

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 |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> The statistical test(s) used AND whether they are one- or two-sided
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> A description of all covariates tested |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> 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) |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> For null hypothesis testing, the test statistic (e.g. F , t , r) with confidence intervals, effect sizes, degrees of freedom and P value noted
<i>Give P values as exact values whenever suitable.</i> |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> 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

The codes generated during this study are available at the OmicShare tools, a free online platform (<https://www.omicshare.com/>).

Data analysis

Statistical analyses were performed using GraphPad Prism. The data are expressed as the mean \pm SEM unless indicated otherwise. Unpaired Student's t -test was used to determine statistically significant differences. A value of $P < 0.05$ was considered significant at the 95% confidence level. Data analysis was performed using the OmicShare tools, a free online platform for data analysis. We used Cell Ranger (<http://support.10xgenomics.com/single-cell/software/overview/welcome>) to complete gene and expression identification, transferred the obtained expression matrix to Seurat (<https://satijalab.org/seurat/index.html>) for subsequent analysis, and used DoubletFinder (<https://github.com/ddiez/DoubletFinder>) to perform polyclonal filtering, thus the result of single cell subcluster classification can be visualized. Monocle2 (<http://cole-trapnell-lab.github.io/monocle-release/>), Monocle3 (<https://cole-trapnell-lab.github.io/monocle3/docs/installation/>), PAGA (<https://github.com/theislab/paga>), and scVelo (<https://scvelo.readthedocs.io/>) were used for pseudotime analysis of single cell subclusters. Use CopyKAT (<https://github.com/navinlabcode/copykat>) to identify malignant cells. CellPhoneDB (<https://github.com/Teichlab/cellphonedb>) was used to analyze cell communication.

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](#)

This study was approved by the ethics committee of the SIR RUN RUN HOSPITAL NANJING MEDICAL UNIVERSITY. The 10 × Genomics datasets generated during this study is publicly available via Genome Sequence Archive (GSA), under the Accession number: HRA002151.

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](https://www.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	No sample size calculation was performed. Cell capture for single cell analysis was aimed to obtain as many cells as possible for each run, which maximum is theoretically 50-60% of 10,000 input cells. The number of cells analyzed (one patient) totalled over 12,000 cells and thus provided robust statistical analysis on clustering and differentially expressed genes.
Data exclusions	For single cell analysis, multiplets or fragmented cells and dying or stressed cells were excluded. Multiplets or fragmented cells during filtration were defined using the numbers of genes and unique molecular index (UMI) counts. Dying or stressed cells were defined by percentages of mitochondrial genes. Thresholds for exclusions in each dataset are defined in Supplementary Fig. S2.
Replication	For in vitro studies, three biological replicates were performed and experiments were repeated at least twice.
Randomization	No randomization was performed.
Blinding	The investigators were not blinded.

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
<input type="checkbox"/>	<input checked="" type="checkbox"/> Antibodies
<input checked="" type="checkbox"/>	<input type="checkbox"/> Eukaryotic cell lines
<input checked="" type="checkbox"/>	<input type="checkbox"/> Palaeontology and archaeology
<input checked="" type="checkbox"/>	<input type="checkbox"/> Animals and other organisms
<input type="checkbox"/>	<input checked="" type="checkbox"/> Human research participants
<input checked="" type="checkbox"/>	<input type="checkbox"/> Clinical data
<input checked="" type="checkbox"/>	<input type="checkbox"/> Dual use research of concern

Methods

n/a	Involved in the study
<input checked="" type="checkbox"/>	<input type="checkbox"/> ChIP-seq
<input checked="" type="checkbox"/>	<input type="checkbox"/> Flow cytometry
<input checked="" type="checkbox"/>	<input type="checkbox"/> MRI-based neuroimaging

Antibodies

Antibodies used	SMA (unnecessary dilution, ZM-0003, ZSGB-BIO), CD68 (unnecessary dilution, ZM-0464, ZSGB-BIO), Ki67 (1:100, ZM-0166, ZSGB-BIO) CD34 (unnecessary dilution, ZM-0046, ZSGB-BIO) Desmin (unnecessary dilution, ZA-0686, ZSGB-BIO), EMA (unnecessary dilution, ZM-0095, ZSGB-BIO), S-100 (unnecessary dilution, ZM-0224, ZSGB-BIO), BCL-2 (unnecessary dilution, ZA-0536, ZSGB-BIO). Servicebio blinded to clinical data independently assessed staining results for CD3 (1:250, GB11014, Servicebio), CD4 (1:400, GB11064-1, Servicebio), CD8 (1:200, GB11068-1, Servicebio).
Validation	All of the above antibodies have been documented and validated by the manufacturer to be specific for the indicated molecules.

Human research participants

Policy information about [studies involving human research participants](#)

Population characteristics

This manuscript describes a rare case of malignant fibrous histiocytoma in the lower femoral segment of the left thigh in a 44-year-old male patient. The patient complained of pain and discomfort in the distal left thigh and went to a local hospital for symptomatic conservative treatment. The result of treatment was not good in the local hospital. The patient suffered from severe pain and found a hard mass at the distal end of his left thigh for one month. Therefore, he came to Sir Run Run Hospital, Nanjing Medical University for further diagnosis and treatment and the pathological result was malignant fibrous histiocytoma. In order to reduce the burden of tumor and relieve the symptoms of leg pain, tumor resection was performed to remove the tumor. In this study, the patient's informed consent was obtained and the surgical residue was used for single-cell RNA sequencing.

Recruitment

The patient came to Sir Run Run Hospital, Nanjing Medical University for further diagnosis and treatment.

Ethics oversight

Sir Run Run Hospital, Nanjing Medical University

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