1. PURPOSE

This Standard Operating Procedure (SOP) describes acceptable procedures for euthanasia of large animal species such as swine, dogs, rabbits, cats, non-human primates (NHP), sheep, goats and cattle. It ensures that animals are euthanized in the most humane way possible.

2. RESPONSIBILITY

Principal investigator (PI) and their research staff, veterinary care staff.

3. CONSIDERATIONS

All animal euthanasia must be performed by appropriately trained personnel approved on the Animal Use Protocol. Euthanasia procedures should not be performed in the same room where animals are housed. All euthanasia procedures must be continuously monitored by the person(s) performing the procedure, until confirmation of euthanasia is complete. Animals must not be left unattended until the procedure is complete.

4. ACCEPTABLE METHODS BY SPECIES

4.1. Rabbit, ferret, cat, dog, non-human primate:
   4.1.1. Barbiturate overdose
   4.1.2. Exsanguination under general anesthesia

4.2. Sheep, swine, goat, cattle:
   4.2.1. Barbiturate overdose
   4.2.2. Exsanguination under general anesthesia
   4.2.3. Physical methods

5. PROCEDURES

5.1. Barbiturate overdose:
   5.1.1. Sedate animals prior to barbiturate overdose.
   5.1.2. Pentobarbital sodium is the main agent of injectable commercial euthanasia solutions.
   5.1.3. Inject pentobarbital intravenously at a dose of 120mg/kg.
       Note: in ferrets, inject pentobarbital intraperitoneally.
   5.1.4. Confirm euthanasia before disposing of the carcass by observing that there is no respiratory movement for at least 3 minutes, and that the heartbeat has ceased.

5.2. Exsanguination:
   5.2.1. Exsanguination is not recommended as a sole means of euthanasia, but may be considered as an adjunct to other agents or methods
   5.2.2. Deeply anesthetize the animal as per SOP.
   5.2.3. Verify that withdrawal reflex is absent by pinching the toes with hemostats of non-hooved animals and pinching leg tendon of a hooved animal.
   5.2.4. Withdraw the maximum volume of blood; volume should be sufficient to cause death.
5.2.5. Confirm euthanasia before disposing of the carcass by observing that there is no respiratory movement for at least 3 minutes, and that the heartbeat has ceased.

5.3. Physical methods:

5.3.1. Licensed slaughterhouse:

5.3.1.1. Use physical methods only on food animals for agricultural use.
5.3.1.2. Animal transportation and slaughter must comply with federal and provincial laws and regulations.

6. REFERENCES

6.2. Canadian Council on Animal Care. CCAC guidelines on euthanasia of animals used in science (2010).

SOP REVISION HISTORY

<table>
<thead>
<tr>
<th>DATE</th>
<th>NEW VERSION</th>
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<tr>
<td>2019.11.20</td>
<td>4.1.1. If necessary, tranquilize Sedate animals in pain or agitated prior to barbiturate overdose.</td>
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<tr>
<td>2019.11.20</td>
<td>4.2.2 Sedate animals prior to exposure to inhalant anesthetics.</td>
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<tr>
<td>2019.11.20</td>
<td>4.3.3 Withdraw the maximum volume of blood; volume should be sufficient to cause death.</td>
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| 2020.12.09 | 3. CONSIDERATIONS
All animal euthanasia must be performed by appropriately trained personnel approved on the Animal Use Protocol. Euthanasia procedures should not be performed in the same room where animals are housed. All euthanasia procedures must be continuously monitored by the person(s) performing the procedure, until confirmation of euthanasia is complete. Animals must not be left unattended until the procedure is complete. |
| 2020.12.09 | 4.1. Rabbit, ferret, cat, dog, non-human primate: |
| 2020.12.09 | 4.2. Sheep, pig, swine, goat, cattle: |
| 2020.12.09 | 5.1.3. Inject pentobarbital intravenously at a dose of 100 to 120mg/kg. Note: in ferrets, inject pentobarbital intraperitoneally. |
| 2020.12.09 | 5.2.6. If the animal is not dead (or for additional security), follow exsanguination by another method of euthanasia, such as overdose of barbiturates. |
| 2021.01.13 | 5.2. Overdose of inhalant anesthetic:
5.2.1. The following gases can be used: halothane, isoflurane, methoxyflurane, other halogenated inhalants.
5.2.2. Sedate animals prior to exposure to inhalant anesthetics.
5.2.3. Expose the animal to a high gas concentration using an anesthetic vaporizer.
5.2.4. Vapors are inhaled until respiration ceases and death ensues.
5.2.5. Confirm euthanasia before disposing of the carcass by observing that there is no respiratory movement for at least 3 minutes, and that the heartbeat has ceased. If the animal is not dead (or for additional security), follow inhalant anesthesia overdose by another method of euthanasia, such as exsanguination. |