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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	Laser confocal imaging system (LSM 900 with Airyscan 2, Zeiss); Dynamic light scattering analyser (Nano-ZS90, Malvern); Automatic chemiluminescence imaging analysis system (5200, Tanon); Fluorescence microscope (Cytation 5, BioTek); Flow cytometry (CytoFLEX S, Beckman Coulter); IVIS spectrum imaging system (IVIS Kinetic, Calliper Life Sciences).
Data analysis	FlowJo (V10) was used for flow cytometry data analysis; GraphPad Prism 8 were used for statistical analysis; The histological section images were analyzed using CaseViewer 2.4.

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 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

The raw data generated in this study are provided in the Supplementary Information/Source Data file. The protein mass spectrometry raw data have been

deposited to the ProteomeXchange Consortium via the iProX repository with the data set identifier Project ID IPX0009378000 [https://www.iprox.cn//page/project.html?id=IPX0009378000]. Source data are provided with this paper.

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	*Use the terms sex (biological attribute) and gender (shaped by social and cultural circumstances) carefully in order to avoid confusing both terms. Indicate if findings apply to only one sex or gender; describe whether sex and gender were considered in study design; whether sex and/or gender was determined based on self-reporting or assigned and methods used.* *Provide in the source data disaggregated sex and gender data, where this information has been collected, and if consent has been obtained for sharing of individual-level data; provide overall numbers in this Reporting Summary. Please state if this information has not been collected.* *Report sex- and gender-based analyses where performed, justify reasons for lack of sex- and gender-based analysis.*
Reporting on race, ethnicity, or other socially relevant groupings	*Please specify the socially constructed or socially relevant categorization variable(s) used in your manuscript and explain why they were used. Please note that such variables should not be used as proxies for other socially constructed/relevant variables (for example, race or ethnicity should not be used as a proxy for socioeconomic status).* *Provide clear definitions of the relevant terms used, how they were provided (by the participants/respondents, the researchers, or third parties), and the method(s) used to classify people into the different categories (e.g. self-report, census or administrative data, social media data, etc.)* *Please provide details about how you controlled for confounding variables in your analyses.*
Population characteristics	*Describe the covariate-relevant population characteristics of the human research participants (e.g. age, genotypic information, past and current diagnosis and treatment categories). If you filled out the behavioural & social sciences study design questions and have nothing to add here, write "See above."*
Recruitment	*Describe how participants were recruited. Outline any potential self-selection bias or other biases that may be present and how these are likely to impact results.*
Ethics oversight	*Identify the organization(s) that approved the study protocol.*

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	Sample size are provided in the figure legends for each experiment and reasonable sample sizes were chosen to ensure they are sufficient for statistical comparison between different groups. For in vitro experiments and analysis, biologically independent experiments of 3 was used to detect a significant difference between different groups. For analysing the biodistribution and tumour accumulation in vivo, 5 mice of each group were used for in vivo imaging and then the mice were euthanized for ex vivo imaging. The in vivo efficacy studies in an H22 tumour-bearing mouse model and B16F10 tumour-bearing mouse model were performed with 6 mice per group. For the immune memory experiment, 5 mice per group were used. For the antitumour activity in an orthotopic H22 tumour model, 5 mice per group were used. Details regarding sample size of all experiments are provided in the methods section and figure legends. In addition, the sample sizes of this study were determined on the basis of similar published studies (Nat Nanotechnol. doi:10.1038/s41565-021-00972-7).
Data exclusions	No data was excluded from the analyses.
Replication	The in vitro experiments were replicated independently for at least 3 times. All in vivo studies were replicated at least 5 biologically independent mice per group. The number of replicates is detailed in the caption of each figures in the main manuscript and supplementary information files. Experiments were repeated and experimental findings were reproducible.
Randomization	In vitro experiments, the cultured cells were randomly assigned to experimental groups. In vivo experiments, the mice were randomized into different groups before treatment.
Blinding	No formal blinding was used due to all data were acquired and analyzed by software with objective standard.

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

1. PerCP/Cyanine5.5 anti-mouse/human CD11b antibody (BioLegend; Catalog number: 101228; Clone name: M1/70; 1:200 dilution)
2. Brilliant Violet 421™ anti-mouse F4/80 antibody (BioLegend; Catalog number: 123132; Clone name: BM8; 1:200 dilution)
3. Alexa Fluor® 647 anti-mouse F4/80 antibody (BioLegend; Catalog number: 123122; Clone name: BM8; 1:400 dilution)
4. PerCP/Cyanine5.5 anti-mouse F4/80 antibody (BioLegend; Catalog number: 123128; Clone name: BM8; 1:50 dilution)
5. PE anti-mouse CD80 antibody (BioLegend; Catalog number: 104708; Clone name: 16-10A1; 1:100 dilution)
6. APC anti-mouse CD206 (MMR) antibody (BioLegend; Catalog number: 141708; Clone name: C068C2; 1:100 dilution)
7. Pacific Blue™ anti-mouse CD45 antibody (BioLegend; Catalog number: 103126; Clone name: 30-F11; 1:400 dilution)
8. APC anti-mouse CD3 antibody (BioLegend; Catalog number: 100236; Clone name: 17A2; 1:100 dilution)
9. PE/Cyanine7 anti-mouse CD69 antibody (BioLegend; Catalog number: 104512; Clone name: H1.2F; 1:50 dilution)
10. FITC anti-mouse CD4 antibody (BioLegend; Catalog number: 100405 ; Clone name: GK1.5; 1:400 dilution)
11. PE anti-mouse CD4 antibody (BioLegend; Catalog number: 100408; Clone name: GK1.5; 1:200 dilution)
12. PE anti-mouse CD8a antibody (BioLegend; Catalog number: 100708; Clone name: 53-6.7; 1:200 dilution)
13. APC anti-mouse CD8a antibody (BioLegend; Catalog number: 100712; Clone name: 53-6.7; 1:200 dilution)
14. PE anti-mouse IFN-γ antibody (BioLegend; Catalog number: 505808; Clone name: XMG1.2; 1:200 dilution)
15. Alexa Fluor® 647 anti-mouse FOXP3 antibody (BioLegend; Catalog number: 126408; Clone name: MF-14; 1:100 dilution)
16. PerCP/Cyanine5.5 anti-mouse/human CD44 antibody (BioLegend; Catalog number: 103032; Clone name: IM7; 1:200 dilution)
17. FITC anti-mouse CD62L antibody (BioLegend; Catalog number: 104406; Clone name: MEL-14; 1:400 dilution)
18. FITC anti-mouse CD206 (MMR) antibody (BioLegend; Catalog number: 141703; Clone name: C068C2; 1:400 dilution)
19. anti-GAPDH (Affinity; Catalog: AF7021; Source: Rabbit; 1:10000 dilution)
20. anti-CD63 (Affinity; Catalog: AF5117; Source: Rabbit; 1:1000 dilution)
21. anti-TSG101 (Affinity; Catalog: DF8427; Source: Rabbit; 1:2000 dilution)
22. anti-alpha 4 (Affinity; Catalog: DF6135; Source: Rabbit; 1:1000 dilution)
23. anti-β-Tubulin (Affinity; Catalog: DF7967; Source: Rabbit; 1:2000 dilution)
24. anti-CCR2 (Affinity; Catalog: DF7507; Source: Rabbit; 1:2000 dilution)

Validation

All antibodies were well-recognized in the field and have their validation statement on their manufactures' websites: https://www.biolegend.com, https://www.affbiotech.com. Furthermore, these antibodies were validated by data provided in the manuscript.
1. PerCP/Cyanine5.5 anti-mouse/human CD11b antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-mouse-human-cd11b-antibody-4257);
2. Brilliant Violet 421™ anti-mouse F4/80 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/brilliant-violet-421-anti-mouse-f4-80-antibody-7199);
3. Alexa Fluor® 647 anti-mouse F4/80 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/alexa-fluor-647-anti-mouse-f4-80-antibody-4074);
4. PerCP/Cyanine5.5 anti-mouse F4/80 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-mouse-f480-antibody-4303);
5. PE anti-mouse CD80 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pe-anti-mouse-cd80-antibody-43);
6. APC anti-mouse CD206 (MMR) antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/apc-anti-mouse-cd206-mmr-antibody-7425);
7. Pacific Blue™ anti-mouse CD45 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pacific-blue-anti-mouse-cd45-antibody-3102);
8. APC anti-mouse CD3 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/apc-anti-mouse-cd3-antibody-8055);
9. PE/Cyanine7 anti-mouse CD69 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pe-cyanine7-anti-mouse-cd69-antibody-3168);
10. FITC anti-mouse CD4 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/fitc-anti-mouse-cd4-antibody-248);
11. PE anti-mouse CD4 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pe-anti-mouse-cd4-antibody-250);
12. PE anti-mouse CD8a antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pe-anti-mouse-cd8a-antibody-155);
13. APC anti-mouse CD8a antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/apc-anti-mouse-cd8a-antibody-150);

14. PE anti-mouse IFN-γ antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/pe-anti-mouse-ifn-gamma-antibody-997);
15. Alexa Fluor® 647 anti-mouse FOXP3 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/alexa-fluor-647-anti-mouse-foxp3-antibody-4662);
16. PerCP/Cyanine5.5 anti-mouse/human CD44 antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/percp-cyanine5-5-anti-mouse-human-cd44-antibody-5605);
17. FITC anti-mouse CD62L antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/fitc-anti-mouse-cd62l-antibody-384);
18. FITC anti-mouse CD206 (MMR) antibody has been validated to be used for flow cytometric analysis and mentioned species reactivity with mouse. (https://www.biolegend.com/en-us/products/fitc-anti-mouse-cd206-mmr-antibody-7318);
19. anti-GAPDH has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-6289-AF7021-GAPDH_Antibody.html);
20. anti-CD63 has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-4424-AF5117-CD63_Antibody.html);
21. anti-TSG101 has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-11944-DF8427-TSG101_Antibody.html);
22. anti-alpha 4 has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-4937-DF6135-Integrin_alpha4_Antibody.html);
23. anti-β-Tubulin has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-11627-DF7967-beta_Tubulin_Antibody.html);
24. anti-CCR2 has been validated to be used for western blotting and mentioned species reactivity with mouse. (https://www.affbiotech.cn/goods-11272-DF7507-CCR2_Antibody.html).

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	The mouse hepatocarcinoma cell strains H22 (catalog number: FH1029) and H22-luc (catalog number: FH0962), mouse malignant melanoma cell strains B16F10 (catalog number: FH0361) and the macrophage cell strain RAW264.7 (catalog number: FH0328) were purchased from Fuheng Biotechnology (Shanghai, China).
Authentication	The cell strains were not validated because the cell strains were used without modification after purchased and the cell morphology and behavior were consistent with expectations.
Mycoplasma contamination	All cell strains showed negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell strains were used in this study.

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals	Female BALB/c mice (6~8 weeks, Stock number: B201) and Female C57BL/6 mice (6~8 weeks, Stock number: B204) were obtained from SPF Biotechnology (Beijing, China). Standard feeding environment (light cycle 12:12, temperature 25 ± 2 °C and humidity 60 ± 10%) for mice.
Wild animals	No wild animals were involved in this study.
Reporting on sex	The sex was not considered in the study design because there was no direct correlation between the selected tumour model and sex.
Field-collected samples	No field-collected samples were involved in this study.
Ethics oversight	All experiments were authorized by the Laboratory Animal Ethical and Welfare Committee of Shandong University Cheeloo College of Medicine (No.19031). All animal experiments were performed under the Guide for Care and the Animal Management Rules of the Ministry of Health of the People's Republic of China.

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

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.*

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	The sample preparation was described in the Methods.
Instrument	BD Accuri C6 Plus and Beckman Coulter CytoFLEX S were were used for flow cytometry data collection.
Software	BD Accuri C6 Plus and Beckman Coulter CytoFLEX S software were used to collect the data. FlowJo V10 software was used to analyse the data.
Cell population abundance	No sorting was performed by flow cytometry.
Gating strategy	Cells were gated on FSC/SSC in general.

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