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Corresponding author(s): Chen Ding

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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	Xcalibur (v4.5)
Data analysis	KSEA (https://casecpb.shinyapps.io/ksea/), STRING (v11.0), Cytoscape (v3.9.1), xCell (https://comphealth.ucsf.edu/app/xcell), ,R (v4.2.3) and R packages [including: factoextra (v1.0.7), ConsensusClusterPlus (v1.62.0), pROC (v1.18.0), survival (v3.5-5), survminer (v0.4.9), GSVA(v1.46.0), msigdbr(v7.5.1), limma (v3.48.3), estimate (v1.0.13), CIBERSORT (v0.1.0), clusterProfiler (v 4.7.0)]

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

All the proteome and phosphoproteome datasets for the cohort study can be accessed through the ProteomeXchange ID: [PXD047297]. For functional studies, all the raw data can be accessed through the ProteomeXchange ID accession: [PXD047429]. The entire proteome and phosphoproteome datasets from these

functional experiments were uploaded to OMIX and can be accessed through the accession: [OMIX005327]. The raw files of Annotated gene sets were collected from GO (https://geneontology.org/docs/download-go-annotations/). For molecular signatures database, KEGG database and Reactome database, we got access to them by the R package: msigdbr (version 7.5.1). The public transcriptomic data for validation were downloaded from supplementary files of published articles (doi:10.1016/j.cell.2017.10.014, doi:10.1074/mcp.M110.000240, doi:10.1038/modpathol.3800794). The information of kinase-substrate relationships was available in PhosphoSite [https://www.phosphosite.org/homeAction.action], Phos-pho.ELM [http://phospho.elm.eu.org/dataset.html], and PhosphoPOINT [http://kinase.bioinformatics.tw/]. Software and publicly available resources used in this study were described in the Methods section. The remaining data are available within the Article, Supplementary Information, or Source Data files.

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	The gender ratio of male/female in our cohort is nearly 50%. Our finding could apply to both two genders. The sex or gender were not considered in study design and the gender of patients in this study was determined based on self-reporting. We collected the gender information of all participants and presented this information in supplementary table1 as one part of clinical data. We didn't perform further sex- or gender- based analyses after calculating the ratio of the gender as we didn't find a predominance of one kind of genders in our cohort and the sarcoma is not a tumor showing different morbidity in different genders.
Reporting on race, ethnicity, or other socially relevant groupings	The participants in our cohort are all Chinese people and they were not classified into subgroups based on their race, ethnicity, or other socially relevant grouping.
Population characteristics	Patients in this cohort ranged from 15-92 years old; the cohort included 128 males, 144 females. 62 cases in FNCLCC I, 133 cases in FNCLCC II, 77 cases in FNCLCC III. We included 17 angiosarcoma, 35 dedifferentiated liposarcoma, 5 epithelioid sarcoma, 52 leiomyosarcoma, 26 myxofibrosarcoma, 11 myxoid liposarcoma, 6 malignant peripheral nerve sheath tumor, 8 otherFS, 15 rhabdomyosarcoma, 18 synovial sarcoma, 43 undifferentiated sarcoma and 36 well-differentiated liposarcoma patients. More detailed clinical information of individual patients are listed in the Supplementary Data 1.
Recruitment	These patients were all newly diagnosed patients with sarcoma who underwent surgical resection and had received no prior treatment for this disease, including chemotherapy, radiotherapy, targeted therapy, or biological therapy. Patients with unclear diagnosis of sarcoma subtypes were excluded.
Ethics oversight	The present study was carried out in compliance with the ethical standards of Helsinki Declaration II and approved by the Institution Review Board of Fudan University Zhongshan Hospital (B2019-200R).Written informed consent was obtained by participants.

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	We conducted proteomics profiling in 272 sarcomas and 91 paired normal adjacent tissues (NATs), phosphoproteomics profiling in 114 sarcomas and 24 NATs. There is no sample-size calculation was performed. We collected the patients with the standard that these patients were newly diagnosed with a clear sarcoma subtype and the five-year following up. Referring to the pan-sarcoma study performed by CPTAC (10.1016/j.cell.2017.10.014), we thought that sample size of 272 is sufficient for this study. No statistical method was used to predetermine sample size. The functional and biological experiments were performed with at least three biological replicates to allow statistical significance testing through student t test. The sample size of glioma patients was based on published papers in the glioma field (PMID: 33577785; PMID: 30343896).
Data exclusions	Several patients without clear diagnosis of sarcoma subtypes were excluded by pathologic experts.
Replication	All experiments were reliably reproduced and indicated in figure legends. The replicated analysis of 293T cell lysates were used for the quality control of the mass spectrometer. 3 biological replicates for every sarcoma cell lines to obtain reliable results of cell proliferation.
Randomization	For ESI-LC-MS/MS analysis, sarcoma samples diagnosed with different sarcoma subtypes were tested with a random order to exclude the bias effects of the mass spectrometry on sarcoma subtypes. When preforming data analysis, samples of sarcoma patients were randomly initialized and allocated into groups to avoid bias from preexisting order. For functional experiments, samples of cell or mice were randomly divided into groups to avoid bias.
Blinding	The investigators who measured protein/phosphosite expression were blinded to patient information. The investigators who performed IHC

Blinding

were blinded to clinical information of sarcoma patients. For consensus clustering analyses, the investigators were blinded to group allocation during data collection. The consensus cluster is unsupervised where no priori knowledge or human factor is involved in the clustering process. For functional experiments, the groups of the samples were maksed when the experimenter is counting the cell number or measuring the tumor size to avoid the prejudices.

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

anti-APEX1 (Cell Signaling Technology Catalog: 10519, dilution 1:100 for IP)
anti-SHC1 (Abcam Catalog: ab33770, dilution 1:100 for IP and 1:1000 for IHC)
anti-MAPK10 (proteintech CatalogCat 17572-1-AP, dilution 1:1000)
anti-NPM1 (Abcam Catalog: ab180607, dilution 1:100)
anti-RIOK1 (proteintech CatalogCat 17222-1-AP, 1:100)
anti-PECAM1 (proteintech Catalog: 66065-2-Ig, dilution 1:1000)
anti-CD36 (proteintech Catalog: 18836-1-AP, dilution 1:1000)
anti-IGFBP6 (proteintech Catalog: 67567-1-Ig, dilution 1:1000)
anti-CD19 (proteintech Catalog: 66298-1-Ig, dilution 1:1000)
anti-IgM (proteintech Catalog: 11016-1-AP, dilution 1:1000)
anti-CD4 (proteintech Catalog: 67786-1-Ig, dilution 1:1000)
anti-ISG20 (proteintech Catalog: 22097-1-AP, dilution 1:1000)
anti-KRT5 (proteintech Catalog: 66727-1-Ig, dilution 1:1000)
anti-KRT9 (Thermo Fisher Catalog: MA5-32396, dilution 1:1000)
anti-CD8 (proteintech Catalog: APC-65069, dilution 1:1000)
ani-CD163 (proteintech Catalog: 16646-1-AP, dilution 1:1000)
anti-CD274 (proteintech Catalog: 66248-1-Ig, dilution 1:1000)

Validation

anti-APEX1 (Cell Signaling Technology Catalog: 10519, dilution 1:100) validated for IP by manufacturer [https://www.cellsignal.cn/products/primary-antibodies/ape1-e5y2c-rabbit-mab/10519].

anti-SHC1 (Abcam Catalog: ab33770, dilution 1:1000) validated for IP and IHC by manufacturer [https://www.abcam.cn/products/primary-antibodies/shc-antibody-ep332y-ab33770.html].

anti-MAPK10 (proteintech CatalogCat 17572-1-AP, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/MAPK10-Antibody-17572-1-AP.htm].

anti-NPM1 (Abcam Catalog: ab180607, dilution 1:1000) validated for IP by manufacturer [https://www.abcam.cn/products/primary-antibodies/npm1alk-antibody-epr11413-ab180607.html].

anti-RIOK1 (proteintech CatalogCat 17222-1-AP, 1:100) validated for IP by manufacturer [https://www.ptgcn.com/products/RIOK1-Antibody-17222-1-AP.htm]

anti-PECAM1 (proteintech Catalog: 66065-2-Ig, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD31-Antibody-66065-2-Ig.htm].

anti-CD36 (proteintech Catalog: 18836-1-AP, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD36-Antibody-18836-1-AP.htm].

anti-IGFBP6 (proteintech Catalog: 67567-1-Ig, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/IGFBP6-Antibody-67567-1-Ig.htm].

anti-CD19 (proteintech Catalog: 66298-1-Ig, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD19-Antibody-66298-1-Ig.htm].

anti-IgM (proteintech Catalog: 11016-1-AP, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/

IGHM-Antibody-11016-1-AP.htm].

anti-CD4 (proteintech Catalog: 67786-1-Ig, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD4-Antibody-67786-1-Ig.htm].
anti-ISG20 (proteintech Catalog: 22097-1-AP, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/ISG20-Antibody-22097-1-AP.htm].

anti-KRT5 (proteintech Catalog: , dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/Cytokeratin-5-Antibody-66727-1-Ig.htm].
anti-KRT9 (Thermo Fisher Catalog: MA5-32396, dilution 1:1000) validated for IHC by manufacturer [https://www.thermofisher.cn/cn/zh/antibody/product/KRT9-Antibody-clone-SD201-09-Recombinant-Monoclonal/MA5-32396].

anti-CD8 (proteintech Catalog: APC-65069, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD8--Antibody-APC-65069.htm].
ani-CD163 (proteintech Catalog: 16646-1-AP, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/CD163-Antibody-16646-1-AP.htm].

anti-CD274 (proteintech Catalog: 66248-1-Ig, dilution 1:1000) validated for IHC by manufacturer [https://www.ptgcn.com/products/PD-L1-CD274-Antibody-66248-1-Ig.htm].

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s)	ISO-HAS was obtained from BioVector Science Lab. VA-ES-BJ (ATCC no. CRL-2138), SU-CCS-1 (ATCC no. CRL-2971), 93T449 (ATCC no. CRL-3043), SW-872 (ATCC no. HTB-92), and SK-UT-1B (ATCC no. HTB-115) were obtained from American Type Culture Collection (ATCC). RKN (Cat ITI04946) was obtained from ITI BioChem. ASM and HEK-293T was obtained from Chinese Academy of Sciences (Shanghai, China).
Authentication	All cell lines were authenticated by Short Tandem repeat (STR) profiling.
Mycoplasma contamination	All cell lines tested negative for mycoplasma contamination.
Commonly misidentified lines (See ICLAC register)	No commonly misidentified cell lines were used.

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	Four-to-six-week-old C57/BL6J nude male mice were obtained from Shanghai SLAC Laboratory Animal Co., Ltd for in vivo xenografts. Mice were housed in pathogen-free, temperature-controlled environment, scheduled with 12-12 h light-dark cycles. The feeding conditions were specific pathogen free animal laboratory with 28 °C and 50% humidity 12/12, providing sufficient water and diet.
Wild animals	This study did not involve wild animals.
Reporting on sex	The mice used in this study include both male and female as the sarcoma could occur without a gender preference. Two genders of mice were randomly distributed into the groups with different treatments.
Field-collected samples	This study did not involve field-collected samples
Ethics oversight	For animal experiment, this study is under the guidelines of the animal care regulations of Fudan University, and was approved by Research Ethics Committee of department of experimental animal science, Fudan University.

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