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Reporting Summary

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Statistical parameters

When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. figure legend, table legend, main text, or Methods section).

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	An indication of whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☐	☒	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistics including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated
☐	☒	Clearly defined error bars *State explicitly what error bars represent (e.g. SD, SE, CI)*

Our web collection on statistics for biologists may be useful.

Software and code

Policy information about availability of computer code

Data collection	MATLAB (R2015a,R2016a), Psych toolbox (v. 3), Plexon recorder (v.2.3), Bruker's Prairie software (v.5.3), MultiClamp (v.700b), pClamp (v.10), Leica Application Suite X (v.3.1.5.16308), LifeCam Software Microsoft (v.3.60.253.0)
Data analysis	MATLAB (R2015a,R2016a), Plexon Offline Sorter (v.2.8.8), Mountain Sort Algorithm (v.1.0.0), ImageJ (v.1.48), Clampfit (v.10.4)

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors/reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

Data available on request from the authors

Field-specific reporting

Please select the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/authors/policies/ReportingSummary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	No statistical methods were used to pre-determine sample size. We used sample sizes similar to literature in the field. For all experiments, we also used sample sizes to provide at least 80% power to detect an effect.
Data exclusions	For single unit recording in the LC: Upon completion of experiments, we verified that targeting of the locus coeruleus region was successful with immunohistochemical techniques. Experiments where electrodes, fiber optics or viral delivery were mis-targeted were excluded from analysis. Recordings session with no ChR2 responsive units were also excluded from the study. We selected portion of extracellular single unit recordings where no obvious drift was detected. For slice electrophysiology: Recordings with a variation of access resistance greater than 15% were exclude. For pupillometry experiment: we excluded trials where baseline pupil size was hyper-dilated or constricted from analysis (<1st or >99th percentile of pupil size distribution). For histology using monosynaptic rabies tracing: Regions adjacent to LC were not considered for analysis due to non-specific expression of virus at the site of injection. These criteria were not pre-established.
Replication	All experiments were reproduced using biological replicates. Attempts at reproduction were successful. We used a range of 3 to 13 mice per conditions for each experiments.
Randomization	Auditory stimuli, and timing of optogenetics activation was randomized. Male or female mice were randomly selected for each experiment.
Blinding	Data collection and analysis was not performed blind. Sorting of neuronal type was performed after data collection.

Reporting for specific materials, systems and methods

Materials & experimental systems

n/a	Involved in the study
☒	☐ Unique biological materials
☐	☒ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology
☐	☒ Animals and other organisms
☒	☐ Human research participants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used	Primary antibodies: Chicken anti-tyrosine hydroxylase (TYH, Aves Labs, lot no. TYH8727985), dilution 1:1000 Rabbit anti-VGAT (131002, Synaptic Systems, lot no. 131002/34), dilution 1:1000 Rabbit anti-GABA (A2052, Sigma, lot no. 126M4791V), dilution 1:1000 Mouse anti-GAD67 (MAB5406 EMD Millipore, lot no. 2923238), dilution 1:200 Rabbit anti-neuropeptide S (ab18252 Abcam), dilution 1:500 Secondary antibodies Streptavidin-488 conjugated antibodies (S32354 ThermoFisher Scientific), dilution 1:200 Goat anti-chicken 647 nm (A21449, ThermoFisher Scientific), dilution 1:500 Goat anti-chicken 488 nm (A11039, ThermoFisher Scientific), dilution 1:500 Goat anti-rabbit 488 nm (A11034, ThermoFisher Scientific), dilution 1:500 Goat anti-mouse 488 nm (A21121, ThermoFisher Scientific), dilution 1:500

Validation

Chicken anti-tyrosine hydroxylase (TYH, Aves Labs)
Validated in: Carter, M.E., et al. Tuning arousal with optogenetic modulation of locus coeruleus neurons. Nature neuroscience 13, 1526-1533 (2010).

Rabbit anti-VGAT (131002, Synaptic Systems),
Validated in: Saunders A, Oldenburg IA, Berezovskii VK, Johnson CA, Kingery ND, Elliott HL, Xie T, Gerfen CR & Sabatini BL (2015). A direct GABAergic output from the basal ganglia to frontal cortex. Nature 521: 85-9. 131 011;

Rabbit anti-GABA (A2052, Sigma)
Validated. R.O. Tasan, A. Bukovac,a Y.N. Peterschmitt, S.B. Sartori, R. Landgraf, N. Singewald, and G. Sperka Altered GABA transmission in a mouse model of increased trait anxiety. Neuroscience. 2011 Jun 2; 183(7): 71–80.

Mouse anti-GAD67 (MAB5406 EMD Millipore)
Validated in J Dimidschstein, Q Chen, R Tremblay, SL Rogers, GA Saldi, et. al. A viral strategy for targeting and manipulating interneurons across vertebrate species. Nature Neuroscience 2016 Dec; 19(2): 1743-1749.

Rabbit anti-neuropeptide S (ab18252 Abcam)
Validated in X Liu, J Zeng, A Zhou, E Theodorsson, J Fahrenkrug, RK Reinscheid. Molecular fingerprint of neuropeptide S-producing neurons in the mouse brain. J Comp Neurol. 2011 Jul 1;519(10):1847-66.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals

Adult mice (> 2 month old) of either sex were used in this study. We used the following mouse lines for the specific expression of various viruses in noradrenergic, GABAergic: TH-Cre (B6.Cg-Tg(Th-cre)1Tmd/J, Jackson Laboratory), Dbh-Cre (B6.FVB(Cg)-Tg(Dbh-cre)KH212Gsat/Mmucd, MMRRC), , GAD2-Cre (Gad2tm2(cre)Zjh/J, Jackson Laboratory). C57Bl/6 wild-type mice were used for control experiments. Optogenetic activation of LC GABAergic neurons (LC-GABA) was done also on VGAT-YFP-ChR2 (B6.Cg-Tg(Slc32a1-COP4*H134R/EYFP)8Gfng/J, Jackson Laboratory).

Wild animals

This study does not involve wild animals.

Field-collected samples

This study does not include field-collected samples.