nature portfolio

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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	Our research team was in charge of collecting the initial biological specimens, all obtained from predetermined experimental conditions and sample sources. We followed a standardized protocol to ensure the integrity and quality of these samples, which included meticulous steps for collection, pre-processing, preservation, and transportation. Samples were meticulously preserved under stringent conditions and subjected to thorough quality assessment prior to dispatch to Magigene for sequencing. The methods employed for the collection and preliminary processing of specimens are detailed in the Methods section of our manuscript. The raw data were generated using Magigene's advanced high-throughput sequencing facilities, which yielded FASTQ files of the sequencing data for further analysis.
Data analysis	The analysis of the high-throughput transcriptomic sequencing data was performed utilizing Magigene's proprietary online analysis platform. This specialized platform encompasses a suite of analysis tools for processing the sequencing data, performing statistical evaluations, and visualizing the results. Due to the proprietary nature of the analysis system, we do not have access to the exact software versions or the underlying code. Nonetheless, we have thoroughly documented each step of our analysis process, including parameter settings and methodological choices, to ensure transparency and reproducibility. Upon request, we can provide additional details concerning our analytical procedures and the settings used, facilitating other researchers to comprehend and replicate our study outcomes.

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 data can be obtained by contacting the corresponding author

Human research participants

Policy information about studies involving human research participants and Sex and Gender in Research.

Reporting on sex and gender	We confirmed that there are no such ethical issues in the countries and regions involved in the study. In the study, we uniformly used biological gender identity.
Population characteristics	Our study does not address this issue
Recruitment	The collection of clinical specimens was meticulously carried out by applying strict patient inclusion criteria. Individuals with a medical history of infection, tuberculosis, or tumors were excluded to eliminate potential confounding effects these conditions might have on the research outcomes. Normal and degenerated intervertebral disc nucleus pulposus samples were distinguished utilizing MRI findings, with the assessment aided by the Pfirrmann grading method. Normal disc samples were procured from patients diagnosed with juvenile idiopathic scoliosis, thereby selecting a cohort with no history of degenerative disc disease, which could confound the comparison. Conversely, degenerated disc samples were obtained from patients undergoing surgery for disc herniation or disc fusion, conditions that are indicative of disc degeneration. By adhering to these selection criteria and distinguishing the samples based on reliable imaging techniques and clinical grading, we aimed to reduce the risk of confounding factors influencing our analysis of disc degeneration.
Ethics oversight	Institutional Research Ethics Committee of the Second Hospital of Lanzhou University

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	Based on the experience and references from previous studies, we designed the sample size to fulfill the research requirements and enable difference analysis.
Data exclusions	No data is excluded
Replication	All experimental results were repeated three times
Randomization	All groups adopted a randomization strategy
Blinding	There was a double-blind relationship between the experimenter and the data analyst

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

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used: P-P53 Antibody (1:1000, ABclonal, China), P21 Antibody (1:1000, ABclonal, China), P16 Antibody (1:1000, ABclonal, China), Aggrecan Antibody (1:1500, Affinity, China), Collagen II Antibody (1:1500, Affinity, China), MMP-9 Antibody (1:1500, Affinity, China), MMP-3 Antibody (1:1500, Affinity, China), Anti-Actin Antibody (1:1500, Abcam, England), and the PI3K/AKT Signaling Pathway Panel (1:1500, Abcam, England).

Validation: All antibodies appearing in the article have been verified as primary antibodies to confirm their species specificity and availability.

Eukaryotic cell lines

Policy information about cell lines and Sex and Gender in Research

Cell line source(s): The cells involved in the experiment were all extracted from primary nucleus pulposus cells of male rats.

Authentication: Primary nucleus pulposus cells have been clearly identified before use

Mycoplasma contamination: Nucleus pulposus primary cells test negative for mycoplasma

Commonly misidentified lines (See ICLAC register): The cells involved in the experiment were all extracted from primary nucleus pulposus cells of male rats.

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: The animals involved in this experiment were all SPF male SD rats.

Wild animals: not involving

Reporting on sex: Firstly, male rats have more stable hormonal levels compared to females, which helps reduce variability in experimental results due to hormonal cycles, thereby improving the repeatability and interpretability of the findings. Secondly, including our research group, most related studies primarily use male rats, and continuing to use males ensures comparability with existing data, facilitating the analysis of new and old research results. Additionally, selecting a single gender reduces the complexity of preliminary exploratory research, allowing us to focus on verifying research hypotheses without increasing the complexity of the experimental design. These reasons collectively form the scientific basis for our choice of male rats as experimental animals.

Field-collected samples: not involving

Ethics oversight: This study received ethical approval from the Institutional Research Ethics Committee of the Second Hospital of Lanzhou University (Ethics Number: 2023A-278).

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

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration: not involving

Study protocol: not involving

Data collection: not involving

Outcomes: not involving