Data supplement for Blaine et al., Association of Prefrontal-Striatal Functional Pathology With Alcohol Abstinence Days at Treatment Initiation and Heavy Drinking After Treatment Initiation. Am J Psychiatry (doi: 10.1176/appi.ajp.2020.19070703).

TABLE S1. Relationships between BOLD activation and (1) group membership, (2) length of abstinence, and (3) early treatment outcome

1.1 Activated Regions from Task x Group*	Lat	BA	F	Coordinates			Volume	AUD vs. HC			
within the Cortico-Striatal- Limbic Pathways				X	Y	Z	(mm^3)	S-N	AC-N	N	S-AC
Prefrontal Cortex											
Anterior Cingulate Cortex			8.4504	0	18	-10	976	↓	↓		↓
Ventromedial Prefrontal Cortex	L	11	9.3322	-8	26	-24	726	↓	↓	↑	
Anterior Prefrontal Cortex	R	10	9.4995	6	54	-6	4512	↓			
Dorsolmedial Frontal Cortex	R	8.9	8.1121	36	35	38	623	↓	↓	↑	
Limbic-Striatal											
Globlus Pallidus	L		9.8029	-7	-2	0	321	↓			↓
Caudate, Putamen	L		10.1108	-27	2	14	1547	↓	↓	↓	↑
Nucleus Accumbens, Caudate, Putamen	L,R		9.6814	20	6	-1	1560	↓	↓	↑	↑
Putamen	R		9.2184	32	12	-3	393	↓			
Midbrain											
Thalamus	L,R		8.7628	-14	-32	10	333	↓	↓		↑
1.2 Activated Regions from Task x Abstinence Days*								ROI associations with Shorter Abstinence			
within the Cortico-Striatal- Limbic Pathways								**S-N**	**AC-N**	**N**	**S-AC**
Prefrontal Cortex											
Dorsomedial Prefrontal Cortex	L,R	9	8.234	-37	33	32	566	↓	↓	↑	↓
Ventromedial Prefrontal Cortex	L	10	10.7579	-5	66	-8	3563	↓	↓	↑	
Limbic-Striatal											
Nucleus Accumbens	L		10.82	12	12	10	1033	↓	↓	↑	↑
Hippocampus, Putamen, Insula	L		9.6259	-25	-16	4	3723				↑
Amygdala, Hippocampus			2.2078	-10	-10	-18	1228	↓	↓	↑	↑
Insula	R		10.3684	40	16	2	1139	↓			↑
Caudate, Putamen, Anterior Cingulate Cortex	R		9.8884	42	54	1	11198	↓	↓	↑	↑
Midbrain											
Brainstem	L		8.9546	0	-31	-44	723	↓	↓	↑	
1.3 Activated Regions from Task x Outcome (HDD); Abstinence Days covaried*								ROI associations with Greater Heavy Drinking Days			
within the Cortico-Striatal- Limbic Pathways								**S-N**	**AC-N**	**N**	**S-AC**
Prefrontal Cortex											
Lateral Anterior Prefrontal Cortex	L	10, 46	9.2045	-29	54	26	872	↓		↑	↓
Ventromedial Prefontal Cortex	L	10	9.2519	-6	64	-5	387	↓	↓	↑	
Dorsomedial Prefontal Cortex	L	8,9	8.326	-10	42	26	3963	↓	↓		↓
Limbic-Striatal											
Caudate, Putamen	L		8.95	-16	20	-3	594	↓	↓	↑	↑
Nucleus Accumbens	L		10.2903	-2	20	-18	1626	↓	↓	↑	↑
Hippcampus, Amygdala	R		2.8078	32	-28	-9	772		↓	↑	
Putame, Insula			9.1776	26	16	-12	706	↓			↑
Midbrain											
Thalamus, Hypothalamus	R		9.3306	-8	-17	2	2632		↓		↑

Note: LAT= left or right lateralization. BA= Broadman area. S-N indicates the stress-neutral contrast, AC-N indicates alcohol cue- neutral contrast, N indicates the neutral condition, and S-AC indicates the stress versus alcohol cue condition. All ROIs are significant after whole brain correction at $p<0.001$, and alphasim cluster correction at $p<0.05$. MNI coordinates indicate peak activation for each ROI. Note: In the 1.2 Task x Abstinence Days analysis and Figure 3 of main paper, the VmPFC-rACC activation extends into the lateral ventricle at this threshold.

TABLE S2. Statistical Models Predicting Heavy Drinking Outcome

MODEL	Standardized B	t/z	p
Abstinence Days	-0.12791	-3.131	0.00174**
Craving: Alcohol Cue	0.423	2.035	0.046*
Craving with Abstinence Days			
Craving: Alcohol Cue	0.01617	0.049	0.9612
Abstinence Days	-0.30618	-3.139	0.0026**
Stress: Neutral	0.6952	2.236	0.028889*
Stress with Abstinence Days			
Stress : Neutral	0.09582	0.487	0.62794
Abstinence Days	-0.19633	-3.188	0.00244**
Craving, Stress, with Abstinence Days			
Craving: Alcohol Cue	-0.032	-0.258	0.798
Stress: Neutral	0.034	0.281	0.78
Abstinence Days	-0.459	-3.949	0.0001***
Cortisol: Neutral Recovery- Provocation	0.277	2.308	0.047*
Cortisol with Abstinence Days			
Cortisol: Neutral Recovery- Provocation	0.98	0.807	0.423
Abstinence Days	-0.401	-3.315	0.002**

Note: Craving during the alcohol cue condition, subjective stress during the neutral condition, and cortisol reactivity in the neutral condition were each separately associated with HDD outcome. However, when abstinence days were included in each of these regression models, these relationships became non-significant**.** Note: B = Beta, t/z= t test value or z value, p = probability value.

FIGURE S1. Manipulation Checks: Cortisol and Heart Rate. (a) AUD participants show elevated cortisol response to neutral cues (recovery-provocation) relative to the healthy control group (t[85]=2.29, p<.03) (shown on left), and corresponding lack of response to stress (stress change-neutral change) and to alcohol cues (alcohol cue change – neutral change) relative to healthy controls (HC) (t(85)=2.057, p=0.0427). The HC cortisol responsivity indicates successful elicitation of stress and alcohol cue response (recovery-provocation), while AUD did not show a normal HPA axis response , despite increased subjective stress and increased heart rate (F(1,170)=6.562, p=0.0113). The y axes on both graphs indicates changes in cortisol measured in micrograms per deciliter of whole blood (ug/dl). (b) AUD participants show greater heart rate increase in the neutral condition relative to baseline (t(85)=6.68, p<0.0001). AUD also show lower average heart rate responses to alcohol cues (alcohol cue-neutral average) than HC (F(1,170)=5.013, p=0.0265), but similar average heart rate responses to stress cues (stress-neutral average), (F(1,170)=3.329, p=0.07). The y axis is in beats/minute.

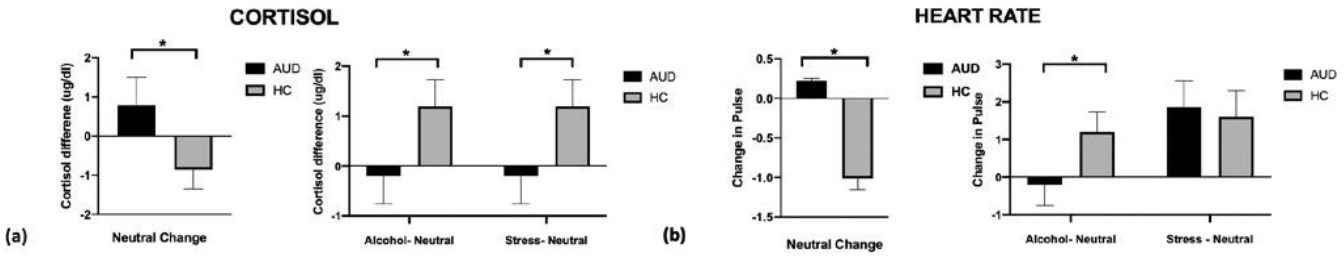
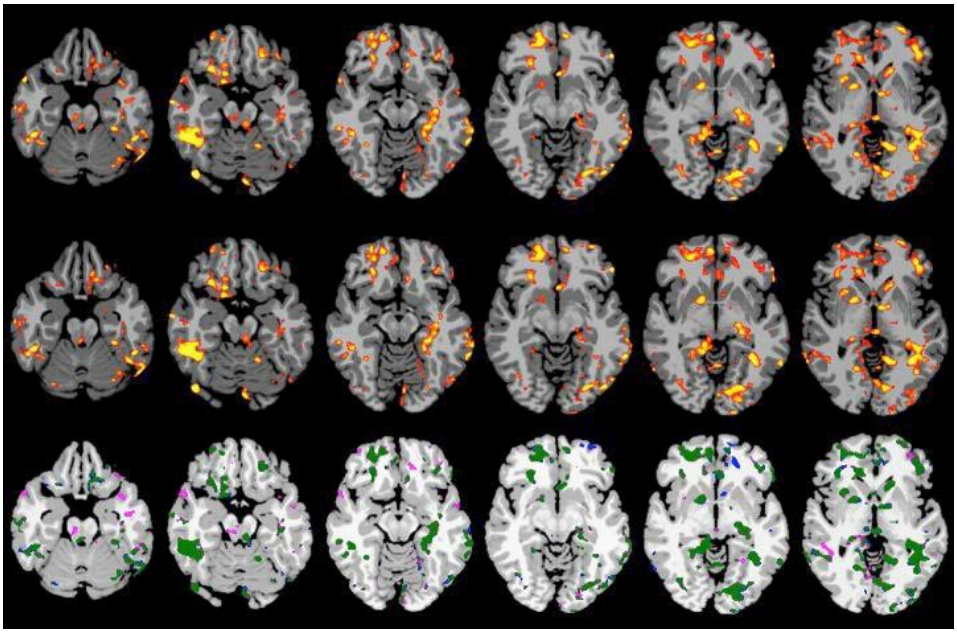
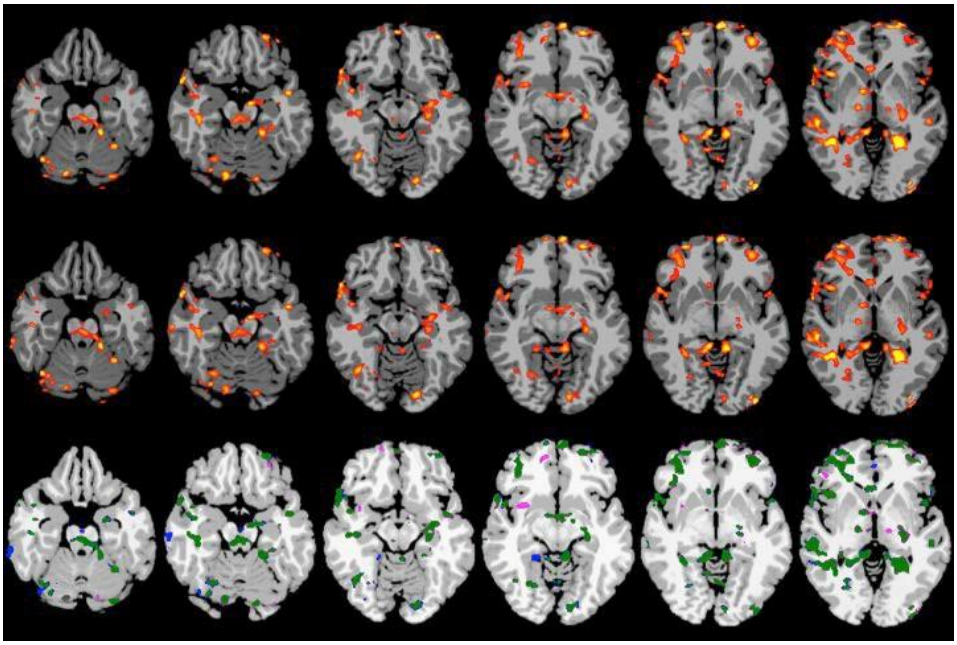


FIGURE S2: fMRI results without and with ARMA Pre-whitening Correction. For each set of images below, row 1 of each column, the non-prewhitened whole brain and cluster corrected results are presented. In row 2, the ARMA prewhitened whole brain and cluster corrected results are presented. In row 3, overlap between the two analyses are shown in GREEN. In Row 3, activation present in the original analysis only (row 1) is shown in pink and activation present only in the ARMA based analysis (Row 2) is shown in blue.

A. Study 1: AUD vs HC: Group X Task Images

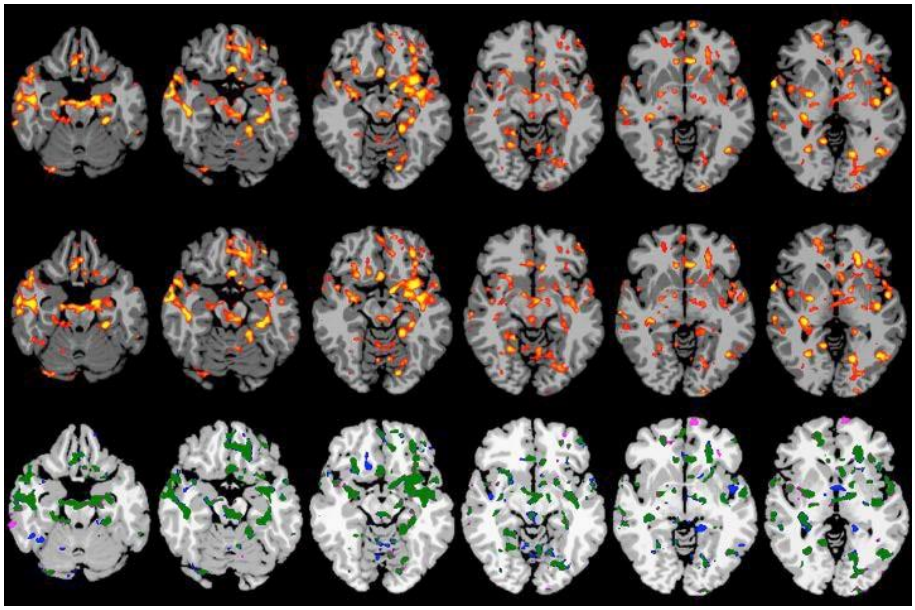


B. Study 2: Task X Abstinence days



(Continued)

C. Study 2: AUD Task X HDD Outcome (Days Abstinence Covaried) Analysis



Supplemental Methods

Cortisol measurement normalization: A number of quality control procedures in cortisol assessment and analysis were implemented. First, all subjects were tested at the same time in the morning with the scan time between 8-10 AM and subject arriving at 7 AM. The IV was inserted at 7:15 AM. We indeed found significant diurnal drops until 8:40 AM, after which we found cortisol levels stabilized during the scanning of the functional response to stress, alcohol cue and neutral blocks between 8:40 and 9:55 AM. Second, as Condition (Stress, alcohol cue and neutral) comparisons were within subjects and condition order was randomized and counterbalanced across subjects, we minimized Between-Subjects cortisol variation for the target outcome of response to each condition. Specific comparisons were conducted at the Within-person levels for Stress and Alcohol Cue relative to Neutral, and Neutral alone condition. Finally, and most importantly, we 'normalized' by adjusting to each subject's own response within each condition, i.e., assessing a change score between

provocation and the recovery timepoint, given the slow and delayed response of Cortisol post-provocation. This allowed us to assess each subjects' response to neutral, stress and alcohol cue images during the post-visual cue recovery relative to provocation, i.e., relative within-subject change for each condition. This careful manipulation allowed for adjusting for any possible Between-person variation during the normal morning diurnal drop in Cortisol, while focusing specifically on the Within-person variation to assess change in Cortisol for each condition relative to the Neutral and also for the active Neutral Control condition.

fMRI Preprocessing: In the fMRI block design of the current studies, the condition data is preprocessed assessing the provocation runs subtracted from baseline fixation runs for each condition, where both baseline and provocation runs are treated exactly the same (same short TR) and without any potential colored noise being subtracted out, and thus, resulting in relative change values per condition and no absolute values being assessed and included. Furthermore, any potential timeseries autocorrelation is also similar across both Groups as well as conditions (Stress, alcohol cue and neutral) within each person's data acquisition, thereby not influencing relative change from baseline data.

Nonetheless, we conducted preprocessing using the AFNI program 3dREMLfit to fit a generalized linear model to the data using a restricted maximum likelihood approach to estimate the temporal auto-correlation structure with an autoregressive moving average (ARMA) as a 'pre-whitening' strategy to address any potential noise due to autocorrelations. As shown in Supplemental Figure 2 above, the second level analysis of group differences in Study 1, and Study 2 prediction models resulted in remarkably similar results to the original analyses presented in the main paper.