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Supplemental Information

**Targeted memory reactivation to augment
treatment in post-traumatic stress disorder**

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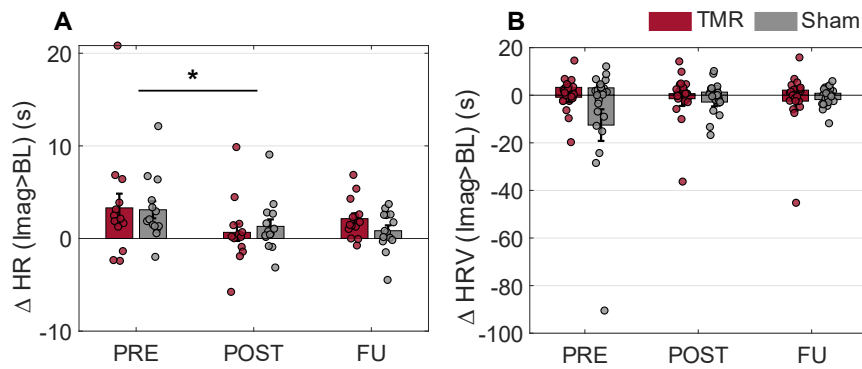


Figure S1. Effect of TMR on heart rate and heart rate variability during script-driven imagery. Related to Figure 3 and STAR Methods.

Autonomic responses were measured with pulse oximetry recording of heart rate (HR) during SDI one day pre-intervention (PRE), one day post-intervention (POST) and one week post-intervention (FOLLOW-UP). Heart rate (A) and heart rate variability (B) were calculated and averaged for the imagine blocks and baseline corrected by subtracting corresponding values of the baseline blocks. To test the effect of TMR on autonomic responses during SDI, we conducted 2x3 factorial ANOVAs with Group (TMR vs Sham) as between subjects factor and Time (PRE vs POST vs FOLLOW-UP) as within subjects factor. Heart rate reduced across groups from pre- to post-treatment (main effect of Time, $F(1.53, 39.83)=3.81$, $p=0.041$). More specific, HR reduced from PRE to POST ($t(27)=2.49$, $p=0.019$), but did not further decrease from POST to FOLLOW-UP. At trend-level, HR remained lower at FOLLOW-UP compared to PRE across groups ($t(27)=1.75$, $p=0.091$). No additive effects of TMR were found for HR. No alterations in HRV across time, nor additive effects of TMR on HRV were found. Error bars represent standard error.

* $p < 0.05$, HR = heart rate, HRV = heart rate variability, Imag > BL = Imagine > Baseline

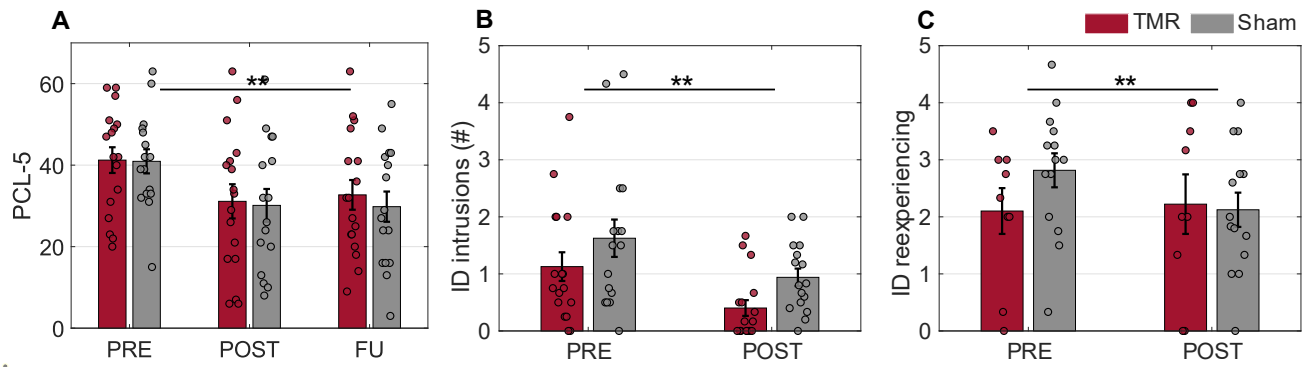


Figure S2. Effect of TMR on PTSD symptoms and daily intrusions. Related to Figure 3 and STAR Methods.

A. Patients self-reported PTSD symptoms one day pre-intervention (PRE), one day post-intervention (POST) and one week post-intervention (FOLLOW-UP) using the PTSD checklist for DSM-5 (PCL-5). A 2X3 ANOVA was performed with Group (TMR vs Sham) as between subjects factor and Time (PRE vs POST vs FOLLOW-UP) as within subjects factor. A main effect of Time was found, showing a reduction of symptoms from pre- to post-intervention ($F(2,62)=20.12, p<0.001$). In more detail, PCL-5 scores reduced from PRE to POST ($t(32)=5.21, p<0.001$). This reduction remained present at FOLLOW-UP relative to PRE ($t(32)=5.08, p<0.001$). PCL-5 symptoms did not further decrease from POST to FOLLOW-UP. No additive effect of TMR on EMDR was found. B. and C. To examine daily intrusions of the targeted traumatic memory, patients kept a diary (integrated in the sleep diary) three days before and one week following intervention. In the diary patients reported the total number of intrusions related to the targeted traumatic memory the previous day, as well as the associated level of subjective distress (the 5 questions of the Re-experiencing subscale of the Responses to Script Driven Imagery Scale (RSDI)). The number of intrusions and subjective distress (mean of 5 Re-experiencing questions) were averaged pre- (PRE), and post-intervention (POST) and compared with a 2X2 ANOVA with Group (TMR vs Sham) as between subjects factor and Time (PRE vs POST) as a within subjects factor. A main effect of Time was found, showing a decrease in the number of intrusions ($F(1,31)=15.93, p<0.001$) and a decrease in the associated level of subjective distress ($F(1, 21)=9.29, p=0.006$) from pre- to post-intervention across groups. No additive effect of TMR was found. Error bars represent standard error.

**** $p < 0.001$, ID = Intrusion Diary, PCL-5 = PTSD checklist for Diagnostic and Statistical Manual of Mental Disorders (DSM- 5)**

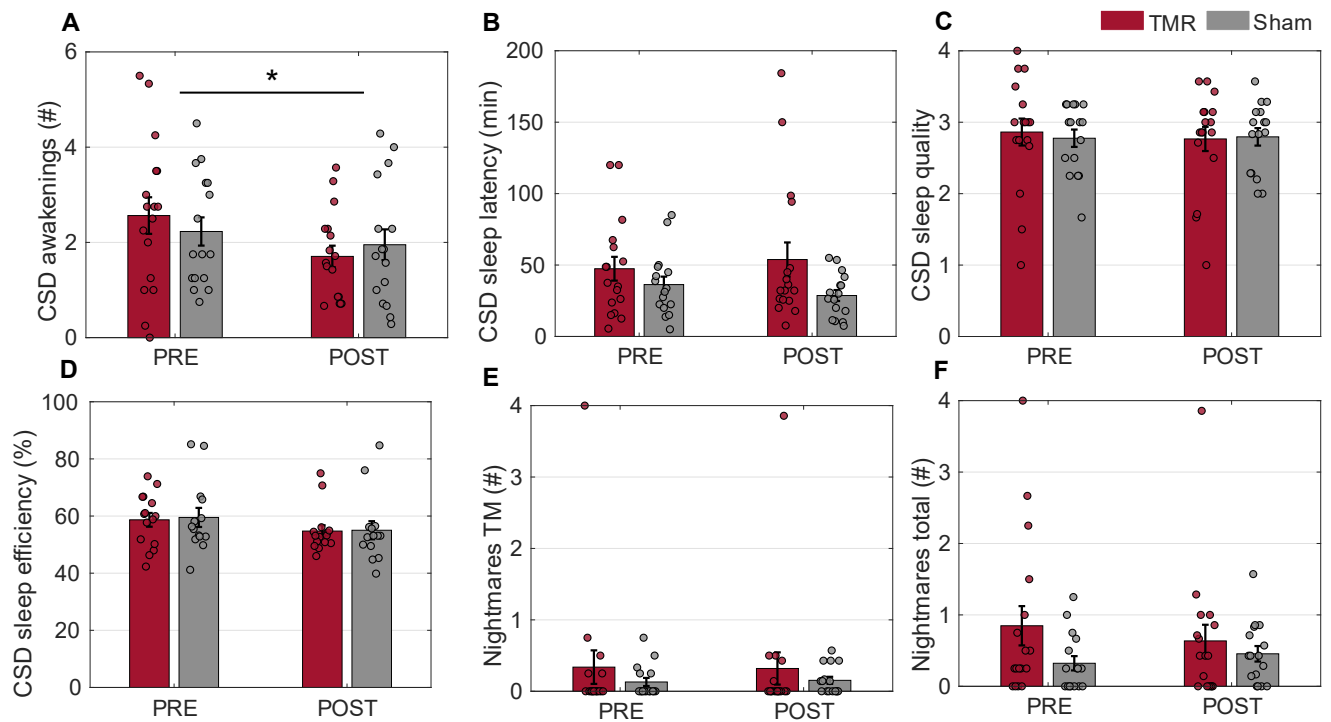


Figure S3. Effect of TMR on sleep diary measures. Related to Figure 3 and STAR Methods.

Diverse subjective sleep measures were evaluated daily by the Consensus Sleep Diary (CSD) from three days pre-intervention (PRE) until one week post-intervention (POST). After being averaged over PRE and POST days, these outcomes were analysed with a 2X2 factorial ANOVA with Group (TMR vs Sham) as between subjects factor and Time (PRE vs POST) as within subject factor. We found that the amount of awakenings decreased from pre- to post-intervention across groups (A, main effect of Time: $F(1,31)=10.92, p=0.002$). No other main effects, nor interaction effects were found for sleep latency (B), sleep quality (C), sleep efficiency (D), nightmares related to the targeted traumatic memory (TM) (E) and nightmares total (F). Error bars represent standard error.

* $p < 0.05$, CSD = Consensus Sleep Diary, # = number of items, # = Number of items, min = Minutes, TM = Target memory

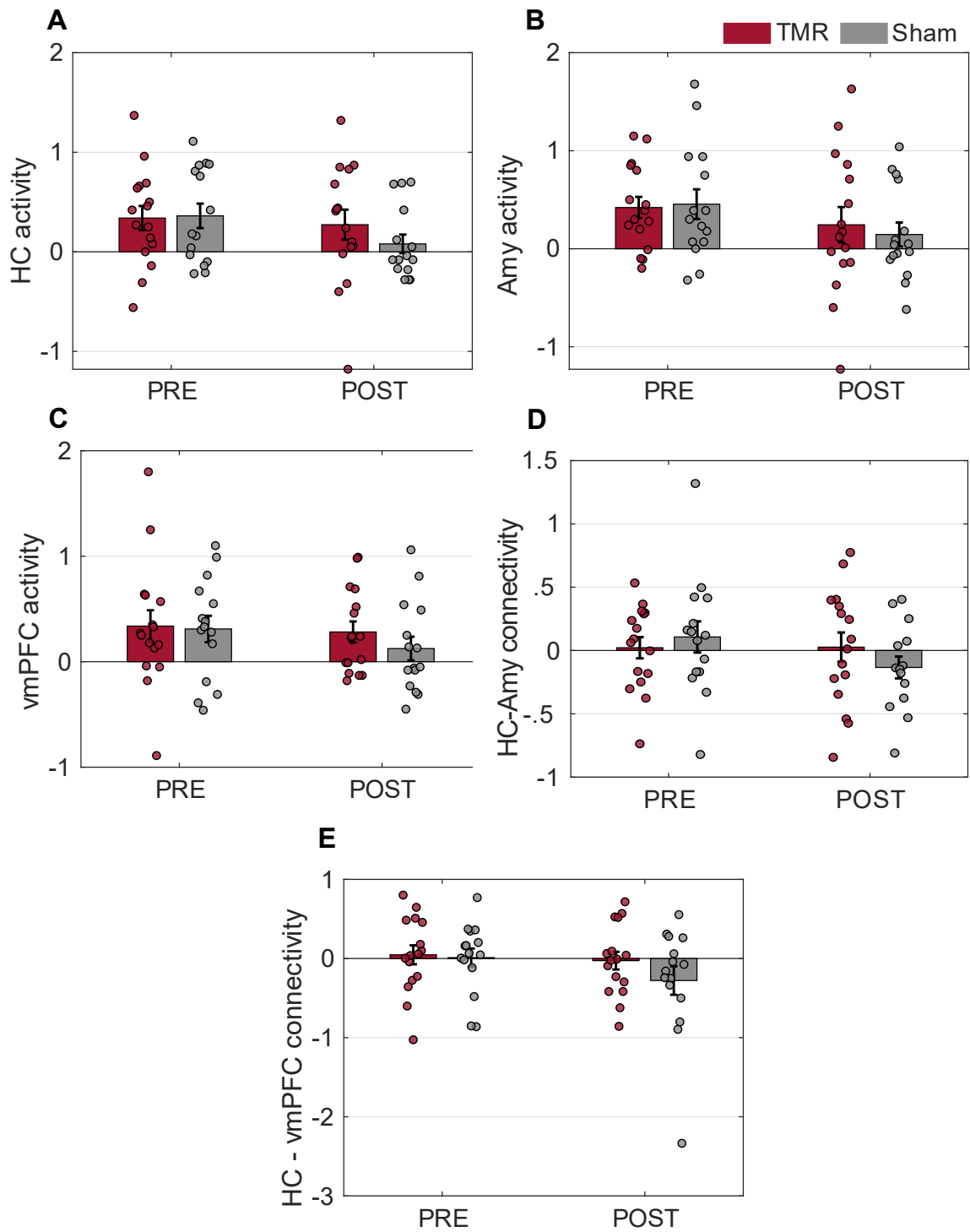


Figure S4. Effect of TMR on fMRI responses to the targeted traumatic memory during script-driven imagery (ROI analysis in SPSS). Related to STAR Methods.

fMRI was obtained during SDI at PRE and POST intervention to visualize the hypothesized enhanced system-level consolidation as a result of TMR. We performed a region of interest (ROI) analysis focusing

on bilateral amygdala, hippocampus and ventromedial PFC, given their documented role in system-level consolidation. We expected both groups to show a reduction in hippocampus and amygdala activation and an increase in vmPFC activation from pre- to post-intervention, representing stronger system-level consolidation and weaker emotional representation of the memory. These effects should then be more pronounced in the TMR group. Beta values representing the imagine versus baseline condition were extracted for these regions using anatomical, AAL-based masks (for amygdala and hippocampus) and a 10 mm sphere around MNI coordinate $x,y,z=0,40,-8$ (for vmPFC^{S1}) and analyzed with SPSS using a 2X2 ANOVA with Group (TMR vs Sham) as between-subjects factor and Time (PRE vs POST) as within-subjects factor (Figure S5A-C). These analyses showed that the amygdala ($t(61)=18.06, p<0.001$), hippocampus ($t(243)=33.51, p<0.001$) and vmPFC ($t(36)=12.81, p<0.001$) were significantly activated during trauma imagery at PRE intervention, indicating involvement of these ROIs in the task. Amygdala activity showed a trend-level reduction across groups from pre- to post-intervention (main effect of time, $F(1,29)=3.53, p=0.070$). No other effects of Time, Group or Group x Time interactions were found. As the neural reorganization accompanying (enhanced) system-level consolidation is best studied using connectivity-based analyses, we performed a generalized form of context-dependent, psychophysiological interaction analysis (gPPI, ^{S2}) with the hippocampus as seed region. Here, we expected both groups to show a reduction in functional coupling between hippocampus and amygdala and hippocampus and vmPFC from pre- to post-intervention, representing stronger system-level consolidation. Again, these effects should then be more pronounced in the TMR group. Results were analyzed with a similar 2X2 ANOVA in SPSS with Group (TMR vs Sham) as between-subjects factor and Time (PRE vs POST) as within-subjects factor. No altered functional coupling of the hippocampus to the amygdala or vmPFC was found as a function of Time, Group or Time x Group interaction (Figure S5D/E). In sum, we did not find evidence of enhanced system-level consolidation as a result of TMR or treatment in general. Error bars represent standard error.

HC = Hippocampus, Amy = Amygdala, vmPFC = ventromedial prefrontal cortex

	Whole night			Cued period		
	TMR	Sham	p	TMR	Sham	p
	M (SD)	M (SD)		M (SD)	M (SD)	
Wake (%)	15.56 (7.49)	11.23 (7.45)	.106	3.98 (3.7)	3.57 (3.14)	.739
N1 (%)	13.19 (4.61)	11.60 (6.97)	.444	8.45 (5.48)	8.78 (8.49)	.896
N2 (%)	34.76 (5.60)	37.48 (6.34)	.202	27.25 (9.98)	29.47 (9.49)	.518
N3 (%)	29.54 (6.92)	28.25 (4.98)	.546	48.63 (15.1)	50.30 (12.7)	.735
REM (%)	22.49 (6.47)	22.65 (6.16)	.941	15.64 (10.43)	11.43 (8.19)	.209
Wake (min)	75.76 (37.31)	57.06 (39.88)	.174	6.55 (6.77)	5.78 (5.34)	.718
N1 (min)	53.14 (16.55)	49.25 (27.71)	.625	13.23 (8.63)	14.34 (14.73)	.792
N2 (min)	143.5 (31.37)	163.75 (36.95)	.099	43.50 (17.9)	46.71 (18.04)	.611
N3 (min)	122.88 (35.63)	123.09 (25.8)	.985	74.32 (18.33)	76.40 (17.23)	.739
REM (min)	93.14 (29.26)	98.59 (28.03)	.590	25.44 (17.63)	17.81 (13.35)	.173
REM latency (min)	136.52 (59.28)	133.34 (59.28)	.878	74.50 (38.36)	85.93 (38.05)	.419
WASO (min)	61.02 (36.93)	39.81 (30.61)	.083	6.55 (6.77)	5.78 (5.34)	.718
TST (min)	412.67 (52.3)	434.68 (46.96)	.214	156.50 (22.99)	155.28(24.77)	.884
Sleep efficiency	83.99 (7.33)	87.24 (8.76)	.256	96.33 (3.71)	96.74 (3.15)	.736
Sleep latency (min)	15.67 (21.09)	17.78 (20.22)	.772	-	-	-

Awakenings (#)	17.11 (6.78)	14.50 (6.35)	.262	4.35 (3.55)	3.81 (3.05)	.644
N2 to Wake (#)	5.47 (4.61)	5.81 (2.58)	.796	1.76 (1.34)	1.31 (1.44)	.360
N3 to Wake (#)	1.64 (1.90)	1.18 (1.42)	.440	0.00 (0.00)	0.00 (0.00)	.825
REM to Wake (#)	1.64 (1.11)	1.43 (1.41)	.638	0.17 (0.39)	0.12 (0.34)	.692
Arousals (#)	121.47 (42.5)	129.06 (59.93)	.676	49.58 (28.12)	52.37 (39.62)	.816
Fragmentation index	24.36 (5.26)	23.96 (7.18)	.856	21.33 (7.03)	23.33 (10.53)	.525
Sleep stage shifts (#)	148.58 (28.75)	156.62 (44.55)	.540	52.29 (19.31)	57.75 (30.52)	.541
Nightmares (#)	0 (1.49)	0 (1.15)	.929	-	-	-
Sleep quality (CSD)	2.71 (0.47)	2.75 (0.683)	.736	-	-	-
SO power (log₁₀ $\mu\text{V}^2/\text{Hz}$)	0.09 (0.002)	0.08 (0.001)	.598	0.087 (0.10)	0.85 (0.10)	.405
Sigma power (log₁₀ $\mu\text{V}^2/\text{Hz}$)	0.19 (0.004)	0.1 (0.006)	.420	0.187 (0.018)	0.179 (0.025)	.345

Table S1. Sleep characteristics of the intervention night. Related to STAR Methods.

During the intervention night, sleep was continuously recorded with polysomnographic EEG- recording. No differences between groups were found for sleep macro architecture measures, including sleep efficiency and continuity (separately for whole night and cued period, i.e. part of the night when TMR was applied). EMDR click presentation did not alter the number of arousals or increase sleep stage shifts from N2 or N3 to wake, indicating that TMR did not disturb sleep continuity. In line with this, subjective sleep quality was not altered by TMR, as evaluated by the Consensus Sleep Diary (CSD). Sleep fragmentation index was calculated as number of awakenings + number of sleep stage shifts divided over the hours asleep^{S3}. Slow oscillation (SO, 0.5-1.5 Hz) and sigma relative power at electrode Fz were calculated over the whole night and cued period separately.

CSD = Consensus Sleep Diary, # = number of items, TST = total sleep time, WASO = Wake after sleep onset, # = Number of items, min = minutes, SO = slow oscillation

Comparison	Region	MNI	K	Z	p (peak)
Main effect of task					
<i>At pre-intervention</i>	R Inferior orbitofrontal gyrus	36, 33, -16	505	6.04	<0.001 ^{a**}
	Superior medial frontal gyrus	0, 54, 24	640	5.97	<0.001 ^{a**}
	Caudate body	0, 0, 10	96	5.25	0.002 ^{a*}
	L Insula	-27, 24, 4	61	5.23	0.002 ^{a*}
	R Amygdala	30, 0, -16	76	5.17	0.002 ^{a*}
	L Fusiform gyrus	-45, -54, -16	25	5.13	0.003 ^{a*}
	R Substantia nigra	9, -15, -9	27	5.12	0.003 ^{a*}
	R Precentral gyrus	39, -12, 50	36	5.05	0.004 ^{a*}
	R Cerebellum	48, -63, 20	21	5.02	0.005 ^{a*}
	L Lingual gyrus	-9, -81, -13	18	4.97	0.006 ^{a*}
	L Putamen	-27, 3, 1	17	4.91	0.007 ^{a*}
	L Middle frontal gyrus	-39, 12, 47	15	4.85	0.010 ^{a*}
	R Calcarine sulcus	24, -60, 17	11	4.84	0.010 ^{a*}
	R Fusiform gyrus	36, -36, -26	11	4.82	0.010 ^{a*}
Main effect of Time					
<i>PRE > POST</i>	No regions	-	-	-	
<i>POST > PRE</i>	No regions	-	-	-	
Main effect of Group					
<i>TMR > Sham</i>	No regions	-	-	-	
<i>Sham > TMR</i>	No regions	-	-	-	
Group x Time interaction					
	No regions	-	-	-	

Table S2. Whole-brain activation patterns in SPM during script-driven imagery of the targeted traumatic memory. Related to STAR Methods.

In addition to the ROI analysis, we performed an exploratory whole brain analysis in SPM using a similar 2X2 ANOVA with Group (TMR vs Sham) as between-subjects factor and Time (PRE vs POST) as within-subjects factor. At the whole brain level, several regions showed involvement in the task at pre-intervention, including right amygdala, left insula and bilateral fusiform gyrus ($p < 0.05$ FWE-corrected, main effect of task). No effects of Time, Group or Group x Time interactions were found.

* $p < 0.05$, ** $p < 0.001$, a = Family wise error-corrected, L = left, R = right, MNI = MNI coordinates in mm, K = cluster size in voxels.

Supplemental References

- S1. Hayes, J.P., Hayes, S.M., and Mikedis, A.M. (2012). Quantitative meta-analysis of neural activity in posttraumatic stress disorder. *Biology of Mood & Anxiety Disorders* 2. <https://doi.org/10.1186/2045-5380-2-9>.
- S2. McLaren, D.G., Ries, M.L., Xu, G., and Johnson, S.C. (2012). A generalized form of context-dependent psychophysiological interactions (gPPI): A comparison to standard approaches. *Neuroimage* 61, 1277–1286. <https://doi.org/10.1016/J.NEUROIMAGE.2012.03.068>.
- S3. Haba-Rubio, J., Darbellay, G., Herrmann, F.R., Frey, J.G., Fernandes, A., Vesin, J.M., Thiran, J.P., and Tschopp, J.M. (2005). Obstructive sleep apnea syndrome: effect of respiratory events and arousal on pulsewave amplitude measured by photoplethysmography in NREM sleep. *Sleep Breath* 9, 73–81. <https://doi.org/10.1007/s11325-005-0017-y>.