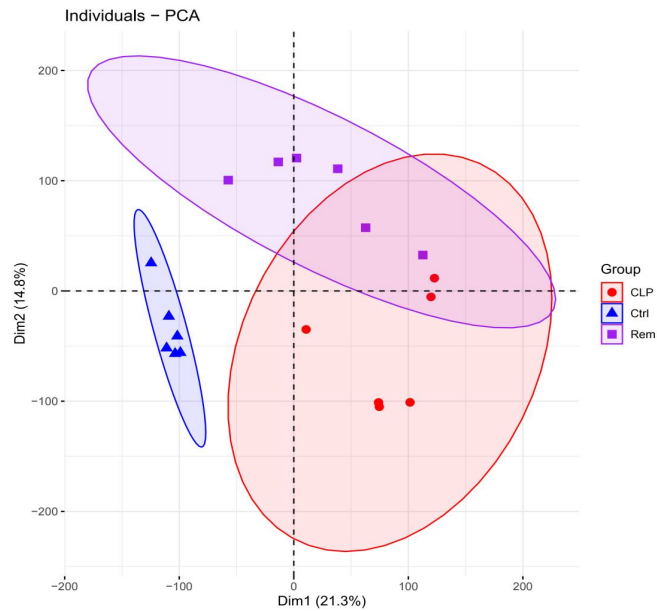


Supplementary Material

1 Supplementary Figures and Tables

1.1 Supplementary Figures



Supplementary Figure 1. Principal component analysis (PCA) of transcriptome profiles from the three experimental groups.

1.2 Supplementary Tables

Supplementary Table 1. Summary of RNA sequencing data and read mapping statistics for all samples.

Sample name	Sample 1								Sample name	Sample 2								Sample name	Sample 3							
	Total reads	Total mapped	Multiple mapped	Unique mapped	Reads mapped to '+'	Reads mapped to '-'	Non-splice reads	Splice reads		Total reads	Total mapped	Multiple mapped	Unique mapped	Reads mapped to '+'	Reads mapped to '-'	Non-splice reads	Splice reads		Total reads	Total mapped	Multiple mapped	Unique mapped	Reads mapped to '+'	Reads mapped to '-'	Non-splice reads	Splice reads
C1	54	5330	3943	4936	2465	2471	3414	1521	R1	68	6780	4522	6328	3162	3165	4343	1985	S1	93	9211	7165	8495	4243	4252	5889	2605
	36	3943	450	0493	0070	0423	9280	1213		99	8462	542	5920	6511	9409	1437	4483		93	9450	232	4218	0325	3893	8781	5437
	61	98.05	7.25	90.79	45.34	45.45	62.81	27.98		59	98.28	6.55	91.72	45.84	45.89	62.95	28.78		05	98.07	7.63	90.44	45.17	45.27	62.70	27.74
	16	%	%	%	%	%	%	%		86	%	%	%	%	%	%	%		76	%	%	%	%	%	%	%
C2	48	4709	3348	4374	2183	2191	2974	1399	R2	56	5502	3700	5132	2564	2567	3520	1612	S2	48	4786	3671	4419	2207	2211	3057	1362
	32	2301	983	3318	0812	2506	7196	6122		06	7256	668	6588	9111	7477	2024	4564		90	7869	925	5944	6900	9044	0109	5835
	33	97.45	6.93	90.52	45.18	45.35	61.56	28.96		61	98.15	6.60	91.55	45.75	45.80	62.79	28.76		98	97.87	7.51	90.36	45.14	45.22	62.50	27.86
	74	%	%	%	%	%	%	%		14	%	%	%	%	%	%	%		70	%	%	%	%	%	%	%
C3	56	5482	4071	5075	2534	2541	3493	1581	R3	53	5253	3531	4900	2448	2451	3381	1519	S3	56	5521	3889	5132	2562	2569	3497	1635
	05	4665	581	3084	0089	2995	7439	5645		56	2138	392	0746	2220	8526	0241	0505		51	3820	396	4424	7381	7043	2334	2090
	95	97.80	7.26	90.53	45.20	45.33	62.32	28.21		03	98.08	6.59	91.49	45.71	45.78	63.13	28.36		21	97.70	6.88	90.82	45.35	45.47	61.88	28.94
	04	%	%	%	%	%	%	%		78	%	%	%	%	%	%	%		92	%	%	%	%	%	%	%
C4	50	4936	3600	4576	2286	2290	3127	1449	R4	47	4686	3135	4372	2184	2187	2944	1427	S4	52	5130	3929	4737	2366	2371	3293	1444
	42	4442	412	4030	1397	2633	3580	0450		80	0284	573	4711	6945	7766	9819	4892		34	9157	196	9961	4447	5514	8813	1148
	01	97.91	7.14	90.77	45.34	45.42	62.03	28.74		89	98.02	6.56	91.46	45.70	45.76	61.60	29.86		84	98.01	7.51	90.51	45.21	45.30	62.92	27.59
	48	%	%	%	%	%	%	%		32	%	%	%	%	%	%	%		76	%	%	%	%	%	%	%
C5	46	4569	3342	4235	2114	2121	2891	1343	R5	59	5843	3722	5471	2733	2737	3745	1726	S5	65	6421	4261	5995	2995	3000	4106	1889
	64	8556	857	5699	4345	1354	8101	7598		66	9014	499	6515	7919	8596	4228	2287		56	8105	067	7038	3184	3854	2010	5028
	90	97.96	7.17	90.80	45.33	45.47	61.99	28.81		78	97.94	6.24	91.70	45.82	45.88	62.77	28.93		27	97.95	6.50	91.45	45.69	45.76	62.63	28.82
	26	%	%	%	%	%	%	%		96	%	%	%	%	%	%	%		14	%	%	%	%	%	%	%
C6	51	5035	3625	4673	2333	2339	3177	1495	R6	54	5364	3506	5013	2504	2508	3422	1591	S7	59	5814	3962	5417	2706	2711	3736	1681
	58	6122	684	0438	9246	1192	7559	2879		68	0198	594	3604	9181	4423	1068	2536		34	0854	470	8384	3445	4939	3640	4744
	81	97.61	7.03	90.58	45.24	45.34	61.60	28.99		03	98.10	6.41	91.68	45.81	45.87	62.58	29.10		97	97.96	6.68	91.29	45.60	45.69	62.95	28.33
	58	%	%	%	%	%	%	%		44	%	%	%	%	%	%	%		78	%	%	%	%	%	%	%

2. Supplementary File 1: ARRIVE 2.0 Checklist

The ARRIVE Essential 10: Compliance Questionnaire

Use this questionnaire to evaluate how well a manuscript complies with the ARRIVE Essential 10. It can be applied to any manuscript describing comparative experiments in living animals, by assessors such as journal staff, editors, or peer reviewers.

Item	Question(s)	Answers
1 Study Design	Are all experimental and control groups clearly identified?	☐ Yes, for at least one experiment ☐ No
	Is the experimental unit (e.g. an animal, litter or cage of animals) clearly identified?	☐ Yes, for at least one experiment ☐ No
2 Sample Size	Is the exact number of experimental units in each group at the start of the study provided (e.g. in the format 'n=')?	☐ Yes, for at least one experiment ☐ No
	Is the method by which the sample size was chosen explained?	☐ Yes, for at least one experiment ☐ No
3 Inclusion & Exclusion Criteria	Are the criteria used for including and excluding animals, experimental units, or data points provided?	☐ Yes, for at least one experiment ☐ No
	Are any exclusions of animals, experimental units, or data points reported, or is there a statement indicating that there were no exclusions?	☐ Yes, for at least one analysis ☐ No
4 Randomisation	Is the method by which experimental units were allocated to control and treatment groups described?	☐ Yes, for at least one experiment ☐ No
5 Blinding	Is it clear whether researchers were aware of, or blinded to, the group allocation at any stage of the experiment or data analysis?	☐ Yes, for at least one experiment ☐ No
6 Outcome Measures	For all experimental outcomes presented, are details provided of exactly what parameter was measured?	☐ Yes, for at least one experiment ☐ No
7 Statistical Methods	Is the statistical approach used to analyse each outcome detailed?	☐ Yes, for at least one analysis ☐ No
	Is there a description of any methods used to assess whether data met statistical assumptions?	☐ Yes, for at least one analysis ☐ No ☐ Not applicable
8 Experimental Animals	Are all species of animal used specified?	☐ Yes, for at least one experiment ☐ No
	Is the sex of the animals specified?	☐ Yes, for at least one experiment ☐ No ☐ Not applicable to species
	Is at least one of age, weight or developmental stage of the animals specified?	☐ Yes, for at least one experiment ☐ No
9 Experimental Procedures	Are both the timing and frequency with which procedures took place specified?	☐ Yes, for at least one experiment ☐ No
	Are details of acclimatisation periods to experimental locations provided?	☐ Yes, for at least one experiment ☐ No
10 Results	Are descriptive statistics for each experimental group provided, with a measure of variability (e.g. mean and SD, or median and range)?	☐ Yes, for at least one experiment ☐ No ☐ Not applicable to the type of data collected
	Is the effect size and confidence interval provided?	☐ Yes, for at least one experiment ☐ No ☐ Not applicable to the type of analysis used

Notes on questionnaire design

The ARRIVE guidelines are a useful resource for authors preparing manuscripts describing animal research, and also provide a framework to evaluate the transparency of those manuscripts. To assess reporting quality, numerous studies have in the past sought to operationalise reporting guidelines (including ARRIVE). Typically, this involves scoring a manuscript's degree of compliance with guideline items in a binary fashion (e.g. an item is either not reported or reported) [1-3], a graded fashion (e.g. not, partially, or completely reported) [4,5], or a combination of the two [6].

This questionnaire has been designed to be as concise and user-friendly as possible. The number of questions used to assess a manuscript's compliance has been kept to a minimum, and in most cases each question is designed to be answered in a binary fashion. Compliance with some Essential 10 sub-items is inherently impossible to judge in this way, instead requiring a subjective judgement on the level of detail provided. For this reason, not all sub-items are represented by a question in this questionnaire.

To facilitate binary answers, it has been necessary to identify the minimum information in a manuscript sufficient to comply with each question. The strengths of this approach include the relatively short length of the questionnaire (and the correspondingly low time burden of using it), and the avoidance of ambiguity that would arise from a graded answering system, in which an intermediate score (e.g. 'partially/insufficiently reported') could denote a number of distinct deficiencies in compliance with an item (e.g. either only part of the item was complied with, or only the reporting of some experiments in the manuscript complied with the item.)

Limitations of this approach centre on the necessity to identify the minimum information sufficient to comply with each question. In some cases, this has resulted in questions that require a guideline sub-item's criteria to have been fulfilled in the reporting of only one experiment in a manuscript. As a result, not all experiments in a manuscript may be described in a way that fulfils that criterion, despite the manuscript being considered to comply with the guidelines overall.

References

1. Hair *et al* (2020). *Res Integ Peer Rev*. doi: [10.1186/s41073-019-0069-3](https://doi.org/10.1186/s41073-019-0069-3)
2. Tihanyi *et al* (2019). *J Surg Res*. doi: [10.1016/j.jss.2018.10.038](https://doi.org/10.1016/j.jss.2018.10.038)
3. Zhao *et al* (2020). *BMC Vet Res*. doi: [10.1186/s12917-020-02664-1](https://doi.org/10.1186/s12917-020-02664-1)
4. Han *et al* (2017). *Plos One*. doi: [10.1371/journal.pone.0183591](https://doi.org/10.1371/journal.pone.0183591)
5. Chatzimanouil *et al* (2019). *J Am Soc Nephrol*. doi: [10.1681/ASN.2018050515](https://doi.org/10.1681/ASN.2018050515)
6. Leung *et al* (2018). *Plos One*. doi: [10.1371/journal.pone.0197882](https://doi.org/10.1371/journal.pone.0197882)