

Reporting Summary

Nature Research 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 Research policies, see [Authors & Referees](#) and the [Editorial Policy Checklist](#).

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

BD FACSDIVA v.6.2 was used for flow cytometry data collection

Data analysis

All the softwares used and their version information, when available, are shown here. Flow-Jo (v10.1) was used for all FACS analyses. IMGT (v3.4.8) was used to analyze TCR V β sequences obtained by Sanger method. ImmunoSEQ Analyzer V3.0 (<http://www.immunoseq.com>) was used for data processing of TCR V β CDR3 sequencing performed at Adaptive Biotechnologies. Scripts written in Java by one of the authors (Mathilde Foglierini) were used to define antigen-specific clonotypes in each donor's repertoire according to identical amino acid sequence in the V β CDR3 and identical V gene usage. The Java scripts are available at <https://bitbucket.org/mathildefog/narcolepsy>. This code is distributed open source under the terms of the GNU Free Documentation License. Graphpad Prism (v7) was used to analyze data and create plots. The Microbeta2 Windows Workstation software, version 2.0.0.87, was used to acquire data of T cell proliferation by [3H]-thymidine incorporation on a Beta counter 2 (Perkin Elmer).

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 availability. The data presented in this manuscript are tabulated in the main paper and in the supplementary materials. TCR sequences from samples listed in Supplementary Table 2 are available as .txt file in Supplementary Information. The sequences have been deposited in the immuneACCESS™ database (URL <http://clients.adaptivebiotech.com/pub/Latorre-2018-nature>, doi:10.21417/B73H0P).

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 sample size calculation was performed. Narcolepsy is a rare disease and we included in the study patients enrolled in different centers in Switzerland.
Data exclusions	Data obtained from healthy controls were included only if there was a T cell response to antigens used as positive controls (Flu antigens for CD4+ T cells and CMV+EBV antigens for CD8+ T cells).
Replication	In a few cases it was possible to analyze separate samples from the same patient with comparable results. In the cases of T cell clones, their reactivity against self-antigens was assessed in multiple occasions (in at least 3 separate experiments) with reproducible results.
Randomization	Narcolepsy patients with HCRT deficiency (CSF HCRT levels < 110 pg/mL) and clinical diagnosis of narcolepsy with cataplexy were allocated to NT1 patients group. Narcolepsy patients with intermediate or normal HCRT levels and clinical diagnosis of narcolepsy with mild or without cataplexy were allocated to NT2 patients group. Patients were 16–53 years of age (median 32) and had different disease duration (range 1–36, median 7 years). Healthy controls were obtained from University Hospital of Bern (of which 1 was a 1st degree family member) and the Swiss Blood Donation Center of Lugano and selected for being HLA-DQB1*0602 positive.
Blinding	Blinding of the investigators was not applied for the screening of autoreactive cells in NT versus C groups, because the healthy donors were included later in the study. The investigators were unaware of the allocation of patients to the NT1 and NT2 groups.

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

Unique biological materials

Policy information about [availability of materials](#)

Obtaining unique materials

Antibodies

Antibodies used

All the commercial antibodies were used following the manufacturer's instructions. Supplier name, catalog number and clone name are indicated here and in the Methods section.

CD4-PE-Texas Red, clone S3.5 from ThermoFisher Scientific, cat n MHCD0417, Lot#1710439A, dil 1:800.

CD45RA-QD655, clone MEM-56 from ThermoFisher Scientific, cat n Q10069, Lot#1785496, dil 1:1000.

CD8-APC, clone B9.11 from Beckman Coulter, cat n IM2469, Lot#51, dil 1:30.

CD14-PE-Cy5, clone RMO52 from Beckman Coulter, cat n A07765, Lot#23, dil 1:30.

CD56-PE-Cy5, clone N901 from Beckman Coulter, cat n A07789, Lot#42, dil 1:30.

CD19-FITC, clone HIB19 from BD Bioscience, cat n 555412, Lot#5097663, dil 1:30.

CD25-PE, clone M-A251 from BD Bioscience, cat n 555432, Lot#5314982, dil 1:20.

CCR7-BV421, clone G043H7 from Biolegend, cat n 353207, Lot#B233487, dil 1:80.

CD19-PE-Cy7, clone SJ25C1 from BD Bioscience, cat n 341113, Lot#7048931, dil 1:30.

CD3-PE, clone UCHT1 from Beckman Coulter, cat n A07747, Lot#26, dil 1:30.

ICOS-Pacific Blue clone C398.4A from Biolegend, cat n 313522, Lot#B170453, dil 1:100.

antiHLA-DR, clone L243, ATCC cat n HB55, produced in house from hybridoma cell line, dil 1:20.

anti-HLA-DQ clone SPVL3 (1), produced in house from hybridoma cell line, dil 1:20.

anti-HLA-DP, clone B7/21 (2), produced in house from hybridoma cell line, dil 1:20.

pan anti-MHC class I, clone W6/32, ATCC cat n HB-95, produced in house from hybridoma cell line, dil 1:20

1) Spits H; Keizer G; Borst J; Terhorst C; Hekman A; de Vries JE. Characterization of monoclonal antibodies against cell surface molecules associated with cytotoxic activity of natural and activated killer cells and cloned CTL lines. Hybridoma, 1983, 2(4):423-37.

2) Watson AJ, DeMars R, Trowbridge IS, Bach FH. Detection of a novel human class II HLA antigen. Nature, 1983, 304:358-61.

Validation

Describe the validation of each primary antibody for the species and application, noting any validation statements on the manufacturer's website, relevant citations, antibody profiles in online databases, or data provided in the manuscript.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)

MHC-class II-negative Type II bare lymphocyte syndrome (BLS II) cells transfected with DRB1*15:01 (DR2b: DRB1*15:01, DRA*01:01) or DRB5*01:01 (DR2a: DRB5*01:01, DRA*01:01) were used as antigen presenting cells (kindly provided by G. Nepom and W. Kwok, University of Washington, Seattle, USA). B and T cell lines were generated from primary human cells and kept short term in culture. EBV-transformed B lymphoblastoid cell lines were generated and used as APCs. Allogeneic PMBCs were used as feeder cells and were isolated from healthy donors obtained from the Swiss Blood Donation Center of Lugano.

Authentication

BSL II cell lines transfected with DRB1*15:01 or DRB5*01:01 were typed for the presence of DRB1*15:01 or DRB5*01:01 allele and for the absence of the DQB1*06:02 allele by PCR amplification. >99% of the cells expressed HLA-class II molecules on their surface as assessed by flow cytometry.

Mycoplasma contamination

EBV-B lymphoblastoid cell lines are routinely tested for mycoplasma contamination and found to be negative.

Commonly misidentified lines (See [ICLAC](#) register)

No cell lines listed in the ICLAC database have been used in the study.

Human research participants

Policy information about studies involving human research participants

Population characteristics

The study included:

- 16 NT1 with HCRT deficiency (CSF HCRT levels < 110 pg/mL) and clinical diagnosis of narcolepsy with cataplexy (narcolepsy type 1, NT1), of which 14 carried the disease-associated HLA-DQB1*06:02 allele;
- 3 NT2 patients, HLA-DQB1*06:02-negative with a clinical diagnosis of narcolepsy with mild or without cataplexy and intermediate or normal HCRT levels (narcolepsy type 2, NT2);
- 13 healthy donors, HLA-DQB1*06:02-positive

Patients were 16–53 years of age (median 32) and had different disease duration (range 1–36, median 7 years). One NT1 patient (P10) suffered also from multiple sclerosis. All healthy donors and 14 out of 19 patients were HLA-DQB1*06:02 positive. All patients included in the study were HLA-typed at the Policlinico San Matteo, University of Pavia. Patients were recruited from University Hospital of Bern, Hospital of Lugano (EOC) and Clinic Barmelweid and healthy donors were obtained from University Hospital of Bern (of which 1 was a 1st degree family member) and the Swiss Blood Donation Center of Lugano. All participants provided written informed consent for participation in the study.

Additional information on human research participants such as age, gender, disease, HLA typing and HCRT levels in the CSF are provided in Extended Data Table 1 and Extended Data Table 3.

Recruitment

The patients were recruited at the University Hospital of Bern, Hospital of Lugano (EOC) and Clinic Barmelweid. Healthy donors were recruited at the University Hospital of Bern and the Swiss Blood Donation Center of Lugano. All participants provided written informed consent for participation in the study. The study was approved by the Ethical committees of Bern (n. 054/15),

ChIP-seq

Data deposition

- ☐ Confirm that both raw and final processed data have been deposited in a public database such as [GEO](#).
- ☐ Confirm that you have deposited or provided access to graph files (e.g. BED files) for the called peaks.

Data access links

May remain private before publication.

For "Initial submission" or "Revised version" documents, provide reviewer access links. For your "Final submission" document, provide a link to the deposited data.

Files in database submission

Provide a list of all files available in the database submission.

Genome browser session

(e.g. [UCSC](#))

Provide a link to an anonymized genome browser session for "Initial submission" and "Revised version" documents only, to enable peer review. Write "no longer applicable" for "Final submission" documents.

Methodology

Replicates

Describe the experimental replicates, specifying number, type and replicate agreement.

Sequencing depth

Describe the sequencing depth for each experiment, providing the total number of reads, uniquely mapped reads, length of reads and whether they were paired- or single-end.

Antibodies

Describe the antibodies used for the ChIP-seq experiments; as applicable, provide supplier name, catalog number, clone name, and lot number.

Peak calling parameters

Specify the command line program and parameters used for read mapping and peak calling, including the ChIP, control and index files used.

Data quality

Describe the methods used to ensure data quality in full detail, including how many peaks are at FDR 5% and above 5-fold enrichment.

Software

Describe the software used to collect and analyze the ChIP-seq data. For custom code that has been deposited into a community repository, provide accession details.

Flow Cytometry

Plots

Confirm that:

- ☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).
- ☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).
- ☒ All plots are contour plots with outliers or pseudocolor plots.
- ☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation

Memory T cells were enriched from PBMCs of narcolepsy patients and healthy controls by positive selection using microbeads. CD4+ and CD8+ total memory cells were further sorted with BD FACSAria excluding CD8+ or CD4+ cells, and CD45RA+, CD25bright, CD14+ and CD56+ cells.

Instrument

BD FACSAria 335119
BD LSRFortessa 647788

Software

FACS Diva (version 6.2) was used for acquisition of samples and Flow-Jo (version 10.1) was used for all the FACS analyses.

Cell population abundance

Purity of the relevant cell populations (CD4+ and CD8+ memory T cells) was checked after sorting at BD LSRFortessa instrument and found to be >98%.

Gating strategy

CD4+ and CD8+ total memory cells were sorted from PBMC using a BD FACSAria III instrument as CD4+ (or CD8+), CD45RA-, CD14-, CD25-/lo, CD14-, CD56- and CD8- (or CD4-) cells. CD4+ and CD8+ T cells from CSF were sorted from T cell lines expanded in vitro using a BD FACSAria III instrument as CD3+CD4+CD8-CD19-CD56- or CD3+ CD8+CD4-CD19-CD56- cells.

- ☐ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.

Magnetic resonance imaging

Experimental design

Design type	<i>Indicate task or resting state; event-related or block design.</i>
Design specifications	<i>Specify the number of blocks, trials or experimental units per session and/or subject, and specify the length of each trial or block (if trials are blocked) and interval between trials.</i>
Behavioral performance measures	<i>State number and/or type of variables recorded (e.g. correct button press, response time) and what statistics were used to establish that the subjects were performing the task as expected (e.g. mean, range, and/or standard deviation across subjects).</i>

Acquisition

Imaging type(s)	<i>Specify: functional, structural, diffusion, perfusion.</i>
Field strength	<i>Specify in Tesla</i>
Sequence & imaging parameters	<i>Specify the pulse sequence type (gradient echo, spin echo, etc.), imaging type (EPI, spiral, etc.), field of view, matrix size, slice thickness, orientation and TE/TR/flip angle.</i>
Area of acquisition	<i>State whether a whole brain scan was used OR define the area of acquisition, describing how the region was determined.</i>
Diffusion MRI	☐ Used ☐ Not used

Preprocessing

Preprocessing software	<i>Provide detail on software version and revision number and on specific parameters (model/functions, brain extraction, segmentation, smoothing kernel size, etc.).</i>
Normalization	<i>If data were normalized/standardized, describe the approach(es): specify linear or non-linear and define image types used for transformation OR indicate that data were not normalized and explain rationale for lack of normalization.</i>
Normalization template	<i>Describe the template used for normalization/transformation, specifying subject space or group standardized space (e.g. original Talairach, MNI305, ICBM152) OR indicate that the data were not normalized.</i>
Noise and artifact removal	<i>Describe your procedure(s) for artifact and structured noise removal, specifying motion parameters, tissue signals and physiological signals (heart rate, respiration).</i>
Volume censoring	<i>Define your software and/or method and criteria for volume censoring, and state the extent of such censoring.</i>

Statistical modeling & inference

Model type and settings	<i>Specify type (mass univariate, multivariate, RSA, predictive, etc.) and describe essential details of the model at the first and second levels (e.g. fixed, random or mixed effects; drift or auto-correlation).</i>
Effect(s) tested	<i>Define precise effect in terms of the task or stimulus conditions instead of psychological concepts and indicate whether ANOVA or factorial designs were used.</i>
Specify type of analysis:	☐ Whole brain ☐ ROI-based ☐ Both
Statistic type for inference (See Eklund et al. 2016)	<i>Specify voxel-wise or cluster-wise and report all relevant parameters for cluster-wise methods.</i>
Correction	<i>Describe the type of correction and how it is obtained for multiple comparisons (e.g. FWE, FDR, permutation or Monte Carlo).</i>

Models & analysis

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