

## 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:	Materials and matheday
		a. The groups being compared, including control groups. If no control group has been used, the rationale should be stated.	Materials and methods: Page 4, lines 138-146
		b. The experimental unit (e.g. a single animal, litter, or 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.	Materials and methods: Page 4, line 142. Also, each figure and table description include this information.
		b. Explain how the sample size was decided. Provide details of any <i>a priori</i> sample size calculation, if done.	The sample size was decided according to previous studies in this area that show that 6-7 animals per group is a relevant number.
Inclusion and exclusion criteria	3	<ul> <li>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.</li> </ul>	Materials and methods: Pages 4-5, lines 132-133, 145-146
		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.	There were no exclusions
		c. For each analysis, report the exact value of $\boldsymbol{n}$ in each experimental group.	Each figure and table include this information
Randomisation	4	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.	Materials and methods: Page 4, lines 141-142
		<ul> <li>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.</li> </ul>	The animals cages were carrefully marked and the animals were manipulated with high attention in order to avoid confusion.
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).	The animals were randomly grouped by MG, the tissue samples were processed by SCM and MG. The data analysis was made by RGF, ERM, JMEC and JVG.
Outcome measures	6	a. Clearly define all outcome measures assessed (e.g. cell death, molecular markers, or behavioural changes).	Materials and methods: Pages 4-7, lines 163-165, 172-273
		b. For hypothesis-testing studies, specify the primary outcome measure, i.e. the outcome measure that was used to determine the sample size.	This proteomic study is a holistic study that is no hypothesis-driven in order to obtain a global informa about protein changes
Statistical methods	7	Provide details of the statistical methods used for each analysis, including software used.	Materials and methods: Page 7, lines 276-301
		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.	Materials and methods: Page 7, lines 297-301
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.	Materials and methods: Pages 3-4, lines 132-137
		b. Provide further relevant information on the provenance of animals, health/immune status, genetic modification status, genotype, and any previous procedures.	Materials and methods: Pages 3-4, lines 132-137
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.	Materials and methods: Pages 3-4, lines 132-176
		b. When and how often.	1 ages 5 4, iiies 152 176
		c. Where (including detail of any acclimatisation periods).	
		d. Why (provide rationale for procedures).	
Results	10	For each experiment conducted, including independent replications, report:	
		<ul> <li>a. Summary/descriptive statistics for each experimental group, with a measure of variability where applicable (e.g. mean and SD, or median and range).</li> </ul>	Each figure and table include this information
		b. If applicable, the effect size with a confidence interval.	

## 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.	Abstract: Pages 1-2, lines 31-49
Background	12	<ul> <li>a. Include sufficient scientific background to understand the rationale and context for the study, and explain the experimental approach.</li> </ul>	Introduction: Pages 2-3, lines 52-128 Introduction: Page 2, lines 52-63
		<ul> <li>Explain how the animal species and model used address the scientific objectives and, where appropriate, the relevance to human biology.</li> </ul>	Our experimental animal model is highly used and accepted in research as an appropriate mode with relevance to human biology
Objectives	13	Clearly describe the research question, research objectives and, where appropriate, specific hypotheses being tested.	Introduction: Page 4, lines 124-128
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.	Materials and methods: Page 4, lines 166-169
Housing and husbandry	15	Provide details of housing and husbandry conditions, including any environmental enrichment.	Materials and methods: Pages 3-4, lines 133-137
Animal care and monitoring	16	Describe any interventions or steps taken in the experimental protocols to reduce pain, suffering and distress.	Materials and methods: Page 4, lines 172-174
· ·		b. Report any expected or unexpected adverse events.	No adverse events were observed during the stud
		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.	Materials and methods: Page 4, line 169
Interpretation/ scientific	17	a. Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature.	Discussion: Pages 12-17, lines 498-703
implications		b. Comment on the study limitations including potential sources of bias, limitations of the animal model, and imprecision associated with the results.	Discussion: Pages 12-17, lines 554-556, 625-627, 684-685, 694-695 Conclusions: Page 18, lines 712-714
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).	Discussion: Pages 12-17, lines 498-518, 524-532, 583-591,629-634,637-643, 650-660, 684-687.
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.	This work was part of an undergraduate thesis, whose protocol was registered in the Research and Postgraduate Coordination-ICB-UACJ
Data access	20	Provide a statement describing if and where study data are available.	Data access: Page 18, lines 734-735
Declaration of interests	21	a. Declare any potential conflicts of interest, including financial and non-financial. If none exist, this should be stated.	Declaration of interests: Page 18, line 732.
		<ul> <li>List all funding sources (including grant identifier) and the role of the funder(s) in the design, analysis and reporting of the study.</li> </ul>	Funding statement: Page 18, lines 726-729.

