



DEPARTMENT OF HEALTH & HUMAN SERVICES

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Office of Laboratory Animal Welfare
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Home Page: <http://grants.nih.gov/grants/olaw/olaw.htm>

FOR EXPRESS MAIL:

Office of Laboratory Animal Welfare
6700B Rockledge Drive, Suite 2500
Bethesda, Maryland 20817
Telephone: (301) 496-7163
Facsimile: (301) 402-7065

May 28, 2019

Re: Animal Welfare Assurance
A3629-01 [OLAW Case J]

Dr. Joseph Heppert
Vice President for Research and Innovation
TTU Institutional Official
Texas Tech University
Box 41075
Lubbock, Texas 79409

Dear Dr. Heppert,


The Office of Laboratory Animal Welfare (OLAW) acknowledges receipt of your initial September 10, 2018 email and your follow-up May 15, 2019 email reporting an incident of noncompliance with the PHS Policy on Humane Care and Use of Laboratory Animals at Texas Tech University. According to the information provided, OLAW understands that your institute identified issues with a survival surgery protocol involving mice. The issues, among others, included a lack of appropriate aseptic technique, inappropriate withholding of prophylactic analgesia, unapproved euthanasia techniques, use of behavioral studies not approved on the protocol, and use of procedures approved on another protocol on the animal population associated with a different protocol. It is understood that the non-compliant activity was not funded by the PHS.

The corrective actions consisted of the Attending Veterinarian suspending the protocol and the IACUC subsequently voting to suspend all of the PI's protocols and place all of the animals on a holding protocol. It is understood from your May 15 email that the PI and laboratory have completed the retraining plan developed by the Attending Veterinarian in coordination with the IACUC. The IACUC has also voted to reinstate the protocol and to lift the probation. The issue has been resolved to the satisfaction of the IACUC.

OLAW believes that the corrective and preventive measures put in place by Texas Tech University are consistent with the provisions of the PHS Policy on Humane Care and Use of Laboratory Animals. We appreciate being informed of this matter and find no cause for further action by this office.

Sincerely,

(b) (6)

 Brent C. Morse, DVM
Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare

cc: IACUC Chair

Morse, Brent (NIH/OD) [E]

From: OLAW Division of Compliance Oversight (NIH/OD)
Sent: Wednesday, May 15, 2019 4:02 PM
To: Heppert, Joseph; OLAW Division of Compliance Oversight (NIH/OD)
Cc: Brooks, Tiffanie; (b) (6)
Subject: RE: Voluntary report of non-compliance

Thank you for this information Dr. Heppert. We will send an official response soon.

Best regards, Brent Morse

Brent C. Morse, DVM, DACLAM
Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health

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From: Heppert, Joseph [mailto:Joseph.Heppert@ttu.edu]
Sent: Wednesday, May 15, 2019 12:53 PM
To: OLAW Division of Compliance Oversight (NIH/OD) <olawdco@od.nih.gov>
Cc: Brooks, Tiffanie <Tiffanie.Brooks@ttu.edu>; (b) (6)
Subject: Re: Voluntary report of non-compliance

Brent,

Thank you for the follow up on this notification.

(b) (6) laboratory went through a substantial period of training. Following this period her protocol was reinstated, though her laboratory remained on probation and under heightened scrutiny. Over the probationary period, (b) (6) lab was visited by post-approval monitoring 18 times. Just yesterday, our IACUC voted to lift the probation effective May 19th 2019.

Consequently, I can report that this issue has been resolved to the satisfaction of our institutional Committee.

Once again, thank you for following up on this notification.

Sincerely,

Joseph A. Heppert
TTU Institutional Official

Joseph A. Heppert, Ph.D.
Vice President for Research and Innovation
Professor of Chemistry

Texas Tech University | Office of Research and Innovation
Box 41075 | (b) (4) Administration Bldg. |
Lubbock, TX 79409-1075
O: (b) (6) | joseph.heppert@ttu.edu

From: "OLAW Division of Compliance Oversight (NIH/OD)" <olawdco@od.nih.gov>
Date: Tuesday, May 14, 2019 at 1:16 PM
To: "Heppert, Joseph" <Joseph.Heppert@ttu.edu>, "OLAW Division of Compliance Oversight (NIH/OD)" <olawdco@od.nih.gov>
Cc: "Brooks, Tiffanie" <Tiffanie.Brooks@ttu.edu>
Subject: RE: Voluntary report of non-compliance

Hello Dr. Heppert,

Has this issue been resolved? We have no record of receiving a final report. Is one available? Your assistance is appreciated.

Sincerely, Brent Morse

Thank you for providing this preliminary report Dr. Heppert. We will open a case file and await further information as you have noted.

Best regards, Brent Morse

Brent C. Morse, DVM, DACLAM
Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health

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From: Heppert, Joseph [<mailto:Joseph.Heppert@ttu.edu>]
Sent: Monday, September 10, 2018 4:33 PM
To: OLAW Division of Compliance Oversight (NIH/OD) <olawdco@od.nih.gov>; accredit@AAALAC.org
Cc: (b) (6) Brooks, Tiffanie <Tiffanie.Brooks@ttu.edu>; Smith, Phil <Phil.Smith@ttu.edu>; (b) (6)
Subject: Voluntary report of non-compliance

Dr. Brent Morse, DVM, DACLAM
Acting Director, Division of Compliance Oversight
OLAW
olawdco@mail.nih.gov

Dr. James R. Swearengen, D.V.M.

Global Director
AAALAC
accredit@AAALAC.org

Dear Dr. Morse and Dr. Swearingen:

In my role as Texas Tech University Institutional Official for Animal Care and Use, I am writing to submit a voluntary report on an instance of non-compliance with our institutional procedures for Animal Care and Use. This situation involves a study using a non-USDA covered species not currently supported by Federal funding in a laboratory not yet supported by Federal funding. However, due to the range and seriousness of the procedural violations identified in this laboratory, our IACUC has recommended and I have concurred that we make this voluntary disclosure. An attachment to this email also outlines the remedies currently underway to retrain the research group and reestablish their animal protocols.

On August 1 2018, an inspection by our (b) (6) our post-approval monitoring officer, identified a number of deficiencies in the animal program of (b) (6) one of our junior faculty in the Department of Biological Sciences. The violations on the survival surgery protocol involving mice, outlined in the attached incident report, included (among other issues) lack of appropriate aseptic surgical technique, inappropriate withholding of prophylactic analgesia, unapproved euthanasia techniques, use of behavioral studies not explicitly approved in the protocol, and use of procedures approved in another animal use protocol on the animal population associated with a different protocol. After identifying the violations, Dr. Tiffanie Brooks, TTU Attending Veterinarian immediately suspended the specific protocol in question. The IACUC subsequently voted to suspend all of (b) (6) protocols and place all of her animals on a holding protocol pending the completion of a corrective course of action, and, subsequent to completion of the plan, to place the laboratory on a 6-month probation.

I have received a final plan for retraining the laboratory, a copy of which is attached to this email. Dr. Brooks developed this plan in coordination with the IACUC, and the investigator has agreed to the retraining plan. The laboratory and investigator have already begun the retraining process.

Please feel free to contact me with any questions or comments on this notification. I will keep you informed as situation is resolved.

Sincerely,

Joseph A. Heppert
TTU Institutional Official for Animal Care and Use

Joseph A. Heppert, Ph.D.
Vice President for Research
Professor of Chemistry
Texas Tech University | Office of the Vice President for Research
Box 41075 | (b) (4) Administration Bldg. |
Lubbock, TX 79409-1075
O: (b) (6) | joseph.heppert@ttu.edu

Continuation of Summary and Plan for reinstatement of protocols for (b) (6)

Re: Subcommittee #2 following Suspension of Protocol 17006-01, 18025-02, 17035-04

On Thursday, August 23, 2018, the IACUC voted unanimously to suspend all three of (b) (6) (b) (6) protocols after learning of multiple non-compliance violations. The violations ranged from significant non-compliance to multiple minor non-compliances. The protocols suspended included:

17006-01 41 mice

18025-02 65 mice

17035-04 12 mice

A second subcommittee was appointed and included, (b) (6) (b) (6) and Dr. Tiffanie Brooks. The subcommittee was charged with determining whether there were additional non-compliances with other protocols and formulate a plan for reinstatement of protocols and thus research activity. The subcommittee convened on 8/24/18, 8/27/18 and 8/29/18. The meetings included the subcommittee alone, the subcommittee with (b) (6) and the subcommittee with (b) (6) s graduate students. It was determined that there were no additional non-compliances associated with 18025-02 and 17035-04.

In addition to the corrective actions imposed by the first subcommittee and the proposed plan formulated by Dr. Brooks, the 2nd committee has determined additional sanctions.

Initial plan from Dr. Brooks (Re-training)

- Resubmit new AUF with updated changes and additions to project. Terminate 17006.
- Formulate and submit for approval SOPs for behavior tests
- Repeat TTU Generic training
- Complete CITI training modules
- Retraining on aseptic technique

Subcommittee #1

6-month probationary period

Consist of announced and unannounced PAMs from the IACUC committee and the ACS staff members. We suggest that each month during the probationary period, one member of the IACUC committee perform a scheduled PAM with the investigator. During this time, ACS staff members will perform random and unannounced PAMs as deemed appropriate. All PAMs will be documented and reported at the monthly IACUC meetings.

Subcommittee #2

PI-written action plan- this plan should include training mechanisms for both new and current students; weekly, monthly and annual methods to monitor compliance within the lab group. The PI has 14 days to respond to the IACUC with this action plan. As per Policy 09, (b) (6) must attend the IACUC meeting on September 19, 2018 for consideration of reinstatement.

Relinquishment of husbandry for all protocols- ACS will take over husbandry indefinitely with the expectation of communication of needs from the PI.

Relinquishment of breeding for all protocols- ACS will take over breeding colony management in consultation with the PI. The PI will incur costs for technician time. At the end of the 6-month probationary period, this action may be re-evaluated by the IACUC.

The subcommittee feels strongly that this plan is vital for the success of the investigator and the University. The PI must adhere to approved protocols and must not deviate in the slightest. Future non-compliances could lead to revocation of animal privileges at Texas Tech University.

Thank you,
Subcommittee #2

Summary of IACUC Subcommittee meeting with (b) (6)

Re: suspension of Protocol #17006-01

As the result of two recent Post Approval Monitoring (PAM) visits (7/31/2018 & 8/1/2018) by (b) (6) of the (b) (6) lab neuroscience research using mouse models, a number of non-compliances were noted (see below). These varied in severity, however if an AAALAC inspection team was visiting Texas Tech at least two of these could result in mandatory changes in the activity in order to maintain university accreditation and several others that would result in Suggestions for Improvement (SFI).

While mandated by federal regulations, PAM visits normally do not result in suspension of a protocol. Typically, the person doing the PAM would note any deviations from the protocol and make oral or written suggestions to the head of the lab and/or students that would bring the procedure back into compliance. It was the number and relative severity of the non-compliances that lead to the suspension of protocol 17006-01 by the Attending Veterinarian.

(b) (6) was understandably very concerned and somewhat distraught over this matter. A critical phone call from the University Attending Veterinarian, Dr. Tiffanie Brooks, to the Chair of Biological Sciences, (b) (6) provided the details of the issues that the PAM discovered, and explained why the protocol was suspended. Then (b) (6) met with (b) (6) to discuss the matter. Between that meeting and one with the designated IACUC subcommittee (see below) any misunderstandings about the requirements for what procedures can be performed have been clarified and any additional training that persons listed on this protocol do need has been identified and it appears to have set (b) (6) on the path to resolving the non-compliances.

Below are the non-compliances discovered from earlier in 2018 and from the PAMs on either 7/31 or 8/1:

Current Non-compliances on protocol 17006-01

Inappropriate breeding (CD1 male to C57BL/6 Female) during intruder test 5/26/18

- Did not self-report the accidental breeding nor euthanize pups immediately.
- Continued to use unapproved mice in experiments.

-Submitted Adverse Event Report per IACUC recommendation 7/18/2018; on IACUC Agenda for August 23, 2018.

PAM 7/31/2018 and 8/1/2018

Procedures performed on 17006-01 which are approved on protocol 18025-02:

- Predator Scent Test
- Social Interaction Test with known and unknown mouse
- 3 Chamber box

Procedures performed that we cannot find approved on any of (b) (6) protocols:

- T maze with a known and unknown mouse (T maze is an alternative to the approved Plus maze, but no mention of mice used as the unknown in the plus maze)
- Novel Object Test
- rectal temperatures-device being used is also not appropriate for rectal temps in mice

Unapproved euthanasia performed: Perfusion of mice

- Protocol states animals should be deeply anesthetized and quickly decapitated
- May need to investigate if this technique is appropriate with the bell jar method of anesthesia or if PI should investigate purchasing an anesthesia machine with nose cone for her lab.
- Dr. Brooks emailed (b) (6) 7/31/2018 about this procedure to see if she knew if it were approved in the protocol and that we were just not finding. After discussion, (b) (6) conceded that this procedure was in fact performed on a live mouse. Dr. Brooks verbally informed (b) (6) she was not to perfuse any more mice.
- (b) (6) was advised to submit an amendment for perfusion and other behavior tests for 17006-01, an SOP for all her behavior tests that will be easy to reference in protocols to help with redundancy of tests. She submitted these two amendments that same evening, 7/31/2018. Dr. Smith, IACUC Chair, called the amendment to Full Committee Review (7/23/2018) due to the breeding issue already on the upcoming IACUC meeting agenda. (b) (6) sent an email requesting the behavior tests not wait until the IACUC meeting due to ongoing studies because they are already approved tests.

Stereotaxic Surgery

-Conversations with one of (b) (6)'s graduate students indicated that all pain meds had previously been given post-operatively. He was subsequently informed that they had to be given pre-emptively and that this was also listed in the protocol as pre-emptive. The mouse undergoing surgery during the PAM was given Buprenex pre-operatively

-It is clear that Banamine was not post-operatively administered as stated in protocol. – During the subcommittee meeting the investigator stated that she did not believe banamine was necessary and buprenex alone was sufficient

-Not shaving the incision site. This is necessary for aseptic technique and was clearly stated in the protocol.

-Not properly scrubbing (one scrub with betadine) as indicated in protocol: alternating betadine and alcohol 3x.

-Instruments were inappropriately sterilized – students were using alcohol which is not a sterilant. Instruments as well as the drill, syringe, and marking pen need to be properly sterilized according to AAALAC.

AAALAC Position Statement: "AAALAC's Council on Accreditation stated that the use of alcohol as a skin disinfectant for rodent survival surgery was acceptable, but that the blanket use of alcohol for surgical instrument preparation was not acceptable. The Guide for the Care and Use of Laboratory Animals (NRC 2011) upholds the position that, "Alcohol is neither a sterilant nor a high-level disinfectant.""

-No supplemental heat during surgery which is also stated in the protocol.

-No drape to maintain a sterile field.

-Not using sterile gloves but exam gloves.

-Not maintaining "sterility" when touching stereotaxic and anesthesia equipment.

-Used vet bond to close the incision not suture as stated in protocol. Vet bond glue is appropriate but was not included in the protocol. The grad student said they use sutures for the viral vectors but not for the short term acute stress test.

-No surgical records were found for any previous surgeries. Graduate student did fill out a surgical card after surgery during which the PAM took place. He was told surgical records could be kept in lab but needed to be available to the IACUC and AAALAC. The only records kept of the surgery were the stereotaxic atlas points, not any drugs given or monitoring the mouse.

- The lack of aseptic techniques has been previously addressed by the attending veterinarian, the ACS post approval monitor and the clinical veterinarian in a PAM performed on 5/22/17. The attending veterinarian and the post approval monitor also reached out to help the investigator determine suitable and appropriate aseptic practices for these types of surgical procedures and even emailed the investigator aseptic techniques videos to review- the investigator was unresponsive to all communication and attempts to help.

Potentially injecting with non-approved drugs. Graduate student said he was injecting with exercise hormone.

-Protocol states viral vectors will be delivered into the brain. – During the meeting the investigator stated that for the acute stress test they injected a protein from the muscle into the brain. This protein is not mentioned in their protocol. The investigator also mentioned that this particular protein is involved in thermogenesis which is why they chose to monitor the mouse's core body temperature (rectal temps)

-The hazardous agent section lists attenuated virus and recombinant DNA to be delivered through intracerebral injection.

-No non-pharmaceutical grade chemicals are listed.

Mice recovering from surgery then underwent the acute stress study approximately 45 min later.

-Protocol states 1-2 weeks after surgery is when animals would undergo electrophysiology or behavior experiments. Not immediately post-op.

-Amendment approved 7/10/2018 has the acute stress study with 3h restraint then memory tasks and euthanasia but does not say would be in mice immediately after surgery.

An IACUC subcommittee made up of (b) (6) was convened and met on Friday, August 3 on then had a meeting with (b) (6) on August 6, 2018.

The subcommittee was happy to find that (b) (6) was already in the process of correcting many of the minor non-compliances. It was also clear that she recognized that the responsibility for training her student researchers was hers; she also acknowledged that as of the days of the PAM the students were not trained adequately. She stated to the subcommittee that 'several lab meetings' would be necessary to clarify what needed to be done and to emphasize the importance of following approved procedures to everyone. We believe that in some cases (b) (6) did not realize that she was not in compliance for example, if a procedure was approved on one protocol, that it was not also approved on a similar, but different protocol. In addition, it is going to be incumbent upon her to write up and submit several SOPs (or LOPs) that she can refer to rather than having to rewrite every procedure each time she submits a protocol.

The subcommittee encouraged (b) (6) to be more proactive in the future and that she should talk to ACS personnel if she has questions about doing a particular procedure or something else concerning animal welfare. In addition, if unexpected outcomes or events do occur (e.g., the inadvertent interbreeding of the two strains of mice she currently has which has produced a "brown strain") she should contact ACS as soon as it is discovered. We also suggested that writing pilot study protocols could avoid long, wordy protocols that might have to be repeatedly revised. Finally (and this point was emphasized by (b) (6) when you get to the end of the procedures that are approved on your protocol, you must stop right there until a future protocol/amendment is approved....

Below we repeat the aforementioned list of non-compliances and indicate in red what (b) (6) has done to resolve each issue to date:

Current Non-compliances on protocol 17006-01

Inappropriate breeding (CD1 male to C57BL/6 Female) during intruder test 5/26/18

-Did not self-report the accidental breeding nor euthanize pups immediately.

-Continued to use unapproved mice in experiments.

-Submitted Adverse Event Report per IACUC recommendation 7/18/2018; on IACUC Agenda for August 23, 2018.

PAM 7/31/2018 and 8/1/2018

Procedures performed on 17006-01 which are approved on protocol 18025-02:

-Predator Scent Test

-Social Interaction Test with known and unknown mouse

-3 Chamber box

(b) (6) has submitted an amendment on July 31 that addresses this issue; it will require some additional modification

Procedures performed that we cannot find approved on any of (b) (6) protocols:

-T maze with a known and unknown mouse (T maze is an alternative to the approved Plus maze, but no mention of mice used as the unknown in the plus maze)

-Novel Object Test

- rectal temperatures-device being used is also not appropriate for rectal temps in mice

The investigator is working to develop an SOP for her behavior testing to be referenced in current and future protocols.

Unapproved euthanasia performed: Perfusion of mice

-Protocol states deeply anesthetized and quickly decapitated.

-May need to investigate if this technique is appropriate with the bell jar method of anesthesia or if PI should investigate purchasing an anesthesia machine with nose cone for her lab.

-Dr. Brooks emailed (b) (6) 7/31/2018 about this procedure to see if she knew if it were approved in the protocol and that we were just not finding. After discussion, (b) (6) conceded that this procedure was in fact performed on a live mouse. Dr. Brooks verbally informed (b) (6) she was not to perfuse any more mice.

-(b) (6) was advised to submit an amendment for perfusion and other behavior tests for 17006-01, an SOP for all her behavior tests that will be easy to reference in protocols to help with redundancy of tests. She submitted these two amendments that same evening, 7/31/2018. Dr. Smith, IACUC Chair, called the amendment to Full Committee Review (7/23/2018) due to the breeding issue already on the upcoming IACUC meeting agenda. (b) (6) sent an email requesting the behavior tests not wait

until the IACUC meeting due to ongoing studies because they are already approved tests.

(b) (6) has submitted an amendment that allows perfusion, which is to be addressed in full committee by the IACUC on 8/23/2018.

(b) (6) second amendment adding behavior tests went out to the committee for review. It does not include all the procedures she was doing off-protocol, so those will need to be added during the amendment review process.

Stereotaxic Surgery

-Conversations with one of (b) (6)'s graduate students indicated that all pain meds had previously been given post-operatively. He was subsequently informed that they had to be given pre-emptively and that this was also listed in the protocol as pre-emptive. The mouse undergoing surgery during the PAM was given Buprenex pre-operatively.

-It is clear that Banamine was not post-operatively administered as stated in protocol.

(b) (6) has instructed her students to give Buprenex pre-emptively and Banamine post-operatively per the protocol.

ACS suggests using the Buprenorphine SR LAB for slow pain release. This may negate the need for the post-operative Banamine injection but will be discussed with Dr. Brooks.

-Not shaving the incision site. This is necessary for aseptic technique and was clearly stated in the protocol.

(b) (6) has purchased a shaver that will allow these surgeries to be performed in accordance with Guide instructions

-Not properly scrubbing (one scrub with betadine) as indicated in protocol: alternating betadine and alcohol 3x.

(b) (6) has instructed her students to follow the directions as listed above

-Instruments were inappropriately sterilized – students were using alcohol which is not a sterilant. This was used to clean the drill, syringe, and pen along with the instruments. All of these need to be properly sterilized according to AAALAC.

AAALAC Position Statement: "AAALAC's Council on Accreditation stated that the use of alcohol as a skin disinfectant for rodent survival surgery was acceptable, but that the blanket use of alcohol for surgical instrument preparation was not acceptable. The Guide for the Care and Use of Laboratory Animals (NRC 2011) upholds the

position that, "Alcohol is neither a sterilant nor a high-level disinfectant."

(b) (6) has purchased a glass bead sterilizer that will allow all instruments to be sterilized at the room where surgeries are being performed; all instruments used in surgeries should be autoclaved initially, with the glass bead sterilizer used in between surgeries performed on the same day

-No supplemental heat during surgery, which is required and alsostated in the protocol.

(b) (6) has purchased a heating pad that will be placed under each mouse undergoing surgery to insure warmth

-No drape to maintain a sterile field.

They will use a sterile drape to help maintain the sterile field

-Not using sterile gloves but exam gloves.

(b) (6) has purchased sterile gloves for the surgery, while exam gloves will be used to move the animals from the cage to the site of surgery

-Not maintaining "sterility" when touching stereotaxic and anesthesia equipment.

The above comment/remedy addresses this matter- the subcommittee did suggest possibly having two people available during the surgical procedures (one to prep the mice and maintain anesthesia, etc. (do the dirty work) and one to perform the surgeries maintained within the sterile field. This will help prevent the surgeon from needing to break the sterile field.

(b) (6) has previously been given ideas to help maintain a sterile field when touching the necessary equipment. ACS would be happy to share those ideas again.

-Used vet bond to close the incision not suture as stated in protocol. Vet bond glue is appropriate but was not included in the protocol. The graduate student said they use sutures for the viral vectors but not for the short-term acute stress test.

There will have to be an amendment submitted if (b) (6)'s group plans to use vet bond glue. We believe that is faster than sutures and may be appropriate for some surgeries

-No surgical records were found for any previous surgeries. Graduate student did fill out a surgical card after surgery during which the PAM took place. He was told surgical records could be kept in lab but needed to be available to the IACUC and AAALAC. The only records kept of the surgery were the stereotaxic atlas points, not any drugs given or monitoring the mouse.

-- The lack of aseptic techniques has been previously addressed by the attending veterinarian, the ACS post approval monitor and the clinical veterinarian in a PAM performed on 4/26/17. The attending veterinarian and the post approval monitor also reached out to help the investigator determine suitable and appropriate aseptic practices for these types of surgical procedures and even emailed the investigator aseptic techniques videos to review- the investigator was unresponsive to all communication and attempts to help.

(b) (6) told us that they do keep surgical records in their research lab and that they can have them available in the facility for ACS, IACUC and AAALAC to view. She also mentioned that she would begin filling out the teal rodent surgery cards and that her students will utilize the green experimental treatment cards as well.

Potentially injecting with non-approved drugs. Graduate student said he was injecting with exercise hormone.

-Protocol states viral vectors will be delivered into the brain.

(b) (6) will submit an amendment that requests that certain proteins (we think muscle) to be injected into brain for the acute stress procedure

-The hazardous agent section lists attenuated virus and recombinant DNA to be delivered through intracerebral injection.

This is apparently being done for some of the mice

-No non-pharmaceutical grade chemicals are listed.

We feel that the muscle protein that is injected will fall under this guideline.

Mice recovering from surgery then underwent the acute stress study approximately 45 min later.

-Protocol states 1-2 weeks after surgery is when animals would undergo electrophysiology or behavior experiments. Not immediately post-op.

-Amendment approved 7/10/2018 has the acute stress study with 3h restraint then memory tasks and euthanasia but does not say would be in mice immediately after surgery.

(b) (6) will have to submit an amendment if she wants to do the acute procedure in a shorter time frame

Additional recommendations from the subcommittee.

(b) (6) will have to address the clarifications for the protocol submitted on 7/31/2018, providing additional detail on the addition of predator scent test and social behavior to 17006-01.

It was discussed that ACS will handle the husbandry of all protocols except for when the mice under 17006-01 are undergoing the stress paradigm; during this portion of the experiments the lab staff will handle the husbandry of the mice. An amendment may be required for the frequency of cage change outs on protocol 18025-02.

Protocol 17006-01 already has numerous previously approved amendments and currently has two outstanding amendments for this protocol awaiting IACUC approval. To fully address all noncompliance issues mentioned another substantial amendment will be required. It might be beneficial to and easier for the investigator to terminate protocol 17006-01, combine all previous amendments and new changes and submit all material in one new protocol.

The subcommittee will send the investigator an email addressing all noncompliance issues that require an amendment and will suggest the termination of protocol 17006-01 to combine all information into a new, more seamless protocol.

The investigator has already been sent two memos from the IACUC office regarding previous minor noncompliances. This current series of noncompliance issues will be the third notification from the IACUC office. IACUC Policy 09 states:

1) Consequences and Resolution of Minor of Non-Compliance Incidents

Resolution of minor issues may be achieved through communication between the ACS personnel, and the individual lab personnel without necessitating IACUC intervention.

If an issue is not resolved or is deemed more serious, it will be reported to the IACUC. The following notification process may be used to obtain compliance or escalate the non-compliance.

First notification: The PI will be required to provide a written response regarding how the incident occurred, how it was corrected and how it will be prevented in the future. This response will be reviewed by the IACUC, and the IACUC may also require additional steps including retraining of investigative staff.

Second notification: Possible revocation of facility access privileges depending upon the circumstances and the response of the PI. The PI will be required to appear before the IACUC or a subcommittee of the IACUC.

Third notification: The non-compliance may be reclassified as significant (see above). If reclassified, it may result in any potential consequences of significant non-

compliance listed above. This may include sanctions up to and including a suspension of activities following a majority vote of the IACUC with a quorum present and/or revocation of a researcher's privileges and any animal activity therein. The PI will be required to meet with the IACUC Chair, Attending Veterinarian, ACS personnel, and/or the Institutional Official whereupon the issue may be referred to the Research Integrity Officer for review as research misconduct in accordance with University Operating Policy 74.08. A third notification may require reporting to the appropriate federal authority and any suspension of animal activity must be reported (see above).

With numerous previous and current non-compliance issues, the subcommittee suggests a probationary period for the investigator. The subcommittee proposes that the probationary period (6 months to one year- as determined by the IACUC committee) consist of announced and unannounced PAMs from the IACUC committee and the ACS staff members. We suggest that each month during the probationary period, one member of the IACUC committee perform a scheduled PAM with the investigator. During this time, ACS staff members will perform random and unannounced PAMs as deemed appropriate. All PAMs will be documented and reported at the monthly IACUC meetings.

Thank you,