Directive 2010/63/EU

EU Guidance Document on Non-technical Project Summaries under Directive 2010/63/EU

Laboratory animals and alternatives: the way forward
Brussels 27th of March 2023
Patri Vergara

Two key documents for the public

Non-technical project summaries:

- *Layman language **summary of the authorised project**, which presents the project following the **project evaluation carried out by CA***

*Updates to the NTS with the **results of RA (RAR):***

- *Layman language **summary of the results** of the **retrospective assessment carried out by CA***

Non-technical project summaries (NTS)

*"To **improve transparency** and **understanding** of the reasons animals are still needed, objective information should be made publicly available."*

Authorised projects:

- *Information on the **objectives** of the project, including the **predicted harm** and **benefits** and the **number and types of animals** to be used*
- *A demonstration of **compliance** with the requirement of **replacement, reduction and refinement***

Content of NTS

Objectives and predicted benefits of the project	
Describe the objectives of the project (for example, addressing certain scientific unknowns, or scientific or clinical needs).	
What are the potential benefits likely to derive from this project? Explain how science could be advanced, or humans, animals or environment may ultimately benefit from the project. Where applicable, differentiate between short-term benefits (within the duration of the project) and long-term benefits (which may accrue after the project is finished).	

Predicted harms
In what procedures will the animals typically be used (for example, injections, surgical procedures)? Indicate the number and duration of these procedures.
What are the expected impacts/ adverse effects on the animals, for example pain, weight loss, inactivity/ reduced mobility, stress, abnormal behaviour, and the duration of those effects?
What species and numbers of animals are expected to be used? What are the expected severities and the numbers of animals in each severity category (per species)?
What will happen to the animals kept alive at the end of the procedure? (5) (6)
Please provide reasons for the planned fate of the animals after the procedure.

Application of the Three Rs
1. Replacement State which non-animal alternatives are available in this field and why they cannot be used for the purposes of the project.
2. Reduction Explain how the numbers of animals for this project were determined. Describe steps that have been taken to reduce the number of animals to be used, and principles used to design studies. Where applicable, describe practices that will be used throughout the project to minimise the number of animals used consistent with scientific objectives. Those practices may include e.g. pilot studies, computer modelling, sharing of tissue and reuse.

3. Refinement Give examples of the specific measures (e.g., increased monitoring, post-operative care, pain management, training of animals) to be taken, in relation to the procedures, to minimise welfare costs (harms) to the animals. Describe the mechanisms to take up emerging refinement techniques during the lifetime of the project.
Explain the choice of species and the related life stages.

https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32020D0569&from=EN

European Commission

ALURES – ANIMAL USE REPORTING - EU SYSTEM

EU NTS DATABASE ON THE USE OF ANIMALS FOR SCIENTIFIC PURPOSES UNDER DIRECTIVE 2010/63/EU

LIST OF RESULTS

Advanced filtering

Country: Country

Title of the project:

EC NTS/RA identifier:

Keyword:

Species: Species

Purpose(s) of the project: Purpose(s) of the project

Year of publication: Year of publication

Language: Language

EU Submission: EU Submission

Clear | Search

Number of entries per page: 10 **Order by column:** (none)

Country	Language	Title	EC NTS/RA identifier	Publication date	Version	EU Submission	Actions
France	French	Identification des cellules responsables de l'élimination des plaquettes lors d'un état réfractaire aux transfusions plaquettaires	NTS-FR-270672	15-03-2023	1	Yes	View
Belgium	Dutch	In-vivo Evaluatie van een linker ventriculair Mechanisch Circulatie ondersteuningssysteem in een schapen model	NTS-BE-658281	15-03-2023	1	Yes	View
Spain	Spanish	Diferencias sexuales en los efectos del desarrollo tumoral y el estrés social sobre la conducta tipo depresiva en ratones.	NTS-ES-447876	15-03-2023	1	Yes	View
Belgium	Dutch	Comedication in an adenoviral animal model for Graves' disease	NTS-BE-138466	15-03-2023	1	Yes	View
France	French	Implications de la protéine ALCAM dans les accidents vasculaires cérébraux ischémiques	NTS-FR-377829	15-03-2023	1	Yes	View

https://webgate.ec.europa.eu/envdataportal/web/resources/alures/submission/nts/list

Guidance document

Non-technical Project Summaries under Directive 2010/63/EU on the protection of animals used for scientific purposes

A working document on Non-technical Project Summaries

- Replacing consensus document of 23-24 January 2013 -

Brussels, 25-26 November 2021

https://ec.europa.eu/environment/chemicals/lab_animals/pdf/posters/Non-Technical_Project_Summaries_EN_556755452.pdf

Objective and document structure

- *To assist **scientists** on drafting quality NTS*

- *Expanded format from 2013 endorsed working document:*
  - *Introduction*
  - *Legal background*
  - *Benefits of NTSs*
  - *General guidance*
  - *Specific guidance (template headings)*
  - ***Illustrative examples (good + poor quality)***

The document in more detail

Specific guidance

Identification of common errors or misunderstandings

Duration of project (in months)

Please enter the anticipated duration of the research project. This should be a whole number: 1-60 months.

Typically, this number will be the duration requested in the project application. Note that competent authorities for project evaluation/project authorisation may amend the requested duration prior to project approval. In all instances however, this number should equate to the total duration of the authorised project. In example 4 in Appendix II, '*90 months*' has been entered for the project duration. However, as per Article 40 (3), project authorisations shall be granted for a period not exceeding 5 years. Therefore, the maximum project duration permissible is 60 months.

The document in more detail

Specific guidance

Guidance + examples

Objectives and predicted benefits of the project

Objectives of the project: Describe the objectives of the project (for example, addressing certain scientific unknowns, or scientific or clinical needs)

Maximum length 2500 characters

This section of the NTS is usually the first to be read by the interested public. Therefore, the overall goal of the project should be described in popular scientific (i.e. non-technical) language. Appropriate background information should be provided in order to give context to the research goal.

For example:

'Duchenne muscular dystrophy (DMD) is a neuromuscular disease of humans that is characterised by severe muscle weakness, including the muscles of breathing. The main muscle of breathing, the diaphragm, is weakened in DMD with consequences for breathing and other functions of the respiratory system including the ability to generate pressures in the chest that allow for effective cough and sneeze, which are important to clear the airways and help to protect against infections.'

Next, the specific research questions that are being addressed should be described, explaining their relevance and why they are of interest.

The document in more detail

Specific guidance

Guidance + examples
How to avoid common errors

Estimated numbers per severity

When completing this section of the NTS for each species used, a value must be entered for each severity classification (otherwise submission of the NTS will be rejected by the EC database). So for example, if 100 mice are to be used in a project comprising 4 groups of 25 mice; one vehicle control and 3 different doses of a test drug, the estimated numbers per severity could be:

- 50 mild (control and low dose groups), and
- 50 moderate (two highest dose groups).

Please note that values of 0 must be entered for each of the two remaining severity categories (non-recovery and severe), otherwise the NTS will fail validation upon submission.

The document in more detail

Specific guidance

Reasons for the planned fate of the animals after the procedure

Please provide reasons for the planned fate of the animals after the procedure.

Max characters 2500

In this section, the reasons underpinning the planned fate of **all** animals (i.e. not only those that are reused/returned to habitat/husbandry/rehomed) intended to be used on the project should be described.

If it is planned to reuse animals, return them to habitat/husbandry system, or rehome them, please provide justification as to why this is the most appropriate option for these animals. For example, reuse of animals, provided their health and welfare is not compromised, may contribute to an overall reduction in the numbers of animals required to be used for scientific/educational purposes.

If animals are planned to be killed during the project, or upon conclusion of the project, then please explain briefly why this is necessary. Explain why reuse/returning to habitat/husbandry/rehoming is not possible. For example, reuse may not be possible as the effects of previous procedures that animals have undergone could potentially introduce unwanted variability/confound the results of any subsequent studies. In cases where animals are being killed in order to harvest their tissues and organs for histology or other analysis, a brief explanation of what this analysis is, and why it is required to achieve the objectives of the study should be provided.

Most procedures end with the animals being killed and it is important to explain to the general public why this is necessary

Examples

- *10 illustrative examples (5 good and 5 bad) in most common areas of research and type of projects*
    - *1. Basic research*
    - *2. Translational and applied research*
    - *3. Regulatory testing*
    - *4. Education and training*
    - *5. Genetically altered animals*

Examples

- *Projects on several species*
  Mice, fish, chicken, ferrets, rabbit, pigs or cattle
- *Single and multiple step procedures*
- *Animals kept alive or killed and the reasons why*
- *Examples include projects of each type of severity*

Example

5. Genetically altered animals

Example 9 (good quality)

Title of the project	Creation, breeding and maintenance of genetically altered mice for cancer research studies
Duration of project (in months)	60 months
Keywords	service provision; cryopreservation; embryo transfer;
Purpose of project (multiple choices possible)	Basic Research: Oncology Translational and Applied Research: Human cancer Maintenance of colonies of established genetically altered animals, not used in other procedures

Example

Objectives and predicted benefits of the project	
Describe the objectives of the project (for example, addressing certain scientific unknowns, or scientific or clinical needs).	The main objective of this project is to offer a high-quality, efficient service to scientists working on tumour therapy projects. This project will enable the creation and breeding of Genetically Altered (GA) mice which will be used on other projects The project will also facilitate breeding and cryopreservation programmes to ensure efficiency and minimisation of animal surplus. If a researcher needs to create a new GA line, to answer a research question this can be carried out under this project. On each occasion, the researcher will hold a relevant project authorisation for the research they intend to perform, the approval of which will have included an evaluation of the justification of the subsequent use of GA lines generated under this project authorisation. The methodology will be selected to have the greatest likelihood of success.
What are the potential benefits likely to derive from this project? Explain how science could be advanced, or humans, animals or environment may ultimately benefit from the project. Where applicable, differentiate between short-term benefits (within the duration of the project) and long-term benefits (which may accrue after the project is finished).	Many researchers that seek to use particular GA lines in their research do not have the technical skills required to create new lines of GA mice, nor to cryopreserve lines as necessary. This group has huge experience in GA methods and has a high likelihood of successfully developing suitable new lines using the fewest number of animals in the most refined manner. This group also aims to offer a cryopreservation (freezing sperm or embryos) service to researchers within the establishment. This service will bring benefits in relation to reducing the number of GA mouse lines required to be maintained on an ongoing basis by "ticking over" breeding, because they are not required for current studies. This strategy minimises the numbers of surplus animals. Cryopreservation also provides an insurance against potential loss of valuable lines should unexpected health issues arise in the animal units. The main benefit of this project is that it will ensure the efficient and streamlined creation of new GA lines, and an overall reduction in the number of GA animals held within the establishment for use in research studies.

Example

Predicted harms	
In what procedures will the animals typically be used	Mice undergo a single step procedure and will be injected, then killed and used as egg donors to generate novel GA mouse lines.
(for example, injections, surgical procedures)? Indicate the number and duration of these procedures.	Other females will be used as recipients (which involves anaesthesia and followed by surgery to implant embryos which have been modified). This may cause surgical pain over few days but it will be controlled by analgesia. These dams will be kept until they have weaned the pups (approximately 6 weeks). Some male mice will be anaesthetised and surgically vasectomised to support the embryo transfer programme. This may cause surgical pain over a day or so but it will be controlled by analgesia. Recovery from this takes only days. They are then used to mate with recipient mice to generate pseudo (false) pregnant animals which is not a procedure *per se*. GA offspring will be bred and maintained under this project for a short period (months) until the line is established and has had an initial welfare assessment, at which time, the mice are transferred to specific experimental project authorisations for use in subsequent procedures as part of cancer research studies. Animals that are genotyped using a tiny piece of ear tissue taken during identification marking are expected to experience such short term pain that analgesia is not thought to be appropriate.
What are the expected impacts/adverse effects on the animals, for example pain, weight loss, inactivity/reduced mobility, stress, abnormal behaviour, and the duration of those effects?	The animals used as embryo recipients and for vasectomy will have some short term discomfort following surgery, managed by pain-killers. We do not expect adverse effects from our gene manipulations in the offspring or the breeding animals, but all animals are monitored carefully.

Example

What species and numbers of animals are expected to be used? What are the expected severities and the numbers of animals in each severity category (per species)?	**Species[4]**	**Estimated total numbers**	**Estimated numbers per severity**			
			Non-recovery	**Mild**	**Moderate**	**Severe**
	Mice	9 500	0	9 000	500	0

What will happen to the animals kept alive at the end of the procedure?[5, 6]	**Estimated number to be reused**	**Estimated number to be returned to habitat/husbandry system**	**Estimated number to be rehomed**
Please provide reasons for the planned fate of the animals after the procedure.	Animals will either be used in the breeding programmes within this project, and killed once they become too old for efficient breeding, or they will be transferred to other projects for further breeding or continued use in tumour-related experiments.		

Example

	continued use in tumour related experiments.
Application of the Three Rs	
1. Replacement State which non-animal alternatives are available in this field and why they cannot be used for the purposes of the project.	All research groups to be supplied with the GA lines generated under this project authorisation use alternative methods as a replacement to the use of animals where possible. However, non-animal methods carried out in the lab with cells/computer simulations cannot adequately model the complete array of molecular, cellular, physiological and pathological actions and interactions necessary to fully understand how genetic alterations result in normal or abnormal processes. The main purpose of this project is to facilitate biomedical research under other project authorisations. As previously discussed, the scientific use of these GA mice for biomedical research will be authorised under the recipient researcher's project authorisation. Descriptions of alternative methods considered by recipient researchers will be reconsidered to ensure no new methods are available.

Example

2. Reduction Explain how the numbers of animals for this project were determined. Describe steps that have been taken to reduce the number of animals to be used, and principles used to design studies. Where applicable, describe practices that will be used throughout the project to minimise the number of animals used consistent with scientific objectives. Those practices may include e.g. pilot studies, computer modelling, sharing of tissue and reuse.	Central creation, breeding and maintenance is the most efficient and effective way to develop new lines for use in scientific procedures. Unnecessary creation/ production of genetically altered mice will be avoided by extensive searching of publications and databases to make sure they are not already in existence. Cryopreservation (freezing) of gametes and embryos to archive (store) lines will minimise surplus. For cryopreservation, typically small numbers of animals (up to 10 females) are required to produce up to 200 embryos, which will make sure each transgenic line is safely stored (can be brought back to life). These 200 embryos can be held in storage meaning that we do not need to continually breed live animals. When the genetic line is needed, re-implantation only requires a small number of mice, typically 2 females. Animals will only be created and/or bred if a user requirement has been established, and the breeding programme will be subject to regular review to optimally meet anticipated demand. When new mouse lines are established as per a request from a user, these will be made available for use on other scientific projects where these is appropriate justification and authority to do so.
3. Refinement Give examples of the specific measures (e.g., increased monitoring, post-operative care, pain management, training of animals) to be taken, in relation to the procedures, to minimise welfare costs (harms) to the animals. Describe the mechanisms to take up emerging refinement techniques during the lifetime of the project.	Aseptic surgical techniques will be strictly followed and if possible non-surgical alternatives for the transfer of embryos and generation of sterile males will be investigated and used. Animals which have immune systems which are weaker than normal will be kept in special cages, given sterile food and water and handled in a special way to protect them from infection. Analgesics are routinely used post-operatively. Tissue sampling is usually combined with identification, but some scientists require electronic chip readers for ID, in which case we use ear biopsy as the genotyping procedure of choice. New technical developments will be sought through internal and external meetings and literature review, and will apply relevant refinements as these become available.
Explain the choice of species and the related life stages.	Mice are the major species used for the generation of transgenic lines so will be used for the purpose of all procedures in this project including embryo transfer and cryopreservation. Life stages will include embryos, young females which have not yet bred, and

Example

This is considered a good quality NTS because:

- **The title is specific and explains the purpose of the project – creation of genetically altered animals is specifically included in the title**
- **Keywords are informative and include reference to the purpose of the GAA project ("provision of service") i.e. the production of GAA for multiple users**
- **Acronym is defined**
- **Predicted harms:**
  - **Harms have been included for each of the procedures. Durations of harms are included**
- **The severity listed is appropriate as surgical procedures under anaesthesia are proposed. As the genetic alteration is not expected to cause significant harms, the classification of the majority seems to be appropriate.**
- **Reasons for the expected fate of the animals have been included.**
- **There is information on the implementation of the 3Rs:**
  - **Alternatives are discussed.**
  - **The numbers of animals to be used are clearly laid out and explained where possible.**
  - **Refinements are described.**
  - **An undertaking is made to keep up with developments over the lifetime of the project.**

Example

This is considered a poor quality NTS because:

- **Keywords are not considered very informative**
- **"Basic Research - Other basic research" has been selected as the project purpose. "Basic research – Oncology" would be more appropriate in this case**
- **Predicted harms:**
  - **The only procedure described is the breeding of mutant fish, however, under adverse effects it is stated that anaesthesia (MS222) will be used. Therefore, it would appear not all procedures/interventions have been captured**
  - **No adverse effects have been described – information on anaesthesia is not appropriate in this section**
- **120 fish have been classified as non-recovery. This does not agree with the other information provided**
- **It is stated that 120 fish will be returned to husbandry. This does not agree with the other information provided. The figures are also inconsistent with the field below where it is stated that the majority of fish will be euthanised.**
- **There is limited information on the implementation of the 3Rs:**
  - **There is no information on available alternatives that were considered, and the use of animals has not been adequately justified**
  - **Detailed information on statistical tests ("*two sample independent t test*") should not be included as it is not helpful to lay people**
  - **It is stated that "statistical analysis *will* be carried out" – no evidence of *a priori* sample size calculations**
  - **No information has been provided on "practices that will be used throughout the project to minimise the number of animals used"**
  - **No information is provided on "mechanisms to take up emerging refinement techniques during the lifetime of the project"**
- **No information is provided to justify the species and the life stage of the animals to be used**

RAR Guidance Document

- *The objective is to develop a guidance document on the update of the NTS to include the Retrospective Assessment Results (RAR).*
- *Target audience: **Competent Authorities** tasked with the RA using the **new template** defined in Annex I Part B of the Decision 2020/569/EU.*
  - ➢ The focus is **not** on the process of RA
  - ➢ Meaningful information on the findings of RA

Content of the draft guidance

- *Introduction*
- *Legal background*
- *Content and publication of the RAR to update the NTS*
- *Benefits of publishing the RAR*
- *Guidance for the drafting of the RAR*
- *Guidance on the content to be included in the template*
- *Appendices*

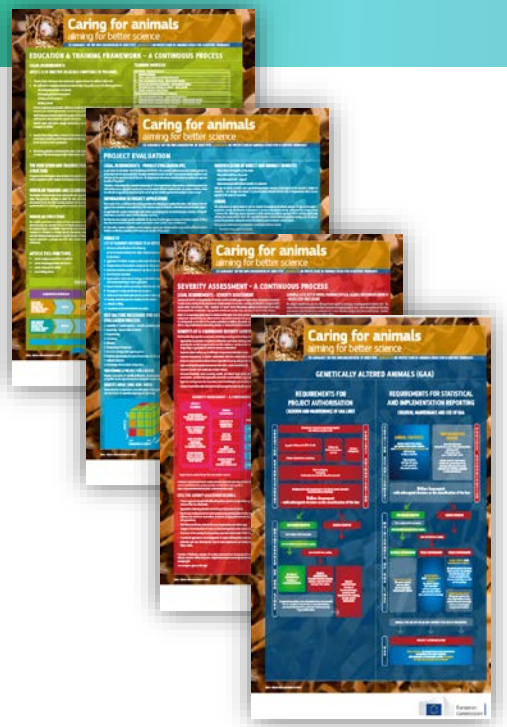
Appendices

- *Appendix I – Template of RAR for NTS update*
- *Appendix II – Illustrative examples of RAR (good and bad quality)*
    1. ***Projects using non-human primates***
    2. ***Severe procedure***
    3. ***Other reasons***
- *Appendix III – List of MS where NTS update with RAR is mandatory (status of 2022)*

Expected endorsement in 2023

patri.vergara@uab.cat

Thank you for your attention!



More information at:

https://ec.europa.eu/animals-in-science

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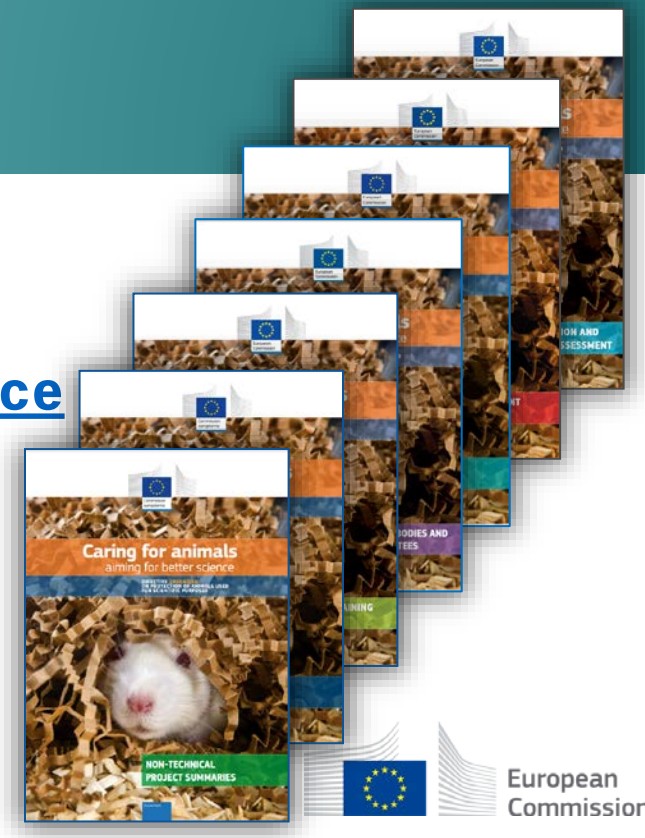

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