

**COMPARATIVE RESEARCH FACILITY
TEXAS STATE UNIVERSITY**

Title: Humane Endpoints for Tumor Growth in Rodents

Purpose: To provide guidance on humane endpoints for tumor growth in rodents.

Scope: This guideline applies to all personnel caring for or using rodents for research at Texas State University.

Background: This guideline addresses humane endpoints for spontaneous (naturally occurring) and/or experimentally induced tumor growth in rodents.

Definitions

- Body Condition Score, as determined by the trained observer with regards to muscle and fat deposition over various areas of the body. This is typically based on a 5-point scale with 1 being emaciated, 3 being ideal, and 5 being obese. See Appendix I.
- Moribund: near death or in the state of dying
- Cachexia: severe loss of weight and muscle mass which cannot be reversed nutritionally
- Ulceration: an inflamed lesion on the skin or internal surface involving tissue destruction

Procedure:

Establishment of tumor models must be done in accordance with an approved IACUC protocol

Where possible, the tumor implantation site should be in an area which minimizes damage to adjacent normal structures (i.e.: intrascapular or flank region for superficial tumors).

It is the responsibility of the principal investigator (PI) to ensure that experimental animal(s) are observed on a regular basis (as stated in the approved IACUC protocol) by lab personnel and that action is taken when endpoints are reached.

Endpoints for euthanasia must be explicitly defined within the approved protocol and animal(s) are to be monitored for such endpoints to minimize the possibility of experiencing pain and distress.

The suggested clinical criteria for determining endpoints include the following:

Rapid or progressive weight loss-Greater than 20% weight loss from baseline weight or age matched controls determined by obtaining the weight of the animal on a regular basis (in general at least two times per week).

Declining BCS - decreasing to 2 (thin) or 1 (emaciated) on a 5-point BCS scale; see Appendix. As weight gain may occur with tumor growth, BCS provides a better assessment of body condition.

Cutaneous tumors should be measured with calipers at sufficient frequency to ensure the maximum tumor size is not exceeded before the animal is euthanized. A single tumor should not exceed a diameter length of 2 cm in adult mice and 4 cm in adult rats in any one dimension.'

In cases where multiple tumors spontaneously occur or are experimentally induced, the size should be correspondingly less and should not exceed the maximum size of a single tumor.

Difficulty ambulating- locomotion is impaired, hind limb weakness, ataxia, or immobile.

Lethargy- decreased activity, loss of interest in environment, slow or reluctant to move when provoked.

Appearance - rough hair coat, progressive dermatitis, hunched posture, abdominal distention, self-mutilation, dry/dull eyes, or dehydration.

Tumor assessment-tumor ulceration, necrosis, or infection, impedes ambulation, elimination, or eating, or if in contact with bedding.

Systemic illness - debilitating diarrhea, cough, nasal discharge, neurologic signs (circling, head pressing, seizure activity), prolonged hyper- or hypothermia, jaundice and/or anemia, or bleeding from any orifice.

If any one of the following criteria are met, the animal is to be euthanized immediately unless otherwise approved in the IACUC protocol:

- Moribund
- Ulceration, infection, or necrosis of the tumor
- Cachexia
- Labored breathing
- Unable to obtain food or water
- Impaired locomotion

Hanahan, D. and Weinberg, R. "Hallmarks of Cancer: The Next Generation." *Cell*, 144(5). (2011): 646 "Ulman-Cullere, Mollie and Foltz, Charmaine. "Body Condition Scoring: A Rapid and Accurate Method for Assessing Health Status in Mice." *Lab Animal Science*. 49(3). (1999): 319-323.

""Merriam-Webster Definition of Moribund." *Merriam-Webster Dictionary*. Merriam-Webster 2013, Incorporated, n.d. Web. 16 July 2013.

IV Workman, P., E. O. Aboagye, F. Balkwill, A. Balmain, G. Bruder, D. J. Chaplin, J. A. Double, J. Everitt, D. A. H. Farnham, M. J. Glennie, L. R. Kelland, V. Robinson, I. J. Stratford, G. M. Tozer, S. Watson, S. R. Wedge, and S. A. Eccles. "Guidelines for the Welfare and Use of Animals in Cancer Research." *British Journal of Cancer*, 102.11 (2010): 1555-577. Print.

"Guidelines for Endpoints in Animal Study Proposals." *ARAC Guidelines*. NIH OACU, Rev. 10 Apr. 2013. "Paster EV, Villines KA, Hickman DL. "Endpoints for Mouse Abdominal Tumor Models: Refinement of Current Criteria". *Journal of Comparative Medicine*. 48.3 (2009): 235-239.

Appendix I The following is described by Ullman-Cullere, et. al. *Body Condition Scoring: A Rapid and Accurate Method for Assessing Health Status in Mice* in the 1999 Journal of Lab Animal Science. 49 (3).



BC 1

- Mouse is emaciated.
- *Skeletal structure extremely prominent; little or no flesh cover.*
- *Vertebrae distinctly segmented.*



BC 2

- Mouse is under conditioned.
- *Segmentation of vertebral column evident. Dorsal pelvic bones are readily palpable.*



BC 3

- Mouse is well-conditioned. *Vertebrae and dorsal pelvis not prominent; palpable with slight pressure.*



BC 4

- Mouse is over conditioned.
- *Spine is a continuous column.*
- *Vertebrae palpable only with firm pressure.*



BC 5

- Mouse is obese.
- *Mouse is smooth and bulky.*
- *Bone structure disappears under flesh and subcutaneous fat.*

A "+" or a "-" can be added to the body condition score

If additional increments **are necessary** (i.e., 2+, 2-...²)



BC 1 – Rat is emaciated

- Segmentation of vertebral column prominent if not visible
- Little or no flesh cover over dorsal pelvis. Pins prominent if not visible.
- Segmentation of caudal vertebrae prominent



BC 2 – Rat is under conditioned

- Segmentation of vertebral column prominent
- Thin flesh over dorsal pelvis, little subcutaneous fat. Pins easily palpable
- Thin flesh cover over caudal vertebrae, segmentation palpable with slight pressure.



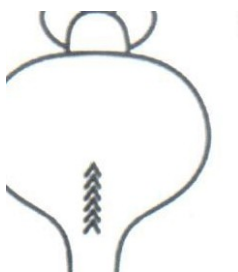
BC 3 – Rat is well conditioned

- Segmentation of vertebral column easily palpable
- Moderate subcutaneous fat store over pelvis. Pins easily palpable with slight pressure.
- Moderate fat store around tail base, caudal vertebrae may be palpable but not segmented.



BC 4 – Rat is over conditioned

- Segmentation of vertebral column palpable with slight pressure.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.



BC 5 – Rat is obese

- Segmentation of vertebral column palpable with firm pressure; may be a continuous column.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis not palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.