| Item No | Item | RECOMMENDATION | Reported on Page Number/Line Number | Reported on Section/Paragraph |
|--------|------|----------------|-------------------------------------|------------------------------|
| 1      | Title | Provide as accurate and concise a description of the content of the article as possible. | P1L1-L2 | Title |
| 2      | Abstract | Provide an accurate summary of the background, research objectives, including details of the species or strain of animal used, key methods, principal findings and conclusions of the study. | P1L26-P2L23 | Abstract |
| 3      | Background | a. Include sufficient scientific background (including relevant references to previous work) to understand the motivation and context for the study, and explain the experimental approach and rationale.  
b. Explain how and why the animal species and model being used can address the scientific objectives and, where appropriate, the study’s relevance to human biology. | P3L2-L29 | Introduction paragraph 1, 2, 3 |
| 4      | Objectives | Clearly describe the primary and any secondary objectives of the study, or specific hypotheses being tested. | P3L30-P4L2 | Introduction paragraph 4 |
| 5      | Ethical statement | Indicate the nature of the ethical review permissions, relevant licences (e.g. Animal [Scientific Procedures] Act 1986), and national or institutional guidelines for the care and use of animals, that cover the research. | P4L17-L18 | Methods paragraph 1 |
| 6      | Study design | For each experiment, give brief details of the study design including:  
a. The number of experimental and control groups.  
b. Any steps taken to minimise the effects of subjective bias when allocating animals to treatment (e.g. randomisation procedure) and when assessing results (e.g. if done, describe who was blinded and when).  
c. The experimental unit (e.g. a single animal, group or cage of animals). A time-line diagram or flow chart can be useful to illustrate how complex study designs were carried out. | P4L4-P8L2 | Methods and Materials section |
| 7      | Experimental procedures | For each experiment and each experimental group, including controls, provide precise details of all procedures carried out. For example:  
a. How (e.g. drug formulation and dose, site and route of administration, anaesthesia and analgesia used [including monitoring], surgical procedure, method of euthanasia). Provide details of any specialist equipment used, including supplier(s).  
b. When (e.g. time of day).  
c. Where (e.g. home cage, laboratory, water maze).  
d. Why (e.g. rationale for choice of specific anaesthetic, route of administration, drug dose used). | P4L4-P8L2 | Methods and Materials section |
| Topic                                      | Section | Page(s)   | Notes       |
|-------------------------------------------|---------|-----------|-------------|
| Experimental animals                      | 8       | P7L14-L22 | Methods paragraph 9 |
| Housing and husbandry                     | 9       | P7L22-L25 | Methods paragraph 9 |
| Sample size                               | 10      | P7L14     | Methods paragraph 9 |
| Allocating animals to experimental groups | 11      | P7L14     | Methods paragraph 9 |
| Experimental outcomes                     | 12      | P8L4-P10L2| Results      |
| Statistical methods                       | 13      | P7L27-P8L2| Statistical analysis |
| RESULTS                                   | 14      | P4L7-L17;P7L14 | Methods paragraph 1,9 |
| Numbers analysed                          | 15      | P4L7-L17;P7L14 | Methods paragraph 1, 9 |
| Outcomes and estimation                   | 16      | P8L4-P10L2| Throughout   |
| Adverse events                            | 17      | none      | none         |
## DISCUSSION

### Interpretation/scientific implications

1. Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature.
2. Comment on the study limitations including any potential sources of bias, any limitations of the animal model, and the imprecision associated with the results.
3. Describe any implications of your experimental methods or findings for the replacement, refinement or reduction (the 3Rs) of the use of animals in research.

### Generalisability/translation

Comment on whether, and how, the findings of this study are likely to translate to other species or systems, including any relevance to human biology.

### Funding

List all funding sources (including grant number) and the role of the funder(s) in the study.

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### References:

1. Kilkenny C, Browne WJ, Cuthill IC, Emerson M, Altman DG (2010) Improving Bioscience Research Reporting: The ARRIVE Guidelines for Reporting Animal Research. PLoS Biol 8(6): e1000412. doi:10.1371/journal.pbio.1000412

2. Schulz KF, Altman DG, Moher D, the CONSORT Group (2010) CONSORT 2010 Statement: updated guidelines for reporting parallel group randomised trials. BMJ 340:c332.

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*As the checklist was provided upon initial submission, the page number/line number reported may be changed due to copyediting and may not be referable in the published version. In this case, the section/paragraph may be used as an alternative reference.*