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Supplementary Tables: 0

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Reporting Checklist for Nature Neuroscience

This checklist is used to ensure good reporting standards and to improve the reproducibility of published results. For more information, please read Reporting Life Sciences Research.

Please note that in the event of publication, it is mandatory that authors include all relevant methodological and statistical information in the manuscript.

▶ Statistics reporting, by figure

- Please specify the following information for each panel reporting quantitative data, and where each item is reported (section, e.g. Results, & paragraph number).

- Each figure legend should ideally contain an exact sample size (n) for each experimental group/condition, where n is an exact number and not a range, a clear definition of how n is defined (for example x cells from x slices from x animals from x litters, collected over x days), a description of the statistical test used, the results of the tests, any descriptive statistics and clearly defined error bars if applicable.

- For any experiments using custom statistics, please indicate the test used and stats obtained for each experiment.

- Each figure legend should include a statement of how many times the experiment shown was replicated in the lab; the details of sample collection should be sufficiently clear so that the replicability of the experiment is obvious to the reader.

- For experiments reported in the text but not in the figures, please use the paragraph number instead of the figure number.

Note: Mean and standard deviation are not appropriate on small samples, and plotting independent data points is usually more informative. When technical replicates are reported, error and significance measures reflect the experimental variability and not the variability of the biological process; it is misleading not to state this clearly.

	TEST USED		n			DESCRIPTIVE STATS (AVERAGE, VARIANCE)		P VALUE		DEGREES OF FREEDOM & F/t/z/R/ETC VALUE	
FIGURE NUMBER	WHICH TEST?	SECTION & PARAGRAPH #	EXACT VALUE	DEFINED?	SECTION & PARAGRAPH #	REPORTED?	SECTION & PARAGRAPH #	EXACT VALUE	SECTION & PARAGRAPH #	VALUE	SECTION & PARAGRAPH #
example 1a	one-way ANOVA	Fig. legend	9, 9, 10, 15	mice from at least 3 litters/group	Methods para 8	error bars are mean +/- SEM	Fig. legend	p = 0.044	Fig. legend	F(3, 36) = 2.97	Fig. legend
example results, para 6	unpaired t-test	Results para 6	15	slices from 10 mice	Results para 6	error bars are mean +/- SEM	Results para 6	p = 0.0006	Results para 6	t(28) = 2.808	Results para 6

	FIGURE NUMBER	TEST USED		n			DESCRIPTIVE STATS (AVERAGE, VARIANCE)		P VALUE		DEGREES OF FREEDOM & F/t/z/R/ETC VALUE	
		WHICH TEST?	SECTION & PARAGRAPH #	EXACT VALUE	DEFINED?	SECTION & PARAGRAPH #	REPORTED?	SECTION & PARAGRAPH #	EXACT VALUE	SECTION & PARAGRAPH #	VALUE	SECTION & PARAGRAPH #
+ −	Fig 2, left column figs	ANOVA	p6, middle para	16	rats in two groups	results, second para	data and SEM plotted in fig 2	fig 2	ANOVA (group × cue × session) revealed significant main effects of cue (F(2,28)=92.3, p<0.0001) and session (F(11,154)=5.26, p<0.0001), and a significant interaction between cue and session (F(22,308)=9.91, p<0.0001).	p 6, middle para	ANOVA (group × cue × session) revealed significant main effects of cue (F(2,28)=92.3, p<0.0001) and session (F(11,154)=5.26, p<0.0001), and a significant interaction between cue and session (F(22,308)=9.91, p<0.0001).	p 6 middle
+ −	fig 2, middle column figs	ANOVA	p7, bottom para	16	rats in two groups	results, second para	data and SEM plotted in fig 2	fig 2	ANOVA's (group × cue × session) revealed significant main effects of cue (reward run: F(2,28)=155.3, p<0.0001; ITI run: F(2,28)=70.1, p<0.0001) and session (reward run: F(3,42)=3.28, p=0.03)	ps 7, bottom para	ANOVA's (group × cue × session) revealed significant main effects of cue (reward run: F(2,28)=155.3, p<0.0001; ITI run: F(2,28)=70.1, p<0.0001) and session (reward run: F(3,42)=3.28, p=0.03)	p 7 bottom

+/−												
+ −	fig 2, left colu mn figs	ANOVA	p8-9	16	rats in two groups	results, second para	data and SEM plotted in fig 2	fig2	ANOVA's (group × cue × trial) revealed significant main effects of cue and trial (reward: cue: $F_{(2,28)}=82.5$, $p<0.0001$; trial: $F_{(7,98)}=20.8$, $p<0.0001$; ITI: cue: $F_{(2,28)}=25.9$, $p<0.0001$; trial: $F_{(7,98)}=23.7$, $p<0.0001$) and significant interactions between cue and trial (reward: $F_{(14,196)}=6.19$, $p<0.0001$; ITI: $F_{(14,196)}=6.39$, $p<0.0001$).	p8-9	ANOVA's (group × cue × trial) revealed significant main effects of cue and trial (reward: cue: $F_{(2,28)}=82.5$, $p<0.0001$; trial: $F_{(7,98)}=20.8$, $p<0.0001$; ITI: cue: $F_{(2,28)}=25.9$, $p<0.0001$; trial: $F_{(7,98)}=23.7$, $p<0.0001$) and significant interactions between cue and trial (reward: $F_{(14,196)}=6.19$, $p<0.0001$; ITI: $F_{(14,196)}=6.39$, $p<0.0001$). .	p8-9
+ −	fig S2, all	ANOVA	p6, top para	16	rats in two groups	results, second para	data and SEM plotted in fig S2	fig S2	p's > 0.93	p6, top para and fig cap	F's < 1.2, df as in main text analyses for fig 2	p6, top para and fig cap
+ −	fig 3, all	Wilcoxon signed rank and rank sum	fig captio n	16	rats in two grups	results, second para	data plotted in fig 3 - from individual subjects so there is no variance relevant to plot	fig 3	multiple, please see fig and cap for relevant p's	fig 3	multiple, two significant were 0.02 and 0.04 as marked on fig	NA
+ −	Fig S3, all	ANOVA	p 17, middle para	16	rats in two groups	results, second para	data and SEM plotted in fig S3	fig S3	p's > 0.92	p24, middle para	F's < 0.25, df as in main text analyses for fig 2	p24, middle para

▶ Representative figures

1. Are any representative images shown (including Western blots and immunohistochemistry/staining) in the paper?

   If so, what figure(s)?

   > yes - fig 1c and S1

2. For each representative image, is there a clear statement of how many times this experiment was successfully repeated and a discussion of any limitations in repeatability?

   If so, where is this reported (section, paragraph #)?

   > the figures are picture of the histology showing expression from individual subjects. there was only one experiment and the methods describe the number of rats and extent of expression is in the results.

▶ Statistics and general methods

1. Is there a justification of the sample size?

   If so, how was it justified?

   Where (section, paragraph #)?

   Even if no sample size calculation was performed, authors should report why the sample size is adequate to measure their effect size.

   > sample size is based on past experience, and is justified with references to prior papers showing the effect we are replicating here.

2. Are statistical tests justified as appropriate for every figure?

   Where (section, paragraph #)?

   | yes. statistical tests are described throughout the results. typically we use ANOVA's with interactions between groups or main effects, with posthocs. and Wilcoxon tests for distributions of index scores within groups and comparing across. |

   a. If there is a section summarizing the statistical methods in the methods, is the statistical test for each experiment clearly defined?

   | yes |

   b. Do the data meet the assumptions of the specific statistical test you chose (e.g. normality for a parametric test)?

   Where is this described (section, paragraph #)?

   | yes. the statistics are described and enumerated throughout the results. |

   c. Is there any estimate of variance within each group of data?

   Is the variance similar between groups that are being statistically compared?

   Where is this described (section, paragraph #)?

   | the variance is similar between groups on all our measures generally. this is evident in figure 2. the statistics we have applied account for the variance. |

   d. Are tests specified as one- or two-sided?

   | tests are two sided unless specified. this is conventional. |

   e. Are there adjustments for multiple comparisons?

   | we do not do multiple comparisons. data from each phase are analyzed via ANOVA. only with appropriate main effects or interactions do we do posthocs. |

3. Are criteria for excluding data points reported?

   Was this criterion established prior to data collection?

   Where is this described (section, paragraph #)?

   | we do not exclude any data points. |

4. Define the method of randomization used to assign subjects (or samples) to the experimental groups and to collect and process data.

   If no randomization was used, state so.

   Where does this appear (section, paragraph #)?

   | we did not randomize subjects. they were assigned to control or expt groups based on whether they received eYFP or NpHR infusions. all data collection was by computer. identity of cues was counterbalanced. all of this is described in our methods. |

5. Is a statement of the extent to which investigator knew the group allocation during the experiment and in assessing outcome included?

   If no blinding was done, state so.

   Where (section, paragraph #)?

   | no. all data collection was by computer. |

6. For experiments in live vertebrates, is a statement of compliance with ethical guidelines/regulations included?

   Where (section, paragraph #)?

   | yes. top of methods. |

7. Is the species of the animals used reported?

   Where (section, paragraph #)?

   | yes. top of methods. |

8. Is the strain of the animals (including background strains of KO/ transgenic animals used) reported?

Where (section, paragraph #)?

all information on animals used and viruses is in the methods in sections on subjects and surgery.

9. Is the sex of the animals/subjects used reported?

Where (section, paragraph #)?

yes. subjects section of methods.

10. Is the age of the animals/subjects reported?

Where (section, paragraph #)?

approx. subjects section of methods.

11. For animals housed in a vivarium, is the light/dark cycle reported?

Where (section, paragraph #)?

yes. subjects section of methods.

12. For animals housed in a vivarium, is the housing group (i.e. number of animals per cage) reported?

Where (section, paragraph #)?

no

13. For behavioral experiments, is the time of day reported (e.g. light or dark cycle)?

Where (section, paragraph #)?

no

14. Is the previous history of the animals/subjects (e.g. prior drug administration, surgery, behavioral testing) reported?

Where (section, paragraph #)?

subjects were naive.

    a. If multiple behavioral tests were conducted in the same group of animals, is this reported?

    Where (section, paragraph #)?

15. If any animals/subjects were excluded from analysis, is this reported?

Where (section, paragraph #)?

no rats were excluded.

    a. How were the criteria for exclusion defined?

    Where is this described (section, paragraph #)?

    b. Specify reasons for any discrepancy between the number of animals at the beginning and end of the study.

    Where is this described (section, paragraph #)?

▶ Reagents

1. Have antibodies been validated for use in the system under study (assay and species)?

   | antibodies were used for the immunohistochemistry. they were obtained from commercial sources by our histology core. |

   a. Is antibody catalog number given?

      Where does this appear (section, paragraph #)?

   b. Where were the validation data reported (citation, supplementary information, Antibodypedia)?

      Where does this appear (section, paragraph #)?

2. Cell line identity

   | n/a |

   a. Are any cell lines used in this paper listed in the database of commonly misidentified cell lines maintained by ICLAC and NCBI Biosample?

      Where (section, paragraph #)?

   b. If yes, include in the Methods section a scientific justification of their use--indicate here in which section and paragraph the justification can be found.

   c. For each cell line, include in the Methods section a statement that specifies:

      - the source of the cell lines

      - have the cell lines been authenticated? If so, by which method?

      - have the cell lines been tested for mycoplasma contamination?

      Where (section, paragraph #)?

▶ Data deposition

Data deposition in a public repository is mandatory for:

   a. Protein, DNA and RNA sequences
   b. Macromolecular structures
   c. Crystallographic data for small molecules
   d. Microarray data

Deposition is strongly recommended for many other datasets for which structured public repositories exist; more details on our data policy are available here. We encourage the provision of other source data in supplementary information or in unstructured repositories such as Figshare and Dryad.

We encourage publication of Data Descriptors (see Scientific Data) to maximize data reuse.

1. Are accession codes for deposit dates provided?

   | n/a |

   Where (section, paragraph #)?

▶ Computer code/software

Any custom algorithm/software that is central to the methods must be supplied by the authors in a usable and readable form for readers at the time of publication. However, referees may ask for this information at any time during the review process.

1. Identify all custom software or scripts that were required to conduct the study and where in the procedures each was used.

we used scripts written to run the behavior. these are in Coulbourn's GS3 system and are available to anyone who wants them. data were analyzed by statistica.

2. If computer code was used to generate results that are central to the paper's conclusions, include a statement in the Methods section under "**Code availability**" to indicate whether and how the code can be accessed. Include version information as necessary and any restrictions on availability.

n/a

▶ Human subjects

1. Which IRB approved the protocol?

   Where is this stated (section, paragraph #)?

   n/a

2. Is demographic information on all subjects provided?

   Where (section, paragraph #)?

   n/a

3. Is the number of human subjects, their age and sex clearly defined?

   Where (section, paragraph #)?

   n/a

4. Are the inclusion and exclusion criteria (if any) clearly specified?

   Where (section, paragraph #)?

   n/a

5. How well were the groups matched?

   Where is this information described (section, paragraph #)?

   n/a

6. Is a statement included confirming that informed consent was obtained from all subjects?

   Where (section, paragraph #)?

   n/a

7. For publication of patient photos, is a statement included confirming that consent to publish was obtained?

   Where (section, paragraph #)?

   n/a

▶ fMRI studies

For papers reporting functional imaging (fMRI) results please ensure that these minimal reporting guidelines are met and that all this information is clearly provided in the methods:

1. Were any subjects scanned but then rejected for the analysis after the data was collected?

   `n/a`

   a. If yes, is the number rejected and reasons for rejection described?

   Where (section, paragraph #)?

2. Is the number of blocks, trials or experimental units per session and/ or subjects specified?

   Where (section, paragraph #)?

   `n/a`

3. Is the length of each trial and interval between trials specified?

   `n/a`

4. Is a blocked, event-related, or mixed design being used? If applicable, please specify the block length or how the event-related or mixed design was optimized.

   `n/a`

5. Is the task design clearly described?

   Where (section, paragraph #)?

   `n/a`

6. How was behavioral performance measured?

   `n/a`

7. Is an ANOVA or factorial design being used?

   `n/a`

8. For data acquisition, is a whole brain scan used?

   If not, state area of acquisition.

   `n/a`

   a. How was this region determined?

9. Is the field strength (in Tesla) of the MRI system stated?

   `n/a`

   a. Is the pulse sequence type (gradient/spin echo, EPI/spiral) stated?

   b. Are the field-of-view, matrix size, slice thickness, and TE/TR/ flip angle clearly stated?

10. Are the software and specific parameters (model/functions, smoothing kernel size if applicable, etc.) used for data processing and pre-processing clearly stated?

    `n/a`

11. Is the coordinate space for the anatomical/functional imaging data clearly defined as subject/native space or standardized stereotaxic space, e.g., original Talairach, MNI305, ICBM152, etc? Where (section, paragraph #)?

n/a

12. If there was data normalization/standardization to a specific space template, are the type of transformation (linear vs. nonlinear) used and image types being transformed clearly described? Where (section, paragraph #)?

n/a

13. How were anatomical locations determined, e.g., via an automated labeling algorithm (AAL), standardized coordinate database (Talairach daemon), probabilistic atlases, etc.?

n/a

14. Were any additional regressors (behavioral covariates, motion etc) used?

n/a

15. Is the contrast construction clearly defined?

n/a

16. Is a mixed/random effects or fixed inference used?

n/a

    a. If fixed effects inference used, is this justified?

17. Were repeated measures used (multiple measurements per subject)?

n/a

    a. If so, are the method to account for within subject correlation and the assumptions made about variance clearly stated?

18. If the threshold used for inference and visualization in figures varies, is this clearly stated?

n/a

19. Are statistical inferences corrected for multiple comparisons?

n/a

    a. If not, is this labeled as uncorrected?

20. Are the results based on an ROI (region of interest) analysis?

n/a

    a. If so, is the rationale clearly described?

    b. How were the ROI's defined (functional vs anatomical localization)?

21. Is there correction for multiple comparisons within each voxel?

n/a

22. For cluster-wise significance, is the cluster-defining threshold and the corrected significance level defined?

n/a

▸ Additional comments

Additional Comments