TITLE: Euthanasia of Rodents

Policy Number: 2014-015

Responsible Department: Institutional Animal Care and Use Committee

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Purpose of Policy: The purpose of this policy is to ensure that euthanasia of laboratory rodents is performed in as humane a manner as possible that does not interfere with the scientific objectives.

Policy Information: For pharmacological methods, WesternU’s Institutional Animal Care and Use Committee (IACUC) requires the use of either pentobarbital, at least 100 mg/kg intraperitoneally (IP); Euthasol (390 mg pentobarbital + 50 mg phenytoin per ml), at least 5 ml/kg IP; or inhalation of 32% isoflurane gas for euthanasia of laboratory rodents. Although the AVMA Guidelines for the Euthanasia of Animals lists carbon dioxide (CO₂) asphyxiation as a conditionally acceptable means of euthanasia, the IACUC does not consider this use of CO₂ to be humane and, therefore, will not approved this use of CO₂ without rigorous scientific justification (Refer to IACUC Policy No. 2014-005; Use of Carbon Dioxide for Euthanasia). Any means of euthanasia not described in this policy must be approved by the IACUC.

In accordance with the AVMA guidelines cited above, anesthesia must be administered prior to euthanasia by a physical method unless scientific evidence is provided that such agents would interfere with the experimental end points.

Physical methods of euthanasia may be accompanied by ultrasonic vocalizations and pheromones released during the process and odors released in the blood afterwards. Such stimuli may induce stress in other animals housed nearby. Therefore, euthanasia using physical methods should be performed outside the animal housing room and away from other animals. If multiple animals are to be euthanized, they should be brought into the sacrifice area one at a time.

Decapitation by guillotine is an inherently dangerous procedure and, for their own safety, personnel must be properly trained in its use. The AVMA guidelines state that “equipment used to perform decapitation should be maintained in good working order and serviced on a regular basis to ensure sharpness of blades.” Thus, when using a guillotine for decapitation, the following will be required:

- The unit should be wiped clean after each use.
• After each cleaning, a few drops of 3 in 1 oil should be applied to the blade surfaces and channels and the blades run together several times to spread the oil evenly over all moving surfaces.
• Blades should be replaced or sharpened as the frequency of use demands.

A guillotine is considered sharp enough if it will cut a thick rubber band without dragging it between the blades and sticking.

Cervical dislocation can cause serious injury and suffering to an animal if not performed properly. Therefore, those carrying out the method must be properly trained to do so. In accordance with the AVMA guidelines, cervical dislocation may not be performed on rodents weighing more than 200 grams since the large physical mass in the cervical region makes the procedure physically more difficult. In such cases, decapitation is recommended.

Fetuses: Neural development in fetuses up to 14 days gestation is minimal and pain perception is unlikely. Euthanasia of the mother or removal of the fetus should ensure rapid death from loss of blood supply. Neural development in fetuses from 15 days gestation to birth is such that fetuses might experience pain. Fetuses at this age are resistant to inhalational anesthetics and it may be preferable to euthanize using parenteral anesthetics, cervical dislocation or decapitation with surgical scissors.

If chemically fixing whole fetuses, they should be anesthetized prior to immersion in or perfusion with fixative solutions by either 1) injecting the fetus with an anesthetic; 2) deeply anesthetizing the mother with a parenteral anesthetic that crosses the placenta (e.g. pentobarbital), or 3) hypothermia of the fetus immediately following removal from the womb.

If fetuses are not required for the study, euthanasia of a pregnant animal must ensure rapid death of the fetus (e.g. pentobarbital).

Neonatal rats and mice (postnatal day 1 to postnatal day 14 inclusive) are resistant to CO₂ asphyxiation and, therefore, pentobarbital at 100 mg/kg IP or 32% isoflurane inhalational gas is recommended. Neonates over 14 days of age may be euthanized per adult guidelines. If CO₂ euthanasia is approved by the IACUC, a secondary physical method of euthanasia is required to ensure death.

Ether is not permitted for anesthesia or euthanasia of rodents as it is highly flammable and hepatotoxic to rodents and humans.