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Last updated by author(s): 10/01/2025

Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Portfolio policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the 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
☐	☒ A statement on 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 statistical parameters 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

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Open-NFT software (http://opennft.org/) for real-time fMRI-Neurofeedback, and in-house online analysis scripts that will be shared before publication via github (https://github.com/nitzanlubi/nf_immune_study)
Data analysis	Python version 3.8.12 and R version 4.2.1. for fMRI preprocessing: fMRIprep version 20.0.2 and FMRIB software library (FSL). Python packages: Scipy package version 1.10.1; scikit-learn package version 1.6.1; nilearn package, version 0.11.1; Code is available via Github: https://github.com/nitzanlubi/nf_immune_study

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 and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio 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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

Requests for de-identified data can be directed at the corresponding author. All requests for data sharing will be reviewed by the Tel-Aviv Sourasky Medical Center

Institutional Review Board (IRB) committee, to verify whether the request is subject to any intellectual property or confidentiality obligations. Requests will be reviewed on the basis of scientific merit, ethical review, available resources and regulatory requirements, and will be responded within 90 days. After approval of a proposal, anonymized individual-level data will be made available for reuse in accordance with the signed consent IRB form. A signed data access agreement with the collaborator is required before accessing shared data.

Research involving human participants, their data, or biological material

Policy information about studies with human participants or human data. See also policy information about sex, gender (identity/presentation), and sexual orientation and race, ethnicity and racism.

Reporting on sex and gender	Sex was determined according to self reporting, and is specified per study group in demographics data in the manuscript. Sex-based analyses were not conducted due to low sample sizes. Gender information was not collected.
Reporting on race, ethnicity, or other socially relevant groupings	Data on race, ethnicity or other social groupings was not collected as part of this study.
Population characteristics	Healthy subjects aged 18-45 that intended to receive HBV vaccination from personal reasons were included in the study (60% females).
Recruitment	Subjects were recruited through approved advertisements, social media, dedicated forums, and bulletin boards. All subjects that met inclusion and exclusion criteria were included in the study. No self-selection bias was expected to impact results.
Ethics oversight	the Tel-Aviv Sourasky Medical Center Institutional Review Board (IRB) committee

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one below that is 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/documents/nr-reporting-summary-flat.pdf

Life sciences study design

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

Sample size	To achieve sufficient statistical power (80%) for our study design, the various expected effects were analysed separately. Results indicated that the most demanding hypothesis, the existence of a moderate correlation (r=0.3) between reward mesolimbic activations during NF training and immunological outcome measures, requires at least 67 data points. As this correlation could be assessed based on subjects from both NF groups, NF group sizes were set to 34 each (with 17 subjects in the no-NF control group, resulting in a 2:2:1 ratio design).
Data exclusions	Neural data: individual runs with excessive head movements were excluded. Runs with more than 20% Framewise-Displacement >0.5 were considered as excessive. for the NF task, no data reached this threshold. For the Monetary Incentive Delay functional localizer task, one subject was excluded based on this criterion. This criterion was not prespecified in the pre-registration, but is a standard stage in fMRI preprocessing and artifacts removal procedures. Immunological data: Twelve subjects (evenly dispersed between groups) did not respond to HBV vaccination according to the standard clinical immunization threshold of HBVab levels below 10 mU/ml) and were therefore excluded from further analyses. This data exclusion criterion was no pre-registered. Nonetheless, as the lack of an immunological response to HBV vaccination is a phenomenon that was documented in the literature (see Qiu, S. et al. https://doi.org/10.1080/21645515.2018.1450122), we assumed our vaccination-induced manipulation failed for these subjects regardless of NF training (indeed, 5 out of 12 were from the no-NF group). Therefore, non-responders were excluded from any immunological or brain-immune association analyses.
Replication	Neural preprocessing was conducted via fMRI-prep, a standardized fMRI analysis software aimed at maximizing reproducability of results. All neural preprocessing and analysis and immunological steps that are required in order to replicate results have been reported in the methods section. All code and outcome variables data is shared via github. All methods including softwares and packages versions have been described in the main text to ensure reproducibility. The findings were not replicated in an additional study.
Randomization	Subjects were randomly assigned to one of three study groups via an in-house Matlab function.
Blinding	Unlike most fMRI-NF studies, both participants and experimenters were blinded to NF group allocation throughout the whole experiment. Specifically, the theoretical hypotheses of the study (the link between motivational and reward-related processes and immune functions) were withheld from subjects, as were the types of neural targets for each group. Therefore, contrary to standard NF training protocols, instructions before training did not mention the specific neural target or its functional definition, nor were preferred process-specific mental strategies mentioned. To ensure the blinding of the experimenter to NF group allocation, the real-time NF software (OpenNFT) was adapted so as to not present ROIs on the experimenter screen. Instead, ROI creation and integration into the NF software were performed algorithmically in a completely blinded fashion, which did not reveal group allocation to experimenters.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☒	☐ Animals and other organisms
☐	☒ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

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

Antibodies

Antibodies used: Hepatitis B surface antigen (anti-HBs) were determined, using a Chemiluminescent Microparticle Immunoassay (CMIA)

Validation: Antibody titers to Hepatitis B surface antigen (anti-HBs) were determined, using a Chemiluminescent Microparticle Immunoassay (CMIA) located in the Virology lab at the Rambam Health Care Campus. In any case antibody titers exceeded the maximal detection range (1000 mU/ml), the sample was diluted in antibody-negative plasma (at a ratio of 1:10-1000). The samples were also quantitatively tested for hepatitis B surface antigen (HBsAg), to exclude any participant who might have been infected with Hepatitis B at the time of vaccination. No participant was excluded based on this criterion.

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration: https://clinicaltrials.gov/study/NCT03951870

Study protocol: The study protocol is included in the submission

Data collection: All experimental sessions and data collection were conducted at the Tel-Aviv Sourasky Medical Center. Recruitment and data collection began in January, 2020, and ended in August, 2022, when reaching the pre-registered N.

Outcomes: The study had one primary neural outcome: reward-mesolimbic ROIs (VTA and Nucleus Accumbens) regulation effects, measured as the difference in BOLD activity between 'upregulation' and 'rest' epochs during NF task across experimental sessions; And one primary immunological outcome: post-vaccination HBV antibodies titer change, measured as the difference between mean log baseline measures (session 1 and 4) and mean post-vaccination measurements 14 and 28 days following HBV vaccination. (session 5 and 6) Secondary outcome measure were long-term HBV antibodies levels measured in a subgroup of 34 subjects 3 months following HBV vaccination (session 7); and measures that could potentially mediate or influence the main neural and immunological outcome measures: neurobehavioural responses to reward anticipation and outcome as measured in the Monetary Incentive Delay task; Effort Expenditure levles as measured in the EEFRT; and motivational tendencies as characterized by K-means clustering of motivational and personality questionnaires.

Plants

Seed stocks: *Report on the source of all seed stocks or other plant material used. If applicable, state the seed stock centre and catalogue number. If plant specimens were collected from the field, describe the collection location, date and sampling procedures.*

Novel plant genotypes: *Describe the methods by which all novel plant genotypes were produced. This includes those generated by transgenic approaches, gene editing, chemical/radiation-based mutagenesis and hybridization. For transgenic lines, describe the transformation method, the number of independent lines analyzed and the generation upon which experiments were performed. For gene-edited lines, describe the editor used, the endogenous sequence targeted for editing, the targeting guide RNA sequence (if applicable) and how the editor was applied.*

Authentication: *Describe any authentication procedures for each seed stock used or novel genotype generated. Describe any experiments used to assess the effect of a mutation and, where applicable, how potential secondary effects (e.g. second site T-DNA insertions, mosiacism, off-target gene editing) were examined.*

Magnetic resonance imaging

Experimental design

Design type	Block design task fMRI (Neurofeedback task)
Design specifications	NF participants performed 8 to 11 fMRI-NF runs (each run consists of five consecutive trials) across three or four fMRI NF training sessions (sessions 1 to 4). During each NF trial, participants were instructed to (i) choose via a key press one out of four mental strategies (predefined by participants before each NF run) for the upcoming 'Upregulate' screen (8 seconds), (ii) rest while passively watching the red dot on the screen (20 seconds), (iii) upregulate neural target activity by employing their chosen strategy (for example, recalling a trip), (40 seconds) and (iv) receive numeric (1-10) and graphic (smiley) feedback proportional to regulation success (difference in neural target activity between the current trial's 'Upregulate' and 'Rest' epochs) (8 seconds). Each run lasted approximately 8 minutes.
Behavioral performance measures	subjects reported their chosen mental strategies via key-presses, but these responses were not used as performance measures.

Acquisition

Imaging type(s)	functional and structural MRI
Field strength	3T
Sequence & imaging parameters	Functional whole-brain scans were performed in interleaved order with a T2*-weighted gradient multi-echo planar imaging pulse sequence (TR/TE=2000/35ms, IPAT acceleration factor=2; flip angle=84º, FOV=220×220mm, matrix size = 96*96, voxel size: 2X2 mm, slice thickness = 2mm, no gap, 56 slices per volume).
Area of acquisition	Whole-brain

Diffusion MRI ☐ Used ☒ Not used

Preprocessing

Preprocessing software	fMRIprep software package (version 20.0.2). Full software-produced automatic report of all parameters and preprocessing steps is reported in the manuscript.
Normalization	Volume-based spatial normalization to one standard space (MNI152NLin2009cAsym) was performed through nonlinear registration with `antsRegistration` (ANTs 2.2.0), using brain-extracted versions of both T1w reference and the T1w template. The following template was selected for spatial normalization: ICBM 152 Nonlinear Asymmetrical template version 2009c;
Normalization template	Template used for anatomical alignment and registration: MNI152NLin2009cAsym.
Noise and artifact removal	The following nuisance regressors were included: six head movement regressors (translation and rotation), framewise displacement predictor, first six anatomical CompCors, as well as mean CSF and white matter global signal predictors.
Volume censoring	We introduced scrubbing regressors into the noise model based on framewise displacement variable, regressing out time points with FD > 0.5.

Statistical modeling & inference

Model type and settings	Time-series statistical analysis was carried out using FILM with local autocorrelation correction21. The general linear model of a NF run included regressors of interest of the four task conditions: (1) Choice (8 s), (2) Rest (20 s), (3) Upregulate (40 s), and (4) feedback (8 s). To account for noise, confound regressors from the fMRIprep preprocessing were included as covariates of no interest. The three main Contrasts of Parameter Estimates (copes) were then computed: "Choice", which represented activity patterns during choice of mental strategies; "Regulate>Rest", which represented regulation effects – the difference in activity patterns between Upregulate and Rest screens; and "Feedback", which represented activity pattern during rewarding feedback regarding regulation success during feedback screens. Effects of each session were averaged using a fixed effects model, by forcing the random effects variance to zero in FLAME (FMRIB's Local Analysis of Mixed Effects)21–23. Finally, group-level effects for session 4 were carried out using FEAT: Z (Gaussianised T/F) statistic images were thresholded using clusters determined by Z>3.1 and a (corrected) cluster significance threshold of P=0.05.
Effect(s) tested	Analysis of Mixed Effects was conducted for "Regulate>Rest" contrast, which represented regulation effects – the difference in activity patterns between Upregulate and Rest screens.

Specify type of analysis: ☐ Whole brain ☐ ROI-based ☒ Both

Anatomical location(s)	All ROIs were defined based on a pre-registered protocol. the reward-mesolimbic ROIs (the VTA and Nac were defined by intersecting a meta-analysis based mask with functional localizer-based task activations during MID task, as specified in the pre-registration and in the Methods section. Control regions used for online feedback calculation for the NF control group were defined anatomically as 5 mm spheres around peak activaiton reported in process-specific meta-analyses. All MNI coordinates for reward-,esolimbic and control regions masks have been pre-registerd.
Statistic type for inference (See Eklund et al. 2016)	Both voxel-wise and cluster-wise corrections were applied in the production of whole-brain images. voxel-wise and and cluster-wise correction was conducted through FSL's Feat custom pipeline with the deafult parameters. All parameters and corrections are reported in the manuscript.
Correction	Z (Gaussianised T/F) statistic images were thresholded using clusters determined by Z>3.1 and a (FWE corrected) cluster significance threshold of P=0.05.

Models & analysis

n/a	Involved in the study
☐	☒ Functional and/or effective connectivity
☒	☐ Graph analysis
☒	☐ Multivariate modeling or predictive analysis

Functional and/or effective connectivity	We appled generalized Psychophysiological Interaction (gPPI) analysis between the VTA and selected regions of interest. VTA BOLD activity was deconvolved and used a regressor in a GLM model to estimate task functional connectivity during task conditions. Then ROI measures were extracted and correlated with other effects of interests using Pearson's correlations.