

Supplemental Material

Research Articles | Neurobiology of Disease

Environmental enrichment preserves retrosplenial parvalbumin density and cognitive function in female 5xFAD mice

<https://doi.org/10.1523/JNEUROSCI.0455-25.2026>

Received: 4 March 2025

Revised: 21 November 2025

Accepted: 5 January 2026

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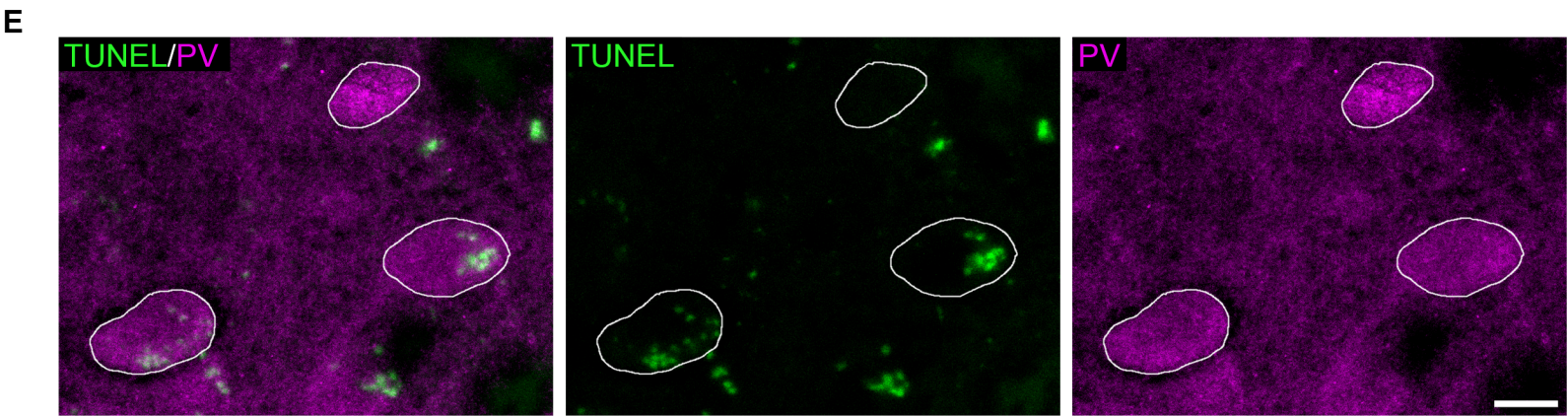
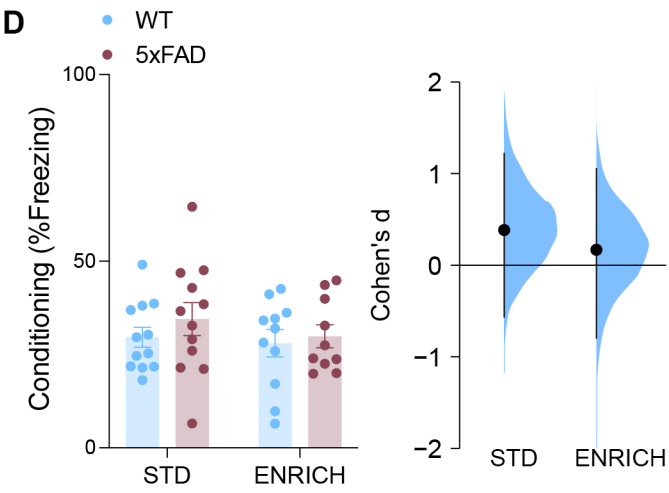
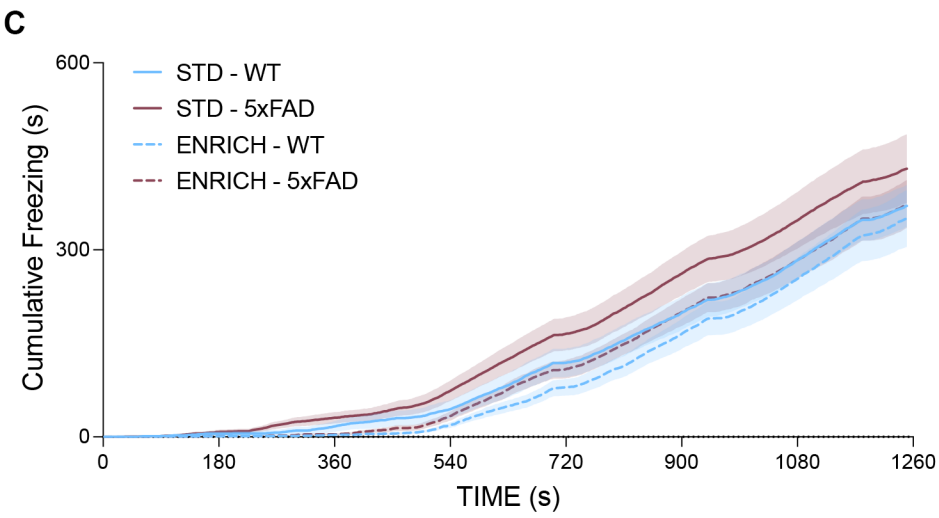
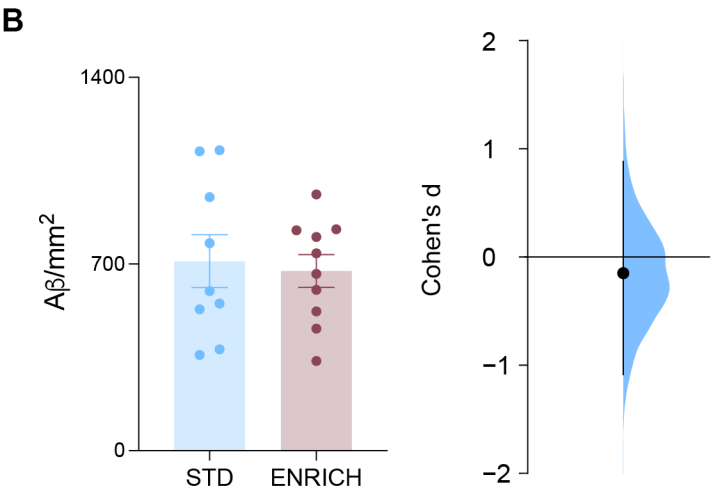
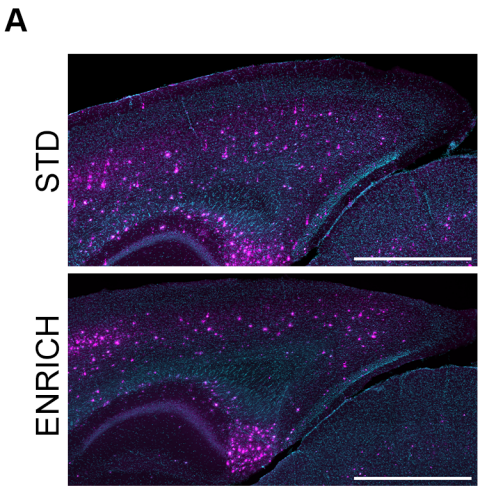


FIGURE S1. Housing conditions do not alter RSC amyloid pathology or behaviour during contextual conditioning.

A Representative photomicrographs of amyloid- β plaque accumulation across the RSC. Scale bars represent 1000 μ m. **B** Density of amyloid- β plaques in the RSC. The density of amyloid- β plaques was not significantly different between 5xFAD mice housed under standard conditions or enriched conditions (n : STD=9; ENRICH=10, two-sample t test, $t_{(17)} = 0.3210$, $P = 0.7521$). Effect size between mice housed in control and enriched conditions (Cohen's $D = 0.745$). **C** Cumulative time spent freezing during the conditioning trial of the trace fear conditioning task. n : WT STD=12; 5xFAD STD=12; WT ENRICH=11; 5xFAD ENRICH=10. **D** Percentage of time spent freezing during the conditioning trial (n : WT STD=12; 5xFAD STD=12; WT ENRICH=11; 5xFAD ENRICH=10). Effect size between WT and 5xFAD mice across control (Cohen's $D = 0.385$) and enriched (Cohen's $D = 0.169$) conditions. **E** Representative photomicrographs of TUNEL staining in RSC PV-INs of 5xFAD mice. Scale bar represents 10 μ m. Data represent mean $\pm$ SEM. All statistical comparisons have been provided as Data S1.

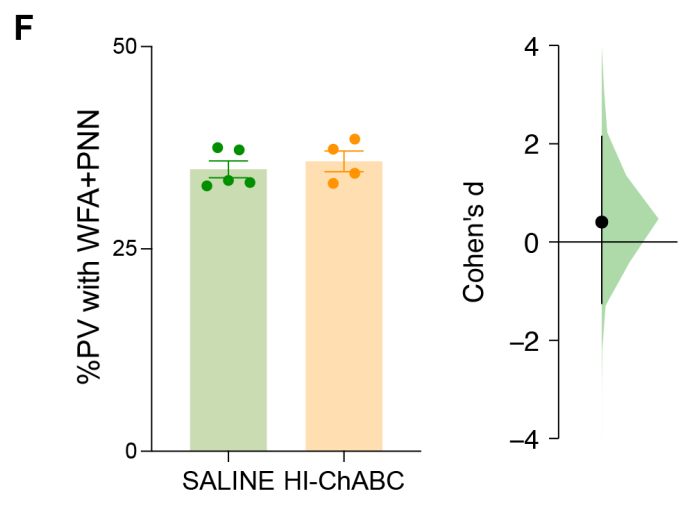
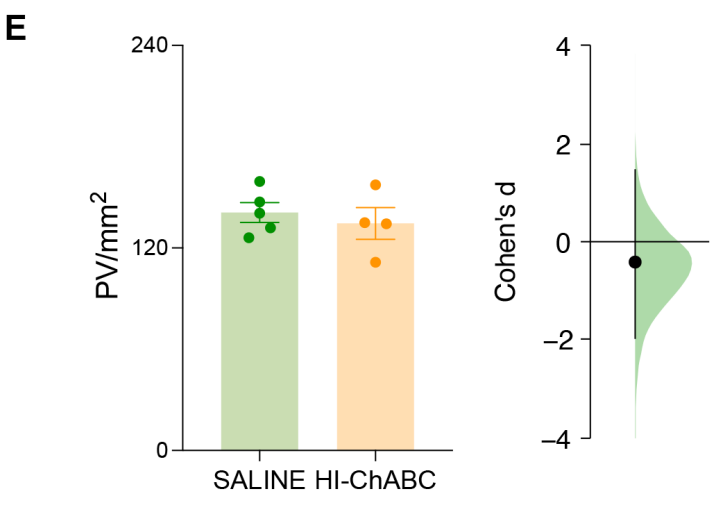
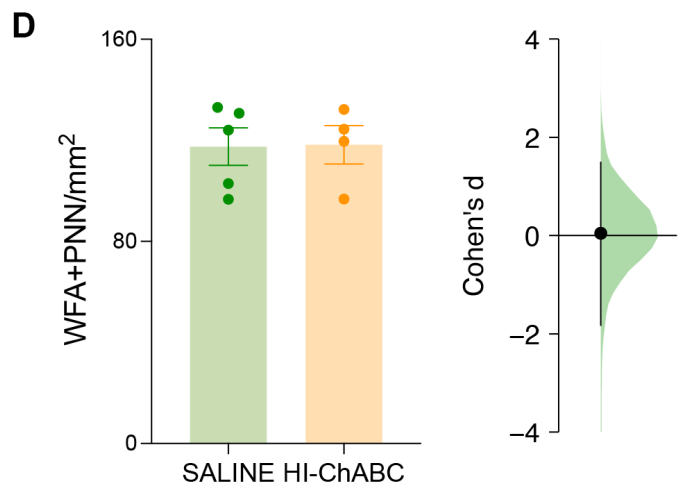
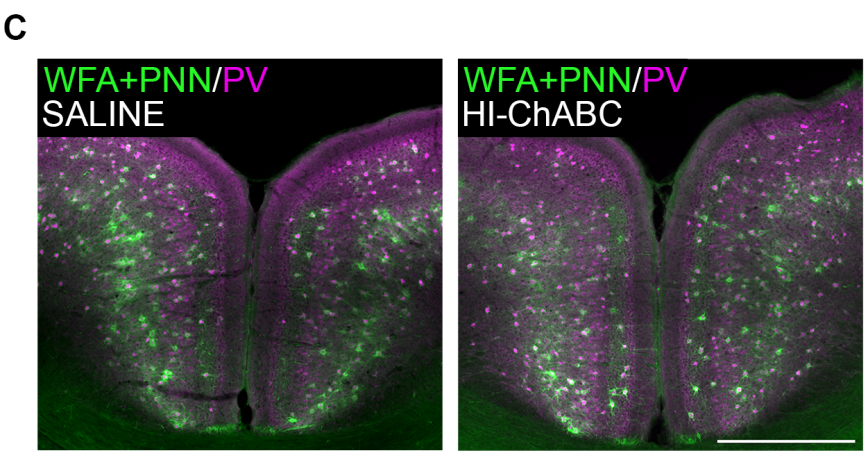
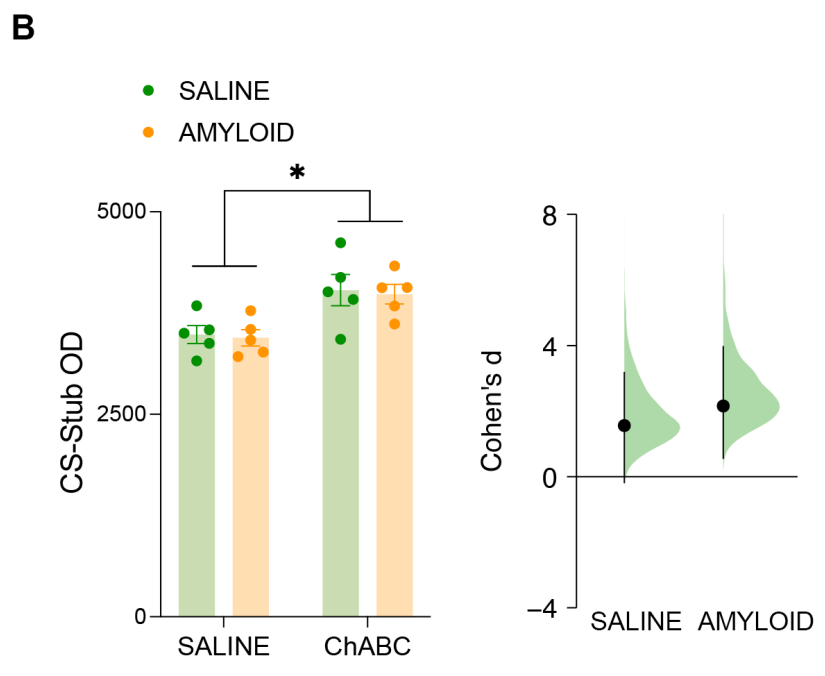
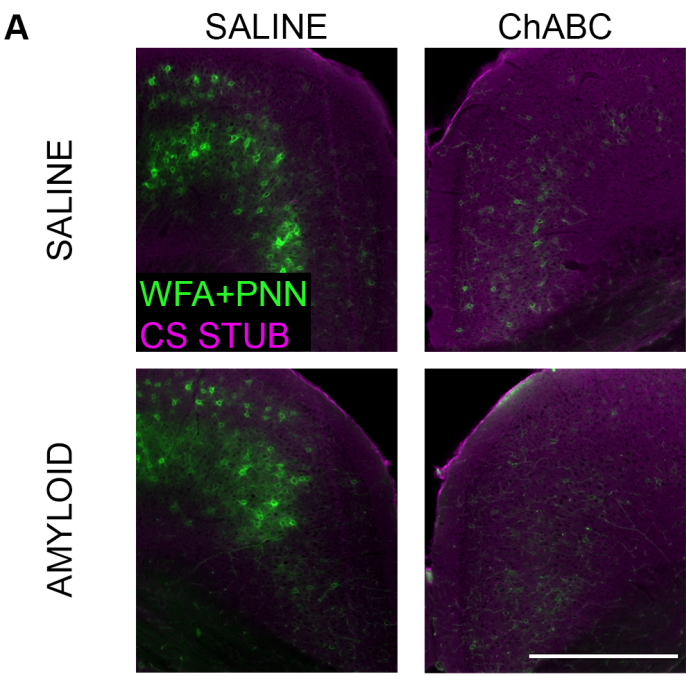
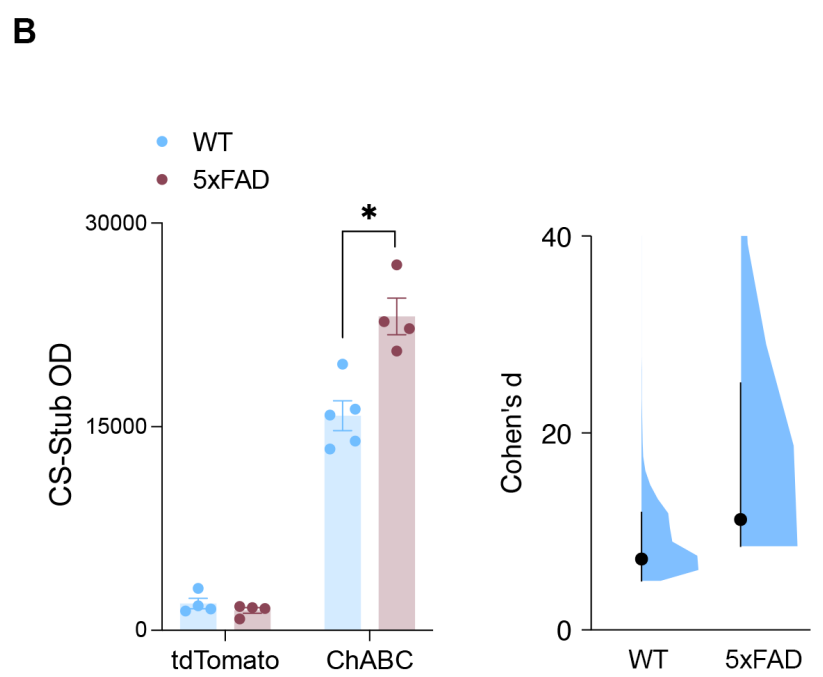
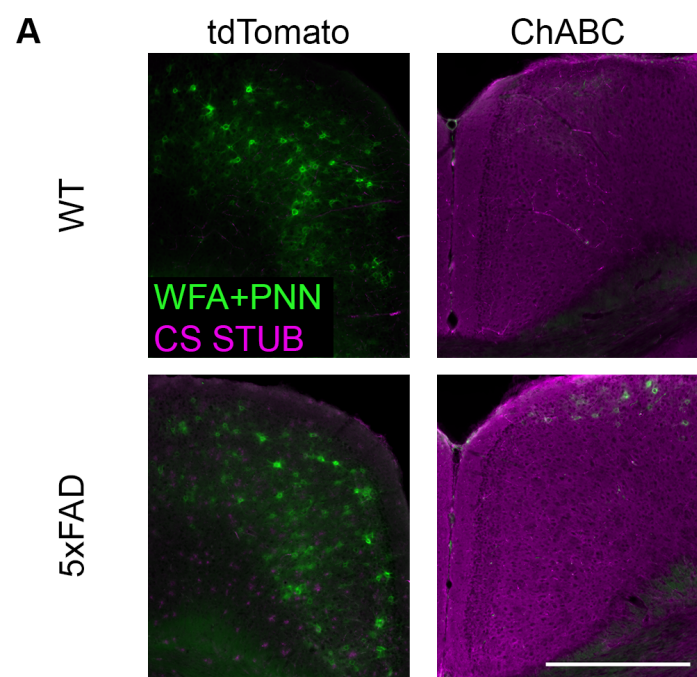


FIGURE S2. Functional chondroitinase ABC infusion impairs expression of PNNs.

A Representative photomicrographs of WFA staining for PNNs and CS-stub labelling across the RSC of mice injected with saline-saline, saline-amyloid, ChABC-saline, and ChABC-amyloid. Scale bar represents 250 μ m. **B** Optical density of CS stubs in the RSC. The mean intensity of CS stub labelling across the RSC increased with ChABC infusion (n : Saline-Saline=5; Saline-ChABC=5; Amyloid-Saline=5; Amyloid-ChABC=5, two-way ANOVA, effect of ChABC, $F_{(16)}=15.78$, $P = 0.0011$). Effect size between saline- and ChABC-infused mice across saline (Cohen's $D=1.55$) and amyloid (Cohen's $D=2.15$) infusion groups. **C** Representative photomicrographs of WFA staining for PNNs and PV labelling across the RSC of saline- and HI-ChABC-injected mice. Scale bar represents 500 μ m. **D** HI-ChABC injection did not alter the density of WFA-labelled PNNs across the RSC (n : saline=4; HI-ChABC=5, two-sample t test, $t_{(7)} = 0.07123$, $P = 0.9452$). Effect size between saline and HI-ChABC injection (Cohen's $D = 0.0478$). **E** HI-ChABC injection did not alter the density of PV-INS across the RSC (n : saline=4; HI-ChABC=5, two-sample t test, $t_{(7)} = 0.6136$, $P = 0.5589$). Effect size between saline and HI-ChABC injection (Cohen's $D = -0.412$). **F** HI-ChABC injection did not alter the percentage of PV-INS enrobed by PNNs across the RSC (n : saline=4; HI-ChABC=5, two-sample t test, $t_{(7)} = 0.6054$, $P = 0.5640$). Effect size between saline and HI-ChABC injection (Cohen's $D = 0.406$). Data represent mean $\pm$ SEM. All statistical comparisons have been provided as Data S1.



1 **FIGURE S3. Viral ChABC expression yields sustained digestion of PNNs.**

2 **A** Representative photomicrographs of WFA staining for PNNs and CS-stub labelling across the
3 RSC of WT and 5xFAD mice injected with viral ChABC or a tdTomato control virus. Scale bar
4 represents 250 μ m. **B** Optical density of CS stubs in the RSC. The mean intensity of CS stub
5 labelling across the RSC increased with viral ChABC expression (n : WT-tdTomato=4; 5xFAD-
6 tdTomato=4; WT-ChABC=5; 5xFAD-ChABC=5, two-way ANOVA, effect of ChABC, $F_{(13)}=357.0$,
7 $P < 0.0001$; Genotype X ChABC interaction, $F_{(13)}=17.31$, $P = 0.0011$; Šídák's test, WT-ChABC
8 vs 5xFAD-ChABC, $P=0.0002$). Effect size between tdTomato- and ChABC-injected mice across
9 WT (Cohen's $D=7.2$) and 5xFAD (Cohen's $D=11.2$) genotypes.

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1 **Table S1. Region List**

2 List of neuroanatomical regions analyzed in brain-wide functional connectivity analyses.

#	Abbr.	Full	#	Abbr.	Full	#	Abbr.	Full
1	MO	Motor cortex	18	CA1	CA1	35	IAD	Interanterodorsal nucleus of the thalamus
2	SS	Somatosensory cortex	19	CA2	CA2	36	LH	Lateral habenula
3	GU	Gustatory cortex	20	CA3	CA3	37	LHA	Lateral hypothalamus
4	VISC	Visceral cortex	21	DG	Dentate gyrus	38	LPO	Lateral preoptic area
5	VIS	Visual cortex	22	ENTI	Entorhinal cortex, lateral part	39	RCH	Retrochiasmatic area
6	ACA	Anterior cingulate	23	ENT	Entorhinal cortex, medial part	40	SNr	Substantia nigra, reticular
7	PL	Prelimbic cortex	24	BLA	Basolateral amygdala	41	VTA	Ventral tegmental area
8	ORB	Orbitofrontal cortex	25	BMA	Basomedial amygdala	42	SC	Superior colliculus
9	AI	Agranular insular cortex	26	ACB	Nucleus accumbens	43	PAG	Periaqueductal gray
10	RSC	Retrosplenial cortex	27	sAMY	Striatum-like amygdalar nuclei	44	NPC	Nucleus of the posterior commissure
11	PTLp	Posterior parietal cortex	28	CEA	Central amygdala nucleus	45	CUN	Cuneiform nucleus
12	ECT	Ectorhinal cortex	29	MEA	Medial amaygdalar nucleus	46	SNc	Substantia nigra, compact part
13	MOB	Main olfactory bulb	30	PALv	Pallidum, ventral part	47	PG	Pontine gray
14	AON	Anterior olfactory nucleus	31	PALm	Pallidum, medial part	48	LC	Locus ceruleus
15	PIR	Piriform cortex	32	VAL	Ventral anterior-lateral thalamus	49	CU	Cuneate nucleus
16	COA	Cortical amygdalar area	33	VPM	Ventral posteromedial thalamus	50	MARN	Magnocellular reticular nucleus
17	PAA	Piriform amygdalar area	34	AV	Anteroventral thalamus			

3 #: position of the region on the correlation matrices; Abbr.: abbreviation; Full: full name of the
4 region.

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1 DATA S1

2 Details of all statistical comparisons presented in the current manuscript.