Document number	AEC 007
Version number	1
Supersedes	0
Implementation date	2016
Review date	2016

Procedure title:

Completion of the Animal Ethic Application Questionnaire

Purpose:

1. To describe the AEC application form

Scope:

1. The SOP is applied to understand the AEC application questionnaire

References:

South African National Standard (SANS 10386-2008):

Terminology / Abbreviations:

1. SAVC – South African Veterinarian Council

Procedure:

The animal ethics questionnaire has been designed to extract the essence of a research project in terms of the Three R's in animal research, for proper evaluation by the committee. This form is based on the specifications in the South African National Standard (SANS 10386-2008): "The Care and Use of Animals for Scientific Purposes", which defines animal as "live, sentient non-human vertebrate, including eggs, foetuses and embryos, that is, fish, amphibians, reptiles, birds and mammals, and encompassing domestic animals, purpose-bred animals, farm animals, wildlife and higher invertebrates such as the advanced members from the Cephalopoda and Decapoda".

Since this entire protocol is not being reviewed (and due to the number of applications can't be reviewed), the questionnaire needs to be completed in its entirety by an applicant so that the committee can have a good working understanding of the research project. While some of the requested information may seem redundant, we also need the keep this information on file from a legal aspect. The committee does realise that some of the question may result in duplication of information being requested. This is however, the only way the committee can gauge the ethical nature of the various different types of projects that are undertaken by staff and students of the UP.

The following sections of the questionnaire are important as:

1. Commencement of the research project

1.1 The AEC cannot grant approval of a research project that has already started or is phase as this is contrary to the process of ethics approval

1.2 All research projects that have already started or have been completed, will be referred to the relevant faculty. Starting a study without ethics approval may result in disciplinary action taken against a staff member or a student registered with the UP.

2. Brief justification

2.1 Every project will need a justification, of 500 words or less

2.2 The justification should briefly give an overview of the problem being investigated.

2.3 Justifications need to be supported by relevant scientific literature.

2.4 The justification needs to tie in with the aims of the study.

2.5 If the studies builds on previous studies or follows on other phases, this needs to be included in the justification.

2.6 If this study disputes previous study findings or needs to repeat a study due to potential external variable being able to influence a study (e.g. climate, breed, food type), this should be properly articulated within the justification.

3. Aim/s of the proposed study

3.1 This section is the important link between the justification and the study hypothesis.

3.2 In completing this section, it is important to ensure that the aims are clear and adequately cover what is trying to be achieved through the study.

4. Potential benefits of the research findings

4.1 For this section the committee would like to see the value prospects of the study.
4.2 One of the underlying principles of animal research, is that the benefit derived from the study must outweigh the potential suffering inflicted on a sentient species.
4.3 The committee needs to be convinced that the research is not completely arbitrary in that it may not have any benefit to people or animals. With this said, the committee does recognise the value of innovative science that may not have immediate impact. For the latter, it is incumbent on the research to convey what they believe will the potential future relevance of a study.

5. Animal/Sample Requirements

5.1 Since the committee keeps records of all animals used by the institution, this section needs to be completed as fully as possible. The committee does understand that not all research types will have the needed information requested.
5.2 Strain: This is applicable to medical models where specific strains of animals are available to study various diseases models.
5.3 Microbial Status: This is an indication of whether the animals will be healthy, diseased or medically defined animals such as Specific Pathogen Free of Gnotobiotic.
5.4 Source of animal: This is an important component in any ethics application, as the source of animals determines if the project is ethical or not e.g. the use of poor quality animals can result in poor results rendering an experiment invalid and this wasteful on animal life. Likewise sourcing of animals that are bought at a market illegally trading in animals may be considered unethical as it has the potential to increase/legitimise illegal wild animal capture/sale. In general animals should first be attained from a reputable breeder, before resorting to open purchase of animals.

6. Justification for the use of sentient animals

6.1 For this section, the committee would like to know that the use of sentient animals have been properly considered. By sentient we imply, that the animal is aware of its environment and as mentioned above are a "live, sentient non-human vertebrate, including eggs, foetuses and embryos, that is, fish, amphibians, reptiles, birds and mammals, and encompassing domestic animals, purpose-bred animals, farm animals, wildlife and higher invertebrates such as the advanced members from the Cephalopoda and Decapoda"

6.2 In this section, the committee would like to know if validated non-sentient alternates have been tried first e.g. the use of cell cultures, bench models or alternative monitoring strategies.

6.3 A core principle in animal ethics is replacement, whereby all attempts needs to be made to replace an animal in research projects when alternative are available.

6.4 In cases where alternates are only available overseas or are not affordable for use at an academic institution or in a developing world, the committee would accept a motivation from the researcher as to why such alternatives are not yet an option.

7. **Reduction of the number of animals** to a minimum to achieve scientific objectives

7.1 In addition to striving to undertake research through the use of alternates to animal research, an important ethical principle is to use the fewest number of animals possible within the project. The committee will look at this aspect of the study very strictly. As such the project's sample size needs to be properly justifiable.

7.1.1 For this section the committee requires a researchers to explain how the intended sample size was attained.

7.1.2 While the factors used in the sample size calculation should be included, it not necessary to include the actual calculations behind the sample size.

7.1.3 While the use of previously published sample sizes are acceptable, it is still incumbent on the researcher to check that the published sample size cannot be further optimised e.g. it is possible that the published study overestimated the number of animals required for the study.

7.1.4 While the committee recognises the importance of a person's prior knowledge in determining the sample size, this needs to be properly articulated e.g. it is not sufficient to say that "Prof xyz has indicated that a sample size of xx is sufficient."

7.2 The committee would specifically look at the following as valid means of gauging adequate application of principles of reduction

7.2.1 The use of a homogenous population, like medically defined animals potentially will result in less variation in the results, thus resulting in smaller sample sizes being needed.

7.2.2 That a particular experiment has not previously undertaken with exactly the same variables, without providing a proper justification for the repeat of the study or why a different response in expected for the repeated study.

7.2.3 For controlled medical, veterinary, physiological, anatomical, behavioural and/or production related research using large sample sizes, it is incumbent on the researcher to justify why a pilot study was not considered as the first step in the research project i.e. pilot studies are very helpful in determining the validity of a study supposition or optimising the research study design to reduce the number of animals inducted into a study (The committee will accept results from previous studies as a surrogate for a pilot study, as long as an indication is provided on the validity of data from the particular study being referred too under local conditions.)

7.2.4 For studies which involve induction of pain (direct or indirect), especially those withholding analgesia, pilot studies are mandatory.

7.2.5 For studies in which no previous knowledge is available to justify the study design, pilot studies will be required, before full approval is granted.

7.3 For studies that require large sample sizes, like zoological/ epidemiological studies:

7.3.1 the use of a phased study approach and/or in-phases statistical analysis could be one manner of reducing the global sample size i.e. While the committee may approve a larger sample size, the research project should still at all times strive to make their conclusions on the smallest possible sample size, as a valid means of reduction.

7.3.2 While the committee accepts that making use of a phase study approach may not always be feasible for all types of studies, this model should still be considered by researchers as a means to reduce overall sample sizes.

7.3.3 the committee would like to see an explanation as to how the researcher will keep the environmental impact of the study to a minimum.

8. **Animal housing and care:**

The committee will look at the housing offered to the animals in terms of current ethical standards.

8.1 For this section, the size of cages, pens, etc need to be described and must follow the SANS code when available (For rodents the type of cage is also important to declare e.g. conventional open topped, individually ventilated, isolation cages). When the SANS code does not cater for a particular species, the reasons for the selection of a particular cage and housing criteria needs justification.

8.2 The temperature lighting conditions, humidity and air changes need to be stated (Please note that there are minimum standards applicable to laboratory animal species to which the committee will default e.g. 12:12 light dark cycle, 15 to 20 air changes, 40 to 70% relative humidity and a temperature of 22±2℃ is required for rats and mice under conventional conditions).

8.3 If the facility is under positive pressure, negative pressure HEPA conditions, this should be stated.

8.4 It is especially important to mention how the normal social structure of the species in question was taken into consideration e.g. animals needing to be pair housed or housed in groups.

8.5 If single housing of animals is necessary, the application needs to state how other sensory stimuli are catered for e.g. can the animals see and smell each other.

8.6 A brief description of the diet and bedding is required. For special diets, mention needs to be made as to how the new diet differs from the normal diet, and the expected changes that can result from this diet change.

8.7 For animals that are to be trapped, a description of the trap, trap placement, trap bedding and period of checking of the trap should be stated.

8.8 Methods of enrichment need to be provided. If enrichment is not provided, a justification needs to be provided, as to the reason for denying such items.

8.9 For facilities outside the UP, it is advisable to provide pictures of the housing environment, so that the committee can better visualise a project.

9. Facility Details:

The physical details of the facility need to be provided

9.1 With laboratory animal facilities and veterinary diagnostic laboratories falling under the jurisdiction of the South African Veterinary Council these facilities need to be registered with the organisation.

9.2 An emergency contact number needs to be provided, per chance that the committee needs to contact the facility in case of an emergency. The facility also needs to be easily contactable, per chance of an ethics complaint being received by the committee.

10. Statement of animal care competence, expertise and experience:

For this section the committee would like to know the degree of animal care of the personnel who are responsible for the care of the animals.

10.1 For this section, the committee requires a short statement of the scientific knowledge competence and experience of the person(s) appointed to ensure the comfort, health and humane treatment of the animal subjects in this study.

10.2 For controlled animal studies, the daily care of the animal should be under the care of a person to be registered as a laboratory animal technologist with the SAVC, authorised to undertake procedures of a laboratory animal technologist by the SAVC, or be registered veterinarian.

10.3 For zoological studies, the person responsible for the care of the animal needs to have appropriate species-specific experience.

10.4 If the study makes use of highly specialised procedures (e.g. surgery) the experience of the person should be stated (Please note that surgery on animals by a medical surgeon is allowable as long as a veterinarian is assisting.)

11. Experimental design:

For this section, the Committee requires an explanation as to how the study will be conducted for any study using animals or samples collected from animals (even if stored samples are in use):

11.1 This should include the rationale behind the specific study design and the necessity of specific study groups.
11.2 The link between the study design and statistical analysis can also be highlighted.
11.3 The manner of allocation animals to the various research groups should also be stated.
11.4 If the study makes use of infectious controls or controls exposed to a particular stressor where treatment is withheld (e.g. pain studies denying analgesia, infection studies that are need to determine the extent of non-treated pathology, psychological studies), the inclusion of these groups need to be properly justified. Furthermore its needs to be clearly articulated as to why historic controls cannot be used in the study.
11.5 For studies involving the testing of a vaccine agent, the committee will need to know the adjuvant in use. Freud's complete adjuvant is not acceptable to the committee, due to severe tissue damage induced by this adjuvant.
11.6 For ease of understanding, the committee would like to have a diagram attached.

12. Restraint of the animals:

For this section the committee would like to know how animals will be restrained for handling, dosing or termination:

12.1 This would require a description of the restraint equipment like the use of mouse restrainer, metabolic cages/chambers, and physical restraint by hand, restraining boards, ropes, etc.
12.2 The time period of restraint needs to be stated. It is not acceptable to restrain animals in a confined area for a long periods of time, without proper justification. The committee would prefer it, if an animal was restrained only when needed and thereafter released into its housing/holding environment until the next point of restraint.
12.3 If animals are chemically immobilised, the method of administration need to be stated.

13. Experimental animal procedures:

For this section, the committee would like a description of how the animals will be handled, sampled and treated:

13.1 If blood is to be collected, the site of sampling needs to be stated, together with the volume of collection. The volume of blood collection needs to be justifiable. The recommendation is no more than 1% of blood volume to be sampled within a period, unless sufficient time is allowed for regeneration.
13.2 If treatments are administered, the route of administration needs to be stated, together with the specifics of the treatment.
13.3 Experimental procedures should also be described in this section e.g. the surgical technique in use, the physiological methods employed.

14. Administration of all medicines/substances:

The committee will require all substance to be administered to an animal in the project, to be listed:

14.1 For each agent/substance listed, the dose and route of administration needs to be stated.
14.2 For studies that involve schedule medicines, the control of the drugs needs to be stated. The applicable legislation of the Medicines and Related Substances Control Act and the Veterinary Act need to be catered for.
14.3 If the animals need to be starved before administration of medicines, this should also be stated. This is important as excessive withdrawal of animal food can result in weight loss or induce abnormal behaviour.

15. Severity of effects of the experimental procedures on the animals:

This section is important for the committee to gauge the degree of deprivation, fear, distress or pain that an animal may experience in a particular study:

15.1 For this section the committee expects a description of the potential distress that a sentient animal may feel upon handling, restraint, drug administration etc. and the duration this degree of distress is expected.
15.2 The information of this section will also be taken as an indication of the researchers' experience with a particular species by the committee.
15.3 The mitigation of this distress can also be included in this section

16. Fate of animals and their disposal at the end of the study:

The committee would like to know what happens to an animal after a study is completed:

16.1 The re-use of laboratory animals is generally not supported. It is also preferred that laboratory rodents are terminated at the completion of the study, since they are not a natural zoological species. For zoological research, the committee would be satisfied with re-use of animals, as long at the initial study was not particularly stressful or if the use of animals already in captivity limits further wild-capture)

16.2 If animals are to be rehomed, this needs to be properly justified e.g. ability of an animal to cope in a home environment, disease spread, etc.

16.3 Wild animals that are removed from their environment, may only be returned with permission from the relevant Nature Conservation organisation.

16.4 For animals that are captured for short periods before releases, the time period for capture to release needs to be stated.

16.5 If animals are to be euthanized the method euthanasia needs to be stated.

16.5.1 In general the committee would prefer euthanasia by use of an anaesthetic overdose.

16.5.2 If another method is used, justification needs to be provided.

16.5.3 The method of euthanasia must be suitable for the species in question.

16.5.4 If methods such as decapitation, cervical dislocation or rapid CNS trauma are the method of euthanasia, the experience of the operator and where necessary the equipment to be used must be stated.

16.6 The method of carcass disposal needs to be provided. This must be in compliance with environmental and municipal legislation.

17. Statistical analysis:

For this section, the committee would like an understanding of the analysis of the data:

17.1 This is important for the committee to understand the particular reasoning behind the inclusion of a particular group of animals in the study.

17.2 Please keep the description basic, as the committee is not supported by persons with advanced statistical skills

17.3 The statistical analysis in use, will also allow the committee to gauge the importance of especially the control groups in the study.

18. Refinement:

This is the last R in animal research. For this section, the committee would like to see how the project has been made a humane as possible:

18.1 In this section, mention can be made towards enrichment, if not already mentioned in the housing section.

18.2 For studies that are painful, mention toward analgesic support can be made. It is unacceptable to deny an animal analgesic support unless the study involves the study of pain or that the painful sequela of the disease are a necessary study parameter (e.g, anti-inflammatories treatment being withheld in a drug discovery study for a new anti-inflammatory). When considering denying analgesic support, consideration must be made towards the management of the human or veterinary patient, where it is very unlikely that the treatment of a disease will occur without concurrent analgesic support when needed. Consideration should also be given to the different types of available for analgesia in animals i.e. if there is concern that the drug could interfere with the study through its mechanism of functioning, a drug working via a different mechanism should be tried.

18.3 For vaccination studies, the committee would expect a research project to show “proof of concept” in terms of increases in protective antibodies, cellular immunity or viral neutralisation in healthy animals before permission to ascend to a challenge animal study will be granted. If this proof of concept cannot be demonstrated without a challenge study, the committee would expect a proper justification from a researcher.

18.4 For animal kept under standard production conditions: Since these animals are inducted into a research project, the committee would still want to see attempts at enrichment.

18.5 If new enrichment is being tested or offered, the committee would like to see some understanding from the researcher for its use.

18.6 The committee has no objection to treats being offered as enrichment aids.

19. Monitoring of experimental animals:

Since animal experimentation can be stressful to an animal, or result in untoward physiological effects, the animals have to be properly monitored twice daily for the duration of the study, to pick up changes from normal. For monitoring the committee needs the name(s) of the responsible person(s) for each activity to be noted (This is important in cases of a complaints, so that the committee would know who the responsible person(s) are). For this monitoring the following are some useful monitoring points (please indicate how frequently the said monitoring will be undertaken):

19.1 Food and water intake

19.2 Changes in General Habitus (e.g. a healthy mice will be calm, move around, be inquisitive and vocalise)

19.3 Posture (e.g. hunched back, and head pressing are signs of pain)

19.4 Condition of the fur/hair (Unkempt fur is usually an indication of stress as the animal has stopped grooming)

19.5 Porphyrin staining as an indication of stress in rodents

19.6 Weight loss or failure to gain weight: is an important parameter in an animal experiment as one of the major impact of stress is loss of appetite. This is particularly of importance in small animals, as their metabolic rates can result in severe catabolism in a short period of time. As a result the committee would expect frequent monitoring of animal weights while within a study. A minimum recommended is weekly for larger animals and twice/three times weekly for rodents.

19.7 Level of activity (e.g. pain will may result in an animal being reluctant to move)

19.8 Body temperature (could indicate hyperthermia or pyrexia)

20. End points for experiments in animals:

The end-points of the study are the conditions that a project will be terminated by the researcher for human reasons. The committee will look at these strictly and may impose certain criteria on a project. The end-points need to link to the monitoring criteria mentioned. The following are general recommendations to consider:

20.1 Percentage weight loss: The committee will expect a rodent in a study to be withdrawn (and if necessary terminated) if the animal loses 10% of body weight over a three day period, while this can be increased to 15% for larger animals (The committee will accept modifications to these with proper motivation).

20.2 Injury: The management of animals with minor and major injuries need consideration as to when a study will be stopped. As a general note, minor injuries can be treated without interfering with the study, while major injuries will require an animal to be withdrawn for the study for further treatment. If it is not possible to treat major injuries, the termination policy needs to be stated.

20.3 Animals showing distress: If animals are severely distress, the management of the animals to overcome this stress needs to be stated (studies involving stress needs to be have other clear end-points towards the termination of the study).

20.4 Pain: Animals that are in pain need to be given proper analgesic support. If an analgesic cannot be administered, reasons need to be provided why the animal are denied pain (if not covered in other sections). Animals that are in severe pain that are not responsive to analgesia need alternate treatment or need to be terminated according to other criteria (studies denying analgesia needs to be have other clear end-points towards the termination of the study).

20.5 Animals becoming moribund: Animals that are moribund can be taken to be in a physiological state whereby they will be unlikely to recover e.g. in a toxicity study, a moribund animal can be considered an unscheduled death, and usually will not offer any further information that will necessitate the animal being taken to death. If the researcher feels that death should be an end-point rather than an animal being moribund, this needs to be substantially motivated.

20.6 Fever: In infectious studies, it is not unusual for an animal to develop a pyrexia during the study. The application will need to clearly state the changes in body temperature expected, and when treatment will be administered. If treated cannot be administered, other end-points need to be provided.

21. General veterinary care:

In terms of animal ethics a veterinarian has to be involved with the study:

21.1 The person has to be registered with the SAVC or authorised as a full veterinarian (Please note that a para-veterinarian cannot fulfil the functions of a veterinarian)

21.2 The veterinarian has to read through the protocol and ensure that the protocol is ethically designed.

21.3 The veterinarian is also the first contact point for the investigator per chance an unexpected finding results in a study or the end-points are reached and there is uncertainty.

21.4 The veterinarian is also needed per chance that an animal is injured and requires treatment.

21.5 For studies in facilities that don't have a full-time veterinarian, the scheduled timetable of animal checks need to be provided. For controlled studies this is expected to be once weekly.

21.6 For zoological studies in the field, the committee understands that it would be impossible to have a project veterinarian available at all times. Under these conditions, the veterinarian needs to be on-call and within reach (e.g. can be called out to deal with an injured animal or is close enough to move the animal to the vet.).

21.7 Any study that requires the immobilisation or anaesthesia of an animal, the veterinarian has to be present for the duration of the procedure and for the reasonable recovery period after anaesthesia.

21.8 Any study involving surgical procedures on an animal, must have a veterinarian present when said procedures are being undertaken by persons other than the said veterinarian (e.g. medical specialist surgeon).

22. Personnel activities:

The committee would like a name of persons involved in the study, and the role that they play. Since this section will contain information already presented in other sections, the activities can be abbreviated.

23. **Biohazard statement:**

As part of its responsibility to the UP, the committee has to ensure that the projects does not pose a hazard to the animal, the environment, staff or students from the use of infective agents, toxic substances, carcinogenic agents or ionising radiation? If the project has the potential to be harmful, the specific safety procedures to be followed to contain these hazards need to be provided and supported by the relevant Institutional Safety Officer.

24. **Declaration for studies needing external approval**:

The committee understand that projects needing external approval will require the committee to provide approval first. However, ethics does require regulatory compliance as well. To allow for these, this approval will be provided by means of a letter with the statement that while the project has been approved, it cannot be start until the necessary permits have been received by the AEC secretariat i.e. the certificate will only be issued at this later point. However, since the committee may not know the steps in the regulatory approval (Albeit unlikely), you will be required to indicate which permits you'll be applying for. If necessary, the committee may further indicate which approvals certificates it deems necessary before the ethical certificate will be issued.