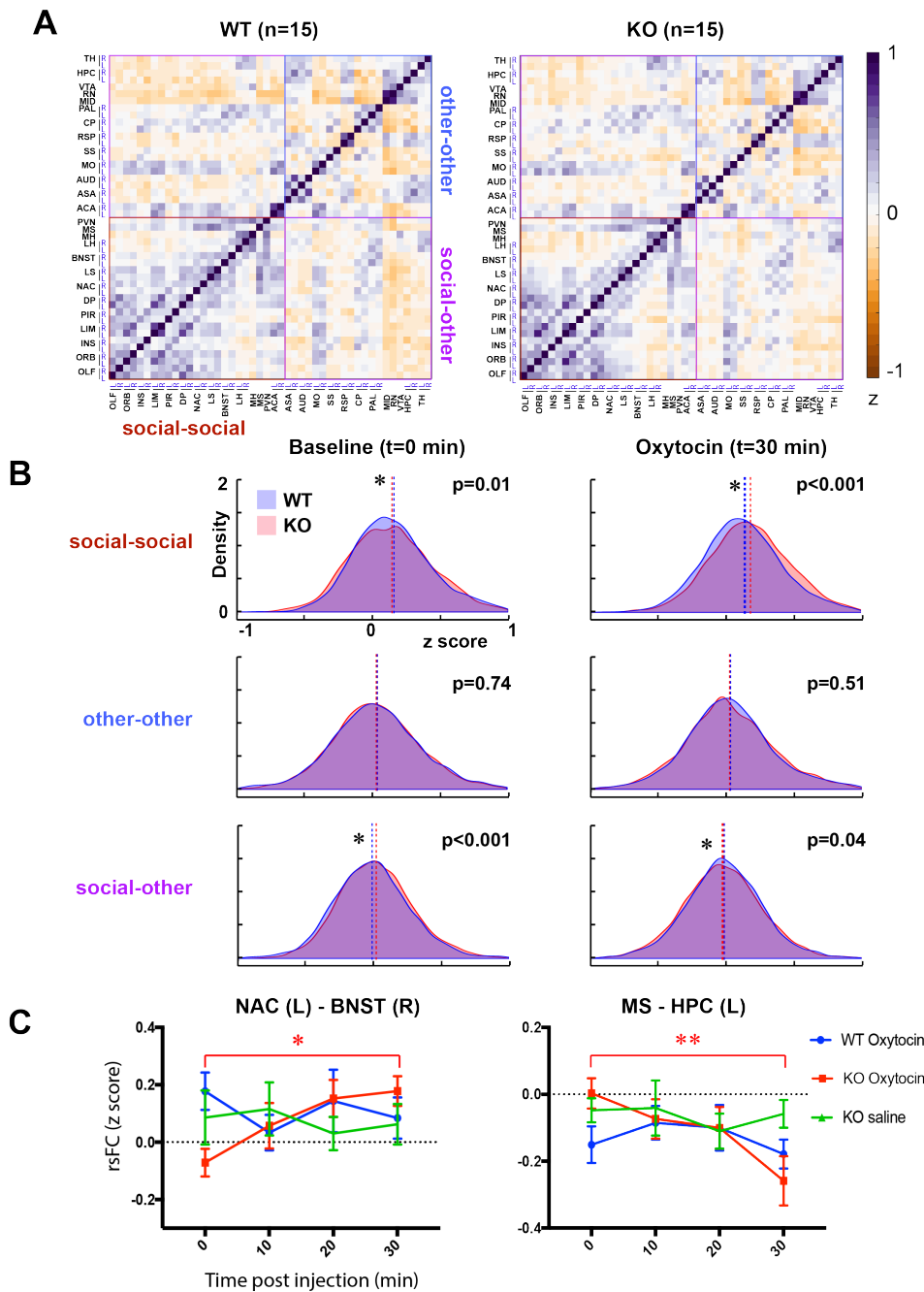
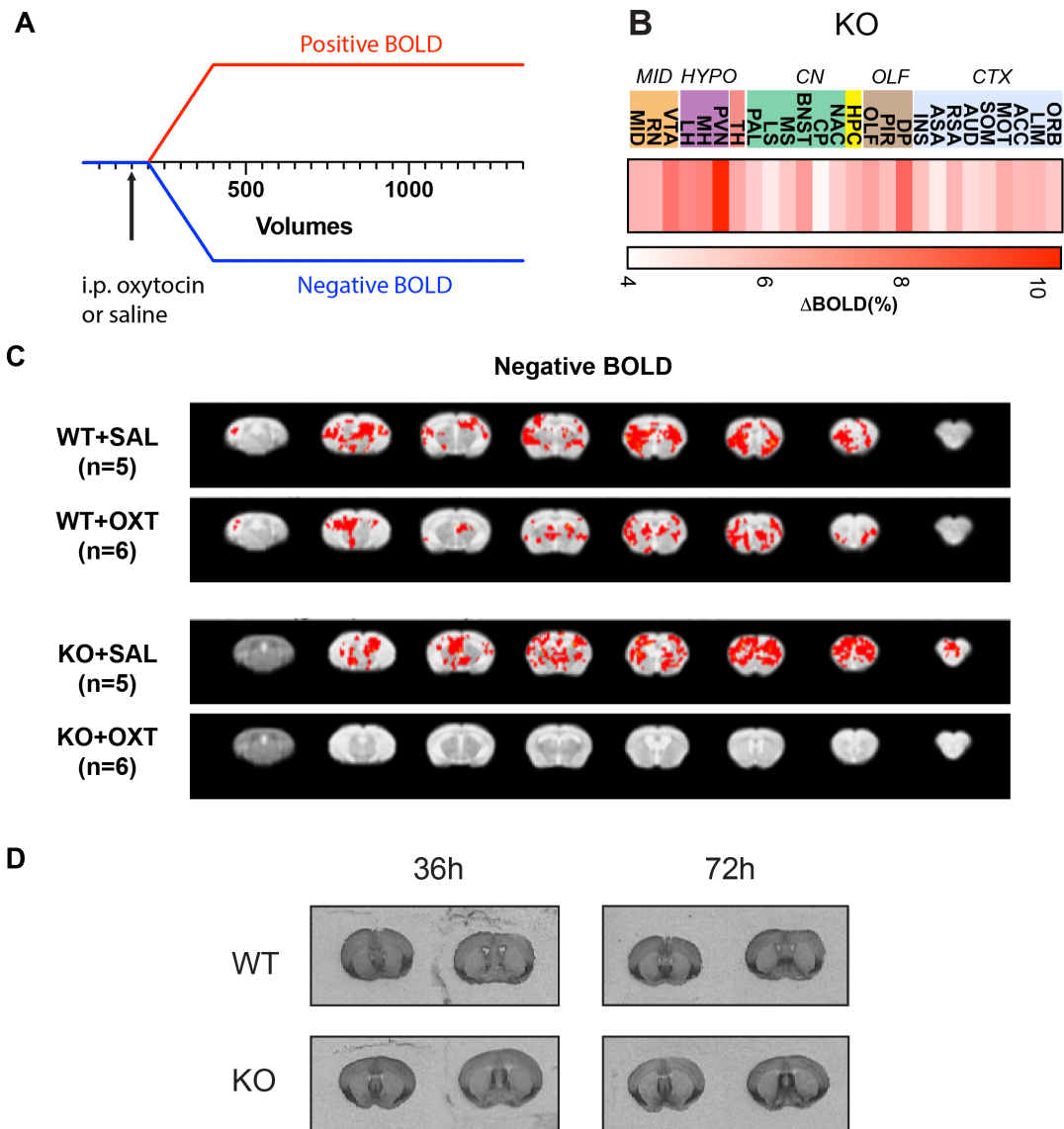


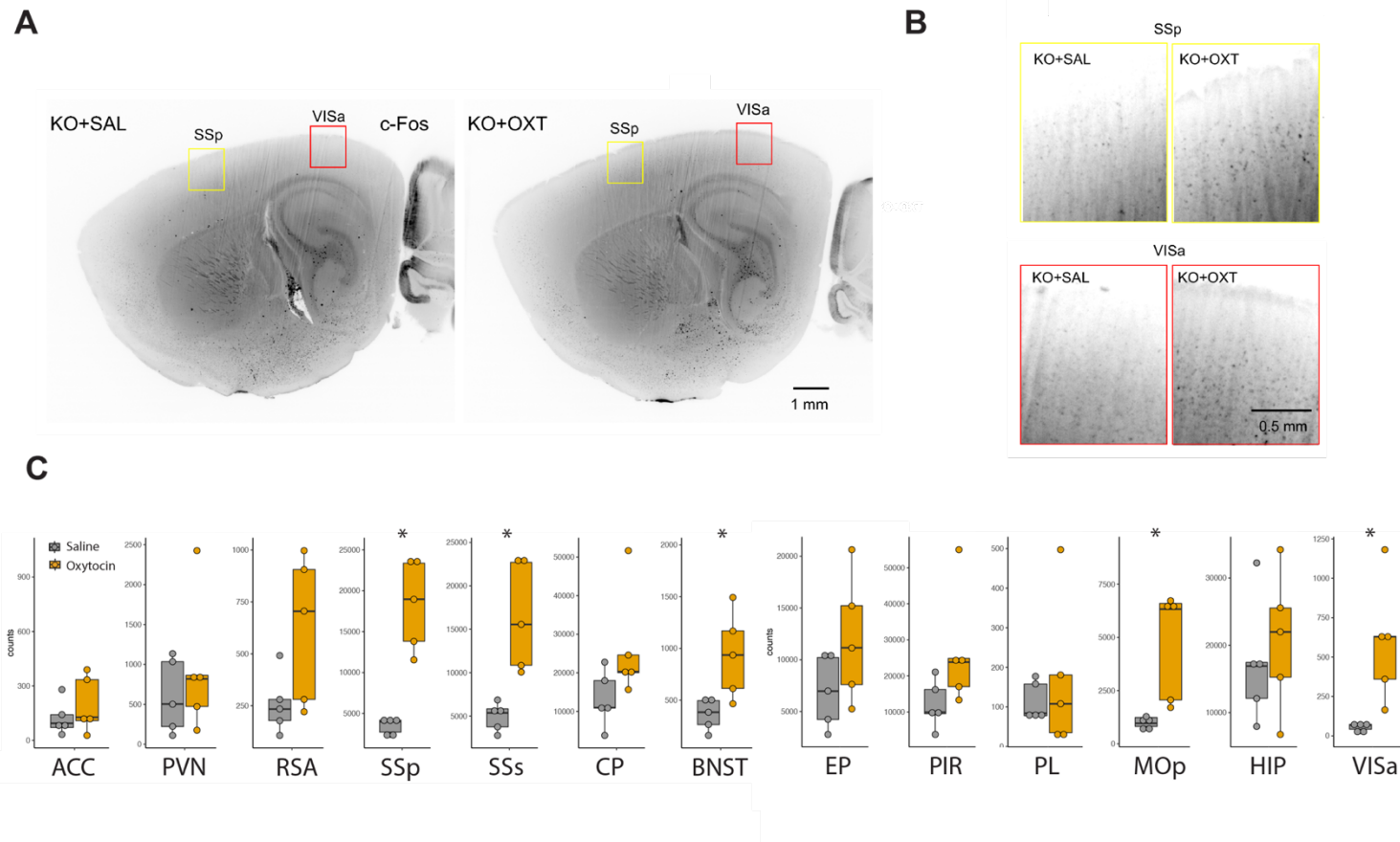
**Fig. S1: fMRI acquisition and analysis schematics, Related to Figure 1.** (A) 3-axis representation of the custom 3D MRI atlas used for this study. Each colored area represents a parcellated region. (B) A schematic of fMRI acquisition and analysis pipeline. (C) Resting-state fMRI acquisition schematic.



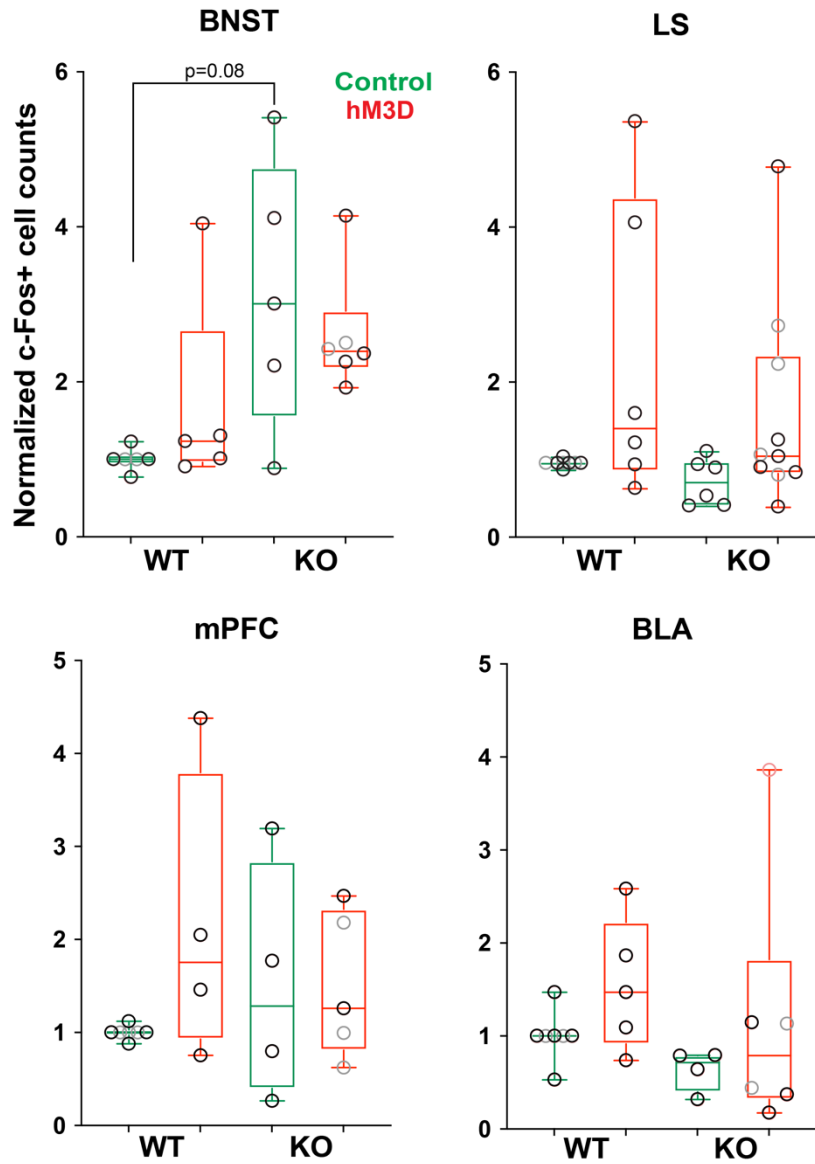
**Fig. S2: Pairwise ROI assessment of rsFC, Related to Figure 1.** (A) Heatmaps representing average baseline rsFC in WT (left) and KO (right) mice. Colored boxes represent pairwise connectivity of social-to-social, social-to-other, and other-to-other regions. (B) Histograms display the differences in mean rsFC between every ROI pair within social (top), other (middle), or between social and other regions (bottom) before and after oxytocin in WT (blue) and KO (red) mice. Monte Carlo exact permutation test (10,000 repetitions),  $p < 0.001$ . (C) Time-course of rsFC shift in 2 ROI pairs exhibiting KO-specific modulation by oxytocin treatment. In both pairs of functionally connected regions, a significant effect is only seen in the KO group receiving oxytocin. Repeated-measures two-way ANOVA comparison of the time effect + Dunn's posthoc test. \*  $p < 0.05$ , \*\*  $p < 0.01$ , ns for all other comparisons.



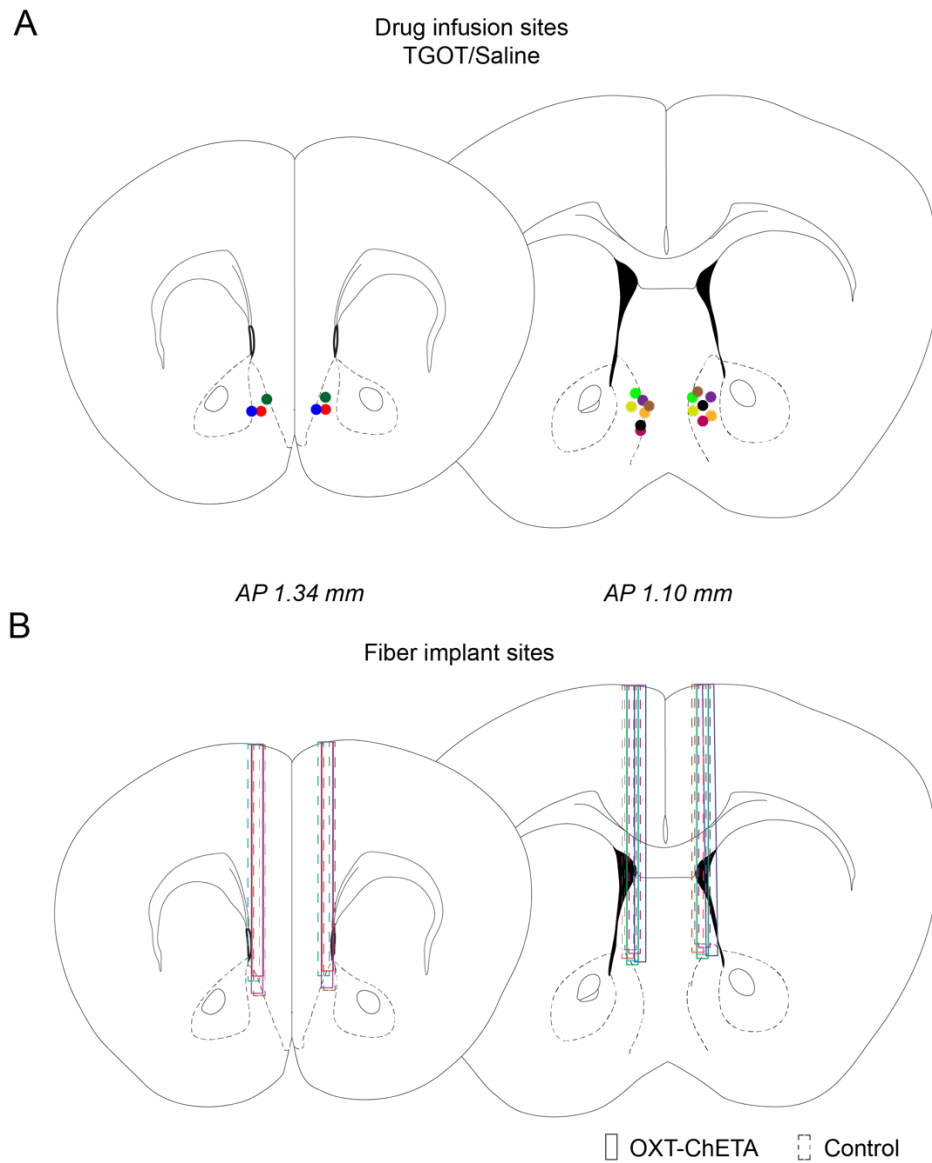
**Fig. S3: BOLD responses to OXT injections in WT and KO mice, Related to Figure 3.** (A) GLM model used to estimate positive or negative BOLD activation. (B) Heatmap illustrates the peak % amplitude change in BOLD signal in the KO mouse for each examined brain region ordered by their regional groupings. (C) Voxels (red) identified as significantly associated ( $p < 0.001$ ) with the Negative BOLD GLM model. (D) Comparison of oxytocin receptor expression between WT and KO mice at two different exposure times (36 and 72 h), revealing a difference in the density of staining in the dorsal lateral septum.



**Fig S4: ROI-wise comparison of c-Fos+ cell counts in iDISCO+ cleared brains, related to Figure 4.** (A) Representative images of c-Fos stained iDISCO+ brains in the sagittal orientation. Yellow and red boxes indicate regions of magnified images in (B), showing the stimulatory effect of oxytocin in SSp (yellow) and VISa (red). Box-whisker plots display quantifications of c-Fos+ cell counts in each region. T-test, \* FDR<0.1. (C) Box-whisker plots display quantifications of c-Fos+ cell counts in a group of brain regions identified as significantly activated by OXT according to pHMRI results. \* FDR<0.1.



**Fig S5: Quantification of c-Fos+ cells in DREADD-stimulated WT and KO mice, Related to Figure 5.** Box-whisker plots represent a summary of cell counts (whisker: min-max values, box: 25-75<sup>th</sup> percentile, lines: individual before-after values, black for males and grey for females). Two-way ANOVA, \*  $p < 0.05$ . ns for all other comparisons.



**Fig S6. Posthoc assessment of *in vivo* NAcSh targeting, related to Figure 6.** (A) Filled circles indicate TGOT/saline infusion sites. (B) Solid (OXT-ChETA) or dotted lines (control) indicate fiber implant sites for optogenetic experiments.

WT		KO	
Region	Appearance	Region	Appearance
MOT	5	NAC	5
LS	4	HPC	5
LIM	4	BNST	5
ACC	4	MS	4
MS	3	MOT	3
SOM	3	MID	3
ORB	2	PIR	3
BNST	2	PVN	2
CP	2	LS	2
DP	2	SOM	2
RN	2	ASA	1
NAC	2	RSA	1
PIR	1	DP	1
		INS	1
		ACC	1

**Table S1. Ranking of the number of regional appearances in ROI pairs exhibiting highest rsFC modifications after OXT, Related to Figure 1.** After pairwise-ROI rsFC analysis, 20 ROI pairs exhibiting top 10 rsFC increases and top 10 decreases were identified for each genotype, then the number of times a brain region appears was counted and ranked.

<b>ROI</b>	<b>OXTR expression (%)</b>	<b><math>\Delta</math>BOLD (%)</b>
AUD	21.20	5.05
BNST	36.85	6.98
HPC	34.30	6.22
LH	22.25	7.47
LIM	37.15	6.19
LS	47.65	4.69
MH	46.03	7.74
MOT	16.45	6.53
NAC	18.30	5.51
OLF	41.40	6.81
PAL	11.60	5.52
PIR	27.65	6.01
PVN	28.10	10.35
SS	19.73	5.40

<b>ROI</b>	<b>OXT fiber density (AU)</b>	<b><math>\Delta</math>BOLD (%)</b>
BNST	3.00	6.98
CP	1.00	4.24
INS	1.00	5.90
LH	4.60	7.47
LIM	2.00	6.19
LS	3.00	4.69
MH	4.33	7.74
MID	2.80	6.25
NAC	1.50	5.51
OLF	1.00	6.81
ORB	2.00	5.66
PAL	4.00	5.52
PIR	1.00	6.01
PVN	6.00	10.35
VTA	1.00	8.05

**Table S2. Comparison of OXT-induced  $\Delta$ BOLD vs. OXT receptor (OXTR) expression (top) and vs. OXT fiber density (bottom) in ROIs that overlap between compared datasets, Related to Figure 3.**



	WT		KO	
	$\Delta$ BOLD (%)	$\Delta$ c-Fos cell count (%)	$\Delta$ BOLD (%)	$\Delta$ c-Fos cell count (%)
OLF	1.48	45.47	5.03	114.98
ORB	-0.44	79.37	4.67	90.02
ACC	1.00	296.91	5.39	109.09
ASA	0.83	-7.19	3.17	116.02
AUD	0.03	-19.56	3.95	264.85
INS	0.56	22.39	4.59	95.21
LIM	1.39	176.44	5.36	40.66
MOT	-0.60	42.71	5.86	308.46
SOM	-0.88	-13.28	3.96	300.87
PIR	1.87	-6.39	5.04	50.88
RS	0.10	52.68	4.99	286.27
DP	2.14	201.21	5.45	23.10
NAC	-1.36	90.37	4.43	125.82
LS	0.64	54.95	3.52	11.31
HPC	1.05	22.17	5.83	170.25
CP	0.38	5.40	3.92	123.68
LH	1.57	33.51	6.79	94.03
TH	-0.11	47.97	6.39	153.76
PAL	-0.02	68.33	5.03	144.66
MH	2.02	163.64	6.45	99.99
MID	1.19	33.55	5.65	112.36
PVN	1.68	175.17	4.70	144.09

**Table S3. Comparison of OXT responses detected by phMRI and c-Fos-iDISCO+ in 22 ROIs that overlap between the two datasets, Related to Figure 4.**

**PVN****WT control**

Mouse#	sex	norm counts	raw counts	cohort
1	F	1.00	19.25	1
2	M	1.00	18.25	2
3	M	1.00	78.50	5
4	M	1.00	12.33	6b
5	M	1.00	12.25	6b

**KO control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	0.83	16.00	1
2	M	0.61	12.25	2
3	F	2.16	39.50	2
4	M	0.67	12.25	2
5	M	2.13	53.33	6a
6	M	0.20	5.00	6a
7	M	3.06	37.67	6b

**WT hm3D**

Mouse#	sex	norm counts	raw counts	cohort
1	M	6.42	123.50	1
2	M	7.47	136.25	2
3	M	4.66	85.00	2
4	M	2.29	179.50	5
5	M	5.47	136.67	6a
6	M	5.39	66.25	6b

**KO hm3D**

Mouse#	sex	norm counts	raw counts	cohort
1	M	5.90	113.50	1
2	M	5.38	5.38	1
3	F	6.90	132.75	1
4	F	7.68	140.25	2
5	M	2.98	233.67	5
6	M	7.36	184.00	6a
7	M	4.50	55.33	6b

**NAC****WT control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	1.00	13.21	3
2	M	1.00	32.26	5
3	F	1.00	1.48	8
4	F	1.00	1.59	9
5	M	1.00	16.21	6a
6	M	1.00	17.05	6b

**KO control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	0.57	7.47	3
2	M	0.10	0.16	9
3	M	1.88	30.40	6a
4	M	0.22	3.67	6b

**WT hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	1.86	24.57	3
2	M	1.03	33.09	5
3	M	1.57	2.31	8
4	M	1.54	24.89	6a
5	M	1.39	23.72	6b

**KO hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	3.95	52.22	3
2	F	3.48	5.15	8
3	F	2.38	3.81	9
4	F	2.48	3.97	9
5	M	1.79	29.03	6a
6	M	3.46	59.00	6b

**BNST****WT control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	1.00	8.05	3
2	M	1.00	29.12	5
3	F	1.00	23.80	8
4	F	1.00	9.80	9
5	M	0.77	26.17	6b
6	M	1.23	41.67	6b

**KO control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	5.41	43.54	3
2	M	4.11	119.74	5
3	M	2.21	42.50	6a
4	M	0.89	30.02	6b
5	M	3.01	29.49	9

**WT hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	4.04	32.52	3
2	M	1.24	36.00	5
3	M	0.91	17.50	6a
4	M	1.01	34.29	6b
5	M	1.30	31.02	8

**KO hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	4.14	33.35	3
2	M	2.37	68.88	5
3	M	2.26	65.76	5
4	M	1.93	37.06	6a
5	F	2.51	24.55	9
6	F	2.42	23.76	9

**Lateral septum****WT control**

Mouse#	sex	norm counts	raw counts	cohort
1	F	1.00	13.13	1
2	M	1.00	64.02	3
3	M	1.00	62.10	5
4	M	1.00	89.08	6a
5	M	0.91	50.26	6b
6	M	1.09	59.70	6b
7	F	1.00	5.73	8
8	F	1.00	17.46	9

**KO control**

Mouse#	sex	norm counts	raw counts	cohort
1	M	0.46	6.00	1
2	M	0.94	60.25	3
3	M	0.98	61.13	5
4	M	0.45	40.31	6a
5	M	1.15	63.46	6b
6	M	0.58	10.09	9

**WT hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	4.10	53.88	1
2	M	1.27	81.06	3
3	M	1.64	102.14	5
4	M	0.98	87.58	6a
5	M	0.68	37.24	6b
6	M	5.41	31.01	8

**KO hm3d**

Mouse#	sex	norm counts	raw counts	cohort
1	M	4.83	63.38	1
2	M	1.09	14.29	1
3	F	2.77	36.38	1
4	M	1.30	83.36	3
5	M	0.88	54.80	5
6	M	0.95	58.94	5
7	M	0.44	39.06	6a
8	F	2.28	13.06	8
9	F	0.85	14.84	9

**Table S4. Raw and normalized counts of c-Fos+ cells in PVN, NAc, BNST, and Lateral Septum after DREADD-stimulated release of endogenous OXT, Related to Figure 5. All raw count values are counts/mm<sup>2</sup> except for the PVN, which reflect raw counts not normalized to area.**

mPFC				
WT control				
Mouse#	sex	norm counts	raw counts	cohort
1	M	1.00	84.90	5
2	M	1.00	29.44	6a
3	M	0.88	55.09	6b
4	M	1.12	70.06	6b
5	F	1.00	4.66	8
6	F	1.00	13.34	9
KO control				
Mouse#	sex	norm counts	raw counts	cohort
1	M	1.77	150.28	5
2	M	3.19	94.01	6a
3	M	0.80	49.90	6b
4	M	0.27	3.55	9
WT hm3d				
Mouse#	sex	norm counts	raw counts	cohort
1	M	0.75	64.00	5
2	M	2.05	60.31	6a
3	M	1.46	91.34	6b
4	M	4.38	20.40	8
KO hm3d				
Mouse#	sex	norm counts	raw counts	cohort
1	M	2.47	209.53	5
2	M	1.26	37.07	6a
3	F	2.18	10.17	8
4	F	0.62	8.33	9
5	F	1.00	13.30	9
BLA				
WT control				
Mouse#	sex	norm counts	raw counts	cohort
1	M	1.00	43.68	3
2	M	1.00	47.29	5
3	F	1.00	10.49	8
4	F	1.00	18.86	9
5	M	1.00	24.09	6a
6	M	0.53	8.78	6b
7	M	1.47	24.44	6b
KO control				
Mouse#	sex	norm counts	raw counts	cohort
1	M	0.79	34.59	3
2	M	0.64	15.41	6a
3	M	0.79	13.09	6b
4	M	0.32	5.99	9
WT hm3d				
Mouse#	sex	norm counts	raw counts	cohort
1	M	1.09	47.64	3
2	M	0.74	32.19	3
3	M	1.86	44.93	6a
4	M	1.47	24.44	6b
5	M	2.58	27.11	8
KO hm3d				
Mouse#	sex	norm counts	raw counts	cohort
1	M	0.37	16.14	3
2	M	0.17	8.16	5
3	M	1.15	27.61	6a
4	F	3.86	40.51	8
5	F	0.44	8.38	9
6	F	1.14	21.42	9

**Table S5. Raw and normalized counts of c-Fos+ cells in mPFC and basolateral amygdala (BLA) after DREADD-stimulated release of endogenous OXT, Related to Figure 5. All raw count values are counts/mm<sup>2</sup>.**