craving and obsessions have shared characteristics; for example, Modell and colleagues (1992) found that those who craved alcohol also reported high obsessional thinking. The same group also identified correlations between compulsive drinking and craving. These findings indicate that obsessions and compulsions may be indicators of problem drinking.

Faces Task

While in the scanner, participants were presented with 45 images of fearful and 45 images of neutral faces obtained from the NimStim set (Tottenham et al., 2009). The faces appeared after a jittered period of time (2 to 14 seconds) in randomized order. Each cue appeared for 2 seconds; participants passively viewed the cues and were not asked to respond. The task took approximately 6 minutes. Following the initial faces run, participants received an IV

infusion of alcohol to achieve a target breath alcohol concentration (BrAC) of 80 mg% over 15 minutes, after which the BrAC was clamped at the target level for an additional 15 min (Ramchandani et al., 1999). Five minutes after achieving the target BrAC, participants were scanned again while the faces task was repeated. Data from the second faces task are not reported due to concerns about alcohol's effects on cerebral blood flow (Marxen et al., 2014; Strang et al., 2015) and order effects because the sober scan always occurred first. Following completion of the scan, the alcohol infusion was stopped, and participants were removed from the scanner. Participants were monitored until their BrAC levels fell below 20 mg% after which they were sent home in a taxi.

Subjective Response to Alcohol Analysis

The subjective response to alcohol was measured via the DEQ. The DEQ was administered immediately before the faces task, and following the conclusion of the alcohol clamp. The difference between baseline and postclamp DEQ scores constituted our measure of the subjective effects of alcohol. Data were analyzed with an analysis of variance (ANOVA) model with medication-type as the between-group variable and DEQ change scores for each question (feeling, wanting more, liking, high, and intoxicated) as the dependent measures. Each model was also tested with age as a covariate as the groups differed on this variable.

fMRI Scanning

The fMRI session was conducted using a General Electric (Fair-field, CT) or Siemens Skyra (Munich, Germany) 3T scanner with a 12 or 20 channel head coil, respectively. Structural scans were also collected for co-registration with functional images using the MPRAGE sequence. The faces task was collected after a modified version of the monetary incentive delay task, which is reported in a separate manuscript (Vatsalya et al., 2015). Two functional runs time-locked to the start of the faces task were acquired using a T2*-EPIRT sequence (T2*-weighted echoplanar imaging; TR = 2,000 ms, TE = 30 ms, FoV = 240 mm, 64 × 64 matrix, 36 axial slices with 0 mm gap, flip angle = 90°, total duration: 6 minute, 20 seconds, 3.75 × 3.75 × 3.8 mm³ voxels) that measured changes in blood oxygen level-dependent (BOLD) contrast.

Imaging Processing

Data were preprocessed using Analysis of Functional Neuro-Images (AFNI) software (Cox, 1996). Echoplanar images were aligned to anatomical images. We inspected data for motion, censoring time points with >0.3 mm motion or 0.3° of rotation relative to the previous time point. Images were spatially smoothed using a 6-mm Gaussian kernel. In the process of aligning the echoplanar images to the anatomical image and warping them to standardized space, voxels were resampled to 3.5 × 3.5 × 3.5 mm³ to generate isotropic voxels of the minimum original dimension, but truncated to 3 significant bits (i.e., 3.75 was truncated to 3.5). Signal for each voxel was scaled by the mean so the average intensity was 100; thus, output could be viewed as percent signal change from baseline. A general linear model fit was performed using AFNI's 3dDeconvolve function, with regressors for alcohol, food, and neutral cues for each phase (cue, target, hit, miss). Six motion parameters were included in the model as regressors of noninterest.

Brain Imaging Analysis

Linear mixed-effects (LME) analyses were conducted using AFNI's 3dLME (Chen et al., 2013). Medication (placebo, varenicline) and face type (fear, neutral) were fixed effects in the model and individual participants were treated as random effects. We also controlled for scanner type to account for the use of 2 different scanners (4 placebo and 5 varenicline were scanned with the Siemens scanner). We also examined the results by scanner to confirm that the results were in the same direction (see Figure S1). LME analysis examined main effect of medication, face type, and medication-by-face-type interaction. Analyses were performed voxel-wise across the entire brain. Volume-threshold adjustment based on Monte Carlo simulations (AFNI's 3dClustSim) was applied to protect family-wise error rate. For a main effect of face type, a more stringent threshold was applied in an attempt to restrict significant clusters to single anatomical regions. Thus, an a priori voxel-wise probability of $p < 0.01$ in a cluster of 1,158 μl (27 voxels) resulted in an a posteriori probability of $p < 0.01$. For medication-by-face-type interaction analyses, which have half the power of a main effect analysis, an a priori voxel-wise probability of $p < 0.05$ in a cluster of 1,402 μl (33 voxels) for the entire brain resulted in an a posteriori probability of $p < 0.05$. Age was tested as a covariate for significant clusters because the groups differed on this variable.

Exploratory Analyses: Drinking Measures and Brain Activity Relationships

We examined whether drinking-related measures correlated with brain activation in response to fearful faces. Specifically, we examined the total number of drinks over the previous 90 days as assessed by the TLFB and the total score on the OCDS. We performed voxel-wise correlations using AFNI's 3dttest++ in a region-of-interest mask of the bilateral amygdala based on a priori hypotheses and the results of the main effect of face-type and the medication-by-face-type interaction results. Volume-threshold adjustment based on Monte Carlo simulations (AFNI's 3dClust-Sim) was applied to protect family-wise error rate. The amygdala mask contained 108 voxels. Based on the simulations, an a priori voxel-wise probability of $p < 0.05$ in a cluster of 3 voxels resulted in an a posteriori probability of $p < 0.05$.

RESULTS

Demographics

The characteristics of participants are shown in Table 1. A *t*-test showed that the groups differed in age (placebo: $\mu = 39.1$, $SD = 13.6$; varenicline: $\mu = 29.5$, $SD = 8.6$; $p = 0.02$). However, as a Shapiro–Wilks test revealed that age was not normally distributed ($W = 0.85$, $p < 0.01$), we also performed a nonparametric analysis. A Mann–Whitney *U*-test showed that the groups did not differ significantly ($p = 0.15$). There were no between-group differences in drinking history, smoking, and physical attributes.

Neuroimaging: Effect of Face-Type and Medication by Face-Type Interaction

Linear mixed-effects analysis revealed that there was a main effect of face type wherein a cluster centered on the left uncus that contained the left amygdala demonstrated greater activation in response to fearful faces relative to neutral faces. A complete list of significant

clusters is available in Table 2. The whole brain analysis examining medication-by-face-type interaction identified a significant cluster containing the left amygdala. As seen in Fig. 1, the placebo group, relative to the varenicline group, showed significantly more amygdalar activation in response to fearful faces, but the groups showed equivalent activation in response to neutral faces. When age was added as a covariate, the medication-by-face-type interaction remained significant, but age was not significantly related to amygdala activity ($p = 0.35$). A complete list of significant clusters is presented in Table 3. The effect remained significant when covarying for scanner type (GE vs. Siemens), but scanner type was not a significant variable in the model, indicating that it did not significantly influence the results.

Subjective Effects of Alcohol

Mean values for each DEQ item at baseline and during the alcohol infusion are reported in Table 4. An ANOVA model revealed no main effect of medication group on change in DEQ measures of feeling ($p = 0.51$), wanting more ($p = 0.13$), liking ($p = 0.11$), high ($p = 0.60$), and intoxicated ($p = 0.43$) from baseline until the end of the alcohol clamp. Age was significantly associated with feeling drug, wanting more, linking, high, and intoxication (all $p < 0.05$), where older individuals reported greater values for each of these measures.

Exploratory Analyses: Brain Activation and Drinking Measures Relationships

Timeline Followback.—As seen in Fig. 2, total drinks (TLFB) during the week prior to the scan were used to measure the relationship between recent drinking and amygdalar activation in response to fearful faces. The natural log (ln) of total drinks was taken to normalize the distribution of this measure. Limbic masking revealed a significant negative correlation between activation of the amygdala bilaterally (right: Brodmann area: 34; $x = 23$, $y = 2$, $z = 16$, Vol = 472 μl ; left: Brodmann area: 28; $x = -20$, $y = -4$, $z = -16$, Vol. = 421 μl) and total drinks consumed (right: $R^2 = 0.19$, $p = 0.01$; left: $R^2 = 0.15$, $p = 0.03$). There was no interaction by medication type ($p = 0.45$).

Obsessive Compulsive Drinking Scale.—There was a significant, negative correlation between activation in the right amygdala (Brodmann area: 34, $x = 24$, $y = -2$, $z = -15$, Vol = 644 μl) in response to fearful faces and the OCDS ($R^2 = 0.31$, $p = .001$). There was no interaction by medication type ($p = 0.71$).

DISCUSSION

This study addressed the hypothesis that chronic varenicline administration would change the neural processing of emotional faces. We show that varenicline administration in heavy drinkers reduces the amygdala response to fearful faces. Further, amygdala activation to fearful faces correlated with drinking measures and indices of obsessive drinking, suggesting that varenicline's effects may be related to aspects of drinking behavior and anxiety. A second goal of this study was to determine whether varenicline reduced subjective response to the acute effects of alcohol. We did not find evidence to support this hypothesis. The reduced activity in the amygdala in response to fearful faces may offer a mechanism that elucidates previous evidence that varenicline can help heavy drinkers reduce their alcohol consumption.

Consistent with previous studies (Hariri et al., 2000, 2002), there was a main effect for participants in this study to show increased amygdalar activation when viewing fearful relative to neutral faces. There was also a medication-by-cue-type interaction in the left amygdala, where participants in the placebo group showed increased activation for fearful relative to neutral faces. This effect was absent for participants in the varenicline group, as they showed similar activity when viewing neutral and fearful faces. This suggests that one of varenicline's effects may be to reduce the magnitude of neural response to emotional stimuli. Given the density of nicotinic AChRs in the amygdala (Wada et al., 1989), it is plausible that varenicline would act here. In a previous study, when participants labeled fearful emotions rather than passively viewing them, they showed more activity in cognitive regions such as medial prefrontal cortex, but reduced amygdala activation (Hariri et al., 2000). Given that one effect of nicotinic agonists is to enhance cognitive functions such as working memory (Levin et al., 2006), varenicline may promote a more cognitive appraisal of fearful faces rather than an emotional one. As stress can trigger alcohol consumption in some individuals (Britton and Bell, 2015), a dampened response to anxiogenic stimuli could be a mechanism for reducing desire to consume alcohol. Amygdala activation correlated with several measures of drinking behavior. Across all participants, individuals with lower amygdala activation reported higher levels of obsessive-compulsive drinking and more alcohol consumption in the previous 90 days. There was no evidence of an interaction with varenicline for any of these relationships, suggesting that they are traits rather than an effect of nicotinic modulation. The correlations between drinking measures and amygdala activation suggest that the faces task may be capturing information relevant to alcohol-related behaviors; however, the direction of these correlations is inconsistent with the direction of the medication effect: individuals with less amygdala activity in response to fearful faces drank more, but the varenicline group showed less activity relative to the control group in response to fearful faces. This discrepancy indicates that further work will be needed to determine how varenicline's effects on neural response to emotional stimuli relate to alcohol consumption.

As previous studies had shown differences between how social and heavy drinking participants responded to fearful faces (Gilman et al., 2012), part of the purpose of this study was to validate that the faces task might be a useful marker of treatment effectiveness for heavy drinkers. The present findings offer mixed support for this hypothesis. The placebo and varenicline groups did not differ on subjective response to alcohol administration, making it difficult to determine varenicline's effectiveness in the present study. Nonetheless, as previous reports have indicated that varenicline can reduce alcohol consumption and craving (Feduccia et al., 2014; Litten et al., 2013; McKee et al., 2009), the present findings—that (i) varenicline diminished differential amygdalar response to emotional faces and that (ii) amygdala activity correlated with drinking measures—indicate that the faces task may help assess medication utility for heavy drinkers.

The placebo and varenicline groups did not differ on subjective response to alcohol administration. Although previous research has shown that nAChR antagonists reduce the stimulating and rewarding effects of alcohol (Gilman et al., 2008, 2012), there has been mixed evidence for the effect nAChR agonists on alcohol response. Some studies have shown that nAChR agonism increases alcohol self-administration and subjective high

(Barrett et al., 2006; Kouri et al., 2004), while others have shown that it leads participants to wait longer before taking a drink (McKee et al., 2008) and reduces subjective high (Ralevski et al., 2012). The absence of a difference in the present study may reflect several factors. First, previous studies showing that nAChR modulation can alter the subjective effects of alcohol used either an antagonist (mecamylamine) or a full agonist (nicotine), whereas varenicline is a partial agonist, and, thus, the effect size may be too small to detect with the sample size in this study. Second, there have been mixed results in the previous studies, suggesting several possibilities: nAChR modulation may not alter the subjective response to alcohol or it may only alter it for select samples. For example, most of the previous research has examined smoking populations, but the present study contained a mix of smokers and nonsmokers. Further, several previous studies have found that nicotine only modulated the subjective response to alcohol in males (Acheson et al., 2006; Kouri et al., 2004), so gender may moderate the effects of nAChR modulation on alcohol response. The absence of an effect of varenicline on the subjective response to alcohol may have resulted because partial agonism of the nAChR does not significantly affect response to alcohol, or it may be due to our sample selection, which included a mix of smoking, nonsmoking, male, and female heavy drinkers. Future studies will need to address whether varenicline modulates subjective response to alcohol in different samples.

This study had several limitations. Participants were scanned at only 1 time point, after 2 weeks on medication, so we cannot assess whether the groups' neural responses during the faces task differed at baseline, that is, prior to varenicline administration. However, the groups were similar on key demographic variables, including drinking history, and because medication was randomized and double-blind, we believe the lack of a baseline scan did not impact our results. Future longitudinal studies are needed help determine how varenicline alters neural processing of emotion across the course of treatment. Further, as our study assessed non-treatment-seeking heavy drinkers, the findings may not generalize to individuals seeking treatment for alcohol use disorder. Lastly, we used pill counts to assess medication adherence, which is a weak measure and introduces the possibility of deception, which cannot be directly assessed without measuring blood levels of varenicline. Future studies should address the generalizability of the present results.

The findings from this study provide initial evidence that a short-term regimen of varenicline can diminish amygdala activity in response to fearful faces in heavy drinkers. This amygdala activity in response to fearful faces may be related to drinking behavior, suggesting that the findings have bearing on varenicline's utility as a therapy for alcohol consumption. Although several considerations apply, these findings support the development of neural markers for the assessment of pharmacotherapies for alcohol use disorders.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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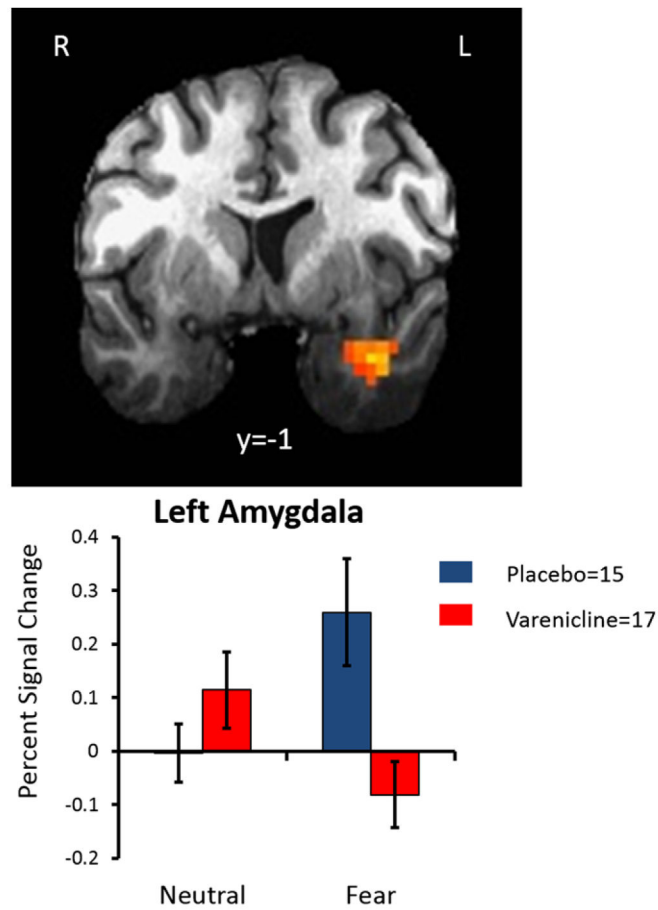


Fig. 1. There was a significant medication group by face-type interaction on blood oxygen level-dependent (BOLD) response in the left amygdala. The placebo group showed significantly more activity in the left amygdala during fearful faces relative to neutral faces, but this effect was not observed in the varenicline group. Error bars represent SEM.

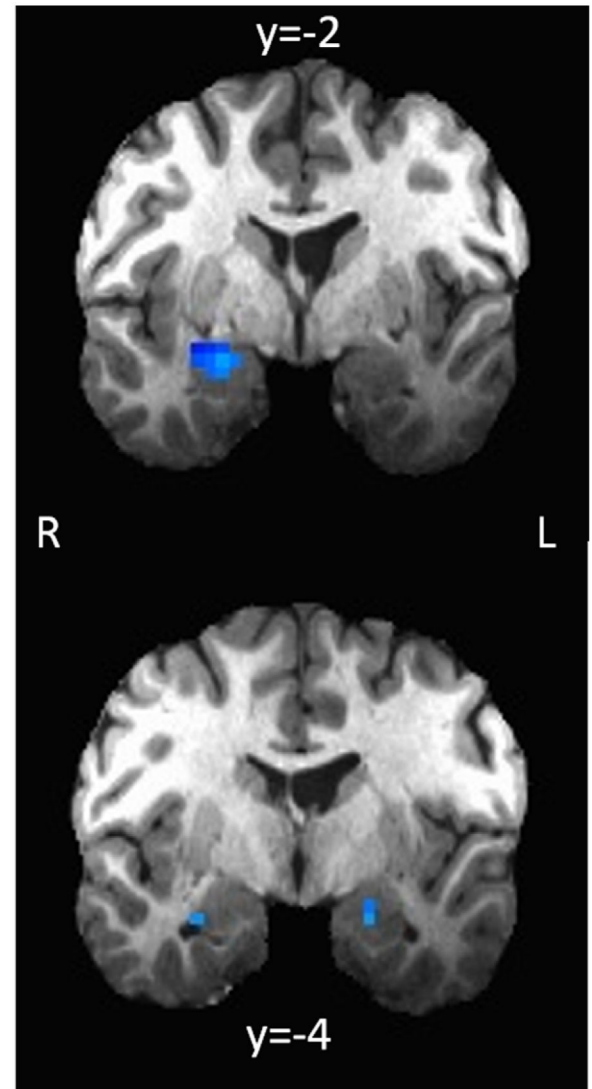
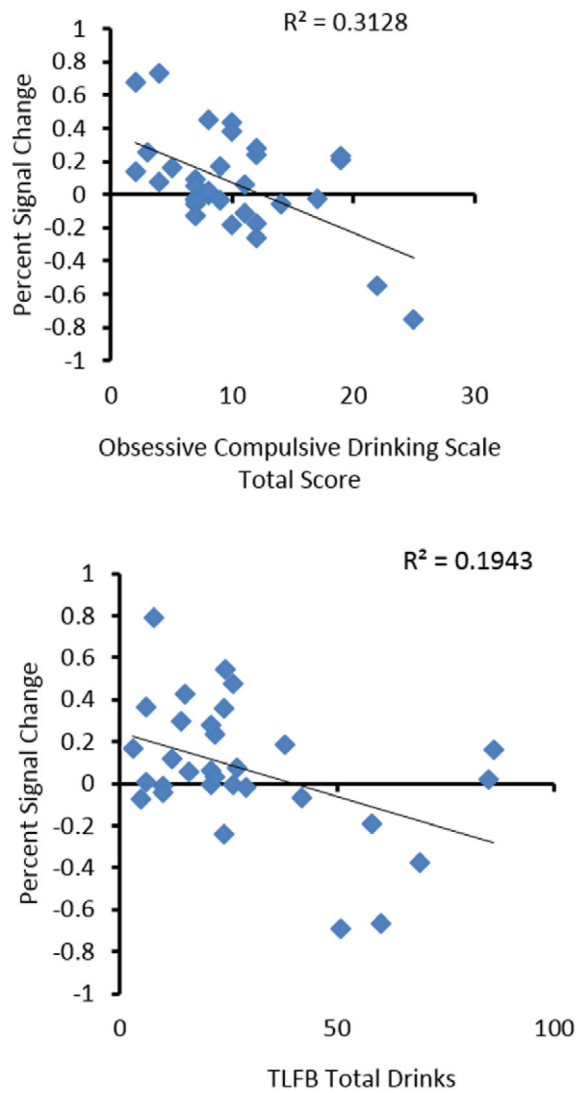


Fig. 2. Amygdala activity when viewing fearful faces was significantly correlated with obsessive compulsive drinking and total drinks in the past 90 days. Individuals with greater amygdala activity had lower levels on these drinking measures.

Table 1.

Participant Characteristics

	Placebo (n = 15) N (%)	Varenicline (n = 17) N (%)
Female	3 (20)	2 (12)
FHP ^a	3 (20)	3 (18)
Current abuse	1 (7)	5 (29)
Current dependence	1 (7)	3 (18)
Current smoker	5 (33)	7 (41)
	Mean (SD)	Mean (SD)
Age ^b (years)	39.1 (13.1)	29.5 (8.4)
Age at first drink (years)	15.6 (4.4)	15.6 (2.0)
90-day Timeline Followback (TLFB)		
Total drinks ^c	449.1 (162.2)	406.4 (201.2)
Drinking days ^c	69.4 (15.5)	67.0 (17.7)
Drinks/drinking day ^c	6.5 (2.2)	6.3 (3.0)
Heavy drinking days ^c	50.7 (22.4)	40.9 (25.2)
Obsessive Compulsive Drinking Scale (OCDS)total ^d	9.9 (5.9)	9.8 (5.1)
OCDS obsessive ^d	3.1 (3.5)	2.9 (2.5)
OCDS compulsive ^d	6.8 (2.8)	6.9 (3.0)
Cigarettes per day (smokers only)	8.2 (4.5)	6.7 (6.5)

The varenicline group was significantly younger than the placebo group as assessed by a *t*-test, but a Shapiro–Wilks test revealed that age was not normally distributed. Therefore, a Mann–Whitney *U*-test was performed, which revealed no significant differences between groups. Based on *t*-tests and chi-square tests, the groups did not differ on any other variable.

^aFamily History Positive for Alcoholism.

^bGroups were significantly different based on a *t*-test. As age was not normally distributed, however, groups were also assessed with a Mann–Whitney *U*-test and did not differ significantly (*p* = 0.15).

^cBased on a 90-day TLFB interview.

^dObsessive Compulsive Drinking Scale.

Table 2.

Main Effect of Face Type. Whole Brain Analysis

Volume (μ l)	x	y	z	L/R	Region	Brodman area
3,216	-18	-94	13	L	Cuneus	18
2,830	-53	28	17	L	Inferior frontal gyrus	46
2,658	-18	-5	-19	L	Uncus/Amygdala	34
2,315	38	55	10	R	Middle frontal gyrus	10
2,187	14	-92	18	R	Cuneus	18
1,801	31	13	-22	R	Superior temporal gyrus	38
1,629	-46	-52	-18	L	Fusiform gyrus	37
1,629	54	-4	2	R	Superior temporal gyrus	22
1,243	51	28	9	R	Inferior frontal gyrus	45
1,157	-61	-19	-8	L	Middle temporal gyrus	21
1,157	2	39	8	R	Anterior cingulate	32

These clusters had a significant main effect of face type (fearful vs. neutral face) with a voxel level $p < 0.01$ and a cluster level $p < 0.01$. Coordinates represent the voxel with the peak activation.

Table 3.

Medication by Face-Type Interaction. Whole Brain Analysis

Volume (μ l)	x	y	z	L/R	Region	Brodmann area
2,615	-46	-54	-17	L	Fusiform gyrus	37
1,544	-26	3	-24	L	Uncus/Amygdala	28
1,501	-12	-96	6	L	Cuneus	17
1,458	12	-94	12	R	Cuneus	18

Group by face-type interaction. These clusters had a significant interaction effect of face type (fearful vs. neutral face) and group (placebo, varenicline) with a voxel level $p < 0.05$ and a cluster level $p < 0.05$. Coordinates represent the voxel with the peak activation.

Table 4.
Subjective Response to Alcohol as Measured by the Drug Effects Questionnaire (DEQ)

	Feel		Like		Want		High		Intoxicated	
	Baseline	Postclamp	Baseline	Postclamp	Baseline	Postclamp	Baseline	Postclamp	Baseline	Postclamp
Placebo Mean (SD)	0.7 (1.2)	2.6 (1.5)	0.8 (1.0)	1.4 (1.5)	0.6 (0.8)	0.9 (1.5)	0.8 (1.4)	2.1 (1.8)	0.7 (1.3)	1.9 (1.9)
Varenicline Mean (SD)	0.4 (0.6)	1.5 (1.1)	0.5 (0.8)	2.3 (1.2)	0.5 (0.9)	1.9 (1.4)	0 (0)	1.1 (0.9)	0.1 (1.3)	2.1 (1.1)

The subjective ratings on the 5 DEQ items are shown at 2 time points: baseline and at the end of the second faces run. Analyses focused on the difference between the timepoints (i.e. the change score) and revealed no significant differences between groups for any of the DEQ items.