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.

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<td>Fig. legend</td>
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<td>p = 0.0359 for NAc</td>
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<td>n = 4 for NAc, n = 3 for dSTR</td>
<td>12 mice pairs for NAc, 3 mice pairs for Str</td>
<td>means +/- SEM</td>
<td>p = 0.0010 for NAc</td>
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</table>
### 3c
Unpaired two-tailed t-test

**Legend page 29**

- $t=3.218$ df=4 for NAc  
- $t=0.6840$ df=4 for dSTR

### 3d
Unpaired two-tailed t-test

**Legend page 29**

- $t=2.727$ df=12 (only for DA)
- $t=0.5411$ df=16 (only for DA)

### 3e
Unpaired two-tailed t-test

**Legend page 29**

- $p=0.5959$ (only for DA)
- $t=0.5411$ df=16 (only for DA)

### 4a
One-way ANOVA, with Dunnett's multiple comparisons test

**Legend page 30**

- $q=4.832$ df=8 for Ret-Flag  
- $q=3.453$ df=8 for Ret-Flag + scrambled  
- $q=0.2174$ df=8 for Ret+shVav2 #1

### 4b
Two-way ANOVA, with Bonferroni's multiple comparisons test

**Legend page 30**

- $q=7.229$ df=9 for 100
- $q=4.009$ df=9 for 10

### 4c
Two-way ANOVA with Dunnett's multiple comparisons test

**Legend page 30**

- $q=2.942$ df=8 between scramble & shVav2 in control group

### 4d
Two-way ANOVA, with Bonferroni's post test

**Legend page 30**

- $t=0.2392$ df=9 between Control & GDNF in shVav2 group
- $t=3.085$ df=8 between Control & GDNF in scramble group

### 4g
Two-way ANOVA, with Bonferroni's multiple comparisons test

**Legend page 30**

- $t=0.9999$ between Control & GDNF in scramble group

### 6b
Two-way ANOVA, with Bonferroni's post test

**Legend page 31**

- $t=4.677$ df=8 between saline & cocaine in WT
- $t=7.803$ df=8 between saline & cocaine in Vav2 KO

### 6c
Two-way ANOVA, with Bonferroni's post test

**Legend page 31**

- $t=7.174$ df=24 between saline & cocaine in WT
- $t=0.7658$ df=24 between saline & cocaine in Vav2 KO

### 6d
Two-way ANOVA, with Bonferroni's multiple comparisons test

**Legend page 32**

- $t=3.599$ df=6 between saline & cocaine in WT
- $t=5.655$ df=6 between saline & cocaine in Vav2 KO

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Nature Neuroscience: doi:10.1038/nn.4060
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<th>#</th>
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<td>Samples from 12 pairs of mice</td>
<td>Mean +/- SEM</td>
<td>Legend page 32</td>
<td>p=0.0391 between saline &amp; cocaine in WT p=0.8013 between saline &amp; cocaine in Vav2 KO</td>
<td>F (1, 10) = 1.804 t=2.777 df=10 between saline &amp; cocaine in WT t=0.8778 df=10 between saline &amp; cocaine in Vav2 KO</td>
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<td>Samples from 16 pairs of mice</td>
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<td>p=0.0034 for WT: saline vs. WT: cocaine p=0.0120 for WT: saline vs. Vav2 KO saline p=0.0392 for WT: saline vs. Vav2 KO: cocaine</td>
<td>F (1, 28) = 8.455 t=3.623 df=15 for WT: saline vs. WT: cocaine t=3.135 df=28 for WT: saline vs. Vav2 KO: saline t=2.645 df=28 for WT: saline vs. Vav2 KO: cocaine</td>
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<td>4,3,6,6 mice/group</td>
<td>Mean +/- SEM</td>
<td>Legend page 32</td>
<td>p=0.0045 for WT: saline vs. WT: cocaine p=0.0090 for Vav3 KO: saline vs. Vav3 KO: cocaine</td>
<td>F (1, 15) = 0.8399 t=3.672 df=15 for WT: saline vs. WT: cocaine t=3.385 df=15 for Vav3 KO: saline vs. Vav3 KO: cocaine</td>
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</table>
| 7a | Unpaired two-tailed t-test                                                 | Fig.  | 17,18 mice per group                    | Mean +/- SEM  | Legend page 33     | p=0.0809                                                       | F (2, 53) = 13.48 P < 0.0001 F (1, 53) = 11.14 P=0.0015 df=19 t=2.242 df=16 |}

Nature Neuroscience: doi:10.1038/nn.4060
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<td>p=0.2182</td>
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<td>p=0.7670</td>
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**Notes:**
- *p* values are standardized for DA only or NAc.
- *t* values are mixed-effects or unpaired *t*-tests.
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### Representative figures

1. Are any representative images shown (including Western blots and immunohistochemistry/staining) in the paper?
   
   If so, what figure(s)?

   Fig. 1a, 1c, 1e, 1g, 1h, 2f, 2h, 2i, 3b, 3g, 3h, 3j, 3k, 4b, 4c, 4e, 4g, 5a-e, 6a, 6f.
   Suppl Fig. 1a, 1c, 1i, 1k, 2a, 2c, 2k, 2m, 3b, 4a-d, 6a-c.

   A clear statement of how many times this experiment is repeated is now shown in the each respective legend.

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 #)?
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.

2. Are statistical tests justified as appropriate for every figure?
   Where (section, paragraph #)?
   a. If there is a section summarizing the statistical methods in the methods, is the statistical test for each experiment clearly defined?
   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 #)?
   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 #)?
   d. Are tests specified as one- or two-sided?
   e. Are there adjustments for multiple comparisons?

3. Are criteria for excluding data points reported?
   Was this criterion established prior to data collection?
   Where is this described (section, paragraph #)?

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 #)?

For in vitro studies, samples size is at least triplicates in 3 independent experiments. For in vivo experiments, at least 5 mice in each group were used. This allows us to perform statistical analysis.
Sample sizes are shown in each respective legend.

Statistical test are justified for each figure.
The statistical method used is shown in each respective legend.

In the Supplementary Methods, a section 'Statistical analysis' described the statistical methods used in the study. For the statistical test used for each individual figure, the information is described in legends.

Yes, they do. They meet the normality tests. Please see the legends.

Mean +/- SEM is a way to describe the variance within each group of data. In most cases, the variance is similar between groups that are being statistically compared.
This is described in the Legends.

Mean +/- SEM is a way to describe the variance within each group of data. In most cases, the variance is similar between groups that are being statistically compared.
This is described in the Legends.

No.

In each experiments, appropriate positive and negative controls were set up and used to make judgment on whether the data is excluded or not. This is reflected in many of the figures shown.

No specific randomization was used.
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 #)?

The present study was performed in a collaborative manner among multiple members of the team, which makes single- or double-blind experiments feasible.

6. For experiments in live vertebrates, is a statement of compliance with ethical guidelines/regulations included? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

7. Is the species of the animals used reported? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

8. Is the strain of the animals (including background strains of KO/transgenic animals used) reported? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

9. Is the sex of the animals/subjects used reported? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

10. Is the age of the animals/subjects reported? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

11. For animals housed in a vivarium, is the light/dark cycle reported? Where (section, paragraph #)?

Yes. page 1, Animal section of the Supplemental Methods.

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

Yes. page 1, Animal section of the Supplemental Methods.

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

Page 17, line 9 in the Supplemental Methods.

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

Yes. page 1, Animal section of the Supplemental Methods.

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.
a. How were the criteria for exclusion defined?
Where is this described (section, paragraph #)?

In our microdialysis experiments, we found that occasionally a few of mice exhibited extremely high levels (e.g. more than 50-fold) of extracellular dopamine under basal conditions compared with the majority of normal mice, with no apparent reasons. In this case, these animals were excluded.

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 #)?

N/A. All animal survived well during the experiments, unless they were sacrificed by investigators.

Reagents

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

   Yes.
   Yes.
   DAT antibody: pg 4, line 3, Supplementary Methods. Vav2 antibody: pg4, line 8, Supplementary Methods.

2. If cell lines were used to reflect the properties of a particular tissue or disease state, is their source identified?
   Where (section, paragraph #)?
   a. Were they recently authenticated?
   Where is this information reported (section, paragraph #)?

   N/A
   N/A

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?
   Where (section, paragraph #)?

   N/A
## 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.  
   - N/A

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:

<p>| | |</p>
<table>
<thead>
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<tbody>
<tr>
<td>1.</td>
<td>Were any subjects scanned but then rejected for the analysis after the data was collected?</td>
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<tr>
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<td>N/A</td>
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<td>a. If yes, is the number rejected and reasons for rejection described?</td>
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<td>Where (section, paragraph #)?</td>
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<td>2.</td>
<td>Is the number of blocks, trials or experimental units per session and/or subjects specified?</td>
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<td>Where (section, paragraph #)?</td>
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<td>3.</td>
<td>Is the length of each trial and interval between trials specified?</td>
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<td>4.</td>
<td>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.</td>
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<td>5.</td>
<td>Is the task design clearly described?</td>
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<td>6.</td>
<td>How was behavioral performance measured?</td>
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<td>7.</td>
<td>Is an ANOVA or factorial design being used?</td>
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<td>8.</td>
<td>For data acquisition, is a whole brain scan used?</td>
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<td>If not, state area of acquisition.</td>
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<td>a. How was this region determined?</td>
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<td>9.</td>
<td>Is the field strength (in Tesla) of the MRI system stated?</td>
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<td>a. Is the pulse sequence type (gradient/spin echo, EPI/echo) stated?</td>
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<td>b. Are the field-of-view, matrix size, slice thickness, and TE/TR/flip angle clearly stated?</td>
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<tr>
<td>10.</td>
<td>Are the software and specific parameters (model/functions, smoothing kernel size if applicable, etc.) used for data processing and pre-processing clearly stated?</td>
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</tbody>
</table>

Nature Neuroscience: doi:10.1038/nn.4060
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 #)?

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 #)?

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

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

15. Is the contrast construction clearly defined?

16. Is a mixed/random effects or fixed inference used?
   a. If fixed effects inference used, is this justified?

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

19. Are statistical inferences corrected for multiple comparisons?
   a. If not, is this labeled as uncorrected?

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

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

Additional Comments