# Application for Project Authorisation concerning

# Regulatory use and Routine production

# *In accordance with Art. 19 of the Royal Decree of 29 May 2013 - Royal Decree on the protection of experimental animals, the application for project authorisation must be submitted using a form available on the Brussels Environment website / platform.*

# *All relevant documents, including project authorisations and the result of the project evaluation, shall be kept for at least three years from the date of expiry of the project authorisation and shall be kept at the disposal of Brussels Environment.*

|  |  |
| --- | --- |
|  | File No. :  Received on: :  Approved on:  Refused on:  Withdrawal of project authorisation:  Appeal against the decision: |

# This form should be sent to the ethical committee to which you are affiliated. If the form is considered complete by the Ethical Committee, they will send the declaration, as well as any subsequent changes made by the user, to Brussels Environment at the following address:

# Brussels Environment

# Inspectorate and Polluted Soils Division

# Animal Welfare Department

# Avenue du Port 86c/3000

# 1000 Brussels

# or by e-mail at the following address: [*labo.bea.dwz@environnement.brussels*](mailto:labo.bea.dwz@environnement.brussels)

|  |  |
| --- | --- |
| **Evaluation of the Harms and Benefits (HBA)**  **(see also the Guideline for Harm-Benefit Analysis)** | |
|  | |
| *The HBA that the ethical committee will conduct is based on several factors that should be taken into account when assessing benefits and harms of animal-based experiments. For the sake of transparency, this form indicates the various criteria (by colour-coding) that will be assessed by the Ethical Committee when conducting their HBA.* | |
|  | |
| **The following colour code was used:** | |
| * Primary benefits |  |
| * Likelihood of achieving the benefits |  |
| * Main harms |  |
| * Modulating factors for harm |  |

*Subject to safeguarding intellectual property and confidential Information (which includes company and manufacturing data), the evaluation shall be performed in an impartial manner and may integrate the opinion of independent parties.*

# Project proposal

# *In accordance with Art. 19 of the Royal Decree of 29 May 2013 - Royal Decree on the protection of experimental animals, for each project subject to authorisation, the person responsible for the project must submit an application which specifies the project proposal (with the information on the elements set out in Annex 6) and a non-technical summary of the project.*

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| TO BE COMPLETED BY THE PERSON RESPONSIBLE FOR THE PROJECT | | | | | | | HBA | |
|  | | | | | | |  | |
| **General project data** | | | | | | |  | |
| **Project** | | | | | | |  | |
| Scientific title of the project | |  | | | | |  | |
| Non-technical title of the project (as stated in the NTS) | |  | | | | |  | |
| Intended start date of the project | | (dd/mm/yyyy) | | | | |  | |
| Intended end date of the project | | (dd/mm/yyyy) | | | | |  | |
| Type of project | |  | New Project | | | |  | |
|  | Amendent (in this case check the Guidelines for project amendment). | | | |  | |
| Amendment (if applicable) | | | | | | |  | |
| Type of amendment | |  | Prolongation of the project (new end date): | | | |  | |
|  | Change in the person responsible for the project / lab director or change in partner establishment | | | |  | |
|  | Procedural change (with no change in the severity or number of animals, unless an exception applies) | | | |  | |
| *Please add the changes directly in the ECD application with track changes or in a different colour. When the amendment is approved, please approve all track changes in the document.* | | | | |  | |
| Motivation of the amendment (incl. exception if it applies) | |  | | | | |  | |
|  | | | | | | |  | |
| **Funding** | | | | | | |  | |
| Code / Reference of the project  *(code assigned by another internal or external body, e.g. financing agency)* | |  | | | | |  | |
| Has the project been peer reviewed by an external funding committee?  If yes, by which funding committee? | | Yes | | No | | |  | |
|  | | | | |  | |
|  | | | | | | |  | |
| Pilot study | | | | | | |  | |
| Have you performed a pilot study for this project?  If yes, indicate the authorisation number here: | | Yes | | No | | |  | |
|  | | | | |  | |
|  | | | | | | |  | |
| User[[1]](#footnote-1) | | | | | | | |  |
|  | | | | | | | |  |
| Identification of the research institution where the research will be submitted and performed | | | | | | | |  |
|  | | | | | | | |  |
| Establishment / Company | | | | | | | |  |
| Name of the establishment / company | |  | | | | | |  |
| Name of institute/department and/or lab (if applicable): | |  | | | | | |  |
| Agreement number: | | LA | | | | | |  |
| Address and contact details of the institute / department / lab | | Street |  | | | | |  |
|  | | N° |  | | Box |  | |  |
| Postal code |  | | City |  | |  |
| Tel | |  | | | | | |  |
| Email | |  | | | | | |  |
| Name of the LA director | |  | | | | | |  |
| Name of the head of the research group or course organiser (if different from the LA director) | |  | | | | | |  |
| Name of the person responsible for the experiment(s) (project manager) | |  | | | | | |  |
|  | | | | | | | |  |
| Safety of researchers and / or the environment | | | | | | | |  |
|  | | | | | | | |  |
| Can the project cause health risks for researchers and/or the environment? If yes, please check the relevant box(es):  The type of risk: | | | | | | | |  |
|  | Biosecurity: genetically modified organisms (GMO) | | | | | | |  |
|  | Biosecurity: biosafety hazard level (L2, L3, …) | | | | | | |  |
|  | Chemical products (toxic, ...) | | | | | | |  |
|  | Physical products (radioactivity, …) | | | | | | |  |
|  | Other, please specify: | | | | | | |  |
| If you checked one of the boxes, please contact your biosecurity officer/coordinator to know which procedures and relevant authorisations are necessary for your project. | | | | | | | |  |
|  | | | | | | | |  |

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Project | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| General description, purpose and justification of the project | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Describe in max. 1000 words the general scientific aspects, the relevance of the project and the final objectives, without the description of the individual experiments, which must be mentioned in chapter 6.  *The project must be described in such a way that it can be understood by all members of the Ethical Commission. In this context, it is essential to specify the following aspects:* | | | | | | | | | | | | | |  |
| 1. Background and state of the art: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| 1. Goals that are specific to the project: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| 1. Scientific, social, socio-economic, educational, environmental, veterinary and/or medical relevance (including who will benefit from this research and when): | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| 1. Safety and efficacy benefits: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| 1. Bibliographical references that contribute to the justification of the proposed research and the references of legal guidelines to support the necessity of the work described and / or benefits and relevant references for specific models that are proposed in your work programme: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Purpose of the project | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Select the goal(s) of the research project concerning Regulatory use and Routine production and complete further. Multiple choices are possible. | | | | | | | | | | | | | |  |
|  | Quality control (including batch safety and potency testing) | | | | | Select an item. | | | | Select an item. | | Select an item. | |  |
|  | Other efficacy and tolerance testing | | | | |  | | | |  | |  | |  |
|  | Toxicity and other safety testing including pharmacology | | | | | Select an item. | | | | Select an item. | | Select an item. | |  |
|  | Routine production by product type | | | | | Select an item. | | | | Select an item. | | Select an item. | |  |
|  | | | | | | | | | | | | | |  |
| Select the Type of legislation | | | | | | | | | | | | | |  |
|  | Legislation on medicinal products for human use | | | | | | | | | | | | |  |
|  | Legislation on medicinal products for veterinary use and their residues | | | | | | | | | | | | |  |
|  | Medical devices legislation | | | | | | | | | | | | |  |
|  | Industrial chemicals legislation | | | | | | | | | | | | |  |
|  | Plant protection product legislation | | | | | | | | | | | | |  |
|  | Biocides legislation | | | | | | | | | | | | |  |
|  | Food legislation including food contact material | | | | | | | | | | | | |  |
|  | Feed legislation including legislation for the safety of target animals, workers and environment | | | | | | | | | | | | |  |
|  | Cosmetics legislation | | | | | | | | | | | | |  |
|  | Other legislation | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Select the Origin of legislation | | | | | | | | | | | | | |  |
|  | Legislation satisfying Union requirements | | | | | | | | | | | | |  |
|  | Legislation satisfying national requirements only (within Union) | | | | | | | | | | | | |  |
|  | Legislation satisfying Non-Union requirements only | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| If this is a test required by legislation, it is essential to specify its legal basis or its regulatory guidelines: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Species and number of animals | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Animal species | | Estimated number | | | Strain | | Genetic status | | | | Criteria of inclusion(age and/or weight, life stages, …) | | Gender |  |
| A[[2]](#footnote-2) | B[[3]](#footnote-3) | C[[4]](#footnote-4) | |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
|  | |  | | |  | |  |  |  | |  | |  |  |
| **Total number of animals used** | |  | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Why did you choose this species? Show the relevance of the selected animal species. The requirement to use animal species that experience less pain, suffer less or suffer less damage with the same reliability must be met. | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| If you plan to use cats, dogs, non-human primates, explain why no other species is suitable or practically available: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| If only one gender of animals was selected, please justify your choice: | | | | | | | | | | | | | |  |
|  | The project concerns a sex-specific disease or biological process | | | | | | | | | | | | |  |
|  | For standardisation and reproducibility reasons, it is not warranted to include animals of different sex | | | | | | | | | | | | |  |
|  | The research is in a relatively early phase, at which it is not yet relevant/needed to include sex as a biological variable | | | | | | | | | | | | |  |
|  | The behaviour of animals of a particular sex is not compatible with the experimental procedures and/or housing conditions | | | | | | | | | | | | |  |
|  | Other, please specify: | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Tissue sampling method for genotyping: is the same method used for identification purposes? | | | | | | | | | | | | | |  |
|  | Yes | | | | | | | | | | | | |  |
|  | No, please justify: | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| **Origin of animals (copy and paste the table below if you use more than one supplier):** | | | | | | | | | | | | | |  |
| Name of the supplier | | | |  | | | | | | | | | |  |
| Country | | | |  | | | | | | | | | |  |
| Agreement number | | | |  | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Are exemptions necessary for this project? | | | | | | | | | | | | | |  |
| *An exemption is needed when it comes to:*   1. *Using animals that were not specifically bred for use in experiments and yet belong to an animal species listed in Annex 1 of the Royal Decree of 29 May 2013* 2. *Using protected animals* 3. *Using non-human primates* 4. *Using protected non-human primates* 5. *Using animals caught in the wild* 6. *Using stray animals or wild animals* 7. *Carrying out procedures outside a user establishment* 8. *Performing procedures with long lasting harm* 9. *Killing animals with methods outside the scope of the Directive* 10. *Providing animals with accommodation, an environment, food, water and care which is not appropriate to their health and well-being or care and accommodation standards set out in Annex III of the Directive* | | | | | | | | | | | | | |  |
|  | | | No | | | | | | | | | | |  |
|  | | | Yes. In this case, has an exemption been requested or already been obtained? If so, please add the authorisation to the project. If not yet available, please include it at a later stage. | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| 3 Rs: REPLACEMENT, REDUCTION, REFINEMENT | | | | | | | | | | | | | |  |
| The principle of the 3 Rs must always be respected. | | | | | | | | | | | | | |  |
| Sourced consulted | | | | | | | | | | | | | |  |
| Which sources did you consult regarding the application of the 3Rs principles? | | | | | | | | | | | | | |  |
|  | Re-Place | | | | | | | <http://www.re-place.be> | | | | | |  |
|  | Norecopa 3R guide | | | | | | | [https://norecopa.no/search?syn=11&sort=name\_s%20asc&q=\*&sf=name&fq=db:%223r%22](https://norecopa.no/search?syn=11&sort=name_s%20asc&q=*&sf=name&fq=db:%223r%22) | | | | | |  |
|  | Netherlands Centre Alternatives to animal use | | | | | | | <https://www.uu.nl/en/research/utrecht-advanced-in-vitro-models-hub-u-aim> | | | | | |  |
|  | Databases and/or editions of ECVAM or FRAME | | | | | | | <https://ec.europa.eu/jrc/en/eurl/ecvam> | | | | | |  |
|  | DB-ALM | | | | | | | https://ec.europa.eu/jrc/en/scientific-tool/database-alternative-methods-animal-experimentation | | | | | |  |
|  | AOP knowledge base | | | | | | | <https://aopkb.oecd.org/search.ashx> | | | | | |  |
|  | Syrf (fully integrated online platform for performing systematic reviews of preclinical studies) | | | | | | | <http://syrf.org.uk/> | | | | | |  |
|  | Pubmed | | | | | | | <https://pubmed.ncbi.nlm.nih.gov/> | | | | | |  |
|  | ISI Web of Science | | | | | | | <http://webofknowledge.com/WOS> | | | | | |  |
|  | Embase | | | | | | | <https://www.embase.com/login> | | | | | |  |
|  | SIS | | | | | | | <http://ihcp.jrc.ec.europa.eu/> | | | | | |  |
|  | OECD | | | | | | | <http://www.oecd.org/> | | | | | |  |
|  | Other, please specify: | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Keywords used for the search: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Application of the 3 Rs[[5]](#footnote-5) | | | | | | | | | | | | | |  |
| Which alternative and/or complementary methods have you considered and why are they not suitable to achieve the objectives of your project without using animals? | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Indicate, if applicable, how you integrate in silico, in vitro and ex vivo work within your in vivo work and the relationship between each part of the project: | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Are special efforts being made to reduce the number of animals used (e.g. collaboration with other researchers, shared use of animals, allowing different laboratories to use the organs of the same animal)? | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
| Are you aware of any identical experiments that were performed in the past? If yes, please explain why these are not a mere duplication of experiments. | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |
|  | | | | | | | | | | | | | |  |

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| SYNTHESIS OF EXPERIMENTS AND PROCEDURES | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| *Definitions:*   * *Experiment: carefully designed sequence of procedures that can be performed to answer a research question.* * *Procedure: any action that can cause an animal a level of pain, suffering or anxiety that is equal to or greater than the insertion of an injection needle. A procedure can be as mild as an injection or as severe as an organ transplant. The breeding of a genetically modified animal is also considered as a procedure if genetic changes in the normal appearance or 'phenotype' of an animal can cause suffering. Procedures are involved in all experiments, but not all procedures are experiments.* | | | | | | | | | | | | | |
|  | | | | | | | | | | | |  | |
| Describe the different experiments that are done to carry out the project: | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| Give a general overview of the different experiments. Adding a timeline or diagram can help to clarify the overview. The details of the procedures per experiment are set out in the next section. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| Detailed description of the experiments | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| Answer the questions in this section for each experiment separately. If you plan on performing more than one experiment, copy and paste this section as many times as needed. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| **Description of experiment no. 1** | | | | | | | | | | | |  | |
| 1. Title of the experiment | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Experimental design and statistical model | | | | | | | | | | | |  | |
| * 1. Describe in detail all actions / procedures performed (e.g. volume and frequency of sampling, etc.). To understand the chronology of the operations, an illustrative timeline is strongly recommended | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| * 1. Number of experimental groups and animals per group | | | | | | | | | | | |  | |
| Clearly indicate the number of animals per group, the number of repetitions of the experiment and the total number of animals. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| * 1. Justification for the number of animals. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Welfare monitoring and refinement | | | | | | | | | | | |  | |
| * 1. Indicate how the monitoring of animal welfare during the experiment will be guaranteed, in particular the frequency of the observations and the monitoring of the inconvenience (and its possible variations through the experiments). | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| * 1. Are animals single housed[[6]](#footnote-6) from the start or during the course of the procedure? | | | | | | | | | | | |  |  |
|  | | | Yes | | | | | | No | | |  | |
| If yes, justify the scientific, animal-welfare or animal-health reasons and the duration of this deviation and specify what measures are taken to limit the discomfort (e.g. enrichment): | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| * 1. Are there other deviations from the standards (e.g., housing, specific diet, fasting…) described in Annex 4 of the Royal Decree of 29 May 2013? If yes, justify the scientific, animal-welfare or animal-health reasons and the duration of this deviation. Explain also the possible negative consequences for the animals and specify what measures are taken to limit those negative effects: | | | | | | | | | | | |  |  |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| * 1. Explain the expected adverse effect of each procedure that is applied. State how you intend to control those effects (e.g. analgesics, anaesthesia, conditioning / training, enrichment, etc.) to minimise the severity. Detail the analgesia protocol, or any other means used to mitigate these adverse effects (anaesthesia, anti-inflammatory drugs, antibiotics…). Provide the list of medication, as well as the dose, route of administration, duration and frequency. Specify which references were consulted to choose the most appropriate method of analgesia / anaesthesia (bibliographic reference or name and position of the person being consulted): | | | | | | | | | | | |  |  |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Re-use of animals | | | | | | | | | | | |  | |
| * 1. Will laboratory animals that have already been used in another animal experiment be used for this experiment? | | | | | | | | | | | |  | |
|  | | | Yes | | | | | | No | | |  | |
| If yes, indicate in which previous experiment these animals were used (authorisation number and short description of the procedure, observed severity level);  *Please note that animals that have already been used in previous procedures cannot be re-used for severe procedures. Under exceptional circumstances, and after a veterinarian has examined the laboratory animal, the Ethical Committee may authorise the re-use of a laboratory animal if it has not been used more than once in an animal experiment involving severe pain, distress or equivalent suffering.* | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| The re-use of animals implies the favourable opinion of the designated veterinarian (or qualified expert) (taking into account the full life cycle of the test animal) and the determination by the designated veterinarian that the general health and welfare status of the animal being re-used has been fully restored. Certificates and/or authorisation documents of the expert in the context of re-use must be kept by the person responsible for the project, and integrated in the retrospective analysis. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Severity classification | | | | | | | | | | | |  | |
| 1. Specify how the severity classification was estimated (motivate). Take into account the duration and repetitions of painful events for the animals in your evaluation, as well as the eventual re-use of the animals. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| *It is recommended to consult the “Severity assessment” document on the European Commission's website: (*[*http://ec.europa.eu/environment/chemicals/lab\_animals/interpretation\_en.htm*](http://ec.europa.eu/environment/chemicals/lab_animals/interpretation_en.htm)*)* | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Expected severity | | | | | | | | | | | |  | |
|  | | **Estimated number per severity** | | | | | | | | | |  | |
| **Species** | | **Non-recovery** | | | **Mild** | | | **Moderate** | | | **Severe** |  | |
|  | |  | | |  | | |  | | |  |  | |
|  | |  | | |  | | |  | | |  |  | |
|  | |  | | |  | | |  | | |  |  | |
|  | | | | | | | | | | | |  | |
| 1. Exemption | | | | | | | | | | | |  | |
| No procedures leading to a severe degree of pain, suffering or anxiety that is likely to be long-lasting and that cannot be reduced is permitted. If the objective of the experiment considers it necessary that a severe degree of pain, suffering or anxiety cannot be reduced or eliminated, then you must describe the scientifically motivated reasons that justify an application for exemption. | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| 1. Humane endpoints | | | | | | | | | | | |  | |
| Indicate the humane endpoints for the animals concerned.  To determine the endpoints, one must rely on the criteria for evaluating animal welfare. It is recommended to use the general score sheet in Annex I for rodents for daily check-ups. In case you do not want to use this score sheet or if you are using another species, justify your choice and provide an alternative score sheet. Other criteria depending on the procedures should be added as specific endpoints, if relevant (see Annex II for examples).  The score on which the humane endpoint decision is based should be adapted to the number of criteria that are evaluated.  **Attach the final proposed score sheet as an Annex to the form (if there are different score sheets for each experiment, please indicate the experiment number above each sheet).** | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| End of the experiments | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| **Fate of the animals kept alive (if applicable)** | | | | | | | | | | | |  | |
| *The final decision to keep an animal alive after use in an animal experiment can only be taken by the expert responsible for the welfare and health status of the animals.* | | | | | | | | | | | |  | |
| Specify the animals concerned and their destination (duplicate the lines of the table if needed): | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  |  | | | Estimated numbers of animals to be re-used, to be returned to habitat / husbandry system or to be rehomed | | | | | |  | |  | |
| Species | Level of discomfort suffered | | | Re-used | | Returned | Rehomed | | | In the case of re-use, return or rehoming of the animal, specify their destination, the procedure followed and, where appropriate, the socialisation programme set up | |  | |
|  |  | | |  | |  |  | | |  | |  | |
|  | | | | | | | | | | | |  | |
| Method(s) of humane killing | | | | | | | | | | | |  | |
| *Only the procedures included in Annex 7 of the Royal Decree of 29 May 2013 are allowed to kill laboratory animals.* | | | | | | | | | | | |  | |
| Specify the animals involved and clearly state the method (s) provided for killing and confirming death: | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| Exemptions / exceptions to Annex 7 of the Royal Decree of 29 May 2013 can only be allowed if the method is considered to be at least as humane; or when the purpose of the procedure cannot be achieved by the use of a method of killing set out in Annex 7. If an exemption must be requested, a detailed scientific justification must be added here: | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| PERSONNEL | | | | | | | | | | | |  | |
|  | | | | | | | | | | | |  | |
| The project manager (person responsible for the experiment(s), see pg. 6) is responsible for the design of the project, its implementation and the proper use of the animals in accordance with the project permit. He/she is the point of contact for the Ethical Commission for all communication concerning the project.  He / she is also responsible for:   * the establishment and updating of the list of people who are involved in the project * making sure that these people have the adequate skills and expertise * keeping a record of the training certificates, as well as the continuous training of the personnel involved in the protocol * ensuring the daily and adequate follow-up of the animals during the experiments * **Provide as Annex III of this form the list of people involved in this project (role, training level, date obtained).** | | | | | | | | | | | |  |  |
|  | | | | | | | | | | | | | |

|  |  |  |
| --- | --- | --- |
| SIGNATURES | |  |
|  | |  |
| The complete questionnaire must be **dated and signed by the head of the research group and the project manager**. Only the original documents are analysed by the committee.  By their signature, the head of the research group and the project manager acknowledge their **full responsibility** and their agreement with the procedures described above.  The signatories confirm when signing this document that the persons involved in the protocol have been adequately trained and that all legal requirements with regard to laboratory animals and biosecurity requirements have been met.  Any amendment to the protocol must be submitted by the signatories to the ethical commission for **prior** evaluation. | |  |
| **Project Manager** | |  |
| Name |  |  |
| Location |  |  |
| Date |  |  |
| Signature (preceded by ‘read and approved’ |  |  |
|  | |  |
| **Head of the research group or LA director** | |  |
| Name |  |  |
| Location |  |  |
| Date |  |  |
| Signature (preceded by ‘read and approved’ |  |  |
|  | |  |

|  |  |  |
| --- | --- | --- |
| ANNEX I : Score sheet to decide on the humane endpoints for Rodents (to be checked daily) | | |
|  | | |
| This score sheet needs to be complemented with relevant parameters depending on the planned experimental procedures and will have to be approved by the Ethical Committee. The total score that should be considered as a humane endpoint depends on the number of parameters. As a general rule of thumb, we propose the following:   * 3 parameters : total score ≥ 5 * 4 parameters : total score ≥ 7 * 5 parameters : total score ≥ 9   HOWEVER: this is still dependent on the type of experiment you perform. Some parameters might weigh more heavily, so it is always advisable to consider your total score carefully and explain your reasoning in your project application. | | |
|  | | |
| Body condition score (BCS)[[7]](#footnote-7) | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | BCS=3 or higher  *Mouse is well conditioned*   * *Vertebrae and dorsal pelvis not prominent; palpable with slight pressure*   *Mouse is over-conditioned*   * *Spine is a continuous column* * *Vertebrae palpable only with firm pressure*   *Mouse is obese*   * *Mouse is smooth and bulky* * *Bone structure disappears under flesh and subcutaneous fat* |  |
| 1 | BCS =2  *Mouse is under-conditioned.*   * *Segmentation of vertebral column evident* * *Dorsal pelvic bones are readily palpable.* | * Put easy, high-energetic food in the cage: diet gel or solid food in petri dish, soaked in water * Weigh mouse every 2 days |
| 2 | BCS=2, lasting for 3 or more days | * Put easy, high-energetic food in the cage: diet gel or solid food in petri dish, soaked in water * Weigh mouse every day * If weight loss was detected and no weight gain within 3 days, contact the veterinarian or the animal should be killed humanely |
| 3 | BCS=1  *Mouse is emaciated.*   * *Skeletal structure extremely prominent; little or no flesh cover* * *Vertebrae distinctly segmented* | * Humane endpoint |
|  |  |  |
| **Body weight changes relative to reference weight**  **(For those who prefer to use weight as an indicator of the health status, the BCS scoring system can be replaced by the following):** | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | Normal |  |
| 1 | > 10%, but ≤ 15% weight loss | * Put easy, high-energetic food in the cage: diet gel or solid food in petri dish, soaked in water * Weigh mouse every 2 days * If weight loss was detected and no weight gain within 3-4 days, contact the veterinarian or the animal should be killed humanely |
| 2 | > 15%, but ≤ 20% weight loss | * Put easy, high-energetic food in the cage: diet gel or solid food in petri dish, soaked in water * Weigh mouse every day * If weight loss was detected and no weight gain within 3 days, contact the veterinarian or the animal should be killed humanely |
| 3 | > 20% weight loss | * Humane endpoint |
|  | | |
| **Physical appearance** | |  |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | Normal | NA |
| 1 | Lack of care, deterioration of physical appearance, superficial wounds | - Follow up daily and contact the veterinarian if no improvement within 3 -4 days  - Apply wound cream without cortisone daily for 1 week. If within 3-4 days no improvement contact the veterinarian. |
| 2 | Rough coat/fur, nasal / ocular excretion, hunched posture | - Put easy, high-energetic food in the cage: diet gel or solid food in petri dish, soaked in water.  - Follow-up daily and contact the veterinarian if no improvement within 2 days or the animal should be killed humanely  - Clean out the eyes and use local antibiotics |
| 3 | Very rough coat/fur, abnormal posture, enlarged pupils, deep wounds, prolapse | Humane endpoint |
|  | | |
| **Animal behaviour** | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | Normal | NA |
| 1 | Minor changes, small reduction in response or excessive response | Follow-up daily and contact the veterinarian if no improvements within 3-4 days |
| 2 | Abnormal reactions, abnormal and reduced mobility, reduced alertness, inactivity | Follow-up daily and contact the veterinarian if no improvements within 2 days |
| 3 | Spontaneous vocalisations, self-mutilation, very restless or immobile, violent reactions or no reaction upon touching | Humane endpoint |
|  | | |
| **Relevant parameter depending on the planned experimental procedures :       (to be completed)** | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 |  |  |
| 1 |  |  |
| 2 |  |  |
| 3 |  |  |
|  | | |
| **TOTAL SCORE** | |  |
| In case of a total score of ≥ 5 or a score of 3 in any variable, killing of the animal will be performed. | | |
|  | | |

|  |  |  |
| --- | --- | --- |
| Annex IIExamples of specific endpoints | | |
|  | | |
| Retro-orbital injection | | |
|  | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | Normal | NA |
| 1 | Minor swelling or protrusion | Check again after 24 h |
| 2 | Minor eye trauma | Consult the veterinarian |
| 3 | Major eye trauma and persistent swelling/protrusion | Humane endpoint |
|  | | |
| Subcutaneous tumour model (1 tumour) | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | No visible tumour present | Daily visual inspection |
| 1 | Tumour visible and < 1000 mm3 | Measure tumour size and weigh the animal every 2-3 days |
| 2 | 1000mm3 < Tumour size < 1500mm3; moderate ulceration (<10% of tumour volume); moderately impacted mobility | Measure tumour size and weigh the animal daily; clean tumour with physiological water and apply isobetadine; provide easily accessible diet gel |
| 3 | Tumour size ≥ 1500 mm3, severe ulceration of the tumour or severely impacted mobility | Humane endpoint |
|  | | |
| Minor surgery (e.g. intranodal injection) | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | Healed skin | NA |
| 1 | Non-infected wound that nicely scars | NA |
| 2 | Minor infection/inflammation or loosening of the sutures | Treat with isobetadine, re-suture the wound or use VetBond |
| 3 | Major infection | Consult veterinarian or the animal should be killed humanely |
|  | | |
| Neurological assessment following stroke (Bederson scale) | | |
|  | **Observation(s)** | **Recommendation(s)** |
| 0 | No observable deficit | NA |
| 1 | Forelimb flexion |  |
| 2 | Decreased resistance to lateral push (and forelimb flexion) without circling |  |
| 3 | Same behaviour as grade 2, with circling | Humane endpoint |
|  | | |

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Annex IIIList of the personnel involved in the project | | | | | | | | |
|  | | | | | | | | |
| **Name** | **First name** | **Education** | | | | **Certificate obtained on (Date)** | **Certificate (Name)** |
| Project manager | Active participant | Animal caretaker (special care) | Animal caretaker  (basic care) |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  | | | | | | | | |

# Project Authorisation

# *In accordance with Art. 22. § 2 of the Royal Decree of 29 May 2013 - Royal Decree on the protection of experimental animals, the form and terms and conditions of the project authorisation are determined by Brussels Environment.*

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **RESERVED FOR THE ETHICAL COMMISSION (EC)** | | | | | | | | |
|  | | | | | | | | |
| **GENERAL INFORMATION** | | | | | | | | |
|  | | | | | | | | |
| User Establishment | | | | LA | | | | |
| Person(s) responsible for the general implementation of the project | | | |  | | | | |
| Person(s) responsible for the compliance with the project authorisation | | | |  | | | | |
| Facilities, if any, where the project will be carried out | | | |  | | | | |
|  | | | | | | | | |
| **PROJECT AUTHORISATION** | | | | | | | | |
|  | | | | | | | | |
| Project | | | | | | | | |
| Project Name | | | |  | | | | |
| Code of the project | | |  | | | Date of EC approval |  | |
| Date of initial application | | |  | | | EC approval applies[[8]](#footnote-8) | From       till | |
| Date of correct application | | |  | | | Submit retrospective analysis at the latest on |  | |
|  | | | | | | | | |
| **Ethical Commission Decision** | | | | | | | | |
|  | Approved | | | | | | | |
|  | Approved under the following specific conditions: | | | | | | | |
|  | Approval of the Ethics Committee to which the external partner institution of the project is affiliated necessary | | | | | | |
|  | Granting of an Exemption by Brussels Environment or Minister necessary | | | | | | |
|  | Granting of an Exemption by the Ethical Commission necessary | | | | | | |
|  | Other: | | | | | | |
|  | Approved temporarily until: | | | | | | | |
|  | Refused | | | | | | | |
|  | | | | | | | | |
| Motivation of the EC: | | | | | | | | |
|  | | | | | | | | |
| **SIGNATURE OF THE ETHICAL COMMISSION** | | | | | | | | |
| (Vice-)President | | | | | | | | |
| Name | | | |  | | | | |
| Function | | | |  | President | | |
|  | Vice-President | | |
| Location | | | |  | | | | |
| Date | | | |  | | | | |
| Signature | | | |  | | | | |
|  | | | | | | | | |

*When using animal test methods to satisfy regulatory requirements, it is essential that measures are in place to ensure that any changes to such requirements are promptly identified and implemented to ensure compliance with obligations on the Three R’s. Project authorisations do not permit continued use of animals, where alternatives have been accepted in or recognised by EU legislation. The project authorisation holder is required to ensure that any new relevant scientifically satisfactory alternative non-animal methods or testing strategies[[9]](#footnote-9) that become available during the validity period of the project authorisation are implemented in compliance with Art. 24. §2 of the Law of 14 August 1986 on the protection and welfare of animals “No animal experiment shall be carried out if another method or testing strategy for obtaining the result sought, not entailing the use of a live animal, is recognised under the legislation of the Union”.*

*When testing is requested for substances produced above 100 tons to satisfy information requirements under Regulation 1907/2006/EC (REACH) [[10]](#footnote-10), the project authorisation holder is required to ensure that an ECHA decision exists on the testing proposal before test on live animals can start.*

*The responsible person must ensure that animal experiments and projects are carried out in accordance with the approval granted. If the responsible person wants to change the project in such a way that it can have a negative impact on animal welfare (new experiments,….), the/she should submit a new project evaluation form to the Ethical Commission (see Guidelines for project amendment).*

*The Ethical Commission or Brussels Environment may withdraw the licence of a project at any time if the project is not carried out in accordance with the approved licence.*

# Authorisation for Minor Amendments

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **RESERVED FOR THE ETHICAL COMMISSION (EC)** | | | | | | | | |
|  | | | | | | | | |
| AMENDMENT AUTHORISATION | | | | | | | | |
|  | | | | | | | | |
| Amendment | | | | | | | | |
| Project Name | | |  | | | | | |
| Code of the Amendment | | |  | | | | | |
| Date of initial submission | |  | | | Date of EC approval |  | | |
| Date of correct submission | |  | | | EC approval applies | From       till | | |
|  | | | | | | | | |
| **Ethical Commission Decision** | | | | | | | | |
|  | Approved | | | | | | | |
|  | Approved under the following conditions: | | | | | | | |
|  | Approved temporarily until: | | | | | | | |
|  | Refused | | | | | | | |
| Motivation of the EC: | | | | | | | | |
|  | | | | | | | | |
| **SIGNATURE OF THE ETHICAL COMMISSION** | | | | | | | |
| (Vice-)President | | | | | | | |
| Name | | |  | | | | |
| Function | | |  | President | | |
|  | Vice-President | | |
| Location | | |  | | | | |
| Date | | |  | | | | |
| Signature | | |  | | | | |
|  | | | | | | | |

# Withdrawal of Project Authorisation

# *In accordance with Art. 23. § 1 of the Royal Decree of 29 May 2013 - Royal Decree on the protection of experimental animals, Brussels Environment has the possibility, with a view to the protection and welfare of experimental animals, of not allowing a project to go ahead (with appropriate justification). Brussels Environment shall inform the user of this decision. The user may appeal against the decision to the Government within thirty days of receipt of the notification.*

|  |  |  |  |
| --- | --- | --- | --- |
| **RESERVED FOR BRUSSELS ENVIRONMENT** | | | |
|  | | | |
| *With regard to the protection and welfare of laboratory animals, Brussels Environment has the possibility to evaluate all projects. In case of a negative evaluation, the project will not be allowed to proceed and the user will be informed.* | | | |
|  | | | |
| **Brussels Environment Decision** | | | |
|  | No Withdrawal of Project authorisation | | |
|  | Withdrawal of Project authorisation | | |
|  | | | |
| Justification of Brussels Environment: | | | |
|  | | | |
| **SIGNATURE OF BRUSSELS ENVIRONMENT** | | | |
|  | | | |
| Name | |  | |
| Function | |  |
| Location | |  | |
| Date | |  | |
| Signature | |  | |
|  | | | |

1. Animal experiments can only be performed at a user's facility. Brussels Environment may grant exemptions based on scientific data to the extent that the user submits an exemption request that contains a scientific justification. [↑](#footnote-ref-1)
2. Not genetically altered [↑](#footnote-ref-2)
3. Genetically altered without harmful phenotype (including animals with spontaneous mutations that are used for the purpose of their mutation) [↑](#footnote-ref-3)
4. Genetically altered with harmful phenotype [↑](#footnote-ref-4)
5. Refinement should be adapted to each experiment and should follow the legislation applicable in the Brussels Capital Region. [↑](#footnote-ref-5)
6. In cases where single housing is foreseen, the duration shall be limited to the minimum period necessary and visual, auditory, olfactory and/or tactile contact shall be maintained. The introduction or re-introduction of animals to established groups shall be carefully monitored to avoid problems of incompatibility and disrupted social relationships. [↑](#footnote-ref-6)
7. As an alternative to the BCS scoring system above, the following categories can also be used. Please note that, in this case, half point scores/incrementals can be used if needed (e.g. BCS 1.5). Score 0: BCS ≥ 3 ; Score 1: BCS ≥ 2, but <3 ; Score 2: BCS > 1, but <2 ; Score 3: BCS = 1 [↑](#footnote-ref-7)
8. The approval of a project by the Ethical Commission is valid for a maximum of 5 years. [↑](#footnote-ref-8)
9. These include methods described, for example, in Test Method Regulation EC/440/2008 and/or related ECHA guidance (<https://echa.europa.eu/support/information-toolkit>) and monographs in European Pharmacopoeia (Ph. Eur.) [↑](#footnote-ref-9)
10. Regulation 1907/2006/EC on the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH), establishing a European Chemicals Agency [↑](#footnote-ref-10)