NOTE: Please save this file locally before filling in the table, DO NOT work on the file within your internet browser as changes will not be saved. Adobe Acrobat Reader (available free here) is recommended for completion.

The ARRIVE guidelines 2.0: author checklist

The ARRIVE Essential 10

These items are the basic minimum to include in a manuscript. Without this information, readers and reviewers cannot assess the reliability of the findings.

Item		Recommendation	Section/line number, or reason for not reporting
Study design	1	For each experiment, provide brief details of study design including: a. The groups being compared, including control groups. If no control group has been used, the rationale should be stated. b. The experimental unit (e.g. a single animal, litter, or cage of animals).	Page 7, Line 85-93 cage of animals
Sample size	2	a. Specify the exact number of experimental units allocated to each group, and the total number in each experiment. Also indicate the total number of animals used. b. Explain how the sample size was decided. Provide details of any *a priori* sample size calculation, if done.	Page 7, Line 85-93 The sample size was calculated based on previous experiments to establish tail s uspension rat models
Inclusion and exclusion criteria	3	a. Describe any criteria used for including and excluding animals (or experimental units) during the experiment, and data points during the analysis. Specify if these criteria were established *a priori.* If no criteria were set, state this explicitly. b. For each experimental group, report any animals, experimental units or data points not included in the analysis and explain why. If there were no exclusions, state so. c. For each analysis, report the exact value of *n* in each experimental group.	Page 7, Line 93-94 There were no exclusions and all experim ental samples were included in the analy sis For each observed index, all experimenta l samples were included in the analysis index. Three to five samples were random ly selected for each group for TEM obser vation.
Randomisation	4	a. State whether randomisation was used to allocate experimental units to control and treatment groups. If done, provide the method used to generate the randomisation sequence. b. Describe the strategy used to minimise potential confounders such as the order of treatments and measurements, or animal/cage location. If confounders were not controlled, state this explicitly.	Randomization was no t used and was balan ced according to bod y weight. The cage location, feeding and tail susp ension of the animals were carried out b y special people to avoid confounding fa ctors affecting the experimental results .
Blinding	5	Describe who was aware of the group allocation at the different stages of the experiment (during the allocation, the conduct of the experiment, the outcome assessment, and the data analysis).	During the rearing phase of the experime nt, the animal experimenter was aware of the group assignment. During the outcom e assessment phase, assessors were unawa re of the group assignments. During the data-analysis phase, the statisticians w ere unaware of the group assignments
Outcome measures	6	a. Clearly define all outcome measures assessed (e.g. cell death, molecular markers, or behavioural changes). b. For hypothesis-testing studies, specify the primary outcome measure, i.e. the outcome measure that was used to determine the sample size.	Tis were assessed by Safranin O stainin g and immunohistochemistry The biomechanical features of cartilage, including microscopic geomorphology, el astic modulus, and hardness, were assess ed using nanoindentation. The microstructure of cartilage and musc le was observed by transmission electron microscopy
Statistical methods	7	a. Provide details of the statistical methods used for each analysis, including software used. b. Describe any methods used to assess whether the data met the assumptions of the statistical approach, and what was done if the assumptions were not met.	Page 11,181-184 Page 11,181-184
Experimental animals	8	a. Provide species-appropriate details of the animals used, including species, strain and substrain, sex, age or developmental stage, and, if relevant, weight. b. Provide further relevant information on the provenance of animals, health/immune status, genetic modification status, genotype, and any previous procedures.	Two-month-old SD rat s at 280g were used Animals were purchased from Beijing Vita he Experimental Animal Technology, and 2 -month-old 280g SD rats with normal geno type were used
Experimental procedures	9	For each experimental group, including controls, describe the procedures in enough detail to allow others to replicate them, including: a. What was done, how it was done and what was used. b. When and how often. c. Where (including detail of any acclimatisation periods). d. Why (provide rationale for procedures).	Page 7,97-104 Page 7,97-104 Page 7,97-104 Page 7,97-104
Results	10	For each experiment conducted, including independent replications, report: a. Summary/descriptive statistics for each experimental group, with a measure of variability where applicable (e.g. mean and SD, or median and range). b. If applicable, the effect size with a confidence interval.	The main results of this study, cartilag e and muscle ultrastructure, are observa tional indicators and do not have means and standard deviations. For other resul ts, means and standard deviations have b een stated in the literature. N/A

The Recommended Set

These items complement the Essential 10 and add important context to the study. Reporting the items in both sets represents best practice.

Item		Recommendation	Section/line number, or reason for not reporting
Abstract	11	Provide an accurate summary of the research objectives, animal species, strain and sex, key methods, principal findings, and study conclusions.	Page 3-4,33-55
Background	12	a. Include sufficient scientific background to understand the rationale and context for the study, and explain the experimental approach. b. Explain how the animal species and model used address the scientific objectives and, where appropriate, the relevance to human biology.	Page 6,63-78 Page 6,63-78
Objectives	13	Clearly describe the research question, research objectives and, where appropriate, specific hypotheses being tested.	Page 6,79-81
Ethical statement	14	Provide the name of the ethical review committee or equivalent that has approved the use of animals in this study, and any relevant licence or protocol numbers (if applicable). If ethical approval was not sought or granted, provide a justification.	Page 7,94-95
Housing and husbandry	15	Provide details of housing and husbandry conditions, including any environmental enrichment.	Page 7, Line 85-93
Animal care and monitoring	16	a. Describe any interventions or steps taken in the experimental protocols to reduce pain, suffering and distress. b. Report any expected or unexpected adverse events. c. Describe the humane endpoints established for the study, the signs that were monitored and the frequency of monitoring. If the study did not have humane endpoints, state this.	Page 7, Line 97-98 Page 8, Line 106-107 Minor tail injury was observed in individual rats during tail suspension. At this time, the gauze was loosened appropriately, but it did not affect the final experimental results This experiment is an animal experiment, which can be completed by the time point, and there is no humane end point.
Interpretation/ scientific implications	17	a. Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature. b. Comment on the study limitations including potential sources of bias, limitations of the animal model, and imprecision associated with the results.	damage. This study adds to the observation that microgravity can cause cartilage and muscle microstructure and other degradation results, and further confirms that microgravity can cause cartilage and muscle damage. Page 15,Line 270-280
Generalisability/ translation	18	Comment on whether, and how, the findings of this study are likely to generalise to other species or experimental conditions, including any relevance to human biology (where appropriate).	The results of this experiment may be related to the fact that long-term bedridden patients may also cause cartilage and muscle damage, and appropriate measures should be taken to avoid these injuries.
Protocol registration	19	Provide a statement indicating whether a protocol (including the research question, key design features, and analysis plan) was prepared before the study, and if and where this protocol was registered.	The study question was to observe the effect of weightlessness on the ultrastructure of knee cartilage and muscle. The main design feature was to simulate weightlessness by tail suspension. The effects of 14 days and 28 days of weightlessness on cartilage and muscle damage were observed. Mitochondrial activity in muscle
Data access	20	Provide a statement describing if and where study data are available.	ntact the corresponding author if required
Declaration of interests	21	a. Declare any potential conflicts of interest, including financial and non-financial. If none exist, this should be stated. b. List all funding sources (including grant identifier) and the role of the funder(s) in the design, analysis and reporting of the study.	There are no conflicts of interest in this study This work was supported by funds from the Research Fund of space health research Foundation of Astronaut Health Center (AHCC2021KF001) and Peking University Third Hospital cohort study (BYSYDL2021014),Funder:Jianquan Wang

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