Want to comment? Participants in the OLAW Online IACUC Staff Seminars have the opportunity to submit questions after the formal presentation. Your input is important, too. OLAW will accept questions and comments from viewers of this recording until June 1, 2012. After the question and comment period closes, OLAW will post the comments, questions, and answers on the OLAW webpage. Please go to the OLAW Education Resources page and click on the seminar title for further information.

Note: Text has been edited for clarity.

Contents:

Use of Non-Pharmaceutical-Grade Chemicals and Other Substances in Research with Animals

Speakers: Patricia Brown, VMD, MS, DACLAM, OLAW, NIH, Carol Clarke, DVM, DACLAM, Animal Care, APHIS, USDA and Christian Newcomer, VMD, DACLAM, AAALAC International Moderator: Jerry Collins, PhD, Division of Policy and Education, OLAW and Yale University. Broadcast Date: March 1, 2012. A recording of the seminar can be viewed at http://grants.nih.gov/grants/olaw/2012-03-01_OLAW_Online_Seminar_Use_of_Non_Pharmaceutical_Grade%20Substances.wmv (Windows Media Player – 42 min).

Slide 1 (Title Slide)
>> Dr. Collins: Hello and welcome to the next in our series of IACUC staff webinars [Online Seminars]. My name is Jerry Collins and I will be the moderator for today's webinar entitled: Use of Non-Pharmaceutical-Grade Chemicals and Other Substances in Research with Animals. As you have probably noticed, we have changed the time of this webinar from 2:00 to 1:00 p.m. Eastern Standard Time. Future webinars will all be presented at 1:00 p.m. Eastern Standard Time. Previously, we broadcast two webinars, one for IOs and one for IACUC Staff. Beginning today we are combining the two into a single webinar. This webinar will be recorded and made available on the OLAW website.

The box in the top right corner of your computer screen is your attendee interface. The audio mode enables you to select how you will listen to a webinar. If you have clicked on the Use Telephone Button the dial-in number, access code and audio PIN are displayed and audio will come to you over the phone. If you choose the Mic and speakers button, the sound will emanate from your computer. In the unlikely event of a problem with the audio, you may select the audio option not in use and listen to the remainder of the presentation using that mode. Your line is muted to help eliminate distracting background noises, so you don't have to worry about other participants hearing sounds from your location.

In December of 2011, NIH announced its decision to adopt the 8th Edition of the *Guide for the Care and Use of Laboratory Animals*. The determination to adopt the *Guide* was made after

OLAW's analysis of comments collected during a 90-day public comment period that ran from February to April 2011. After completing that analysis, OLAW developed and posted Position Statements to clarify how we expect Assured institutions to implement the *Guide*. A second comment period was initiated when the Position Statements were posted. This one was 65 days long -- it closed on February 3 of this year -- and enabled the public to comment on their understanding of the Position Statements. Today's webinar has been developed in response to the questions and concerns that you, our participants, expressed in both public comment periods.

We are delighted to have our colleagues from USDA Animal Care and AAALAC with us today. We will work together to explain each organization's policies, guidance and regulations, and the ways that research organizations can meet the expectations of all three organizations. During the first part of our webinar, each speaker will explain his or her organization's policy on the use of non-pharmaceutical-grade compounds in research with animals. The explanations will include definitions. This is important because our policies include words that are used in everyday English but have specialized meanings when used in the context of these regulations.

Then we will move on to the question and answer section. Usually at our webinars, we accept and answer your questions in real-time. But today things will be a little different. Questions have been selected and answers have been prepared in advance by staff here in the OLAW office. The questions reflect the detailed, thoughtful comments submitted during both public comment periods. We hope that this advance preparation will provide useful answers to the immediate questions and also give you insight into how we apply policy, guidance and regulations to the issue. However, if you have additional questions after the webinar ends, you may email them to olawdpe@od.nih.gov. Later on there will be a slide with that address on it.

You may also use the submit question box on the top right corner of your screen to send questions to us now. Those questions will only be visible to staff in the OLAW office. We won't have time to respond to those questions during the broadcast, but OLAW, USDA and AAALAC will work together to answer your questions. A compilation of these additional questions and answers will be amended to the webinar transcript and posted on the OLAW website under Educational Resources. You will be able to access a recording of this webinar, a PDF version of the slides and other background material at the same site. We will prepare all this material as quickly as we can but are not able to promise a specific date at this time.

Our first speaker, representing the USDA, will be Dr. Carol Clarke. Dr. Clarke received her Bachelor's degree from Johns Hopkins University and her DVM from Tuskegee School of Veterinary Medicine. She completed laboratory animal medicine training at SmithKline Beecham Pharmaceuticals and joined NIH, serving in a number of laboratory animal positions. She is a Diplomate of the American College of Laboratory Animal Medicine [ACLAM]. Dr. Clarke joined the US Department of Agriculture in 2011, and currently serves as the Research Specialist Staff Officer at APHIS [Animal and Plant Health Inspection Service], Animal Care headquarters located in Riverdale, MD.

Our second speaker, representing AAALAC, will be Dr. Christian Newcomer. Dr. Newcomer is the Executive Director of the Association for the Assessment and Accreditation of Laboratory Animal Care International, usually referred to as AAALAC. He is a graduate of the School of Veterinary Medicine at the University of Pennsylvania. Dr. Newcomer completed post-doctoral training in laboratory animal medicine at the University of Michigan and is board certified by ACLAM. Prior to his appointment at AAALAC International, he held positions in laboratory animal medicine at the Massachusetts Institute of Technology, Tufts New England Medical Center, the University of North Carolina at Chapel Hill, the Veterinary Resources Program here at the National Institutes of Health and the Johns Hopkins University.

Our third speaker of the day will be Dr. Patricia Brown. Dr. Brown currently serves as the Director of the Office of Laboratory Animal Welfare (OLAW) at NIH. She received her veterinary degree from the University of Pennsylvania and completed Laboratory Animal Medicine training at Penn State. She served in a variety of positions at the NIH before joining OLAW. Dr. Brown is a Diplomate of ACLAM.

Slide 2 (USDA Regulations Regarding the Use of Non-Pharmaceutical Grade Compounds in Research)
Now let's begin. Our first speaker is Dr. Carol Clarke from USDA. Dr. Clarke, may investigators use non-pharmaceutical-grade compounds in their research with animals?

Slide 3 (Policy 3: Veterinary Care)
>> Dr. Clarke: Yes, and good afternoon. Our Policy 3 on veterinary care addresses this issue. Investigators are expected to use pharmaceutical-grade medications whenever they are available, even in acute procedures. Non-pharmaceutical-grade chemical compounds should only be used in regulated animals after specific review and approval by the IACUC, for reasons such as scientific necessity or non-availability of an acceptable veterinary or human pharmaceutical-grade product. Cost savings is not a justification for using non-pharmaceutical-grade compounds in regulated animals. Next slide.

Slide 4 (Pharmaceutical-grade Compound)
Pharmaceutical-grade compound is any active or inactive drug, biologic, reagent, et cetera, which is approved by the FDA [or] for which a chemical purity standard has been written or established by any recognized pharmacopeia, which is a book or a compendia, such as the US Pharmacopeia [USP], the National Formulary [NF], the British Pharmacopoeia [BP], the Pharmacopoeia of the Council of Europe [EP]. Note the USP and the NF have combined their standards into one compendia and I have enclosed the link [http://www.usp.org/usp-nf]. Next slide.

Slide 5 (Guidance to IACUCs)
Some guidance to the IACUCs when reviewing a protocol. Consider the following: Consider the goals. The justification. The level of pain and distress. And with that, will purity differences result in toxic and adverse effects? Will there be an increase in pain and distress? If so, consider alternatives. And I would refer you to the National Agricultural Library, the Animal

Welfare [Information] Center. Our website is enclosed. Also, look at the procedure. Is it a survival or a non-survival procedure? Sterility would be a consideration for survival procedures. Next slide.

Slide 6 (USDA Expectations)
USDA expectations are thus: The wording of the regulations provides latitude for professional judgment; hence, it is expected that the system of protocol evaluation serves as an opportunity for IACUC and investigators to work together in determining an optimal way to conduct experiments within the context of humane animal care. Next slide.

Slide 7 (Summary)
To summarize, Policy 3 states that pharmaceutical-grade compounds are to be used whenever available. Non-pharmaceutical-grade compounds are only used when the pharmaceutical-grade is unavailable or when there is a scientific justification which has IACUC approval. Cost savings is not an acceptable justification. The USDA accepts the FDA definition of a pharmaceutical-grade compound. Finally, the regulations provide latitude for IACUC and investigators to work together in determining an optimal way to conduct experiments within the context of humane animal care. Next slide.

Slide 8 (Acknowledgements)
I would like to acknowledge Dr. Kay Carter-Corker, D'Anna Jensen, Dr. Rafaat Fahmy and Dr. Pamela Chamberlain for their contributions to this presentation. Next slide.

Slide 9 (AAALAC's Consideration of the Use of Non-Pharmaceutical-Grade Chemicals and Other Substances in Research with Animals)
>> Dr. Collins: Our next speaker is Dr. Chris Newcomer from AAALAC. Chris, may investigators use non-pharmaceutical-grade compounds in their research with animals?

>> Dr. Newcomer: Hello everyone; thank you for joining us today. At the outset of my brief remarks, I would like to remind you that AAALAC International is participating in today's webinar not as regulatory agency, but as a non-profit, volunteer accrediting organization, and I invite those of you who are not familiar with the accreditation program to learn more at www.aaalac.org.

Now, to the question posed by Dr. Collins about the use of non-pharmaceutical-grade compounds in their research animal studies: There are clearly circumstances where it may be necessary to use a non-pharmaceutical-grade compound to meet the requirements of a scientific investigation. For example, and here are two obvious ones, studies on the pharmacology or toxicology of ethnobotanical compounds or of street drugs of abuse, may involve chemical blends that are not fully characterized or available in a pharmaceutical-grade. The use of these compounds would be fully acceptable with IACUC oversight and review to establish and assure that the preparation and use of the compounds are appropriate for the unique scientific objectives of the study. Next slide.

Slide 10 (AAALAC's Position)
Also I would like to note that AAALAC International's Council on Accreditation has appointed a committee to prepare a Frequently Asked Question on the subject of the use of non-pharmaceutical-grade substances in research animal studies. This FAQ is under active development and I anticipate that the final review and adoption of our advisory on this topic will be available in the spring of 2012, tentatively in the May time-frame. I have no intention of preempting the effort or prerogative of that committee, but I am pleased to make a few general remarks based upon AAALAC's experience on this subject over the past two decades.

AAALAC site visitors have frequently encountered instances where non-pharmaceutical-grade compounds were used in research for therapeutic and/or investigational purposes. Our standards of assessment in these instances have been framed from applicable regulatory requirements and guidelines and from contemporary veterinary practices. Next slide.

Slide 11 (AAALAC's Assessment of NPG Drug Use: General Factors of Importance)
As I noted earlier, there are clearly instances where the use of non-pharmaceutical-grade compounds may be essential to meet experimental objectives and may be permitted with appropriate institutional oversight. AAALAC International expects the IACUC to play a central role in the oversight and approval of the use of non-pharmaceutical-grade compounds, beginning with the review of the scientific justification for such use. If the compound is available commercially in a pharmaceutical-grade counterpart, the IACUC should establish why this product is unacceptable, discounting cost as a factor relevant to this determination.

The IACUC process should ensure that applicable regulations and guidelines are taken into consideration during the review. When circumstances require that a compound must be formulated within the institution or one of its laboratories, the chemical purity of the compound or compounds should be US Pharmacopeia or higher for therapeutic applications and whenever possible for research applications. It is also important for the IACUC to ascertain key quality control and assurance factors as follows:

- Provisions for drug reconstitution, preparation and/or compounding should be appropriate.
- Attention should be paid to drug purity, sterility, osmolality, concentration and other parameters impacting subject response and animal welfare.
- Drug safety, efficacy and shelf-life measures should be established to protect subject welfare and promote valid experimental outcomes.
- And personnel responsible for the preparation, administration and experimental evaluation of compounds at the institutional level should have adequate training, experience and performance.

Next slide.

Slide 12 (AAALAC's Position)
In consideration of the broad areas of concern mentioned in the previous slide, it is important to note that AAALAC's approach to the assessment of non-pharmaceutical-grade use has been based upon performance standards affording institutions considerable latitude in the path to

research outcomes that are non-injurious to animal subjects. Typically, when problems related to non-pharmaceutical drug use have been detected, AAALAC has ranked these findings as Suggestions for Improvement and not as Mandatory items in accreditation reports. However, when clear, demonstrable adverse animal welfare outcomes are detected that are related to the use of non-pharmaceutical-grade compounds and are not the expected experimental consequences of the compounds under study, AAALAC International may elevate the finding to a Mandatory item that requires correction for successful accreditation.

Slide 13 (Use of Non-Pharmaceutical-Grade Chemicals and Other Substances in Research with Animals)
>> Dr. Collins: Our next speaker is Dr. Patricia Brown from OLAW. Pat, what does OLAW have to say? May investigators use non-pharmaceutical-grade compounds in their research with animals?

Slide 14 (OLAW's Position)
>> Dr. Brown: Yes, OLAW agrees with USDA's Policy 3 that investigators must use pharmaceutical-grade substances for both investigational and clinical purposes in their PHS-funded studies with animals, unless there is a reason to use non-pharmaceutical-grade substances. The use of non-pharmaceutical-grade substances has been, and will continue to be, a necessary and acceptable component of biomedical research. Use of non-pharmaceutical-grade substances must be justified and that justification must be reviewed and approved by the IACUC.

>> Dr. Collins: Has OLAW's position changed?
>> Dr. Brown: Not really. OLAW provided guidance on the use of non-pharmaceutical-grade substances as early as 1993 in a letter authored by the director of the Division of Animal Welfare, OPRR [Office for Protection from Research Risks]. OPRR was the predecessor of OLAW. In 2003, OLAW published our policy on the use of non-pharmaceutical-grade chemical compounds in experimental animals in Lab Animal Magazine [*Lab Animal* 2003; 32(9):33-36.]; and, in 2006 as a Frequently Asked Question on the OLAW website. This Frequently Asked Question was updated in December 2011 and it is the guidance that is in effect today [FAQ F4].

Slide 15 (IACUC Responsibility)
This slide provides specifics about how the IACUC can meet its responsibilities in the oversight of the use of non-pharmaceutical drugs in research with animals. The IACUC may use a variety of administrative methods to review and approve the use of such agents. For example, the IACUC may establish acceptable scientific criteria within the institution, rather than on a case-by-case basis. It is important for investigators and IACUCs to consider safety, efficacy and availability of pharmaceutical-grade compounds when deciding what is best for the animals and the research. Cost savings alone are not an adequate justification for the use of non-pharmaceutical-grade or compounded drugs in animals. The same principles and need for professional judgment apply to non-survival studies.

Agents for sedation, analgesia or anesthesia agents should be veterinary or human

pharmaceutical-grade compounds, unless the use of an investigational chemical or formulation is scientifically necessary, appropriately justified and approved by the IACUC. The use of a non-pharmaceutical-grade euthanasia agent must meet the same standards.

Slide 16 (IACUC Evaluates Potential Adverse Consequences)
The IACUC is responsible for evaluating the potential adverse consequences of such agents when used in animals. In making its evaluation, the IACUC may consider factors including, for example, the grade, purity, sterility and acid-base balance, pyrogenicity, osmolality and stability, the site and route of administration, compatibility of components, side effects and adverse reactions, storage and pharmacokinetics. This is a very comprehensive list. Not all of these factors may be applicable in all circumstances and the IACUC may not need to request all of this information in every situation.

Slide 17 (PI Responsibilities)
This slide explains the investigator's responsibility to the IACUC. In the animal study proposal, the investigator should identify any drugs, biologics or reagents that will be administered to animals. If these agents are not human or veterinary pharmaceutical-grade substances, the investigator must provide a scientific justification for their use and describe the methods that will be used to ensure appropriate preparation and administration.

Slide 18 (Specific Guidance to IACUCs)
IACUCs have responsibilities to the investigators, too. The next four slides provide examples of situations in which it would be reasonable for the IACUC to review and approve the use of non-pharmaceutical-grade substances. We will provide a Word document of these examples in this webinar location on the OLAW website so that IACUCs may use these in developing their own criteria, if they wish.

Slide 19 (Example 1)
Example 1: When no equivalent veterinary or human drug is available for experimental use, then the highest-grade equivalent chemical reagent should be used and formulated aseptically, with a non-toxic vehicle, as appropriate for the route of administration.

Slide 20 (Example 2)
Example 2: Although an equivalent veterinary or human drug is available for experimental use, the chemical-grade reagent is required to replicate methods from previous studies because results are directly compared to those of replicated studies.

Slide 21 (Example 3)
Example 3: Although an equivalent veterinary or human drug is available, dilution or change in formulation is required.

- If adulteration by dilution, addition or other change in formulation is required, there may be no additional advantage to be gained by using the USP formulation. Note that here we are using the term adulteration as a technical term. Nothing negative is implied by the use of the word "adulteration".

- In this situation, use of the highest grade reagent may have the advantage of single-stage formulation and also result in purity that is equal to or higher than the human or veterinary drug.
- Professional judgment should be used to determine the appropriate test material and to ensure use of an agent with the least likelihood for causing adverse effects.

It is important to remember that veterinary and human drugs that are reconstituted according to the product insert remain pharmaceutical-grade drugs.

Slide 22 (Examples 4 and 5)
Example 4: The available human or veterinary drug is not concentrated enough to meet experimental requirements.

Example 5: The available human or veterinary drug does not meet the non-toxic vehicle requirements for the specified route of administration.

>> Dr. Collins: Do USDA and AAALAC concur or have any further thoughts on the examples where the IACUC may consider a non-pharmaceutical-grade compound acceptable in lieu of a veterinary or human product? Dr. Clarke?
>> Dr. Clarke: As per Animal Care Policy 3, a scientific justification is required.

>> Dr. Collins: Dr. Newcomer?
>> Dr. Newcomer: AAALAC would be in general agreement with the thoughtful and thorough discussion offered by Dr. Brown on this issue.

Slide 23 (Questions?)
>> Dr. Collins: Okay. Let's move on to questions. OLAW has received some very specific questions regarding the use of non-pharmaceutical-grade substances. We want to share guidance developed in response to these questions with the community. As I said in my opening comments, we were not able to include all questions in this presentation, but it is our hope that guidance from OLAW, USDA and AAALAC -- provided in this webinar -- will aid each IACUC in using a performance based approach to questions unique to their program.

>> Dr. Brown: I'd like to remind everyone that all of us are responsible for ensuring that animals used in research, teaching and testing are treated humanely. OLAW recognizes the challenge investigators face and we fully support the essential work that they do.
We were mindful of those challenges as we prepared answers to the questions that you are about to hear. Ultimately, it is up to the each IACUC, practicing local self-monitoring, to ensure that pain and distress are avoided or minimized, while at the same time supporting justified scientific research. It is our hope that this webinar will support that essential activity by providing examples of how our guidance may be applied at the local level.

Slide 24 (Questions)
>> Dr. Collins: How does compounding of drugs factor into this issue? Are investigators required to use a compounding pharmacy when it is necessary to use a specific mixture of

experimental drugs, chemicals or other formulations?
>> Dr. Brown: OLAW considers the compounding of investigational agents or the customized manipulation by dilution or addition of vehicles to pharmaceutical-grade substances for administration to animals as necessary and acceptable scientific activities carried out by researchers. However, these activities should be described in the animal study and reviewed and approved by the IACUC.

>> Dr. Collins: To carry this thread further, may investigators use a commercial compounding pharmacy to prepare specific mixtures of experimental drugs?
>> Dr. Brown: Yes. There may be circumstances where the customized manipulation of an FDA approved drug by a licensed pharmacist is needed to meet the needs of a research study. Examples include mixing two injectable drugs into a specialized formulation, preparing a paste from crushed tablets or adding flavoring to a drug. FDA states that for compounding by a pharmacy or veterinarian to be legal, it cannot be from bulk or raw active ingredients, although FDA does under specific circumstances allow this practice.

>> Dr. Collins: May investigators prepare the specific mixture themselves in the laboratory?
>> Dr. Brown: Yes. However, these activities should be described in the animal study and reviewed and approved by the IACUC.

Slide 25 (Question)
>> Dr. Collins: Do USDA and AAALAC concur or have any further thoughts on the issue of compounding? Dr. Clarke?
>> Dr. Clarke: The Animal Welfare Act and the Animal Welfare Regulations do not address that level of detail, however their wording provides for latitude of thought; hence, the system of protocol evaluation allows the IACUC and investigators to work together to determine an optimal way to conduct experiments within the context of humane animal care.

>> Dr. Collins: Dr. Newcomer?
>> Dr. Newcomer: AAALAC International understands that compounding pharmacies may be a useful resource to animal care and use programs seeking to acquire pharmaceutical-grade drugs in unique combinations or concentrations for therapeutic or research purposes. Institutions are expected to comply with the state and federal regulations applicable to compounding and would be wise to evaluate the compounding pharmacies under consideration to determine if they are quality providers.

Slide 26 (Question)
>> Dr. Collins: When it is necessary to add a vehicle or diluent to a chemical or substance that will be administered to an animal, is it required to use a pharmaceutical-grade material?
>> Dr. Brown: It depends on the route of administration and the need to maintain sterility. Our concern is that it doesn't injure the animals and is appropriate for the science. Professional judgment should be used in making this determination. For oral administration, the vehicle or diluent should be food grade. For injections such as IntraMuscular, IntraPeritoneal, or subcutaneous, the diluent or vehicle should be sterile and physiologic.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: The USDA concurs with OLAW's position.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: And AAALAC concurs with the answers offered by OLAW.

Slide 27
>> Dr. Collins: Our next question. Does the requirement distinguish between the use of non-pharmaceutical-grade substances for clinical, that is medical or veterinary, or research use? Dr. Newcomer?
>> Dr. Newcomer: In AAALAC's view, the general philosophy concerning the selection and use of compounds for clinical or therapeutic applications and for research applications in laboratory animals is the same. The *Guide* recognizes that pharmaceutical-grade compounds afford the subjects protection against toxic or unwanted side effects potentially minimizing important variables in scientific studies. Compounds used in veterinary and human clinical applications are routinely available in a pharmaceutical-grade and should be used whenever possible. However, scientific studies may require the use of compounds that are not available in pharmaceutical-grade, or that may only be available in a pharmaceutical-grade that is legitimately deemed unacceptable for particular scientific reasons. In these cases, with appropriate IACUC oversight, non-pharmaceutical-grade compounds may be acceptable when prepared and maintained using sound pharmaceutical practices.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: Policy 3 addresses the use of non-pharmaceutical-grade compounds only within the context of research and experimentation.

>> Dr. Collins: And Dr. Brown?
>> Dr. Brown: OLAW concurs with USDA and AAALAC.

>> Dr. Collins: I would like to confirm this point since it is one that is of great interest to the community. Am I correct in stating that USDA and OLAW apply equivalent guidance about the use of pharmaceutical-grade drugs to all compounds administered to research animals, not just to medicine that would be used to provide veterinary medical care for a research animal? Dr. Brown?
>> Dr. Brown: Yes, you are correct, Jerry.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: Yes, you are correct.

>> Dr. Collins: And Dr. Newcomer, any comment from AAALAC?
>> Dr. Newcomer: Yes. AAALAC would agree with that.

Slide 28 (Question)
>> Dr. Collins: Our next question: Does the requirement force us to use a more expensive substance that does not confer any additional benefits over a less expensive substance? Dr. Newcomer?
>> Dr. Newcomer: The question as stated does not reference any quality factors of the substance other than expense and AAALAC supports the prudent use of scientific resources. If both have acceptable efficacy, there is no need to spend any more money than is necessary.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: According to Policy 3, cost savings is not a justification for using non-pharmaceutical-grade compounds in regulated animals.

>> Dr. Collins: Dr. Brown?
>> Dr. Brown: OLAW concurs with USDA and AAALAC.

Slide 29 (Question)
>> Dr. Collins: Our next question: Is dilution of a drug, such as Ketamine with saline for use in the mouse considered compounding? Dr. Brown?
>> Dr. Brown: No, it is considered an adulteration of the original product, but one that is necessary to ensure that the appropriate dosage is administered. As long as proper sterile technique and a sterile diluent are used, there should be no issues of concern for the IACUC.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: The USDA concurs with OLAW's position.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: AAALAC also concurs with this answer.

Slide 30 (Question)
>> Dr. Collins: On to our next question. Can the IACUC approve the use of tribromoethanol, also known as Avertin? Dr. Brown?
>> Dr. Brown: Avertin is the trade name for the injectable anesthetic agent 2,2,2-tribromoethanol. Avertin was once manufactured as a pharmaceutical-grade drug. It is no longer available commercially. The preparation and use of tribromoethanol for anesthesia needs to be scientifically necessary, appropriately justified and approved by the IACUC, taking into consideration the side effects, stability, storage requirements and other considerations associated with the preparation of this agent. There are multiple reports in the literature of physiologic harm to animals including ileus, adhesions and mortality from the use of tribromoethanol. OLAW would advise IACUCs to critically evaluate the proposed use of tribromoethanol and the consideration of alternative methods that avoid or minimize discomfort, distress and pain. OLAW has recently learned of journals turning down studies for publication that described use of tribromoethanol.

Slide 31 (Avertin References)
For the benefit of the IACUCs and investigators, we will provide five references from the literature, including three published in 2005 that highlight some of the problems with use of tribromoethanol in the transcript of this presentation [at the end of this document].

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: The USDA concurs with OLAW's statement.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: Despite the well-documented pitfalls previously referenced by Dr. Brown on the use of tribromoethanol, AAALAC has recorded the safe, effective and acceptable use of this agent for short-term rodent anesthesia in many animal care and use programs. However, on this subject, along with a whole host of others, AAALAC encourages institutions to heed the following advice from the 8th Edition of the *Guide* and I quote: "The body of literature related to animal science and the use of animals is constantly evolving, requiring programs to remain current with the information and best practices." End quote.

Slide 32 (Question)
>> Dr. Collins: Our next question: Is it necessary to use USP or Grade A, that is medical CO2, to euthanize rodents? Dr. Brown?
>> Dr. Brown: Either USP Grade A (medical) or Grade B (industrial) carbon dioxide may be considered acceptable as they each provide a minimum purity for carbon dioxide of 99.0%. Carbon dioxide should be supplied in compressed gas in cylinders. The use of dry ice is unacceptable according to AVMA Euthanasia Guidelines (PDF).

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: The USDA concurs with this statement.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: And AAALAC also concurs with this opinion.

Slide 33 (Questions)
>> Dr. Collins: Our next question. Can commonly available euthanasia solutions like Euthasol or Fatal Plus be diluted and used as an anesthetic for survival surgery? Dr. Brown?
>> Dr. Brown: No. Typically these solutions are not sterile and contain drugs other than anesthetic agents that could harm or kill the animals even if diluted.

>> Dr. Collins: Dr. Clarke?
>> Dr. Clarke: The USDA concurs with OLAW's position.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: And AAALAC also concurs with this answer.

>> Dr. Collins: Can a euthanasia solution be used as an anesthetic in non-survival surgery? Dr. Brown?
>> Dr. Brown: A euthanasia solution may not be used as an anesthetic for survival or non-survival procedures. OLAW in concert with USDA agree that a procedure may be performed as a part of euthanasia. And this would be limited to terminal perfusion or exsanguination. In both cases, death is an immediate outcome of the procedure.

>> Dr. Collins: Dr. Clarke, any additional comments?
>> Dr. Clarke: No additional comments.

>> Dr. Collins: Dr. Newcomer?
>> Dr. Newcomer: AAALAC would also agree with this answer.

Slide 34 (Question)
>> Dr. Collins: Our next question: Is it okay to use non-sterile euthanasia solution for euthanasia? Dr. Clarke?
>> Dr. Clarke: Yes. Yes it is consistent with the adequate veterinary care under the Animal Welfare Act, Paragraph 2143, Subparagraph (a)(3)(A) and the Animal Welfare Regulations 9 CFR Chapter 1, Paragraph 2.33, Subparagraph (a), (b)(2) and (4) in which a humane death is achieved.

>> Dr. Collins: Dr. Brown?
>> Dr. Brown: OLAW would agree with USDA.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: AAALAC is also in agreement.

Slide 35 (Question)
>> Dr. Collins: Our next question: Can non-pharmaceutical-grade pentobarbital be used for euthanasia? It is of higher purity than the veterinary product. Dr. Clarke?
>> Dr. Clarke: It can be used if scientifically justified and satisfies the paragraphs from the Act and the Regulations previously cited.

>> Dr. Collins: Dr. Brown?
>> Dr. Brown: We agree with USDA that it can be used if scientifically justified and administered appropriately.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: AAALAC also agrees on this matter.

Slide 36 (Question)
>> Dr. Collins: Our next question: Does the OLAW non-pharmaceutical-grade substance policy apply to aquatic species? Dr. Brown?
>> Dr. Brown: Yes. The guidance is applicable to aquatic species because the composition of

the drug either -- both the purity, solubility and toxicity -- are just as relevant in the case of aquatic species as well as other animals. And special attention needs to be given to drug concentrations in the volume of water in which the animal is placed.

>> Dr. Collins: Dr. Newcomer, any comments on this question?
>> Dr. Newcomer: Using the *Guide* 8th Edition as a standard and recognizing that it covers aquatic vertebrates, AAALAC would also agree on this issue.

Slide 37 (Question)
>> Dr. Collins: And our final question: OLAW Frequently Asked Question F4 states: "...the IACUC may establish acceptable scientific criteria for use of these agents within the institution, rather than on a case-by-case basis." And these agents are the non-pharmaceutical-grade compounds. The question then is: Can you give an example of how this might be used?
Dr. Brown?
>> Dr. Brown: Well, this is similar to an IACUC approving a standard operating procedure for a surgical procedure, which then can be referenced by the PI in the protocol. So those approvals should be periodically reviewed by the IACUC at least every three years. If the IACUC bases its approval on a scientifically justified reason -- for example, non-availability or greater purity of product and has clear guidelines on issues like reconstitution, handling, storage -- then use of a non-pharmaceutical-grade compound may be considered for an institution-wide approval.

>> Dr. Collins: Dr. Clarke, any additional comments?
>> Dr. Clarke: No additional comments. We agree with this statement.

>> Dr. Collins: And Dr. Newcomer?
>> Dr. Newcomer: AAALAC would accept the approach voiced by Dr. Brown.

Slide 38 (olawdpe@od.nih.gov)
>> Dr. Collins: Okay. That's the end of our questions. I would certainly like to thank Dr. Clarke, Dr. Newcomer and Dr. Brown for taking their time and extending their effort in this webinar. If you have additional questions, as participants, please email them to olawdpe@od.nih.gov by March 30 of this year [2012]. OLAW, USDA and AAALAC will prepare answers that will be posted on the OLAW website with the transcript and recording of this webinar.

Slide 39 (Presenters)
This slide contains contact information for each of our presenters. And we'll pause for a moment so you can jot that information down. And I'm going to assume you all are very quick writers, so we'll go on to our last slide.

Slide 40 (OLAW Online Seminar Series Upcoming Schedule)
The upcoming seminar series schedule includes webinars in June, September and December. Topics will be announced at a later date. And as always, we encourage you to submit suggestions or requests for topics for those seminars.

Thanks to each of you, our participants, for taking time from what we know are busy schedules to tune in to the broadcast. Your ongoing efforts to ensure humane care of the animals entrusted to you continue to facilitate research, teaching and testing that is an essential part of improving human and animal health. Thank you and we hope you have a very pleasant spring.

Additional Submitted Questions Not Addressed During the Webinar

Introduction

On March 1, 2012, OLAW, USDA AC and AAALAC participated in the OLAW Online Seminar "**Use of Non-Pharmaceutical-Grade Chemicals and Other Substances in Research with Animals**". Below, we answer questions that were submitted during and after the webinar. We hope you will take away not only a specific answer, but also the logic applied to interpreting OLAW, USDA and AAALAC guidance so that you will be empowered to apply this reasoning to other questions that arise at your institution. For additional information please view the recorded webinar and read the supporting materials that can be found on the OLAW Education Resources webpage.

The following are important concepts for IACUCs and investigators considering the use of pharmaceutical- and non-pharmaceutical-grade substances in research with animals:

- Investigators must use pharmaceutical-grade substances in biomedical and behavioral research with animals when they are available. Non-pharmaceutical-grade substances may be used if justified by the researcher and approved by the IACUC.
- The use of non-pharmaceutical-grade substances has been, and will continue to be, a necessary and acceptable component of biomedical research.
- IACUCs, veterinarians, animal care personnel and investigators are responsible for ensuring that animals used in research, teaching, and testing are treated humanely.
- OLAW recognizes that investigators face many challenges and we fully support the essential work that they do. Ultimately, it is up to each IACUC, practicing local self-monitoring, to ensure that any pain and distress experienced by animals is avoided or minimized, while at the same time supporting justified scientific research.

IACUCs at PHS Assured institutions have the authority and the responsibility to make decisions about the research conducted at their institution. These decisions must be in compliance with the PHS Policy, as determined by OLAW. This document provides examples to help IACUCs understand how OLAW guidance may be applied at the local level.

Questions

1. Question. How does one determine whether a particular drug is available in a pharmaceutical-grade? Is there one place to determine what the source(s) might be?

Answer. You may determine what is available by consulting the FDA database. The Orange Book is the reference for FDA-approved human drugs. The Green Book is the reference for FDA-approved veterinary drugs.

2. Question. We seek guidance on the use of: diluted Fatal Plus for perfusions; chemical/analytical-grade pentobarbital; oral pharmaceutical-grade tamoxifen vs. chemical-grade tamoxifen mixed in oil; chemical-grade doxycycline; non-pharmaceutical-grade PMSG; and tribromoethanol.

Answer. This list does not provide the context necessary for us to answer as individual questions. However, we will comment on recent issues that have been raised about some items in the list.

- **Diluted Fatal-Plus for perfusions**: OLAW and USDA received inquiries as to whether investigators may use diluted Fatal-Plus as an anesthetic. The use of Fatal-Plus for anesthesia is specifically prohibited by FDA in the instructions included on the label of the product. The extra-label use of a euthanasia product for its pentobarbital content is unacceptable and violates the PHS Policy and Animal Welfare Act and Regulations. Such proposed use may not be approved by an IACUC or used by investigators at Assured institutions or used on regulated species. We note that some euthanasia procedures include perfusion of the animal prior to death. FDA approved euthanasia solutions may be used in those procedures in combination with the perfusion agent to perform perfusion and euthanasia as a single procedure.
- **Chemical /analytical-grade pentobarbital**: Agents for sedation, analgesia, or anesthesia should be veterinary or human pharmaceutical-grade compounds, when available, unless the use of a non-pharmaceutical chemical or formulation is scientifically necessary, appropriately justified and approved by the IACUC. The use of a non-pharmaceutical-grade euthanasia agent must meet the same standards. If no equivalent veterinary or human drug is available for experimental use, then the highest-grade equivalent chemical reagent should be used and formulated aseptically and with a non-toxic vehicle as appropriate for the route of administration. Recent exorbitant cost increases of pentobarbital have placed it logistically into the unavailable category. Pentobarbital from a reagent or analytical-grade powder, properly prepared by a pharmacist or other knowledgeable individual (e.g., chemist, veterinarian, researcher), with assurance of appropriate storage and handling, and approval by the IACUC is acceptable. IACUC approval can be institution-wide for the drug prepared in this fashion for all approved users.

- **Oral pharmaceutical-grade tamoxifen vs. chemical-grade tamoxifen mixed in oil; chemical-grade doxycycline; tribromoethanol**: We are unable to provide information because we don't have a context for the question. The IACUC has the duty and the responsibility to decide this issue based on the use being proposed and the needs of the science. During the webinar, Dr. Brown provided examples of situations in which it would be reasonable for the IACUC to review and approve the use of non-pharmaceutical-grade substances:

  1. If no equivalent veterinary or human drug is available for experimental use, then the highest-grade equivalent chemical reagent should be used and formulated aseptically and with a non-toxic vehicle as appropriate for the route of administration.

  2. Although an equivalent veterinary or human drug is available for experimental use, the chemical-grade reagent is required to replicate methods from previous studies because results are directly compared to those of replicated studies.

  3. Although an equivalent veterinary or human drug is available, dilution or change in formulation is required.

     - If adulteration by dilution, addition, or other change in formulation is required, there may be no additional advantage to be gained by using the USP formulation.
     - Use of the highest-grade reagent may have the advantage of single-stage formulation and also result in purity that is equal to or higher than the human or veterinary drug.
     - Professional judgment should be used to determine the appropriate test material and to ensure use of an agent with the least likelihood for causing adverse effects.

  4. The available human or veterinary drug is not concentrated enough to meet experimental requirements.

  5. The available human or veterinary drug does not meet the non-toxic vehicle requirements for the specified route of injection.

The IACUC may be guided by these examples in determining if the proposed use of non-pharmaceutical-grade tamoxifen, doxycycline, or tribromoethanol is appropriate to consider and approve. (E.g., example 3 may apply to the use of chemical-grade tamoxifen diluted in oil; use of non-pharmaceutical grade doxycycline when given parenterally may not be acceptable as this use does not meet the conditions of example 1; tribromoethanol, used as an anesthetic in the production of genetically altered mice, may not meet the requirement of number 2, as an anesthetic with fewer side effects may be equally effective.)

If the IACUC decides that use of a non-pharmaceutical-grade compound is indicated, they will want to consider potential adverse consequences that may result if the compound is

used in animals. In making its evaluation, the IACUC may consider factors including, for example, the grade, purity, sterility and acid-base balance, pyrogenicity, osmolality and stability, the site and route of administration, compatibility of components, side effects and adverse reactions, storage and pharmacokinetics. This is a very comprehensive list. Not all of these factors apply in all circumstances and the IACUC may use its own judgment to determine what, if any, of this information is useful and necessary for making a decision.

3a. Question. Dr. Brown stated that investigators must "identify any drugs, biologics, or reagents that will be administered to animals" in IACUC applications. Does this mean they must identify ALL substances being administered to animals? (E.g., eye ointment, fluids, heparin, antibiotics, atropine, mannitol, tattoo ink.)

3b. Question. My question does not apply to use of non-pharmaceutical-grade compounds for therapeutic purposes but rather for basic research. Do OLAW, USDA and AAALAC expect that every time an investigator proposes to use a[ny] compound for basic research in or on an animal (e.g., extract of plant root; biological material that has just been identified such as a gene, siRNA, or protein; a newly synthesized chemical compound), such a use must be justified to the IACUC?

3c. Question. The food animals eat, the bedding they sleep in and enrichment devices are not pharmaceutical-grade and animals ingest them. What does the wind blow in for outdoor-housed animals to ingest or inhale? Should the IACUC require justification for these not being pharmaceutical-grade too?

Answer. Any drugs, biologics, or reagents that are administered to animals as part of a study or experiment must be included on the animal study proposal and reviewed and approved by the IACUC. Drugs or substances used in clinical practice on the research animals must be pharmaceutical-grade, if possible, and do not have to be listed in the animal study proposal. It is up to the IACUC, practicing local self-monitoring, to ensure that pain and distress are avoided or minimized, while at the same time supporting justified scientific research. The system of protocol evaluation allows the IACUC and investigators to work together to determine an optimal way to conduct experiments within the context of humane animal care. OLAW and the *Guide* have repeatedly stressed that investigators and IACUCs are empowered to use professional judgment. Food, bedding and wind blown particulate matter are not drugs, biologics, or reagents and do not require scientific justification.

4. Question. We test non-pharmaceutical-grade chemicals formulated by scientists, sometimes using unusual vehicles (e.g., Cremophor, DMSO, DMF, PEG400) to get the compounds into solution. To what extent do you expect IACUC's to evaluate non-pharmaceutical drug use? Please clarify how an average IACUC is supposed to evaluate these situations. Will each IACUC require a medicinal chemist or pharmacist?

Answer. Vehicles administered to animals in biomedical research must be pharmaceutical-grade, if available. Use of non-pharmaceutical-grade vehicles must be justified and that justification must be reviewed and approved by the IACUC. The IACUC and investigator must consider the route of administration; products administered orally should be food-grade.

The current guidance on use of non-pharmaceutical-grade substances when they are available remains substantially the same as was written by the Office for Protection from Research Risks in 1993, published in Lab Animal Magazine in 2003, and added to the OLAW website as FAQ F4 in 2006. IACUCs are not required to include a medicinal chemist or pharmacist. IACUCs and investigators should use professional judgment, common sense, and standard references, as they have done in the past, in making a determination about the use of non-pharmaceutical-grade substances in biomedical research with animals.

5. Comment. You seem to be equating "pharmaceutical grade" with "commercially available" and placing "compounded drug products" as non-pharmaceutical. The USP has an extensive chapter on compounding of drugs (usually by licensed pharmacists) that provides the basis for the preparation of drug products for human patients. A more precise definition of terms would be helpful. 1. Do not use "drug" when you mean "drug product." 2. If you mean "commercially available" say so and do not use "pharmaceutical" and "non-pharmaceutical" because by definition in state pharmacy practice acts, any compounded product prepared via USP standards by a pharmacist is "pharmaceutical grade". Further your statement that “compounding with bulk materials is prohibited” is false.

Response. OLAW and USDA used the terms pharmaceutical-grade as early as 1993 and 1997 in guidance issued on this topic. These terms have also been adopted in the 8th Edition of the *Guide*. Dr. Clarke used the definition that was supplied by FDA Center for Veterinary Medicine (CVM). CVM is the regulatory authority on drugs given to animals and veterinary compounding. Regarding bulk compounding, FDA has published a list of bulk drug substitutes for compounding and subsequent use in animals to which FDA would exercise enforcement discretion and not ordinarily object.

6. Question. Please provide guidance on the acceptability or non-acceptability of tribromoethanol or Avertin in mice.

Answer. Avertin is the trade name for the injectable anesthetic 2,2,2-tribromoethanol. Avertin was once manufactured as a pharmaceutical-grade drug, but is no longer available. For compliant use of tribromoethanol, the preparation and use of this anesthetic must be scientifically necessary, appropriately justified and approved by the IACUC. In making its decision the IACUC must consider the side effects, stability, storage requirements and other considerations associated with the preparation of this agent. There are multiple reports in the literature of physiologic harm to animals including ileus,

adhesions and mortality from the use of tribromoethanol. OLAW advises IACUCs to critically evaluate the proposed use of tribromoethanol and consider alternative anesthetics that avoid or minimize discomfort, distress and pain. OLAW's webinar provided references that document problems with the use of tribromoethanol (References provided at the end of this document; or see Slide 31).

7. Question. During the webinar, Dr. Brown said, "OLAW has recently learned of journals turning down studies for publication that described use of tribromoethanol." Can you give more details about that?

Answer. Yes. An editorial published by *Cardiovascular Research* (2012) 93(1):1–3 includes the information that "6% of the total articles received in the past year for evaluation in *Cardiovascular Research* were rejected for ethical reasons. One of the most frequent causes of rejection on ethical grounds is the improper choice of anesthetic drugs for major surgical procedures." OLAW has been informed that the use of tribromoethanol was a factor in rejection of a study.

8. Question. Can the use of certain non-pharmaceutical-grade chemicals such as Avertin or Tricaine be approved by the IACUC for specific types of studies (blanket approval) or would it have to be on a case-by-case basis moving forward?

Answer. An institution-wide policy could be developed as a standard operating procedure (SOP). OLAW FAQ F4 states: "...the IACUC may establish acceptable scientific criteria for use of a non-pharmaceutical-grade substance within the institution, rather than on a protocol-by-protocol basis." This is similar to an IACUC approving a standard operating procedure for a surgical procedure, which then can be referenced by the PI in a protocol. Those approvals should be periodically reviewed by the IACUC (at least every three years).

An institution-wide policy for the use of a non-pharmaceutical-grade chemical must be based on scientific justification and must include clear guidelines on relevant issues such as reconstitution, handling and storage. The following are examples of scientific justifications that an IACUC might choose to consider and approve: (1) A substance is needed for an experiment but pharmaceutical-grade is not available; (2) A reagent-grade product of greater purity than the pharmaceutical-grade is needed for the experiment. See also transcript slide 37.

9. Question. Dr. Clarke agreed with OLAW on the 3-year review interval for standard operating procedures (SOPs). Is 3-year review of SOPs acceptable to the USDA for SOPs involving USDA-regulated species?

Answer. Dr. Clarke's statement was not accurate. In Lab Animal 2012; 41 (02), OLAW and USDA state, "Some IACUCs allow investigators to reference SOPs in their protocols rather than provide a written narrative of common animal use procedures. Such SOPs

should be reviewed by the IACUC at appropriate intervals for proposed activity review (at least once every three years according to PHS Policy or semi-annually, if they involve USDA-regulated species) to ensure they are up-to-date and accurate."

10. Question. Do you recommend that institutions have written guidelines to cover their policies regarding non-pharmaceutical-grade chemicals?

Answer. The IACUC is free to select suitable administrative methods for conducting official business as long as the methods are compliant with the PHS Policy and the *Guide*.

11. Question. Many of the drugs that we use do not say if they are pharmaceutical-grade. Many of these are ordered from Sigma. We cannot find a reliable method for determining if these are or are not pharmaceutical-grade. Can you tell us a place or way that we can find that information on MSDS, website?

Answer. OLAW provides the following definition (Position Statement 3): A pharmaceutical-grade compound is a drug, biologic, or reagent that is approved by the Food and Drug Administration (FDA) or for which a chemical purity standard has been established by the United States Pharmacopeia-National Formulary (USP-NF), or British Pharmacopeia. If the product does not meet these standards, it is not a pharmaceutical-grade product. Reagents are not drugs. Drugs are manufactured by a pharmaceutical producer under good manufacturing practices and approved by the FDA. OLAW recommends that the investigator contact the manufacturer, if the information cannot be found on a commercial website. Also, the Green Book (veterinary) and the Orange Book (human) available on the FDA website, provide a list of FDA-approved, pharmaceutical-grade drugs.

12. Question. Some investigators use chemicals from SIGMA for various compounds (e.g., tribromoethanol) for anesthetics that are not made in a pharmaceutical grade. However these chemical bottles have no expiration date labeled. They appear to have a date of manufacture only. I do not know how long these chemicals can be expected to be "in date". Do you have any guidance on this?

Answer. If you cannot find information that you need about a product on a commercial website, contact the manufacturer. It would be reasonable for the IACUC to ask the investigator to provide information about stability of a compound prepared in the laboratory. OLAW Position Statement 3 states: "The IACUC is responsible for evaluating the potential adverse consequences of such agents when used for research. In making its evaluation, the IACUC may consider factors including, for example: grade, purity, sterility, acid-base balance, pyrogenicity, osmolality, **stability**, site and route of administration, compatibility of components, side effects and adverse reactions, storage and pharmacokinetics."

13. Question. Are Sigma USP reagent offerings considered pharmaceutical-grade (such that no special justification is required on a protocol to use them in animals)?

Answer. No. Reagents are not pharmaceutical-grade. Use of reagents must be scientifically justified. The institution may develop an institution-wide policy on the use of reagents.

14. Question. Your comment that the IACUC ***may*** consider a number of factors (grade, purity, sterility, acid-base balance, pyrogenicity, osmolality, stability, site & route of administration, compatibility of components, side effects and adverse reactions, storage and pharmacokinetics) is clear from slide 16. I certainly appreciate your pointing out the use of the word "may" in this context. In my opinion, confusion might arise from the language in the 8th edition of the *Guide.* On page 31, the *Guide* states, "…[T]he use of a non-pharmaceutical-grade chemical or substance may be necessary to meet the scientific goals of a project or when a veterinary or human pharmaceutical-grade product is unavailable. In such instances, ***consideration should be given*** [my emphasis] to the grade, purity, sterility, pH, pyrogenicity, osmolality, stability, site and route of administration, formulation, compatibility and pharmacokinetics of the chemical or substance to be administered, as well as animal welfare and scientific issues relating to its use."

Answer. The IACUC is responsible for evaluating the potential adverse consequences of non-pharmaceutical-grade agents when used in animals. In making its evaluation, the IACUC **may** consider factors including, for example, the grade, purity, sterility and acid-base balance, pyrogenicity, osmolality and stability, the site and route of administration, compatibility of components, side effects and adverse reactions, storage and pharmacokinetics. This is a very comprehensive list. Not all of these factors may be applicable in all circumstances and the IACUC may not need to request all of this information in every situation.

15. Question. How do non-pharmaceutical grade requirements apply to diluents used in drug discovery? What is OLAW's stance on the use of pharmaceutical-grade substances infused within diets?

Answer. It depends on the route of administration and the need to maintain sterility. OLAW's concern is that the substance doesn't injure the animals and is appropriate for the science. Professional judgment should be used in making this determination. For oral administration, the vehicle or diluent should be food grade. For injections such as IntraMuscular, IntraPeritoneal, or subcutaneous, the diluent or vehicle should be sterile and physiologic.

16. Question. Is there guidance with drug use in animals with regard to the JCAHO requirement of the 28-day lifespan of a multi-use vial? Since our location is a medical center, we fall under JCAHO jurisdiction, but no exceptions have been made for animal

research.

Answer. The Joint Commission, formerly called the Joint Commission on the Accreditation of Healthcare Organizations (JCAHO) is an independent, not-for-profit organization that accredits and certifies health care organizations and programs in the United States. They do not accredit, certify, or oversee biomedical research with animals.

17. Question. Please clarify whether this guidance only includes medical/veterinary substances or if it also included substances that are used in the vehicle for test and control articles.

Answer. USDA and OLAW agree substances administered to research animal subjects must be pharmaceutical-grade unless scientifically justified. This applies to research, medical and veterinary substances and vehicles for test and control substances. For many test or novel agents, a pharmaceutical-grade may not exist so the IACUC needs to approve this use based on scientific justification. The PI should give as much information as possible about any adverse reactions of the proposed class of agents.

18. Comment. Dr. Brown's slide that states that the use of non-pharmaceutical-grade drugs may be justified for the purpose of replicating previous studies seems to be overly broad and provide a loophole for PIs who refuse to consider alternative (perhaps better) available drugs that can achieve the same or equivalent results. I wish that the importance of encouraging PIs to consider the use of ALTERNATIVE drugs that are equally effective or more effective and are available in pharmaceutical-grade form would be addressed in this discussion. That is, if a PI proposes the use of a specific compound to anesthetize an animal which is not available in pharmaceutical-grade form, but alternative anesthetics proven to be as effective without compromise to the research are available, the IACUC should give serious consideration to "requiring modifications" for the use of the pharmaceutical-grade equivalent.

Response. The IACUC has a responsibility to ensure that animals used in research, testing, and training are treated humanely and a responsibility to empower good science. The IACUC should discharge both of these responsibilities with honesty, fairness and respect for both the scientist and the animals. Ultimately, the IACUC must ensure that the investigator uses a pharmaceutical-grade compound when one is available that is as effective as the non-pharmaceutical-grade substance and fulfills the requirements of the research.

19. Question. By the definition of pharmaceutical-grade compounds, I would assume that a **test article** (not yet FDA approved) produced by a pharmaceutical company would not be in the Pharmacopeia and would therefore be considered non-pharmaceutical grade. Is it acceptable for the IACUC to accept a chemical compound to be of high quality when it comes from a pharmaceutical company or should they request documentation on purity and sterility?

Answer. The IACUC is responsible for evaluating the potential adverse consequences of such agents using the information available from the provider and should make the decision on a case-by-case basis. For many test or novel agents, a pharmaceutical-grade may not exist so the IACUC needs to approve this use based on scientific justification. The PI should give as much information as possible about any adverse reactions of the proposed class of agents.

20. Question. If I generate antibodies from a rabbit, will I be able to use them in my studies?

Answer. This decision will be up to the IACUC. The proposed study should be reviewed by the IACUC in the same way that any proposed study would be. There is no overriding prohibition against using antibodies that were generated in an animal in such studies. However, antibodies should be made *in vitro*, rather than *in vivo*, if possible.

21. Question. Do we need to use pharmaceutical-grade sterile saline as a diluent for anesthetics or other drugs? Some drugs require sterile water or other chemicals to make up the diluents. Do all solutions/chemicals put in the diluents need to be of pharmaceutical-grade?

Answer. OLAW considers the compounding of investigational agents or the customized manipulation by dilution or addition of vehicles to pharmaceutical-grade substances for administration to animals as necessary and acceptable scientific activities carried out by researchers in their laboratory. However, these activities should be described in the animal study and reviewed and approved by the IACUC using the same criteria as previously discussed in the webinar and Question 15. The diluent requirement is the same as the primary agent, that is, pharmaceutical-grade and sterile unless an alternative is needed based on scientific justification and approval by IACUC.

22. Question. Dr. Clarke responded that “the USDA concurs with OLAW position” on the mouse/ketamine anesthesia question. In other panel discussions to which I have listened, the USDA panelists have recused themselves on rodent questions.

Answer. Dr. Clarke was concurring on the general concept of drug dilution as an adulteration of the original product, but one that is necessary to ensure that the appropriate dosage is administered. This would apply to dilution of a drug for use in a similar sized regulated species (e.g., hamster). Additionally, USDA has jurisdiction over rodents other than the genus *Mus* and *Rattus*. Again, as long as proper sterile technique and sterile diluent are used, there should be no issues of concern for the IACUC.

23a. Question. Many researchers use silastic tubing packed with hormone and sealed with medical adhesive for surgical implantation in chronic administration studies. One touted benefit of this type of implant is that crystalline products as opposed to liquid can

then be utilized. Alternatives such as implantable osmotic pumps charged with liquids often cannot accomplish the same duration of administration thus resulting in the need for additional implantation surgeries. Is there a source for pharmaceutical-grade crystalline hormones? An additional limitation to the use of silastic tubing is the difficulty of sterilizing the implants. Researchers are reluctant to use gas sterilization when they do not know for sure how the process might alter the hormone being implanted and studies on this subject are lacking.

23b. Question. When it is necessary to administer a reagent in a high salt content vehicle, which would be the preferred delivery route: intraperitoneal or intravenous? What is the acceptable salt content of a vehicle?

Answer. The Health Research Extension Act of 1985 is the statute that confers the authority to oversee PHS-supported animal activities to NIH. The Congressional Committee report that accompanied the law stated, "It is far preferable to place primary responsibility for assuring compliance with NIH guidelines on committees within institutions rather than relying on intrusive Federal inspections." OLAW interprets this to mean that Congress intended for IACUCs to have the authority and the responsibility to make meaningful decisions about biomedical research conducted with animal models. The PHS Policy (IV.C.1.a.-g.) restates the concept. "In order to approve proposed research projects…the IACUC shall conduct a review of those components related to the care and use of animals and determine that the proposed research projects are in accordance with this [PHS] Policy. Questions 23a and 23b are examples of questions that the IACUC has the responsibility and the authority to decide.

24. Comment. I am writing to update OLAW on recent developments in the pentobarbital problem. Currently there is only one company producing Nembutal. This is licensed for human use but the distributor will sell it for animal use. Their current price is $1113 per 50 ml bottle. Not too many years ago, the cost was less than $50, then it increased to $300, then to $800 just a few months ago, now over $1000. The cost may continue to escalate as the company that acquired rights to manufacture it has a monopoly. Over the last year as prices increased and availability declined, researchers who were able converted to alternative anesthetics. A number of researchers are simply unable to switch due to scientific reasons. For them to pay these extreme prices would be a significant financial burden that may impact their research objectives.

Response. Regulatory guidance on this matter specifically allows for use of non-pharmaceutical-grade compounds due to non-availability and with IACUC approval. The exorbitant cost of this product has placed it logistically into the unavailable category. Pentobarbital from a reagent or analytical-grade powder, properly prepared by your pharmacist or other knowledgeable individual (e.g., chemist, veterinarian, researcher), with assurance of appropriate storage and handling, and approval by the IACUC is acceptable. IACUC approval can be institution-wide for the drug prepared in this fashion for all approved users.

25. Question. Are “pharmaceutical secondary standards” manufactured by chemical companies considered pharmaceutical-grade?

Answer. FDA: If the secondary standard material is obtained from a reputable source and complies with compendia standards, this material should be fine for use in clinical studies.

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