Data supplement for White et al., Complementary Features of Attention Bias Modification Therapy and Cognitive-Behavioral Therapy in Pediatric Anxiety Disorders. Am J Psychiatry (doi: 10.1176/appi.ajp.2017.16070847)

SUPPLEMENTAL METHODS

Participants

A total of 58 patients completed the pre-treatment dot-probe task in the magnetic resonance imaging (MRI) scanner. Data from four patients did not meet quality-control criteria (accuracy < 70% n=2; excess movement n=1; technical errors n=1). Twenty-five patients completed a non-fMRI version of the dot-probe task in the clinic prior to treatment. Of the 54 patients with usable pre-assessment fMRI data, 47 had pre-treatment PARS ratings, 45 had mid-treatment PARS ratings, and 40 had post-treatment PARS ratings, and all patients had SCARED data at baseline.

Of the 54 youths assessed with fMRI pre-treatment, 47 were diagnosed with Generalized Anxiety Disorder (GAD) and/or Social Anxiety Disorder (SAD); the seven patients with neither GAD nor SAD had Separation Anxiety Disorder. Other comorbid diagnoses included: Separation Anxiety Disorder (n=18), Specific Phobia (n=21), Attention-Deficit Hyperactivity Disorder (ADHD) (n=9), Selective Mutism (n=3), Tic Disorder (n=2), and Enuresis (n=1).

Of the 54 patients who had baseline fMRI data, 40 completed a post-treatment dot-probe scan. However, data from 5 patients did not meet quality control criteria (accuracy < 70% n=2, excess movement n=3). Twenty-three patients completed the post-treatment dot-probe task in the clinic. Of these 23 patients, three had usable fMRI pre-treatment data for the dot-probe task, and the remaining 20 completed both the pre- and post-treatment dot-probe task in the clinic. A total of 21 patients did not complete a post-treatment dot-probe assessment.

To provide comparison data for the n=54 patients with pre-treatment fMRI data, 51 healthy comparisons were selected from a larger pool of n=62 subjects to create a sample group-matched with patients on IQ, age, and sex, all *p*s >.1. PARS and CGI-I clinician ratings were not assessed for the healthy comparison group. However, baseline symptoms across patients and healthy comparisons were assessed using parent- and child-completed scales (SCARED)(1); the total scores from parent and child ratings were averaged together to provide a total anxiety score. Among these 51 healthy comparisons, 48 had data on the SCARED collected within 6 weeks of the pre-assessment fMRI scan.

Secondary analyses examined regional neural changes across time associated with treatment. To create a matched healthy comparison data set for the n=31 patients with both pre- and post-scan fMRI data, data were assembled from 31 of the 51 healthy comparisons with two dot-probe fMRI assessments, group matched with patients on IQ, age, and sex. Of note, no data reported in the current study from patients appear in prior publications. For the 51 healthy comparisons, some data in a subset of these subjects appear in a prior report on reliability of the dot-probe task (2).

Dot-Probe Pre-processing

On the dot-probe task, RT-based bias scores were calculated using methods from prior research (3,4). For both behavioral and fMRI data, incorrect trials and trials in which RTs were <150 ms or >2000 ms were removed from analyses. Additionally, for each participant, trials with RT >2.5 standard deviations of the mean RT for that condition (Congruent, Incongruent, Neutral) were also removed. RT-based Attention Bias scores were created by subtracting mean RT on Congruent trials from mean RT on Incongruent trials.

Additional Treatment Information

All patients were treated with cognitive behavioral therapy (CBT). CBT treatment followed procedures in the two treatment manuals from the Child and Adolescent Multimodal Study (CAMS), one for patients 13 years-old or younger and the other for patients 14 years-old or older (5,6). Patients were treated by one of two licensed psychologists, both of whom had at least five years of experience using CBT in the treatment of pediatric anxiety disorders. One of these psychologists (EB) had been a supervising CBT therapist in the CAMS study and served as a resource when questions arose about procedures for implementing the CAMS manuals.

Supplemental Data Analyses

The main text highlighted task-based connectivity, specifically the findings that emerged both at baseline for diagnosis and for treatment-related results. The supplement reports on all significant connectivity findings that survived whole-brain correction and corrections for the PFC and insula ROIs. Additionally, the supplemental material reports on significant findings in regional neural activation for the blood-oxygen level dependent response (BOLD). Additionally, ROI analyses that examined task-related differences in baseline amygdala activation between patients and healthy comparisons, as well as in relation to treatment response are reported. These analyses examined differences in the average level of activation in all voxels lying within each of the two anatomically-defined amygdala ROIs.

Finally, a set of three exploratory analyses were examined. First, effects were examined for age and sex on the main interactions of interest. These included baseline brain function differences related to anxiety, overall treatment in patients, and ABMT-specific treatment effects. Second, for both functional connectivity and regional activation, analyses examined

changes in task-based fMRI response before and after treatment in 1) patients and a healthy comparison group and 2) active and placebo AMBT patient groups. To implement the first analysis, pre- and post-treatment imaging data in patients, as well as two scans approximately nine weeks apart in healthy comparisons, were compared using AFNI's 3dLME. Time (Pre, Post) x Condition (Congruent, Incongruent, Neutral) were entered as within subject factors and Group (Patients, Healthy Comparisons) was entered as a between subjects factor. To examine changes across time as a function of ABMT, a similar analysis was conducted within patients, where ABMT Group (Active, Placebo) was substituted as the between subjects factor and PARS ratings were entered as covariates. Third, baseline anxiety differences using a dimensional approach were examined. This fourth set of analyses utilized data in both patients and healthy comparisons, treated as a single group, and examined associations in the combined sample with anxiety using SCARED scores. These analyses utilized data from n=103 participants, as SCARED scores were missing for 2 healthy comparison subjects. These data were subjected AFNI's 3dMVM program with SCARED total scores as a covariate of interest and task condition (Congruent, Incongruent, Neutral) as the within-subject variable. Next, to specifically model associations between specific symptoms of generalized anxiety (GAD) or social anxiety (SAD), similar associations were examined between the two relevant SCARED subscales and brain function. Due to the high correlation ($r = .70$; $p < .001$) between the GAD and SAD subscales and concerns about multi-collinearity, analyses using a single scale (GAD or SAD) are presented

SUPPLEMENTAL RESULTS

Additional findings for pre-treatment anxiety-related differences in amygdala-based connectivity and neural activation

In addition to the significant results reported in the main text, ROI results revealed patients and healthy comparisons also differed in connectivity between the right amygdala and left insula [cluster size = 219 mm^3, peak activation = -36, -14, 14]. No findings with left amygdala or regional activation approached significance.

Additional findings for amygdala-based connectivity and neural activation associated with overall treatment response

See Table S2 for a list of all significant clusters. Additional right-amygdala based findings emerged beyond those reported in the main text. Specifically, treatment response was associated with task-based right amygdala connectivity differences in six additional clusters surviving whole brain correction. These included clusters in the PCC/precuneus and bilateral striatum. An additional cluster in the left insula survived the insula ROI threshold. For left amygdala-based connectivity analysis of treatment response, two clusters in the temporal gyrus survived whole brain correction.

The findings with BOLD signals revealed that treatment response was related to differences in regional activation across task conditions in several regions (see Table S2). Clusters in the dorsolateral prefrontal cortex (dlPFC) and left postcental gyrus survived whole brain correction and a cluster in the right middle frontal gyrus (premotor cortex area) survived the PFC ROI correction.

Additional findings for amygdala-based connectivity and neural activation associated with ABMT-specific treatment response

In addition to the right amygdala–right insula connectivity finding reported in the main text, ABMT-specific response was also associated with right amygdala-left insula connectivity [cluster size = 281mm^3, peak activation = -44, -1, -4].

Pre-treatment amygdala activation

The following analyses examined diagnostic differences in baseline amygdala activation on the dot-probe task. Diagnostic differences for activation in the left amygdala, F(2,206)=3.21, p =.042, reflected a task-related difference in activation for the patient but not the comparison group. Specifically, in patients, amygdala activation was significantly greater in the neutral compared to the incongruent condition, t(53) = 2.45 p=.012; in the comparison group, amygdala activation did not differ among task conditions. Moreover, post-hoc analyses directly contrasted the two groups also showed a trend for the patient group to manifest greater amygdala activation than the comparison group for both the Congruent and Neutral conditions, $t(104)=1.80$, $p=.07$; $t(104)=1.82$, $p=.07$. Finally, the group-by-condition interaction was not statistically significant for the right amygdala, $F(2,206)=2.84$, $p=.06$. Of note, as reported in the main text, the right amygdala is the location where the main between-group connectivity findings emerged.

Exploratory correlation analyses found no relation between task-related activation in the right (*p*s>.10) or left (*p*s>.21) amygdala at baseline and overall patient treatment outcome. Examining each AMBT group separately, neither the active or placebo groups displayed a significant relation between treatment response and baseline activation in the right (*p*s>.13) or left (*p*s>.09) amygdala.

Additional Exploratory Supplemental Analyses

Age and Sex Effects

The significant interactions with age and sex on the main contrasts of interest are presented in Table S1. As briefly noted in the main text, no age-related findings manifested in the right amygdala. However, left amygdala-left insula connectivity differed among age and diagnostic groups at baseline (see Figure S1a) and predicted treatment outcome. For the baseline findings, in patients, age negatively correlated with connectivity on the Attention Bias contrast (Incongruent - Congruent), $r(54)= -.35$, $p=.009$. For healthy comparisons an opposite pattern emerged: age positively correlated with the Attention Bias connectivity contrast, $r(51)=.30$, $p=.03$.

For the treatment-related left amygdala-left insula finding (see Figure S1b), in adolescent patients, higher symptoms after treatment negatively correlated with connectivity on the Attention Bias contrast, $r(16)= -.64$, $p=.004$ (partial correlation controlling for ABMT group and pre-treatment PARS ratings). No such correlation manifested in the younger patient group, $r(16)= .36$, $p=.14$. Of note, left amygdala-left insula was also significant for the Age X ABMT Group X Condition X Treatment Response interaction; however, given the small sample size the four-way interactions are not interpreted. Similarly, four-way interactions emerged with sex (see Table S1), but are also not interpreted. No other interactions with sex emerged.

Differences in amygdala-based connectivity and neural activation before and after treatment

For the analyses that examined differences in brain function across time, no clusters in either the connectivity or regional activation results surpassed any correction thresholds. This

was true for the analyses that examined differences between patients and healthy comparisons, as well as the analyses that examined differences between patients in the active and placebo ABMT groups.

A dimensional approach to examine pre-treatment anxiety-related differences in amygdala-based connectivity and neural activation

Total SCARED Anxiety Scores. The final set of analyses treated patients and comparison youths as a single group and examined associations with levels of anxiety on the SCARED. For task-based functional connectivity with the right-amygdala seed, whole brain corrected analyses revealed associations in the posterior cingulate cortex (PCC)/precuneus [cluster size = 4141 mm^3; peak activation =-1,-64,29] and medial PFC [cluster size=1250 mm^3; peak activation = 6,54,-1]. In ROI-based analyses, level of anxiety on the SCARED also correlated with connectivity in the right insula [cluster size = 234 mm^3; peak activation = 41, -6, 14] and left insula [cluster size = 234 mm^3; peak activation= -34, -14, 16)]. No findings emerged with the connectivity for the left amygdala seed or for the regional activation analyses.

Generalized and Social Anxiety Subscales. Using SCARED subscales, GAD but not SAD symptoms predicted connectivity (see Table S3). Many associations were similar to those seen in both the categorical (patients vs. healthy comparisons) and dimensional approaches (total SCARED scores). For example, there was a large association between levels of GAD symptoms and connectivity between the right amygdala and right insula, as detected with the between group analysis focused on diagnostic status and the analysis of the total SCARED scores. There was also strong amygdala-PCC/precuneus connectivity that resembled that detected with SCARED Total Scores and right amygdala-mPFC connectivity survived the PFC threshold

[cluster size =1000mm^3; peak activation= -6, 54, 11]. There was also amygdala-dACC connectivity that survived whole brain correction, a finding that also emerged between patients and healthy comparisons at baseline [cluster size = 813; peak activation= 9, -9, 41], but failed to surpass the ROI threshold (as many voxels fell outside the mask).

Although no associations between GAD symptoms and regional activation emerged, variation in SAD symptoms was related to activation, generating a large cluster encompassing large portions of the amygdala and adjacent structures (Table S3). There was also a second cluster in the amygdala [cluster size=438 mm^3; peak activation =-24, -4, -21], but it did not survive the study's statistical thresholds. In the larger cluster, the high relative to low social anxiety group showed increased activation on the Neutral condition. The high social anxiety group also showed greater activation on Congruent and Neutral trials relative to Incongruent trials.

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TABLE S1. Regions of Differential Amygdala Connectivity and Activation by Age and Sex on the Dot-Probe Task for all Main Analyses

	Peak TLRC Coordinates (LPI)			Cluster Size in mm^3	Location
	x	y	z		
AGE x CONDITION x ANXIETY GROUP					
Neural Activation Functional Connectivity	-1	23	42	734[b]	dorsomedial PFC
Left Amygdala Seed	51	-14	1	1047[b]	R. Insula/Superior Temporal Gyrus
	46	19	-6	906[b]	R. ventrolateral PFC
	36	6	41	656[b]	R. dorsolateral PFC/Premortor Cortex
	-32	13	1	563[b]	L. Insula
Right Amygdala Seed	-	-	-		*no significant clusters*
AGE x CONDITION x POST-TREATMENT PARS RATINGS					
Neural Activation Functional Connectivity	-	-	-		*no significant clusters*
Left Amygdala Seed	-51	11	11	1391[a]	L. Insula/ventolateral PFC
	31	9	-11	1047[b]	R. Inferior Frontal Gyrus
	-29	14	16	234[b]	L. Insula
Right Amygdala Seed	-39	4	16	406[b]	L. Insula
	46	-21	14	250[b]	R. Insula
AGE x ABMT GROUP x CONDITION x POST-TREATMENT PARS RATINGS					
Neural Activation Functional Connectivity	-	-	-		*no significant clusters*
Left Amygdala Seed	-11	-44	46	4,063[a]	L. Posterior Cingulate Cortex/Precuneus
	-16	-61	24	2,641[a]	L. Precuneus
	-56	-36	31	2,297[a]	L. Inferior Parietal Lobule
	59	-26	19	1,531[a]	R. Postcentral Gyrus
	31	9	-14	1,438[a]	R. Inferior Frontal Gyrus
	59	-36	29	1,422[a]	R. Inferior Parietal Lobule
	-4	36	14	1,281[a]	medial PFC/rostral ACC
	44	16	16	1,172[a]	R. ventrolateral PFC
	-4	14	31	1,078[a]	L. dorsal ACC
	41	24	21	1,031[b]	R. dorsolateral PFC
	-44	-9	9	891[b]	L. Insula

	36	9	1	219 [b]	R. Insula
Right Amygdala Seed	6	41	9	4,953 [a]	medial PFC/rostral ACC
	4	6	-1	1,734[a]	rostral ACC/Caudate
	-24	24	-6	844 [b]	L. Insula/Inferior Frontal Gyrus
	-31	9	-4	281 [b]	L. Insula
	41	-14	6	234 [b]	R. Insula
SEX x ABMT GROUP x CONDITION x POST-TREATMENT PARS RATINGS					
Neural Activation	-18	56	27	1,250 [a]	L. dorsolateral PFC
	-4	36	41	875[b]	dorsomedial PFC
	-26	31	44	797 [b]	L. dorsolateral PFC
	-31	16	3	781 [b]	L. Insula/Clasutrum
Functional Connectivity					
Left Amygdala Seed	-	-	-		*no significant clusters*
Right Amygdala Seed	-	-	-		*no significant clusters*

TLRC = Talairach; ACC= anterior cingulate cortex; PFC = prefrontal cortex; [a] indicates clusters that surpassed the whole brain correction, [b] indicates the findings surpassed the ROI threshold correction (i.e., PFC or Insula); Gender did not interact with any other contrasts of interest

TABLE S2. Regions of Differential Amygdala Connectivity on the Dot-Probe Task that Predict Treatment Response in Anxious Youths

	Peak TLRC Coordinates (LPI)			Cluster Size in mm^3	Location
	x	y	z		
CONDITION x POST-TREATMENT PARS RATINGS					
Neural Activation					
	34	24	41	2,141[a]	R. dorsolateral PFC
	-34	-34	49	1,172[a]	L. Postcentral Gyrus
	41	9	51	1,047[b]	R. Premotor Cortex
Functional Connectivity					
Left Amygdala Seed	-44	-51	21	1,781 [a]	L. Superior Temporal Gyrus
	-61	-44	1	1,656 [a]	L. Middle Temporal Gyrus
Right Amygdala Seed	-1	-69	14	9,203 [a]	L. Posterior Cingulate Cortex
	-29	1	-1	1,906 [a]	L. Striatum/Lentiform Nucleus/Putamen
	54	-24	9	1,859 [a]	R. Insula/Superior Temporal Gyrus
	-49	-14	44	1,578 [a]	L. Postcentral Gyrus
	-41	-54	16	1,438 [a]	L. Superior Temporal Gyrus
	16	-1	54	1,281 [a]	R. Supplemental Motor Area
	21	6	9	1,219[a]	R. Striatum/Lentiform Nucleus/Putamen
	-51	-19	19	641[b]	L. Insula/Postcentral Gyrus

TLRC = Talairach; PFC = prefrontal cortex; [a]indicates clusters that surpassed the whole brain correction, [b]indicates the findings
surpassed the ROI threshold correction (i.e., PFC or Insula)

TABLE S3. Regions of Differential Amygdala Connectivity and Activation as a Function of Generalized and Social Anxiety During the Dot-Probe Task

	Peak TLRC Coordinates (LPI)			Cluster Size in mm^3	Location
	x	**y**	**z**		
CONDITION X GAD SCARED ANXIETY SCORES					
Neural Activation	-	-	-	-	*no significant clusters*
Functional Connectivity					
Left Amygdala Seed	-	-	-	-	*no significant clusters*
Right Amygdala Seed	46	6	14	3,016[a]	R. Insula
	-1	-56	44	2,016 [a]	L. Precuneus
	-41	-71	26	1,406 [a]	L. Midle Temporal Gyrus
	-4	4	36	1,078 [a]	dorsal ACC
	-6	54	11	1,000[b]	medial PFC
	-34	-14	16	484[b]	L. Insula
CONDITION X SAD SCARED ANXIETY SCORES					
Neural Activation	1	-16	-14	2,641 [a]	R. Amygdala/ Red Nucleus
Functional Connectivity					
Left Amygdala Seed	-	-	-	-	*no significant clusters*
Right Amygdala Seed	-	-	-	-	*no significant clusters*

TLRC = Talairach; GAD = Generalized Anxiety; SAD = Social Anxiety Disorder; ACC= anterior cingulate cortex; PFC = prefrontal cortex; [a] indicates clusters that surpassed the whole brain correction, [b] indicates the findings surpassed the ROI threshold correction (i.e., PFC or Insula)

FIGURE S1. Left amygdala-left insula functional connectivity associated age on the Dot-Probe Task. **a.** Age differences between anxious and healthy comparisons across task condition were detected in connectivity between the left amygdala and left insula [cluster size =563 mm^3, peak activation = -32,13,1]. **b**. Treatment analyses also showed age-related effects in left amygdala-left insula connectivity [cluster size= 1391mm^3, peak activation= -51,11,11] that emerged from the Condition-by-Age-by-Post-treatment PARS interaction. Images displayed in radiological convention (left-right).