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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 <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

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n/a	Confirmed
	$oxed{x}$ 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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted Give <i>P</i> values as exact values whenever suitable.
x	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
x	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), 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 Microsoft Excel and SPSS statistics 22 version R22.0 were used for data collection.

Data analysis SPSS Statistics version 22.0

SPSS Statistics version 22.0, R software (v4.3.1), SAS version 9.4, TissueFAXS SL system (7.1.120), HALO™ software (v3.5), and FCAP Array™ Software (version 3.0) were used for data analysis and figure generation.

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 <u>data availability statement</u>. 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 <u>policy</u>

All data requests will undergo review by Sichuan Cancer Hospital and the study sponsor, Shanghai Junshi Biosciences Co., Ltd., to assess any potential intellectual property or confidentiality obligations. A proposal detailing the study objectives and statistical analysis plan will be required for evaluation. Additional materials may also be requested during the evaluation process. Data will be available upon request 12 months after the publication of this article. Requests for access to deidentified participant data from this study can be submitted via email to wangqifeng@scszlyy.org.cn, accompanied by a detailed proposal for approval. Please allow

1 month for the response to the request. Access to the shared data will require signing a data access agreement with the sponsor. The study protocol is available as Supplementary Note in the Supplementary Information file. The remaining data are available within the Article, Supplementary Information, or Source Data file. Source data are provided with this paper.

Research involving human participants, their data, or biological material

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

Reporting on sex and gender

This study included both male and female participants based on self-report and body characteristics. Sex and/or gender was not considered in the study design. Both male and female patients who met the inclusion criteria were eligible for inclusion in this study. The results of the study apply to both male and female participants. No sex analysis was therefore carried out. The sex distribution was 29 men (87.9%) and 4 women (12.1%).

Reporting on race, ethnicity, or other socially relevant groupings

Race, ethnicity, or other socially relevant groupings were not considered in the research design.

Population characteristics

We assessed 56 patients for eligibility; among whom, 33 (median age: 59 years, range: 43–74; 29 men) were enrolled. The most common sites for oligometastasis were distant lymph nodes (63.6%), followed by the lungs (15.2%), and bones (9.1%) (Table 1). We observed that 27 (81.8%) patients had a total of 32 oligometastatic lesions, of which 12 were in distant organs and 15 in non-regional lymph nodes, whereas 6 patients (18.2%) had only regional lymph node metastases (cTanyN3M0). Patients were enrolled in the study at Sichuan Cancer Hospital and were treated with toripalimab (anti-PD1) plus chemotherapy and radiotherapy.

Recruitment

Patients were offered the opportunity to participate in the clinical trial by investigators during routine visits at Sichuan Cancer Hospital. All patients who met the criteria were included in the study, with no potential for self-selection bias. Investigators at Sichuan Cancer Hospital screened and enrolled participants who met all the inclusion criteria and none of the exclusion criteria as defined in the protocol. All participants provided written informed consent prior to enrollment in the study.

Ethics oversight

The study was performed in compliance with the Declaration of Helsinki and Good Clinical Practice guidelines, and was approved by the Institutional Review Board of Sichuan Cancer Hospital, Sichuan, China (Ethics number: SCCHEC-02-2021-021). An interim analysis, authorized by the Institutional Review Board of Sichuan Cancer Hospital at a later stage, included data on some secondary endpoints (ORR, DCR, toxicity) to provide an early assessment of efficacy and to help identify and address safety issues early. All patients provided written informed consent before any procedure. No participation compensation was provided. The trial was registered with chictr.org.cn (ChiCTR2100046715) on May 27, 2021. Analyses were conducted as planned in the preregistration. There were some minor deviations from the preregistration regarding the inclusion criteria, exclusion criteria, statistical analyses, and other aspects. These deviations are explicitly indicated at the end of the Supplementary Note.

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	w that is the best fit for your research.	If you are not sure, read the appropriate sections before making your selection. $ \\$
X 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

n=33. The necessary sample sizes were assessed using a one-sided test based on an increase of median PFS from 6.3 months to 12 months with chemo-immunotherapy, as previously described. This calculation assumed a type I error of 0.05, a statistical power of 80%, and a 20% dropout rate. The follow-up duration was calculated from enrollment to the date of the last follow-up. To observe 16 PFS events, we calculated that 32 patients were needed.

Data exclusions

All patients enrolled in this study were included for analysis.

Replication

Not applicable. This is a single-arm phase 2 clinical trial, involving the assessment of radiotherapy in combination with chemo-immunotherapy in human participants. No preclinical data that could be replicated is provided in the manuscript.

Randomization

Not applicable. We did not include a control arm in this study. This was a single-arm study with no randomization.

Blinding

Not applicable. This was an open label clinical trial. Given that there was no randomization, and patients were included in one consecutive cohort, the blinding did not apply.

Reporting for specific materials, systems and methods

•	authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, evant 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 & experime	ental systems Methods		
n/a Involved in the study	n/a Involved in the study		
Antibodies	ChiP-seq		
x Eukaryotic cell lines	Flow cytometry		
Palaeontology and a	archaeology MRI-based neuroimaging		
Animals and other of	organisms		
Clinical data			
Dual use research o	f concern		
x Plants			
Antibodies			
Antibodies used	Anti-PDL1 (ZA-0629, 1:50; ZSGB Biotech, Beijing, China), Anti-CD68 (ZM-0060, 1:100, dye480; ZSGB Biotech), CD8 (ZA-0508, 1:100, dye570; ZSGB Biotech), CD11c (45581, 1:400, dye520; Abcam, Cambridge, UK), CD4 (ZA-0509, 1:100, dye690; ZSGB Biotech), Pancytokeratin (ZM-0067, 1:100, dye780; ZSGB Biotech).		
Validation	No new antibodies were generated in this study. All antibodies are commercially available with detailed descriptions available in the manufacturer's websites (Supplemental Data 1).		
Clinical data Policy information about cl	inical studies		
All manuscripts should comply	with the ICMJEguidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.		
Clinical trial registration	The trial was registered with the Chinese Clinical Trial Registry, number ChiCTR2100046715.		
Study protocol	The study protocol is presented as Supplementary Note in the Supplementary information file.		
Data collection	Data were collected using Microsoft Excel and SPSS Statistics 22 at Sichuan Cancer Hospital. Recruitment for the study began on June 1, 2021, with the first patient enrolled on June 30, 2021, and the last patient enrolled on September 30, 2022. Data collection commenced with the enrollment of the first patient in June 2021 and continued until the cut-off date of October 12, 2023. Follow-up is ongoing.		
Outcomes	The primary endpoint was progression-free survival (PFS), defined as the time between beginning treatment and tumor progression, patient death, or the last follow-up. Secondary endpoints included the objective response rate (ORR), disease control rate (DCR), duration of remission (DoR), 1- and 2-year OS rates, patient-reported health-related quality of life (HRQoL), and adverse events. Objective responses included complete remission (CR) and partial response (PR). Disease control represented CR, PR, and stable disease (SD) while DoR was determined as the interval between the first objective response to the first documentation of progression or all-cause death. OS was assessed from the start of therapy to all-cause death. Exploratory outcomes included the relationship between clinical outcomes with immune cell types in the tumor microenvironment, and biomarkers in peripheral blood.		
Plants			
Seed stocks	The study did not involve any plants.		

Novel plant genotypes

Authentication

Not applicable.

Not applicable.

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

Serum samples were obtained at baseline and during treatment (after two cycles of chemo-immunotherapy and before radiotherapy). Blood samples were obtained from subjects via venipuncture into anticoagulant-free vacuum tubes, followed by clot formation at room temperature for 30 minutes to 1 hour. The clotted blood samples were then centrifuged at $1000-2000 \times g$ for 10-15 minutes at 4°C to separate the serum from cellular components. The resulting serum supernatant was carefully collected, aliquoted, and stored at -80°C until analysis. The assessment of serum levels of cytokines including L-2, L-4, L-6, L-10, L-17, $TNF-\alpha$, and $IFN-\lambda$ was performed using a magnetic beads kit (281004; Wellgrow, Beijing, China). Briefly, 50 μ l of the serum sample or reference standards were added to 50 μ l of capture beads suspension, mixed thoroughly, and incubated at RT in the dark for 1 hour. The supernatant was removed after magnetic precipitation, and the beads were incubated with 100 µl of a fluorescent-labeled antibody at RT for 1 hour. After washing, the beads with binding cytokines and antibodies were ready for flow cytometry analysis.

Instrument

The instrument is BD FACSCanto™ flow cytometry (BD Biosciences, San Jose, CA, USA).

Software

The flow cytometry data was analyzed by FCAP ArrayTM Software (Version 3.0).

Cell population abundance

The analysis of periphery cytokines was performed by using a commercial magnetic beads kit following the manufacturer's instructions (Wellgrow, cat#281004, Beijing, China). The analyzed 'cells' are actually magnetic beads with a uniform size and high purity.

Gating strategy

Cytokine detection in this experiment was performed using the CBA method with the beads of uniform size and gating by FSC and SSC. The number of each fluorescent beads acquired was 300 per sample. The seven fluorescent beads coated with different cytokine antibodies showed different fluorescence intensities in the APC channel, so the precise clustering of the seven cytokines can be realized. Each cytokine detection antibody was coupled to PE fluorescence, so its concentration is related to the PE channel fluorescence intensity, by substituting the specific values into the corresponding standard curve to get an accurate cytokine concentration.

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