

How to improve scientific quality and animal welfare when planning animal studies

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[@adrian_3r](#)

norecopa.no/3radvances



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3rd International Conference Series on 3R's Research & Progress
Advances in Animal Models and Cutting-Edge Research in Alternatives

17-18 November 2022, Visakhapatnam, AP, India

Organised by Oncoseek Bio-Acasta Health



3radvances.com

Norecopa: PREPARE for better Science

Norway's National Consensus Platform for the
Three Rs: Replacement, Reduction and Refinement
and a source of *global* 3R resources



<https://norecopa.no>

Established in 2007

Norecopa: PREPARE for better Science

norecopa.no : an updated overview of global 3R resources



The screenshot shows the norecopa.no website interface. The header features the norecopa logo and navigation links: About Norecopa, Alternatives, Databases & Guidelines, Education & training, Legislation, Meetings, More resources, News, and PREPARE. A horizontal menu lists various topics such as Anaesthesia and analgesia, Animal facilities, Animal welfare organisations, Blood sampling, Culture, Email discussion lists, Environmental enrichment, Ethics, Experimental design and reporting, Harm, Health and safety, Health monitoring, Humane, Literature searches and systematic reviews, and Organisations. A search filters sidebar on the right includes sections for Search filters (Order by: Relevance, Typo tolerance: Default), Database (listing 11 databases with counts), Browse the databases (listing 7 categories with counts), and Search in the databases (listing 7 search criteria).

approx. 9,000 webpages
350,000 hits annually
7-8 detailed newsletters per year

Design and reporting of animal experiments

This page supplements advice given in [Section 4 of the PREPARE guidelines](#). PREPARE covers all aspects of design (including animal and facility related issues).

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NORSK ENGLISH

Search:

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[Fish 2005](#) | [Wildlife 2008](#) | [Fish 2009](#) | [Agricultural animals 2012](#) | [Field research 2017](#) | [Past meetings](#) | [Meetings Calendar](#) | [An informal guide to arranging a scientific meeting](#) | [Presentations](#)

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norecopa.no/meetings/meetings-calendar

Webinar and Meetings calendar

November 2022

- ▶ [Introduction to microbiological monitoring in rodents facilities](#), FGB course, 17 November 2022
- ▶ [Advances in Animal Models and Cutting-Edge Technologies](#), online meeting, 17-18 November 2022
- ▶ [Case Surgery](#), webinar (Rebecca LaFleur), 18 November 2022
- ▶ [FELASA Laboratory Animal Course on Primates](#), Göttingen, 21-25 November 2022
- ▶ [21st International Congress of the European Society of Toxicology In Vitro \(ESTIV\)](#), Barcelona, 21-25 November 2022
- ▶ [Recognition, prevention and alleviation of pain and distress in laboratory animals](#), online

+ webpages for past meetings and recorded meetings

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we welcome more from you!

Centres

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- [Culture of Care Network](#) ⓘ
- [ecopa](#) ⓘ
- [EU-NETVAL](#) ⓘ
- [EU3Rnet](#) ⓘ
- [FELASA](#) ⓘ
- [FESSACAL](#) ⓘ
- [Scand-LAS](#) ⓘ
- [Concordat on Openness](#) ⓘ

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Databases & Guidelines

Published lists of resources are difficult to search and quickly become outdated. Lists on a website are easier to search, but do not enable the use of filters or intelligent search engines.

Norecopa has therefore constructed four databases, which together with all the text on this website can be searched simultaneously using the search field at the top of every page.

- > [3R Guide](#): a global overview of **databases, guidelines, information centres, journals, email lists, regulations and policies** which may be of use when planning experiments which might include animals. [A quick overview of all the guidelines can be accessed here.](#) Norecopa has written several of these, including [the PREPARE guidelines for planning animal research and testing.](#)
- > [NORINA](#): a global overview of audiovisual aids and other items which may be used as **alternatives or supplements to animals in education and training** at all levels from junior school to University, including [dissection alternatives](#) and surgical simulators.
- > [TextBase](#): a global overview of **textbooks and other literature within laboratory animal science** and related topics.
- > [Classic AVs](#): a subset of NORINA covering **audiovisual aids that are based on older technology.**

These databases are updated regularly. [Please give us feedback](#) if you discover errors or omissions.

The Norecopa website also includes four other collections:

- > [NAL](#): a collection of literature references relating to [the 3Rs](#) from the US National Agricultural Library
- > European Commission datasets:
 - ▶ [3Rs Knowledge Sources](#): over 800 resources collected by the Commission in 2016
 - ▶ [3Rs Education and Training Resources](#), over 560 items collected in 2018
 - ▶ [Non-animal models for respiratory tract diseases](#), over 280 models identified in a literature review of over 21,000 publications

Here is [an alphabetical global list of all the databases](#) cited on the Norecopa website.

norecopa.no/databases-guidelines

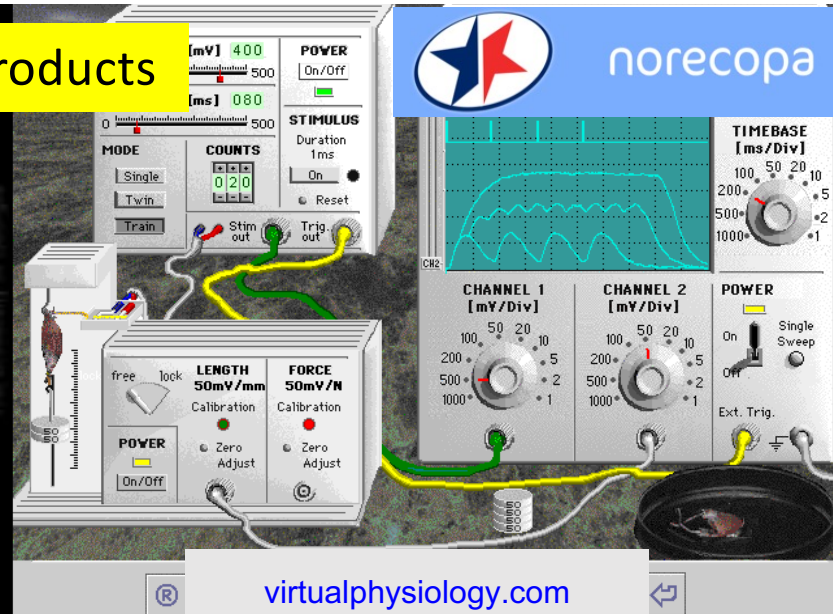
links to over 70 other databases

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NORINA database: approx. 3,000 products



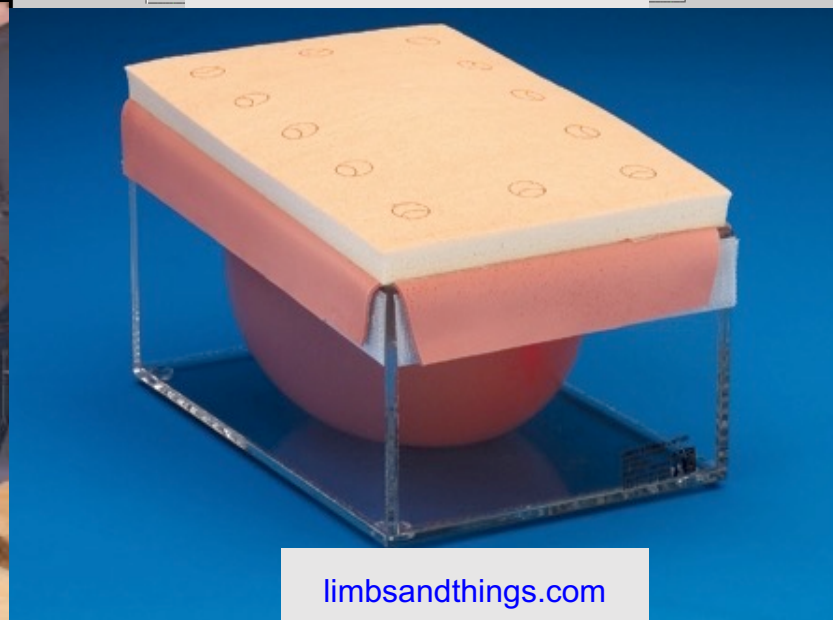
3dglasshorse.com



virtualphysiology.com



rescuecritters.com



limbsandthings.com

norecopa.no/education-training/homemade-educational-materials



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norecopa.no/education-training/films-and-slide-shows



Rat s.c. injection
Norecopa | 1,380 views



Testing anaesthetic depth in the chicken
Norecopa | 598 views



Blood sam
Norecop



Subcutaneous injection in the rabbit
Norecopa | 1,479 views



Rat i.p. injection (method 2)
Norecopa | 1,280 views



Blood collection from the saphenous vein in the mouse
Norecopa | 6,777 views



Blood sam
Norecop



Subcutaneous injection in the chicken
Norecopa | 1,806 views

ANATOMÍA DE LA RATA
Dra. Dolores Vallejo Ruiz
Departamento de Biología de Sistemas, Universidad de Alcalá (Madrid)
Asesoría Científica: Dr. José María Orellana Moriana
Centro de Experimentación Animal, CAI Medicina-Biología, Universidad de Alcalá

Anatomía de la rata
Norecopa | 977 views



Subcutaneous injection in the rat - Technique 1
Norecopa | 2,249 views



Blood sam
Norecop









Immobilisation of the rabbit
Norecopa | 2,072 views
















Lifting a rabbit
Norecopa | 2,420 views

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Training resources for animal research

 <p>National Legislation (EU1) Understand the national and international legal and regulatory framework within which projects involving animals are constructed and managed and of the legal responsibilities of the people involved.</p>	 <p>Ethics, Animal Welfare and the 3Rs (EU2) Identify the ethical and welfare issues raised by the use of animals in scientific procedures and understand the basic principles of the 3Rs.</p>
 <p>Basic and Appropriate Biology (EU3) Discover the basic principles of animal behaviour, care, biology and husbandry.</p>	 <p>Animal Care, Health and Management (EU4) Examine information on various aspects of animal health, care and management including: environmental controls, husbandry practices, diet, health status and disease.</p>
 <p>Recognition of Pain, Suffering and Distress (EU5) Identify the normal condition and behaviour of experimental animals and differentiate between a normal animal and one which is showing signs of pain, suffering or distress.</p>	 <p>Humane Methods of Killing (EU6.1) Learn the principles of humane killing including descriptions of the different methods available and information to help you compare the methods permitted to determine the most appropriate method.</p>
 <p>Minor Procedures without Anaesthesia (EU7) An introduction to the theory relating to minor procedures and information about appropriate methods of handling, restraint, appropriate techniques for injection, dosing and sampling relevant to the species.</p>	 <p>Anaesthesia for Minor Procedures (EU20) Guidance and information for individuals who, during their work with animals, will need to apply sedation or short-term anaesthesia for a brief period and mild pain level procedure.</p>

eModules

 <p>Click to access</p> <p>eModule – Recognition and Prevention of Pain, Suffering and Distress (EU5)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Humane Methods of Killing (EU6)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Design of procedures and projects (level 1) (EU10)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Design of procedures and projects (level 2) (EU11)</p> <p>ACCESS</p>
 <p>Click to access</p> <p>eModule – The Severity Assessment Framework (EU12)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Anaesthesia for Minor Procedures (EU20)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Pre-Anaesthetic Preparations (EU21-1)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Choosing an Anaesthetic (EU21-2)</p> <p>ACCESS</p>
 <p>Click to access</p> <p>eModule – Anaesthetic Monitoring and Intraoperative Care (EU21-3)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Anaesthetic Breathing Systems, Airway Management and Neuromuscular Blocking Agents (EU21-4)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Anaesthetic Management and Preventing Problems (EU21-5)</p> <p>ACCESS</p>	 <p>Click to access</p> <p>eModule – Post Anaesthetic Care (EU21-6)</p> <p>ACCESS</p>
 <p>Click to access</p> <p>eModule – Project Evaluation (EU25)</p>			

TextBase:

1,500 books related to LAS:

norecopa.no/textbase

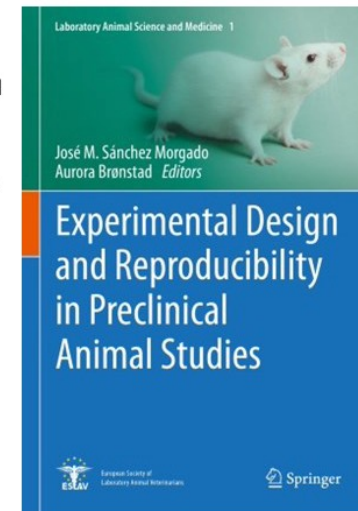
Experimental Design and Reproducibility in Preclinical Animal Studies

By José M. Sánchez Morgado & Aurora Brønstad (Eds.)

Record number: 8619d

This book provides grounds on how to plan and conduct animal experiments that can be reproduced by others. It touches on factors that may impact the reproducibility of animal studies including: the animal genetic background, the animal microbial flora, environmental and physiological variables affecting the animal, animal welfare, statistics and experimental design, systematic reviews of animal studies, and the publishing process.

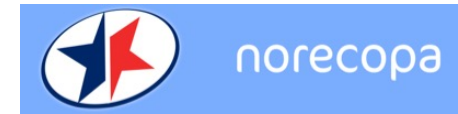
The book addresses advanced undergraduates, graduate students and all scientists working with animals.



norecopa.no/textbase/experimental-design-and-reproducibility-in-preclinical-animal-studies

3R-Guide (400 guidelines for animal research and testing)

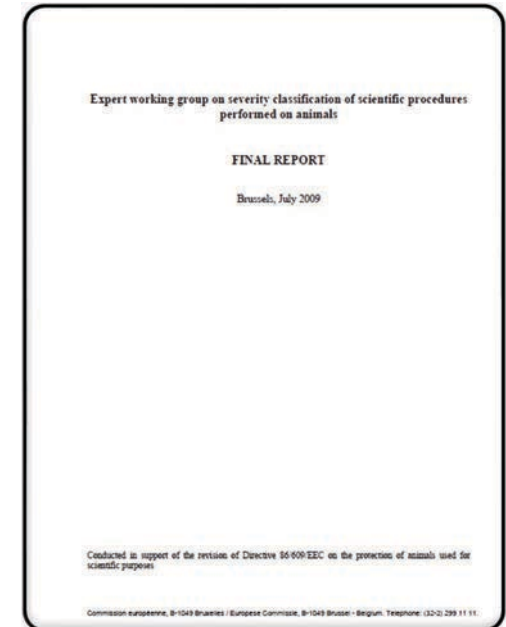
norecopa.no/3r-guide



Guidance on the severity classification of procedures involving fish

Report from a Working Group convened by Norecopa

Laboratory Animals, 2011
norecopa.no/categories



http://ec.europa.eu/environment/chemicals/lab_animals/pdf/report_ewg.pdf

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"...improve scientific quality and animal welfare?"

- valid data (a true treatment effect)
- reproducible and translatable experiments
- best possible animal welfare
- health & safety (of animals and people)
- a culture of care in the research group
- communication of best practice to others



colourbox.com

Scientists are becoming increasingly concerned about the validity of animal experiments

NATURE | NEWS

Swiss survey highlights potential flaws in animal studies

Poor experimental design and statistical analysis could contribute to widespread problems in reproducing preclinical animal experiments

Pain management in pigs undergoing experimental surgery; a literature review (2012–4) FREE

A. G. Bradbury, M. Eddleston, R. E. Clutton

Br J Anaesth (2016) 116 (1): 37-45. DOI: <https://doi.org/10.1093/bja/aev301>

Published: 03 October 2015

selection criteria. Most articles (193/233, 83%) described use of drugs with analgesic properties, but only 87/233 (37%) described postoperative analgesia. No article provided justification for the analgesic chosen, despite the lack of guidelines for analgesia in porcine surgical models and the lack of formal studies on this subject. Postoperative pain assessment was reported in only 23/233 (10%) articles. It was found that the reporting of postoperative pain management in the studies was remarkably low, reflecting either under-reporting or under-use. Analgesic description, when given, was frequently too limited to enable reproducibility. Development of a

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NATURE | NEWS FEATURE

1,500 scientists lift the lid on reproducibility


Survey sheds light on the 'crisis' rocking research.

Monya Baker

25 May 2016 | Corrected: 28 July 2016

More than 70% of researchers have tried and failed to reproduce another scientist's experiments, and more than half have failed to reproduce their own experiments. Those are some of the telling figures that emerged from *Nature's* survey of 1,576 researchers who took a brief online questionnaire on reproducibility in research.

We cannot improve our research by
better reporting alone...



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[reddit.com](https://www.reddit.com)



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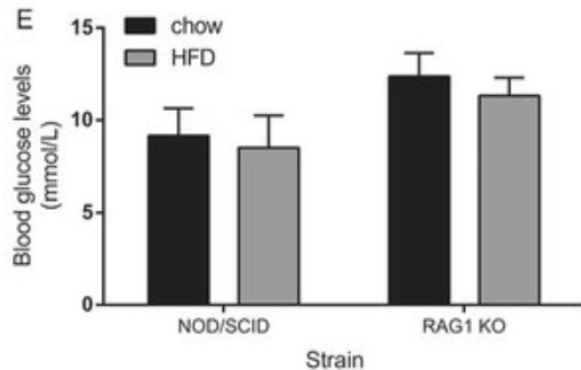
Frequently highlighted causes of the "reproducibility crisis"

1. **Publication bias** (reporting only positive results)
2. **Lack of randomisation and blinding**
3. **Low statistical power**
4. **P-value hacking** (manipulating data to obtain significance)
5. **HARKing** (Hypothesizing after the results are known)

norecopa.no/concerns



The scientist



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The mouse

Breeding

New social groups

Transportation

Acclimation to research facility

Allocation to experimental group

Adaptation to new diet

Handling and immobilisation

Blood sampling

often also:

injections, gavaging, surgery

pain and distress

developing illness and death

Contingent suffering



animalcaresystems.com

(not just the direct suffering caused by the procedure)

Fear, boredom and discomfort

Caused by, for example:

Transport, or changes in housing, husbandry and social groups

Single-housed male mice show symptoms of what in humans would be characterised as depression

<http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0111065>

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photo: colourbox.com

Stress caused by capture and handling



News > Science

Scores of scientific studies based on mice thrown into doubt because they were

Mice pick naturally

Ian Johnston

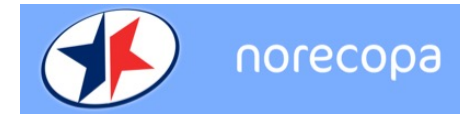


't act

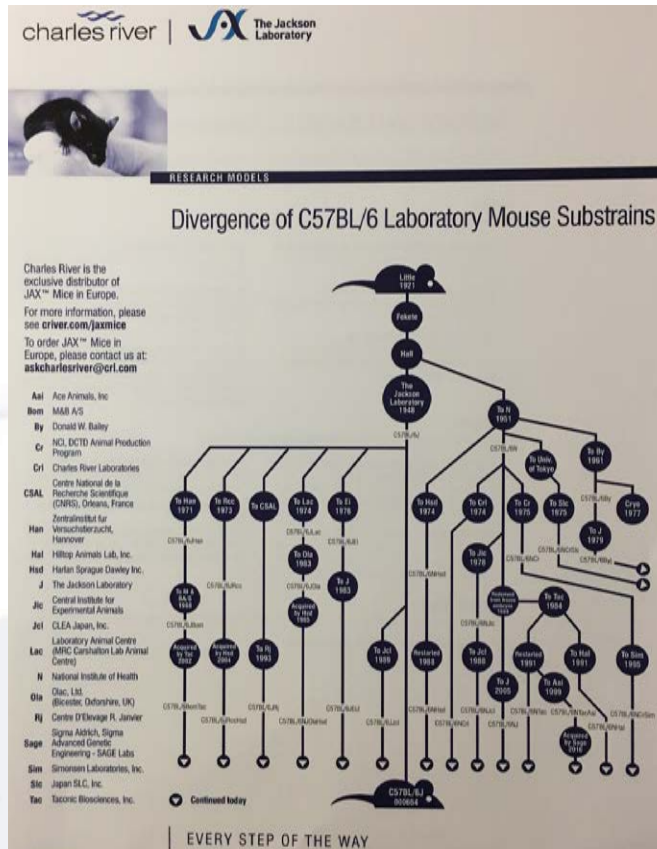
nc3rs.org.uk/3rs-resources/mouse-handling

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Some of the animal-related issues...

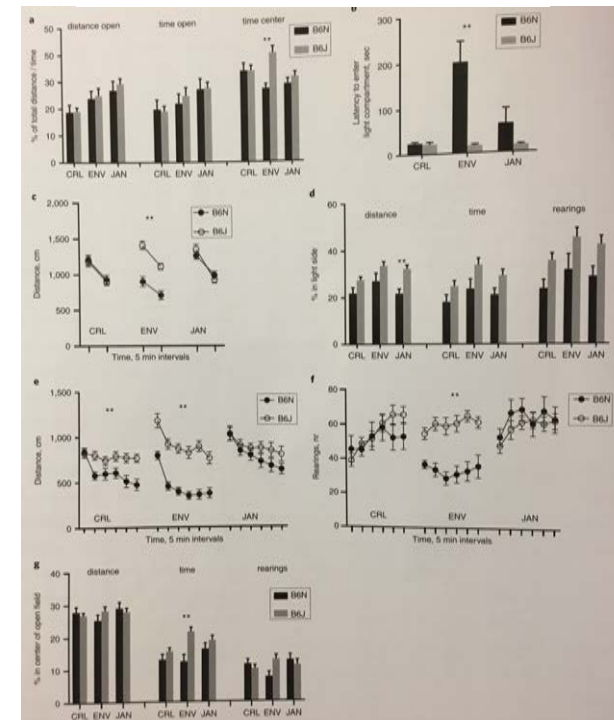


The C57BL/6 mouse



Åhlgren & Voikar (2019): Behavioural differences between /6J and /6N mice

nature.com/articles/s41684-019-0288-8



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Artefacts caused by poor administration techniques

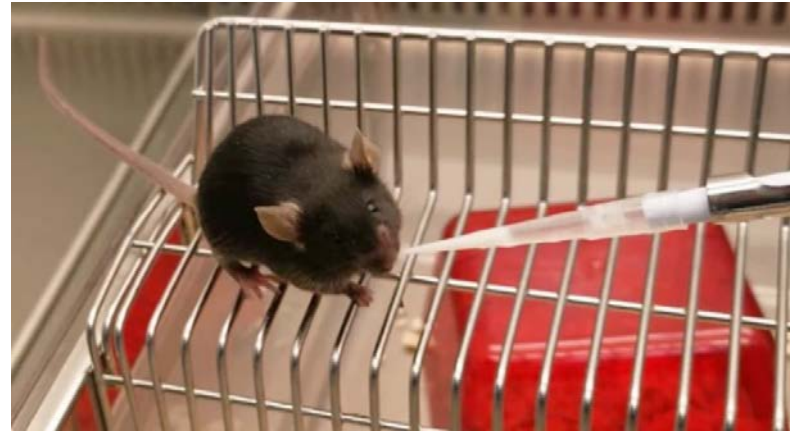


Photo: NMBU

- *Are you sure that your injection ends up in the same place each time?*
- *Are they realistic? (intramuscular injections in small animals)*



photo: NMBU



[nature.com/articles/s41684-021-00723-0.epdf](https://www.nature.com/articles/s41684-021-00723-0.epdf)



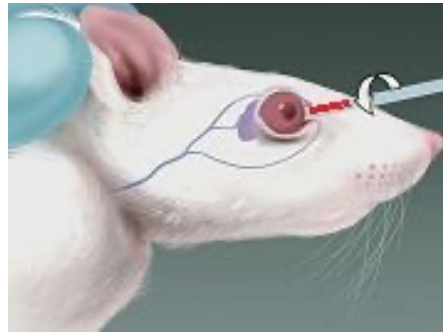
[youtube.com/watch?v=bdtVZtrr69c](https://www.youtube.com/watch?v=bdtVZtrr69c)

<https://norecopa.no/education-training/films-and-slide-shows>

'A simple' case: a researcher wants a blood sample



medipoint.com/html/for_use_on_mice.html



theodora.com/rodent_laboratory/blood_collection.html



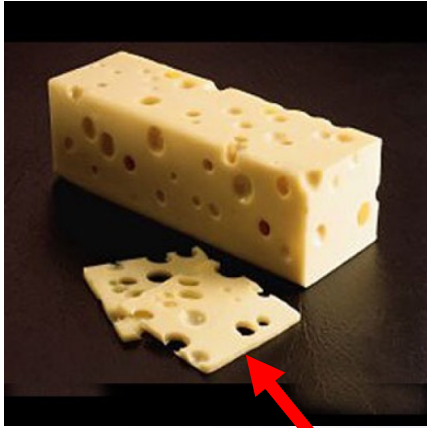
photo:NMBU

vimeo.com/486368886

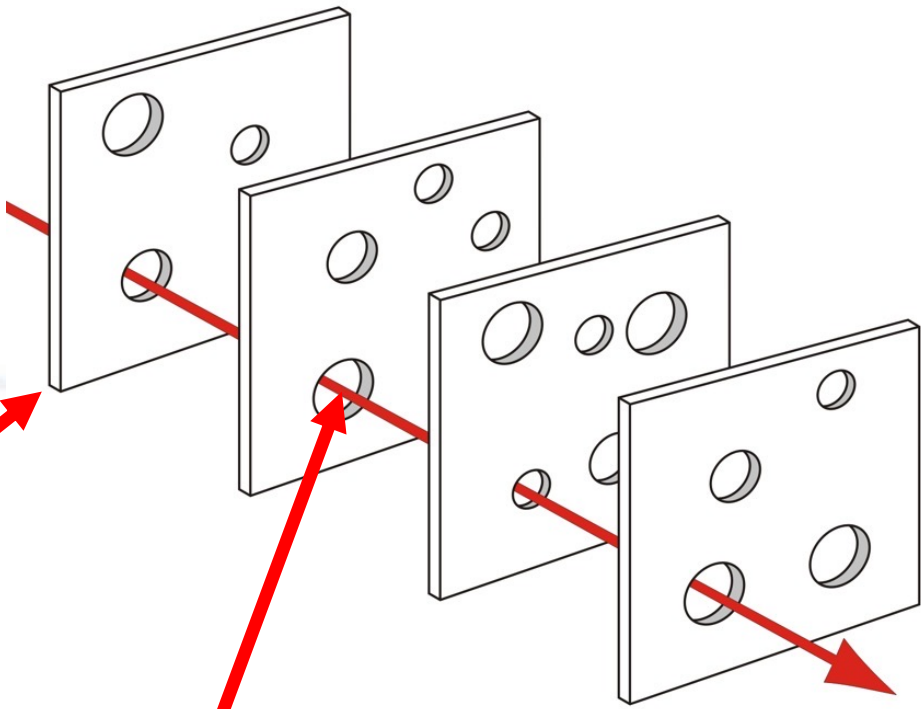
The best blood sampling techniques are those where you can:

- ✓ see the blood vessel
- ✓ regulate the amount of blood you remove
- ✓ stop the bleeding easily (including internal bleeding)
- ✓ avoid damage to the surrounding tissue
- ✓ collect samples rapidly, to avoid artefacts due to mechanical stress, temperature changes, differing lengths of sampling time

Threat and Error Management



eaugallecheese.com/Swiss-Cheese



"Layer of defence"
or redundancy

Weakness / hazard

Loss

wikipedia.org/wiki/Swiss_cheese_model



- A. Animal Care and Use Program**
- B. Animal environment, Housing and Management**
- C. Veterinary Care**
- D. Physical plant**

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A simple but effective Master Plan



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A contract between the animal facility and the research group

Division of labour, responsibilities and cost

Clarifying all stages of the experiment

Ensuring that all necessary data are recorded

	Animal facility	Researcher	Not applicable
Animal:			
Arrival date			
Species			
Strain/stock and substrain			
Supplier (full name and address) or bred on the premises			
Number and sex			
Age, weight, stage of life cycle on arrival			
Pre-treatment (surgical or medical) from supplier			
Quality (e.g. SPF, germ-free, gnotobiotic, conventional)			
Acclimation time before the start of the experiment			
Time and duration of fasting (with/without water and bedding)			
Environment:			
Type of housing: barrier/conventional			
Temperature (mean ± variation)			
Light schedule			
Relative humidity (mean ± variation)			
Number of air changes in the animal room/cabinet per hour			
Environmental enrichment			
Housing:			
Free-range, shelf, cabinet, isolator			
Cage type and size			
Number and method of distribution of animals per cage			

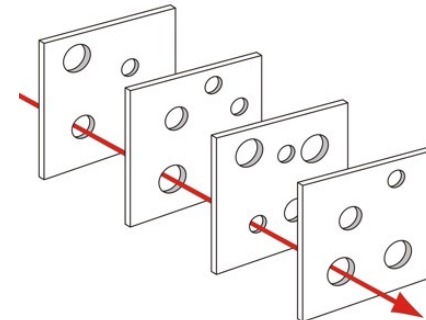
A Contingency Plan, based upon risk assessment

- Access to emergency services (police, fire, medical and veterinary help, security guards, personnel transport in cases of acute illness)
- Means of communication with staff members at all levels
- SOPs for acute illness, including
 - serious haemorrhages
 - fainting
 - allergic reactions

Many of these needed revision in the light of Covid-19
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- corrosive injuries
- and forms for reporting such injuries
- Firefighting, evacuation of personnel and animals
- Access to specialist services (e.g. ventilation system, plumbing, electrical installations, suppliers of equipment)
- Routines in cases of power failure, water leaks and (if applicable) natural disasters such as flooding
- Routines for emergency killing of animals
- Routines in cases of threats to the facility or personnel

<https://norecopa.no/prepare/6-facility-evaluation/master-plan-and-sops/contingency-plan>



Temporary staff at weekends and holidays

The pathway to better science



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norecopa.no/PREPARE and ivd-utrecht.nl/en/news/better-animal-research-through-open-science-1



Original Article

Laboratory Animals
0011-7
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sagepub.co.uk/journalsPermissions.nav
DOI: 10.1177/0023677217724823
journals.sagepub.com/home/lan
SAGE

PREPARE: guidelines for planning animal research and testing

Adrian J Smith¹, R Eddie Clutton², Elliot Litley³, Kristine E Aa Hansen⁴ and Trond Brattelid⁵

Abstract
There is widespread concern about the quality, reproducibility and translatability of studies involving research animals. Although there are a number of reporting guidelines available, there is very little overarching guidance on how to plan animal experiments, despite the fact that this is the logical place to start ensuring quality. In this paper we present the PREPARE guidelines: Planning Research and Experimental Procedures on Animals: Recommendations for Excellence. PREPARE covers the three broad areas which determine the quality of the preparation for animal studies: formulation, dialogue between scientists and the animal facility, and quality control of the various components in the study. Some topics overlap and the PREPARE checklist should be adapted to suit specific needs, for example in field research. Advice on use of the checklist is available on the Norecoba website, with links to guidelines for animal research and testing, at <https://norecoba.no/PREPARE>.

Keywords
guidelines, planning, design, animal experiments, animal research
Date received: 5 April 2017; accepted: 27 June 2017

Introduction
The quality of animal-based studies is under increasing scrutiny, for good scientific and ethical reasons. Studies of papers reporting animal experiments have revealed alarming deficiencies in the information provided,^{1,2} even after the production and journal endorsement of reporting guidelines.³ There is also widespread concern about the lack of reproducibility and translatability of laboratory animal research.⁴⁻⁷ This can, for example, contribute towards the failure of drugs when they enter human trials.⁸ These issues come in addition to other concerns, not unique to animal research, about publication bias, which tends to favour the reporting of positive results and can lead to the acceptance of claims as fact.⁹ This has understandably sparked a demand for reduced waste when planning experiments involving animals.¹⁰⁻¹² Reporting guidelines alone cannot solve the problem of wasteful experimentation, but thorough planning will increase the likelihood of success and is an important step in the implementation of the 3Rs of Russell & Burch (replacement, reduction, refinement).¹³ The importance of attention to detail at all stages is, in our experience, often underestimated by scientists. Even small practical details can cause omissions or artefacts that can ruin experiments which in all other respects have been well-designed, and generate health risks for all involved. There is therefore, in our opinion, an urgent need for detailed but overarching guidelines for researchers on how to plan animal experiments which are safe and scientifically sound, address animal

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Pre-published under Open Access on 3 August 2017, sponsored by the Universities Federation for Animal Welfare (UFAW), UK

<https://doi.org/10.1177/0023677217724823>



Over 25,000 downloads from the journal website so far

Also downloadable from norecoba.no/PREPARE

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How do others achieve reproducibility?



<https://www.meonuk.com/runway-markings-explained>



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...and precision in a variable environment?



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10-15 checklists even on short routine flights



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Checklists

- Reduce risk of **forgetting** to carry out vital actions
- Ensure checks are carried out in the **correct sequence**
- Encourage **cooperation** and **cross-checking** between crew members
- Make sure that everyone is "**on the same page**"

Too late to read the checklists when you have ARRIVED!



colourbox.com

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Hudson River, 2009

en.wikipedia.org

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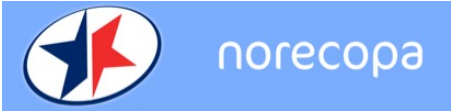


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travelandleisure.com/airlines-airports/what-happens-when-planes-hit-birds


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15.25.33	-01.38	Kaptein	Cockpit	V one, rotate
15.25.38	-01.33	Kaptein	Cockpit	positive rate
15.25.39	-01.32	Styrmann	Cockpit	Gear up please
15.25.39	-01.32	Kaptein	Cockpit	Gear up
15.26.37	-00.34	Kaptein	Cockpit	Uh what a view of the Hudson today
15.26.42	-00.29	Styrmann	Cockpit	Yeah
15.27.07	-00.04	Kaptein	Cockpit	After takeoff checklist complete
15.27.10	-00.01	Kaptein	Cockpit	Birds
15.27.11	-00.00	Styrmann	Cockpit	Whoa
15.27.11	00.00			
15.27.12	+00.01	Kaptein	Cockpit	Oh ---
15.27.13	+00.02	Styrmann	Cockpit	Oh yeah
15.27.14	+00.03	Styrmann	Cockpit	Uh oh
15.27.15	+00.04	Kaptein	Cockpit	We got one rol... both of 'em rolling back
15.27.18	+00.07	Kaptein	Cockpit	Ignition, start
15.27.21	+00.10	Kaptein	Cockpit	I'm starting the APU
15.27.23	+00.12	Kaptein	Cockpit	My aircraft
15.27.24	+00.13	Styrmann	Cockpit	Your aircraft
15.27.28	+00.17	Kaptein	Cockpit	Get the QRH... loss of thrust on both engines
15.27.32	+00.21	Kaptein	Radio	Mayday mayday mayday. Uh this is Cactus fifteen thirty [sic] nine, hit birds. We've lost thrust on both engines. We're turning back towards LaGuardia.

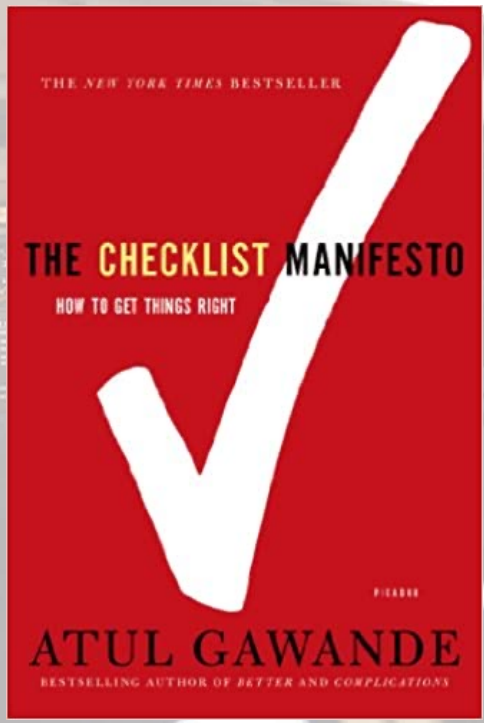
All 155 passengers and crew saved

Surgical Safety Checklist



World Health Organization | Patient Safety
A World Alliance for Safer Health Care

Before induction of anaesthesia <small>(with at least nurse and anaesthetist)</small>	Before skin incision <small>(with nurse, anaesthetist and surgeon)</small>	Before patient leaves operating room <small>(with nurse, anaesthetist and surgeon)</small>
Has the patient confirmed his/her identity, site, procedure, and consent? <input type="checkbox"/> Yes	<input type="checkbox"/> Confirm all team members have introduced themselves by name and role. <input type="checkbox"/> Confirm the patient's name, procedure, and where the incision will be made.	Nurse Verbally Confirms: <input type="checkbox"/> The name of the procedure <input type="checkbox"/> Completion of instrument, sponge and needle counts <input type="checkbox"/> Specimen labelling (read specimen labels aloud, including patient name) <input type="checkbox"/> Whether there are any equipment problems to be addressed
Is the site marked? <input type="checkbox"/> Yes <input type="checkbox"/> Not applicable	Has antibiotic prophylaxis been given within the last 60 minutes? <input type="checkbox"/> Yes <input type="checkbox"/> Not applicable	To Surgeon, Anaesthetist and Nurse: <input type="checkbox"/> What are the key concerns for recovery and management of this patient?
Is the anaesthesia machine and medication check complete? <input type="checkbox"/> Yes	Anticipated Critical Events To Surgeon: <input type="checkbox"/> What are the critical or non-routine steps? <input type="checkbox"/> How long will the case take? <input type="checkbox"/> What is the anticipated blood loss? To Anaesthetist: <input type="checkbox"/> Are there any patient-specific concerns? To Nursing Team: <input type="checkbox"/> Has sterility (including indicator results) been confirmed? <input type="checkbox"/> Are there equipment issues or any concerns?	
Is the pulse oximeter on the patient and functioning? <input type="checkbox"/> Yes	Is essential imaging displayed? <input type="checkbox"/> Yes <input type="checkbox"/> Not applicable	
Does the patient have a: Known allergy? <input type="checkbox"/> No <input type="checkbox"/> Yes Difficult airway or aspiration risk? <input type="checkbox"/> No <input type="checkbox"/> Yes, and equipment/assistance available Risk of >500ml blood loss (7ml/kg in children)? <input type="checkbox"/> No <input type="checkbox"/> Yes, and two IVs/central access and fluids planned		



This checklist is not intended to be comprehensive. Additions and modifications to fit local practice are encouraged.

Revised 1 / 2009

© WHO, 2009

who.int/patientsafety/topics/safe-surgery/checklist/en

amazon.com/gp/product/0312430000

PREPARE:

Planning Research and Experimental Procedures on Animals: Recommendations for Excellence

PREPARE covers 15 topics:

Formulation of the study

1. Literature searches
2. Legal issues
3. Ethical issues, harm-benefit assessment and humane endpoints
4. Experimental design and statistical analysis

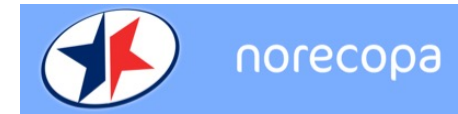
Dialogue between scientists and the animal facility

5. Objectives and timescale, funding and division of labour
6. Facility evaluation
7. Education and training
8. Health risks, waste disposal and decontamination

Methods

9. Test substances and procedures
10. Experimental animals
11. Quarantine and health monitoring
12. Housing and husbandry
13. Experimental procedures
14. Humane killing, release, reuse or rehoming
15. Necropsy

Items in pink are
not typically
highlighted in
reporting guidelines



PREPARE



The PREPARE Guidelines Checklist Planning Research and Experimental Procedures on Animals: Recommendations for Excellence

Adrian J. Smith¹, R. Eddie Clutton², Elliot Lilley³, Kristine E. Aa. Hansen⁴ & Trond Bratteli⁵
¹Norecopa, c/o Norwegian Veterinary Institute, P.O. Box 750 Sentrum, 0106 Oslo, Norway; ²Royal (Dick) School of Veterinary Studies, Easter Bush, Midlothian, EH25 9RG, U.K.; ³Research Animals Department, Science Group, RSPCA, Wilberforce Way, Southwater, Horsham, West Sussex, RH13 9RS, U.K.; ⁴Section of Experimental Biomedicine, Department of Production Animal Clinical Sciences, Faculty of Veterinary Medicine, Norwegian University of Life Sciences, P.O. Box 8146 Dep., 0033 Oslo, Norway; ⁵Division for Research Management and External Funding, Western Norway University of Applied Sciences, 5020 Bergen, Norway.

PREPARE[®] consists of planning guidelines which are complementary to reporting guidelines. PREPARE covers the three broad areas which determine the quality of animal research: the 3Rs (Replacement, Reduction, Refinement) and the 3Ss (Good Science, Good Sense, Good Sensibilities).

1. Formulation of the study
 2. Dialogue between scientists and the animal facility

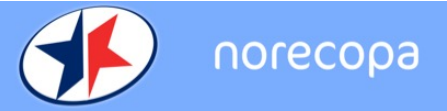
Topic	Recommendation
(A) Formulation of the study	
1. Literature searches	<input type="checkbox"/> Form a clear hypothesis, with primary and secondary outcomes. <input type="checkbox"/> Consider the use of systematic reviews. <input type="checkbox"/> Decide upon databases and information specialists to be consulted, and construct search terms. <input type="checkbox"/> Assess the relevance of the species to be used, its biology and suitability to answer the experimental questions with the least suffering and to welfare needs. <input type="checkbox"/> Assess the reproducibility and translatability of the project.
2. Legal issues	<input type="checkbox"/> Consider how the research is affected by relevant legislation for animal research and other areas, e.g. animal transport, occupational health and safety. <input type="checkbox"/> Locate relevant guidance documents (e.g. EU guidance on project evaluation).
3. Ethical issues, harm-benefit assessment and humane endpoints	<input type="checkbox"/> Construct a lay summary. <input type="checkbox"/> In dialogue with ethics committees, consider whether statements about this type of research have already been produced. <input type="checkbox"/> Address the 3Rs (replacement, reduction, refinement) and the 3Ss (good science, good sense, good sensibilities). <input type="checkbox"/> Consider pre-Registration and the publication of negative results. <input type="checkbox"/> Perform a harm-benefit assessment and justify any likely animal harm. <input type="checkbox"/> Discuss the learning objectives, if the animal use is for educational or training purposes. <input type="checkbox"/> Allocate a severity classification to the project. <input type="checkbox"/> Define objective, easily measurable and unequivocal humane endpoints. <input type="checkbox"/> Discuss the justification, if any, for death as an end-point.
4. Experimental design and statistical analysis	<input type="checkbox"/> Consider pilot studies, statistical power and significance levels. <input type="checkbox"/> Define the experimental unit and decide upon animal numbers. <input type="checkbox"/> Choose methods of randomisation, prevent observer bias, and decide upon inclusion and exclusion criteria.

Animal welfare and Three Rs!

Topic	Recommendation
(B) Dialogue between scientists and the animal facility	
5. Objectives and timescale, funding and division of labour	<input type="checkbox"/> Arrange meetings with all relevant staff when early plans for the project exist. <input type="checkbox"/> Construct an approximate timescale for the project, indicating the need for assistance with preparation, animal care, procedures and waste disposal/decontamination. <input type="checkbox"/> Discuss and disclose all expected and potential costs. <input type="checkbox"/> Construct a detailed plan for division of labour and expenses at all stages of the study.
6. Health risks, waste disposal and decontamination	<input type="checkbox"/> Perform a risk assessment, in collaboration with the animal facility, for all persons and animals affected directly or indirectly by the study. <input type="checkbox"/> Assess, and if necessary produce, specific guidance for all stages of the project. <input type="checkbox"/> Discuss means for containment, decontamination, and disposal of all items in the study.
(C) Quality control of the components in the study	
9. Test substances and procedures	<input type="checkbox"/> Provide as much information as possible about test substances. <input type="checkbox"/> Consider the feasibility and validity of test procedures and the skills needed to perform them.
10. Experimental animals	<input type="checkbox"/> Decide upon the characteristics of the animals that are essential for the study and for reporting. <input type="checkbox"/> Avoid generation of surplus animals.
11. Quarantine and health monitoring	<input type="checkbox"/> Discuss the animals' likely health status, any needs for transport, quarantine and isolation, health monitoring and consequences for the personnel.
12. Housing and husbandry	<input type="checkbox"/> Attend to the animals' specific instincts and needs, in collaboration with expert staff. <input type="checkbox"/> Discuss acclimatisation, optimal housing conditions and procedures, environmental factors and any experimental limitations on these (e.g. food deprivation, solitary housing).
13. Experimental procedures	<input type="checkbox"/> Develop refined procedures for capture, immobilisation, marking, and release or rehoming. <input type="checkbox"/> Develop refined procedures for substance administration, sampling, sedation and anaesthesia, surgery and other techniques.
14. Humane killing, release, reuse or rehoming	<input type="checkbox"/> Consult relevant legislation and guidelines well in advance of the study. <input type="checkbox"/> Define primary and emergency methods for humane killing. <input type="checkbox"/> Assess the competence of those who may have to perform these tasks.
15. Necropsy	<input type="checkbox"/> Construct a systematic plan for all stages of necropsy, including location, and identification of all animals and samples.

References
 1. Smith AJ, Clutton RE, Lilley E, Hansen KEA & Bratteli T. PREPARE: Guidelines for Planning Animal Research and Testing. *Laboratory Animals*, 2017, DOI: 10.1177/0023677217724823.
 2. Kilkenny C, Browne WJ, Cuthill IC et al. Improving Bioscience Research Reporting: The ARRIVE Guidelines for Reporting Animal Research. *PLoS Biology*, 2010; DOI: 10.1371/journal.pbio.1000412.

Further information
<https://norecopa.no/PREPARE> | post@norecopa.no | [@norecopa](https://twitter.com/norecopa)



In addition to the checklist, much more information is available on:

norecopa.no/PREPARE



A screenshot of the norecopa.no website. The header is blue with the Norecopa logo and the word "norecopa" in white. To the right, there are language options for "NORSK" and "ENGLISH", and a search bar with a magnifying glass icon. Below the header is a navigation menu with several items: "About Norecopa", "Alternatives", "Databases & Guidelines", "Education & training", "Legislation", "Meetings", "More resources", "News", "PREPARE" (which is circled in red), and "Species". Below the navigation menu is a list of links for the PREPARE Checklist, including: "1-Literature searches", "2-Legal issues", "3-Ethical issues, Harm-Benefit Assessment and humane endpoints", "4-Experimental design and statistical analysis", "5-Objectives and timescale, funding and division of labour", "6-Facility evaluation", "7-Education and training", "8-Health risks, waste disposal and decontamination", "9-Test substances and procedures", "10-Experimental animals", "11-Quarantine and health monitoring", "12-Housing and husbandry", "13-Experimental procedures", "14-Humane killing, release, re-use or re-homing", and "15-Necropsy". At the bottom of the page, there is a breadcrumb trail "norecopa.no / PREPARE" and social media icons for Facebook, Twitter, Email, and a plus sign for more options.

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norecopa.no/PREPARE

- 3-Ethical issues, harm-benefit assessment and humane endpoints
 - 3a Construct a lay summary.
 - 3b In dialogue with ethics committees, consider whether statements about this type of research have already been produced.
 - 3c Address the 3Rs (Replacement, Reduction, Refinement) and the 3Ss (Good Science, Good Sense, Good Sensibilities).
 - 3d Assessment and justify any likely animal harm.
 - 3f Discuss the learning objectives, if the animal use is for educational or training purposes.
 - 3g Allocate a severity classification to the project.
 - 3h Define objective, easily measurable and unequivocal humane endpoints.
 - 3i Discuss the justification, if any, for death as an end-point.
- 4-Experimental design and statistical analysis

5. Have the experiments been carried out before, and is any repetition justifiable?
6. What [approaches to reduce distress](#) have been considered?

3a Construct a lay summary.

- General principles
- For fish researchers**

1. Have national or local research ethics committees already produced statements relevant to the research being planned? Consideration should also be paid to the broader context of the research. For example, research directed at increasing the productivity of farming at the expense of (or without improving) individual animal welfare, or wildlife research whose primary aim is population management.

Links to quality guidelines and scientific papers worldwide on e.g. blood sampling, injection volumes, housing and husbandry, analgesia, humane endpoints, experimental design

and will any advances in this research only index the title and abstract? Will the project be rejected?

3. Have the Three S's ([Good Science, Good Sense and Good Sensibilities](#)) been addressed? Sufficient time should be allocated to this point, since two of the three S's are highly subjective, but equally important. The use of commonsense and critical anthropomorphism are justifiably part of the work to assess the impact of research on animals, not least when a scientific evidence base does not exist.
4. Does the proposed study have a clear rationale and scientific relevance, and what will be the next step if the hypothesis is supported or rejected?
5. Have the experiments been carried out before and is any repetition justifiable?
6. What [approaches to reduce distress](#) have been considered?
7. Will the project undergo [pre-registration](#) and will negative results be published, to avoid publication bias?

Many more [links to resources on ethics are available here](#). Details about pre-registration of animal studies and reporting of critical incidents are to be found in the section on [Experimental Design and Statistical Analysis](#).

Harm-Benefit Assessment

PREPARE encourages scientists to collaborate with animal carers and technicians from Day 1

- they have a right to know and will be more motivated
- they know the possibilities (and limitations) in the animal facility
- they often possess a large range of practical skills and are good at lateral thinking
- they know the animals best
- the animals know them best
- lack of involvement creates anxiety, depression and opposition to animal research, as well as limiting creativity which might improve the experiments

While we are waiting for the scientific evidence...

Carol M. Newton (1925-2014)



National Library of Medicine

The three S's

- *Good Science*
- *Good Sense*
- *Good Sensibilities*

<https://norecopa.no/3S>

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Culture of Care

The International Culture of Care Network
norecopa.no/coc

A demonstrable commitment, throughout the establishment, to improving:

- animal welfare
- scientific quality
- care of staff
- transparency for all stakeholders, including the public

It goes beyond simply complying with the law!

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Communication and the Culture of Care

Penny Hawkins, RSPCA Research Animals Department
on behalf of the International Culture of Care Network*

Effective two-way communication between scientists and animal technologists is essential for a good Culture of Care
The European Commission suggests the 'development of formal and informal communication channels, for mutual benefit with respect to science and animal welfare'
Here are some examples from International Culture of Care network members

Regular meetings

Scheduled meetings for scientists, animal technologists, vets, unit managers and AWERB members



Regular refresher/update meetings for all organised by NTCO



Special events

Duo-talks: researcher talks about their science, and animal technologists talk about techniques and animal care within the project



ELH organises an informal meeting for all, in which anyone can raise welfare issues



Building communication into existing processes

Each study has a pre-start and wash-up meeting involving everybody



Three Rs improvements reported to AWERB & shared at external user meetings



Other ideas

A 'boxless' event: anyone can submit 'out of the box' ideas to improve practice



A staff survey for all e.g. how much do you agree with statements such as 'in our group we listen to each others' ideas about animal welfare'



*norecopa.no/culture-of-care

Culture of Care facilitates honest discussion



"because we've always done it that way"

"as often as necessary"

"there are no alternatives"

Closely related to a culture of care is

a **Culture of Challenge** (Louhimies, 2015).

Look for the acceptable, rather than choosing the accepted.



Culture of Care Network

norecopa.no/coc



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norecopa.no/PREPARE and ivd-utrecht.nl/en/news/better-animal-research-through-open-science-1

arriveguidelines.org

The ARRIVE guidelines 2.0

This section of the website provides detailed explanations about each item of the guidelines. Use the left-hand side menu to navigate to each item.

To facilitate a step-wise approach to improving reporting, the guidelines are organised into two prioritised sets:

ARRIVE Essential 10

These ten items are the basic minimum that must be included in any manuscript describing animal research. Without this information readers and reviewers cannot assess the reliability of the findings.

Recommended Set

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

Norecopa: PREPARE for better Science

The screenshot shows the ARRIVE guidelines website. The top navigation bar includes 'Home', 'About', 'ARRIVE guidelines', 'Supporters', 'Resources', 'Publications', and 'News'. The left sidebar contains a list of guidelines, with '11. Abstract' highlighted under the 'Recommended Set' category. The main content area is titled '11. Abstract' and includes a summary box: '11 Provide an accurate summary of the research objectives, animal species, strain and sex, key methods, principal findings, and study conclusions.' Below this are tabs for 'Explanation' and 'Examples'. The 'Explanation' tab is active, showing a paragraph: 'A transparent and accurate abstract increases the utility and impact of the manuscript, and allows readers to assess the reliability of the study [1]. The abstract is often used as a screening tool by readers to decide whether to read the full article or whether to select an article for inclusion in a systematic review. However, abstracts often either do not contain enough information for this purpose [2], or contain information that is inconsistent with the results in the rest of the manuscript [3,4]. In systematic reviews, initial screens to identify papers are based on titles, abstracts and keywords [5]. Leaving out of the abstract information such as the species of animal used or the drugs being tested, limits the value of preclinical systematic reviews as relevant studies cannot be identified and included. For example, in a systematic review of the effect of the MVA85A vaccine on tuberculosis challenge in animals, the largest preclinical trial did not include the vaccine name in the abstract or keywords of the publication, the paper was only included in the systematic review following discussions with experts in the field [6]. To maximise utility, include details of the species, sex and strain of animals used, and accurately report the methods, results and conclusions of the study. Also describe the objectives of the study, including whether it was designed to either test a specific hypothesis or to generate a new hypothesis (see item 13 – Objectives). Incorporating this information will enable readers to interpret the strength of evidence, and judge how the study fits within the wider knowledge base.' Below the text is a 'References' section with two entries: 1. Haynes RB, Mulrow CD, Huth EJ, Altman DG and Gardner MJ (1990). More informative abstracts revisited. *Ann Intern Med.* doi: 10.7326/0003-4819-113-1-69; 2. Hair K, Macleod MR, Sena ES, Sena ES, Hair K, Macleod MR, Howells D, Bath P, Irvine C, MacCallum C, Morrison G, ...

There are three broad areas which need to be considered when planning animal studies:

1. The suitability of the species or strain as a model of the target organism
2. The ethical issues surrounding their use: '[choosing the right animal for the right reason](#)'. The large increase in use of genetically altered lines has created increasing [concern about the suitability of these animals as models of human conditions](#).
3. Characterisation of the animals. Items to be considered, in collaboration with the supplier, include:
 - > Species, strain, line and phenotype (with an explanation of any genetic modifications)
 - > Age, developmental stage, sex and weight
 - > Stage of oestrous cycle and any previous breeding history
 - > Any necessary pre-treatment (e.g. castration) for this
 - > Name and address of the supplier/breeder, method of capture and transport
 - > [Health status](#) (e.g. germ-free, gnotobiotic, SPF)
 - > Re-use of animals, which should be justified by legislation
 - > Any plans for release or re-homing, which must be justified

More resources

- > [Examples and references](#) from the NC3Rs
- > [Information on inbred strains of mice and rats](#)
- > [Strategies to minimise genetic drift and maximise experimental reproducibility in mouse research](#)
- > [Mouse Locator, UK](#)
- > [The Collaborative Cross panel of inbred mouse strains](#)
- > [Nude mice - more than what meets the eye](#)
- > [The Rat Guide](#)
- > [Rat Behavior and Biology](#)



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"We ARRIVED, because we were PREPARED"

- ✓ *Better Science*
- ✓ *Improved animal welfare*
- ✓ *Advancement of the 3Rs*
- ✓ *Safer working environment*

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vimeo.com/358069203 or norecopa.no/PREPARE/film
 3-minute cartoon film



Norecopa: PREPARE for better Science

norecopa.no/Europhysiology

 norecopa
PREPARE for better Science

Let's PREPARE together to ARRIVE in better shape: how to plan animal experiments

Adrian Smith, Norecopa, c/o Norwegian Veterinary Institute, P.O. Box 64, 1431 Ås, Norway
adrian.smith@norecopa.no

What's the problem?

Preclinical *in vivo* research needs to be reproducible and translatable, while maximising the animals' welfare and replacing them with alternatives wherever possible. This can be summed up in the 3Rs of Russell & Burch: **Replace, Reduce & Refine**.

Scientists are usually well aware of **reporting** guidelines when publishing research. These are important, but a sub-standard study, like a burnt cake, cannot be improved by a better description. Guidelines for **planning**, although not mandatory, are of great help in designing better experiments.



The path to better science



norecopa.no/PREPARE

What can Norecopa offer?

Norecopa maintains a comprehensive database of resources for scientists, which include:

- 8,900 searchable webpages of quality 3R resources, with filters to facilitate searching
- the PREPARE guidelines for planning animal experiments, with a checklist in over 30 languages
- links to recordings of webinars covering all aspects of animal research
- an International Webinars & Meetings Calendar
- a collection of 400 guidelines for planning and conducting animal research
- an English-language newsletter with the latest developments within experimental design
- the NORINA database of alternatives to animal use in education and training
- a Refinement Wiki



Examples of Norecopa's resources:

PREPARE covers:

- ✓ Formulation of a study
- ✓ Dialogue between scientists and the animal facility
- ✓ Quality control of the components in the study



The Refinement Wiki
wiki.norecopa.no

Norecopa gratefully acknowledges financial support from:
The Norwegian Parliament, the Ministry of Agriculture & Food and the Ministry of Trade, Industry & Fisheries; the Nordic Society against Painful Experiments (NSMSO), Novo Nordisk, the Norwegian Animal Protection Alliance (Dyrevernalliansen), the Norwegian Society for Protection of Animals (Dyreskytelseen Norge), the Research Council of Norway, Laboratory Animals Ltd., the Royal Society for the Prevention of Cruelty to Animals (RSPCA), Sanofi, the Scottish Accreditation Board, the Stånsen Foundation, the Universities Federation of Animal Welfare (UFAW) and the US Department of Agriculture (USDA).

Boxbox graphics: colorbox.com

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Thanks to Norecopa's main sponsors:

- Standing Committee on Business Affairs, Norwegian Parliament
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- Laboratory Animals Ltd.
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- Nordic Society Against Painful Experiments (NSMSD)
- Norwegian Society for Animal Protection (Dyrebeskyttelsen Norge)
- Norwegian Animal Protection Alliance (Dyrevernalliansen)
- Novo Nordisk
- Sanofi
- Scottish Accreditation Board (SAB)
- Stiansen Foundation
- Universities Federation for Animal Welfare (UFAW)
- US Department of Agriculture (USDA)

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Dyrevernalliansen

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Newsletter no. 3-2022 from Norecopa

Welcome to Norecopa's third newsletter in 2022!

Please share this newsletter with your colleagues and friends!

Norecopa maintains [an international Webinars and Meetings Calendar](#), which is updated several times a week, with links to [recorded webinars and events here](#).

You will find shortcuts to several other key resources on [our front page](#).

We continue to maintain a list of resources related to the Covid-19 pandemic and about preparedness in general: [Be PREPAREd](#). Let us know if you have additions.

You can tip a friend, subscribe or unsubscribe, and share the newsletter on social media using the links above. We are on [Facebook](#), [Twitter](#) and [LinkedIn](#).

[All Norecopa's newsletters can be read here](#) and their content is indexed by the search engine on [Norecopa's website](#).

This newsletter contains the following items (if some links do not work, check that your mail program has opened the whole of the newsletter):

- [General update on Norecopa's activities](#)
- [Overview: The rise of European 3R centres](#)
- [Quality assurance of Norecopa's website](#)
- [Infographics about non-animal methods](#)
- [Resources from the RSPCA](#)
- [European overview: Non-animal methods in science](#)
- [Framework proposal for intensive 3R courses](#)
- [News from other 3R Centres](#)
- [Resources from Understanding Animal Research](#)
- [Prizes and Grants](#)
- [New books on animal welfare](#)
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Thank you for listening!