The ARRIVE Guidelines Checklist

| Item No | Item No | RECOMMENDATION | Reported on Page Number/Line Number | Reported on Section/Paragraph |
|---------|---------|----------------|------------------------------------|-----------------------------|
| Title   | 1       | Provide as accurate and concise a description of the content of the article as possible. | Page 1                        | Title                       |
| Abstract| 2       | 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. | Page 2                        | Abstract                    |

**INTRODUCTION**

| Background | 3 | 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. | Page 4 -5 | Introduction |

| Objectives | 4 | Clearly describe the primary and any secondary objectives of the study, or specific hypotheses being tested. | N/A | N/A |

**METHODS**

| Ethical statement | 5 | 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. | Page5/line 13-14 Page6/line 27-29 | Method/ paragraph 1 Method/ paragraph 6 |
|-------------------|---|-----------------------------------------------------------------|------------------------------------|----------------------------------------|
| Study design      | 6 | 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. | Page5/line 3-4 Page6/line 21-22 Page9/line 12-14 | Method/ paragraph 1 Method/ paragraph 6 Method/ paragraph 9 |
| Experimental procedures | 7 | 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). | Page5/line 3-12 Page6/line 16-27 Page9/line 1-15 | Method/ paragraph 1 Method/ paragraph 6 Method/ paragraph 9 |
| Section                                      | Page Range | Method/paragraph |
|----------------------------------------------|------------|------------------|
| Experimental animals                         | 3-4        | 1                 |
| Housing and husbandry                        | 13-16      | 6                 |
| Sample size                                  | 3-12       | 1, 6              |
| Allocating animals to experimental groups    | 3-12, 9-15 | 1, 6, 9           |
| Experimental outcomes                       | 8-12       | 1                 |
| Statistical methods                          | 8-12       | 1                 |
| RESULTS                                      |            |                  |
| Baseline data                                |            | N/A              |
| Numbers analysed                             | 19-21      | Figure legend/paragraph 1-6 |
| Outcomes and estimation                      | 5-33       | Result/all paragraphs |
| Adverse events                               |            | N/A              |
DISCUSSION

Interpretation/scientific implications 18  
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 19  
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 20  
List all funding sources (including grant number) and the role of the funder(s) in the study.

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.