Delaying the Inevitable: a Justified Means to an End?

Great Eastern University's (GEU's) IACUC was presented with an allegation of non-compliance. The post-approval monitor, Mr. Cooper, identified that Dr. Guaio's lab staff were not adhering to the humane endpoints approved in his IACUC protocol. Dr. Guaio's IACUC protocol indicated that mice with tumors exceeding two centimeters of growth would be humanely euthanized. Cooper found over five cages containing *Peromyscus* (deer mice) with tumors exceeding these approved endpoints. Consequently, Dr. Guaio and his junior post-doc, Dr. Yin, were both required to attend the IACUC meeting to explain the protocol deviation. Guaio and Yin admitted no wrongdoing since the protocol deviation occurred only after consultation with the attending veterinarian, Dr. Tracy Thompson. They explained that their research included the use of novel therapeutics that reduce cancer growth. Yin said that even though the tumors exceeded the defined endpoints, they needed at least one extra week to administer the therapeutics and collect sufficient data for analysis. Although the protocol defined endpoints were exceeded, Yin indicated that based on his discussion with Dr. Thompson, the deer mice could be maintained for at least another week. Thompson examined the animals and granted the extension provided that monitoring was increased to twice a day and palliative care was provided (i.e., moving food to the cage floor, providing diet gel). Thompson attended the meeting and informed the IACUC that she felt the animals could continue in the study. She indicated that allowing the animals to continue in the study for a few extra days was much better that wasting the animals by mandating euthanasia just to satisfy the defined humane endpoints. If Guaio could get data out of these animals, then protocols could be amended to align the humane endpoints with what the animals are experiencing. GEU's IACUC agreed with Thompson's logic that precious animal resources should be preserved, but the committee indicated measures should be taken to ensure future processes would adhere with the regulatory expectations. The committee members were certain that deviating from an approved protocol was non-compliance, but struggled with the need to preserve animal resources. Can a veterinarian, PI, IACUC Administrator, or anyone else unilaterally permit animals to exceed the IACUC approved humane endpoints for the purpose of research activities and without IACUC approval? ❐

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A WORD FROM USDA AND OLAW

In this scenario, after consultation with the veterinarian, a researcher extends the euthanasia endpoint of a group of deer mice, a species covered under the Animal Welfare Act (AWA)[1], beyond the IACUC-approved humane endpoint of two centimeters of tumor growth. The justification was based on the need to continue data collection and not waste the animals unnecessarily.

Response from the USDA

The change in an endpoint of a study constitutes a significant change to an ongoing animal activity. Under the AWA regulations, a significant change requires Institutional Animal Care and Use Committee (IACUC) review and approval[2]. The Attending Veterinarian in this scenario did not have the individual authority to approve the tumor growth beyond what was written in the protocol as an endpoint. The Principal Investigator is required to submit the proposed significant change to the IACUC for review and approval before implementation. The submission should include the rationale for changing the endpoint, and address measures to minimize the pain and distress associated with the proposed change[3]. The incident is not reportable to the USDA; however it would be cited on the inspection report as a noncompliance with the regulatory requirement for IACUC approval of significant changes to an animal activity. Note: An institution may have additional approval processes besides the IACUC. The officials of these additional approval mechanisms do not have the authority to approve an animal activity or a significant change to an activity that has not been approved by the IACUC[4].

Response from OLAW

In their unique role, the veterinarian may always intervene when a clinical situation arises that requires treatment[5]. However, in this case, the veterinarian is not providing clinical care for the affected animals. Because the change in the endpoint may result in greater pain or distress, it constitutes a significant change that requires IACUC review and approval[6]. As a result, the veterinarian's guidance, and the subsequent delay in euthanasia without IACUC approval is noncompliant with the PHS Policy and reportable to OLAW[6–8]. ❐

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Opportunity for an improved animal care program

We understand deer mice to be a USDA-regulated species and assume applicability of the PHS Policy. The Animal Welfare Regulations and PHS Policy (see IV.A.3.b.(1)) state the composition of the IACUC will include a veterinarian with direct/delegated program authority and responsibility for animal activities[1,2]. *The Guide* further states that "Veterinary consultation must occur when pain or distress is beyond the level anticipated in the protocol description"[3].

The Protocol Review coordinators offer the following compliance considerations:

1. **What is a humane endpoint?**
Per the *Guide*, a humane endpoint is "the point at which pain or distress in an experimental animal is prevented, terminated, or relieved. Euthanizing an animal at the humane endpoint prevents unalleviated pain and distress"[1].

2. **How do humane endpoints relate to the 3Rs (and preserve animal resources)?**

- Per the *Guide*, "The use of humane endpoints contributes to refinement by providing an alternative to experimental endpoints that result in unrelieved or severe animal pain and distress, including death"[1].
- Dr. Thomson's attempt to address the 3Rs, presumably, referred to reduction, which "involves strategies for obtaining comparable levels of information from the use of fewer animals or for maximizing the information obtained from a given number of animals (*without increasing pain or distress*) so that in the long run fewer animals are needed to acquire the same scientific information. This approach relies on an analysis of experimental design, applications of newer technologies, the use of appropriate statistical methods, and control of environmentally related variability in animal housing and study areas" [1].
- Dr. Thomson's decision to allow animals to continue in the study past the IACUC approved humane endpoints does not align with the concepts of the 3Rs or the relationship between humane endpoints and the 3Rs.

3. **Can a veterinarian, PI, IACUC Administrator, or anyone else unilaterally permit animals to exceed the IACUC approved humane endpoints for the purpose of research activities and without IACUC approval?**
Only the IACUC can change approved humane endpoints since that change would be considered significant. A significant change to protocol requires IACUC review by either Designated Member Review (DMR) or Full Committee Review (FCR). Veterinary Verification and Consultation (VVC) could not be used to approve this significant change because exceeding humane endpoints, by definition, results in "greater pain, distress, or degree of invasiveness"[2].

4. **Was the protocol deviation non-compliance?**
Deviating from an approved IACUC protocol is always considered non-compliance.

5. **What should the IACUC have done?**
This scenario represents a protocol non-compliance and, if not unique to the described situation, a programmatic deficiency. Members of GEU's Animal Care and Use Program require education and retraining. ❐

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The attending veterinarian (AV), Dr. Thompson, did appropriately evaluate the mice as would be expected from the program of veterinary care. The animals must have been in overall good health as she determined that with increased monitoring and palliative care the animals would not endure unnecessary discomfort if kept for the additional week, under continued veterinary (and lab) oversight. Although Dr. Thompson is an integral member of the IACUC and program, she should have promptly reported the matter to the IACUC, rather than it being discovered by Mr. Cooper during their post-approval monitoring activities. Doing so would have allowed the appropriate body, the IACUC, to deliberate on the incident in a timely manner and to conduct continued/ongoing review of the approved protocol, as stated in the PHS Policy (see IV.C.5)[2]. It would also prevent setting an institutional precedent where the AV could unilaterally be permitted to allow exceptions to protocol endpoints, which is not allowed.

Another concern is the confusion demonstrated by Dr. Guaio and his lab staff. The AV temporarily allowing continued use of the animals did not abdicate them of any wrongdoing. If they were following their protocol monitoring plan and endpoints, a non-compliance could have been avoided. The incident would be promptly reported to OLAW in accordance with their guidance on prompt reporting[4]. As OLAW holds a memorandum of understanding (MOU) with the USDA, we understand that the USDA would be notified. Grant funds should not be used during the period of non-compliance and the lab encouraged to use the data only to inform future experiments.

This is also a lesson in improved protocol review and consideration of properly defined humane and experimental endpoints. Tumor size alone is often not adequate in determining the overall health of an animal. IACUC reviewers (e.g., veterinary pre-review of the protocol) should ensure the intervention plan is reasonable to the model proposed. Research staff should be educated concerning the approaches that can be used when describing intervention plans. For example, could the animal assessment include a body condition score[5]? Could pharmacological or non-pharmacological means of supplemental care and increasing

monitoring occur as a first measure? If in uncharted territory, *The Guide* recommends that "the use of pilot studies is an effective method for identifying and defining humane endpoints and reaching consensus among the PI, IACUC, and veterinarian"[6]. We want to ensure animal welfare while preventing premature euthanasia of an animal.
Dr. Thompson likely recognized the need for refining the Guaio protocol, but misstepped in her failure to promptly defer the matter to the IACUC. ❐

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Endpoints and veterinary authority

This scenario relates to the scope of authority of the Attending Veterinarian (AV).

While we recognize, and partially agree, with Dr. Thompson's consideration of preservation of animal resources and the need to obtain valuable research data, however, the AV does not have the authority to unilaterally allow an exception for the Principal Investigator (PI) to exceed the IACUC-approved humane endpoints.

The responsibility of the AV is to implement a comprehensive program of veterinary care and serve as a voting member of the IACUC in accordance with the requirements of *The Guide for the Care and Use of Laboratory Animals*, the *PHS Policy* and the Animal Welfare Act and Regulations. Such responsibility requires that the AVs have sufficient program authority to uphold that responsibility. With respect to humane endpoints, the AV does have the authority to intervene in situations involving animal pain and distress and treat the animal, or even require humane euthanasia if necessary[1]. In other words, the AV has the authority to implement humane endpoints to minimize pain and distress, even if that occurs before the endpoints described in the protocol are reached. But the AV does not have the authority to extend predetermined humane endpoints that have been established and approved by the IACUC.

The *Guide* indicates that there are several key aspects to establishing humane endpoints: humane endpoint determination should include the PI, the veterinarian, and the IACUC – prior to the start of the study (i.e., during protocol review); the humane endpoint should be precisely defined (including assessment criteria); the frequency of animal observations should be described; the personnel monitoring the animals should be experienced and/or trained to recognize the endpoint, and; the required response, or action upon reaching the humane endpoint[2].

Given that establishing humane endpoints is a key component of animal studies and that the IACUC is tasked with ensuring that appropriate endpoints are in place, it would be inappropriate for a single member of the IACUC, or animal program, to later grant an exemption that exceeds IACUC-established limits.

While the regulations clearly require that a veterinarian serve on the IACUC, provide a veterinary care program to ensure the health and well-being of animals used in biomedical research, and must have the authority to implement such a program, the regulations are silent on granting the AV unilateral authority to exceed the IACUC-approved endpoints.

The PI had a clearly defined endpoint of tumor size greater than 2 cm approved in the protocol. To change the endpoint, would qualify as a "significant change" as per OLAW's definition; "significant changes include changes that have, or have the potential to have a negative impact of animal welfare. Significant changes must be approved by one of the valid IACUC approval methods described in the PHS Policy"[3]. Dr. Thompson must have assumed that the increasing tumor size may have a negative impact on the animals, as she required increased monitoring and the addition of palliative care. In this scenario, the AV cannot approve this significant change; IACUC review and approval is needed. ❐

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