Exhibit 1

1) Letter dated September 12, 2012 from PETA, presenting the allegation and requesting disciplinary action against the University. The supporting documents included clinical records, an unrelated USDA report, and a 2008 version of the protocol. This letter was also sent to the NIH and NIDCD Directors.

AN INTERNATIONAL ORGANIZATION DEDICATED TO PROTECTING THE RIGHTS OF ALL ANIMALS

PETA

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September 12, 2012

A3368-4P

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Office of Laboratory Animal Welfare
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James F. Battey, Jr., M.D., Ph.D., Director
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31 Center Drive, MSC 2320
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Francis S. Collins, M.D., Ph.D., Director
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Via e-mail: wolffa@od.nih.gov; batteyj@nidcd.nih.gov; francis.collins@nih.hhs.gov

Dear Dr. Wolff, Dr. Battey, and Dr. Collins:

I am writing on behalf of People for the Ethical Treatment of Animals (PETA) and our more than 3 million members and supporters to request that the Office of Laboratory Animal Welfare (OLAW) and the National Institute on Deafness and Other Communication Disorders (NIDCD) investigate what we believe to be egregious violations of the Public Health Service's *Guide to the Care and Use of Laboratory Animals* (the *Guide*) related to the treatment of cats used in invasive brain experiments at the University of Wisconsin-Madison (UW-Madison, PHS Assurance #A3368-01). We urge you to give this case top priority and if our allegations are substantiated, to take swift and decisive disciplinary action against UW-Madison–which has a long and sordid history of failing to comply with the *Guide* and the Animal Welfare Act–including terminating any active National Institutes of Health (NIH) grants for the project and demanding repayment of past funds awarded.

Background

PETA has obtained extensive internal documentation, including veterinary records, daily care logs, surgical reports, protocols, and dozens of disturbing photographs, related to the use of cats in a project at UW-Madison funded by the NIDCD. All of the photographs and many of the records are related specifically to a cat named Double Trouble (a.k.a. "G07" and "L005") who was used and killed in this experiment in 2008.

Affiliates
• PETA Foundation (U.K.)
• PETA Asia-Pacific
• PETA India
• PETA Germany
• PETA Netherlands

Double Trouble was subjected to several invasive surgeries, including the implantation of a steel post in her head, steel coils in her eyes, electrodes in her brain, and cochlear implants in her ears. Because Double Trouble developed a serious and chronic bacterial infection, and the experiment was deemed a failure by UW staff, Double Trouble was killed in December 2008.

UW-Madison's Noncompliance with the *Guide* and U.S. Government Principles

The aforementioned materials compiled by PETA indicate that UW-Madison exhibited problems in a number of areas, including the following:

- **Veterinary Care**: UW-Madison failed to provide adequate veterinary care, failed to observe animals on a daily basis, failed to ensure adequate surveillance, diagnosis, treatment, and control of disease, and failed to minimize discomfort, distress, and pain of animals (U.S. Government Principle IV and National Research Council [NRC] 10, 56, 59 and 61).
- **Adherence to the protocol approved by the Institutional Animal Care and Use Committee (IACUC)**: The Principal Investigator significantly deviated from the protocol that had been approved by the UW-Madison IACUC, and the IACUC failed to sufficiently monitor experimental activities to prevent this deviation (NRC 9).
- **Anesthesia**: UW-Madison failed to ensure that appropriate, adequate, and/or appropriately-administered anesthesia was provided to animals, and failed to ensure competence of personnel responsible for conducting surgical procedures on animals (U.S. Government Principles IV, V, and VIII and NRC 61 and 64).
- **Animal Numbers**: UW-Madison failed to ensure that justifications provided for the numbers of animals used were scientifically or statistically based (U.S. Government Principle III and NRC 10).
- **Least Invasive Methods**: UW-Madison failed to ensure that the least invasive procedures had been considered (NRC 10).

Despite these numerous apparent areas of noncompliance with the *Guide* and violations of the U.S. Government Principles, there is no record of UW-Madison having reported these problems to OLAW.[1] Other cats are still being used and killed for this study at UW-Madison.

1. Failure of UW-Madison to provide adequate veterinary care to animals by failing to ensure that procedures involving animals minimized discomfort, distress, and pain to animals

The *Guide* states unequivocally that "veterinary care is an essential part of an animal care and use program"[2] and further specifies that the attending veterinarian is responsible for providing guidance to laboratory personnel in issues associated with "animal care, behavior, and well-being."[3] The *Guide* stipulates that all animals "should be observed for signs of illness, injury, or abnormal behavior by a person trained to recognize such signs [and that such observation] should occur daily, but more-frequent observations might be warranted, such as during postoperative

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[1] Based on records secured through the Freedom of Information Act and Wisconsin's state open records law.
[2] NRC [National Research Council]. *Guide for the Care and Use of Laboratory Animals.* National Academy Press: Washington, D.C. (1996): 56. (NRC 56). The 1996 edition of *The Guide* is referenced in this complaint because most of the incidents discussed here occurred prior to the adoption of the new edition.
[3] *Ibid.*

recovery or when animals are ill or have a physical deficit."[4] Further, the *Guide* cautions that occasionally, "protocols include procedures that have not been previously encountered or that have the potential to cause pain or distress that cannot be reliably controlled,"[5] including protocols involving physical restraint, multiple major survival surgery, and food restriction—all of which were components of the experiments in which Double Trouble was used—and advises greater scrutiny of these protocols. Additionally, the *Guide* recommends that "signs of illness, distress, or other deviations from normal in animals should be reported promptly to ensure appropriate and timely delivery of veterinary medical care."[6] These guidelines were routinely violated at UW-Madison.

According to the records obtained by PETA, Double Trouble was subjected to several invasive surgeries on her eyes, ears and brain. Following these multiple surgeries, Double Trouble's health rapidly deteriorated. Records state that she was observed twitching, which the clinical notes indicate was a "neurological sign."[7] Further, according to veterinary records, Double Trouble's head wound never healed. More than *six months* after a steel head post was implanted in Double Trouble, records describe her wound as "open, moist w/bloody purulent discharge, [with] moderate swelling."[8] Even after this observation, Double Trouble was still used in an invasive procedure where a recording chamber was implanted into her head and electrodes were inserted into her brain.[9]

Double Trouble's records indicate that she was *not* being observed on a daily basis (Exhibit A):

- During a 19-month period between August 29, 2006 and March 24, 2008, there are only 14 documented observations for Double Trouble. On one date during this period, personnel observed vomit in her cage, but she was not observed again for 7 days.
- There are only three documented observations for Double Trouble in July 2008, even though Double Trouble had been deafened and implanted with cochlear devices the month prior.
- There are no documented observations of Double Trouble between 8/25/08 and 9/19/08 when lab staff observed Double Trouble pacing and an "asymmetry to the face," indicating that she had likely suffered damage to her facial nerve and possible paralysis.[10]
- After the 9/19/08 observation, there are no documented observations for Double Trouble for over two weeks, when UW staff noted that Double Trouble had a "wire loose from right side of jacket" and noted "playing with cord overnight/this a.m.? small plastic piece found on cage floor; wire placed back into jacket."[11]

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[4] NRC 59.
[5] NRC 10.
[6] NRC 59.
[7] UW-Madison, Treatment and Progress Record for Double Trouble, entry dated 13 Aug 2008. Attached as Exhibit A; page 8.
[8] UW-Madison. Treatment and Progress Record for Double Trouble, entry dated 24 Oct 2008. Attached as Exhibit A; page 9.
[9] UW-Madison. Procedure Log for Double Trouble, dated 21 Nov 2008. Attached as Exhibit E.
[10] http://www.petmd.com/cat/conditions/neurological/c_ct_facial_nerve_paresis
[11] UW-Madison. Treatment and Progress Record for Double Trouble, entry dated 6 Oct 2008. Attached as Exhibit A; page 8.

According to records, a resilient bacterial infection developed in Double Trouble's surgical wound starting on or about October 22, 2008, and she was placed on an antibiotic regimen.[12] Despite this, experimenters continued to use her for the experiment for several weeks. The records indicate that the infection was never brought under control and one of the last entries in Double Trouble's records states that she "appear[ed]…depressed."[13]

Since the protocol specified that Double Trouble would be subjected to multiple invasive, survival surgeries, UW staff should have diligently monitored Double Trouble's health status because of the increased risk of adverse health outcomes. Because UW-Madison staff failed to observe Double Trouble *at least daily*, the cat's worsening condition was not communicated to veterinary staff in a timely manner and the efficacy of any medical care provided to her was greatly compromised.

Furthermore, allowing Double Trouble to remain on this experiment for several months while undergoing repeated treatments for chronic complications produced by the study is not consistent with the *Guide's* recommendation that: "A continuing and thorough assessment of surgical outcomes should be performed to ensure that appropriate procedures are followed and timely corrective changes instituted."[14] Fundamentally, the failure of the UW-Madison IACUC to ensure that the protocol included "criteria and process for timely intervention [or] removal of animals from a study"[15] contributed to the exacerbated pain, discomfort, and distress suffered by Double Trouble, in violation of U.S. Government Principle IV, which stipulates the "minimization of discomfort, distress, and pain"[16] in animals.

Finally, in the absence of any meaningful relief provided by the experimenters to Double Trouble during her protracted suffering through the multiple invasive surgeries and the persistent infections which spanned months, it is possible that humane euthanasia would have provided the only viable option for relieving Double Trouble's pain and distress. If this is true, the experimenters' failure to euthanize Double Trouble would constitute a noncompliance with the *Guide's* recommendation that euthanasia be used "as a means to relieve pain or distress that cannot be alleviated by analgesics, sedatives, or other treatments."[17] This is reinforced in U.S. Government Principle VI, which specifies that: "Animals [who] would otherwise suffer severe or chronic pain or distress that cannot be relieved should be painlessly killed at the end of the procedure or, **if appropriate, during the procedure**"[18] [emphasis added].

2. Failure of the PI to adhere to the IACUC-approved protocol and failure of UW-Madison to sufficiently monitor animal activity to prevent the PI's deviation from the approved protocol

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[12] UW-Madison. Treatment and Progress Record for Double Trouble, entry dated 22 Oct 2008. Attached as Exhibit A; page 9..
[13] UW-Madison. Treatment and Progress Record for Double Trouble, entry dated 27 Oct 2008. Attached as Exhibit A; page 10.
[14] NRC 61.
[15] NRC 10.
[16] Office of Laboratory Animal Welfare [OLAW], Public Health Service Policy on Humane Care and Use of Laboratory Animals, *U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training*. Available at: http://grants2.nih.gov/grants/olaw/references/phspol.htm#USGovPrinciples.
[17] NRC 65.
[18] OLAW. Principle VI.

The *Guide* stipulates that the IACUC is responsible for the "review of proposed uses of animals in research, testing, or education (i.e., protocols); and establishment of a mechanism for receipt and review of concerns involving the care and use of animals at the institution."[19] Thus, any activity involving animals must be approved by the IACUC, and any significant deviation from that approved activity must be approved by the IACUC before it can be performed. Moreover, *continuing* IACUC oversight of animal activities is required by federal regulations and policies.

The experimental protocol in which Double Trouble was used states:

> "If any surgical procedure is longer than about 30 minutes (e.g. for the surgeries to implant head restraint device, eye or ear coils, **or recording chambers**), [the experimenters] insert an endotracheal tube to administer gas (isoflurane) anesthesia" [emphasis added].[20]

However, Double Trouble underwent a surgery to implant a recording chamber on November 21, 2008, in which her scalp was incised, a hole was drilled into her skull, and a recording chamber was glued over the hole. Double Trouble's records indicate that she was only intramuscularly administered ketamine and acepromazine, even though the duration of the surgery was over two hours (Exhibit C). Because acepromazine is a sedative and intramuscular ketamine is not capable of producing surgical-plane anesthesia, halfway through the surgery the experimenters noticed that the anesthesia "was light" and they administered an additional 40 milligrams of ketamine.

The failure to administer gas anesthesia during this invasive procedure constitutes a significant deviation from the IACUC-approved protocol for which the PI never submitted an amendment, according to the public records provided to PETA by UW-Madison. This failure to adhere to the protocol directly—and substantially—impacted the welfare of Double Trouble. The incident also suggests inadequacies in UW-Madison's postapproval monitoring program and in the university's training program, which are discussed at greater length in the following section.

3. <u>Failure to use appropriate methods of anesthesia and failure to ensure competence of personnel responsible for conducting surgical procedures on animals</u>

The *Guide* clearly stipulates that: "The selection of the most appropriate analgesic or anesthetic should reflect professional judgment as to which best meets clinical and humane requirements without compromising the scientific aspects of the research protocol. Preoperative or intraoperative administration of analgesics might enhance postsurgical analgesia. The selection depends on many factors, such as the species and age of the animal, the type and degree of pain, the likely effects of particular agents on specific organ systems, the length of the operative procedure, and the safety of an agent for an animal, particularly if a physiologic deficit is induced by a surgical or other experimental procedure."[21]

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[19] NRC 9

[20] UW-Madison IACUC-Approved Protocol "Behavioral and Physiological Studies of Sound Localization." Question 17. Attached as Exhibit B; page 6.

[21] NRC 64

However, the incident described in the earlier section constitutes an undermining of the professional veterinary judgment provided through the IACUC's review of the protocol, and the use of inappropriate and inadequate pain relief during an invasive, painful surgery. The incident also constitutes a blatant violation of U.S. Government Principles IV and V, which stipulate, respectively, that: "Proper use of animals, including the avoidance or minimization of discomfort, distress, and pain when consistent with sound scientific practices, is imperative"[22] and "Procedures with animals that may cause more than momentary or slight pain or distress should be performed with appropriate sedation, analgesia, or anesthesia."[23]

UW-Madison records reveal three other similar incidents where inappropriate, insufficient, or improperly administered anesthesia was used in conjunction with this protocol:

a. On April 24, 2008, Double Trouble underwent a procedure measuring her auditory brainstem response (ABR) which required that she be anesthetized with a mixture of ketamine and acepromazine. Her records indicate that during this procedure she was "kicking" and "waking up" and personnel wrote "anesthesia too light?"[24]
b. On June 11, 2008, Double Trouble underwent a highly invasive surgery where experimenters implanted cochlear devices in her inner ears. Surgical logs indicate that immediately after experimenters had cut into her head, Double Trouble's anesthetic mask came off and she began to wake up.[25] PETA has obtained photographs of Double Trouble during this surgical procedure.[26] These graphic photographs indicate that Double Trouble would have experienced enormous suffering when she awoke out of anesthesia in the middle of the surgery.
c. On December 17, 2008, another cat used in this same project, known only as "cat 33," was undergoing an invasive surgery to implant coils into her eyes. During the procedure, fluid began to fill her lungs and she stopped breathing. The cat's isoflurane was removed to clear her trachea and as a result, she woke up during the surgery.[27] A properly sized and placed endotracheal tube will block the tracheobronchial tree to keep liquid from getting into the lungs.

The pattern created by the four incidents presented here, wherein cats undergoing invasive surgeries were given inappropriate, inadequate, and/or improperly administered anesthesia suggests serious deficiencies in UW-Madison's training of personnel performing surgical procedures—in noncompliance with the *Guide,* which states explicitly that: "The PHS Policy and the AWRs [Animal Welfare Regulations] place responsibility with the IACUC for determining that personnel performing surgical procedures are appropriately qualified and trained in the procedures to be performed."[28] The deficiencies also violate U.S. Government Principle VIII, which states: "Investigators and other personnel shall be appropriately qualified and experienced for conducting procedures on living animals. Adequate arrangements shall be

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[22] OLAW. Principles IV and V.
[23] *Ibid.*
[24] UW-Madison. Procedure Log for Double Trouble, dated 24 Apr 2008. Attached as Exhibit D.
[25] UW-Madison. Procedure Log for Double Trouble, dated 11 Jun 2008. Attached as Exhibit G; page 2.
[26] See Appendix.
[27] UW-Madison. Procedure Log for Cat 33, dated 17 Dec 2008. Attached as Exhibit F.
[28] NRC 61.

made for their in-service training, including the proper and humane care and use of laboratory animals."[29]

4. Failure to provide scientifically based justification for number of animals used in protocol

The *Guide* advises that the PI is responsible for including justification for the number of animals proposed in a protocol and that the IACUC is responsible for ensuring that a proposed protocol includes "justification of the species and number of animals requested"[30] and that "whenever possible, the number of animals requested should be justified statistically."[31] This is reinforced in U.S. Government Principle III: "The animals selected for a procedure should be of an appropriate species and quality and the minimum number required to obtain valid results."[32]

However, in the IACUC-approved experimental protocol, the PI justified the use of 30 cats per year for this experiment "based upon past experience" representing an "average" number of cats used over the past 30 years.[33] In glaring contradiction, the PI stated elsewhere in the protocol that he had historically used 15 cats per year, not 30. Further, the PI did not give any scientific or statistical basis for either of these arbitrary figures and merely stated that "this number allows [the experimenters] to collect enough data to keep up a productive publication record that ensures our constant funding from NIH over these 30 years."

Neither appealing to past animal use nor citing requirements for publication of manuscripts are adequate explanations for the need to use a specific number of animals in an experiment. Moreover, the USDA has previously determined that these kinds of non-scientific statements are not adequate justifications for animal use. The University of California-San Francisco was cited for a violation of the AWA earlier this year for precisely this reason.[34]

In spite of the inconsistency in the numbers reported by the PI and in spite of the PI's failure to offer a scientific justification for the number of cats required for the protocol, the IACUC approved this protocol's use of 30 cats—raising serious concerns about the thoroughness of IACUC reviews at UW-Madison.

5. Failure to ensure that least invasive procedures had been considered

The *Guide* requires that when the IACUC reviews a proposed protocol, the protocol should include a discussion of the "availability or appropriateness of the use of less-invasive procedures, other species, isolated organ preparation, cell or tissue culture, or computer simulation."[35]

However, the alternatives search included in the protocol for the set of experiments in which Double Trouble was used does not consider any of the specific procedures covered by the

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[29] OLAW. Principle VIII.
[30] NRC 10
[31] *Ibid.*
[32] OLAW. Principle III.
[33] UW-Madison IACUC-Approved Protocol "Behavioral and Physiological Studies of Sound Localization." Question 12. Attached as Exhibit B; page 3.
[34] United States Department of Agriculture Plant Health Inspection Service Inspection Report for University of California-San Francisco dated 24 Jan 2012. Attached as Exhibit C.
[35] NRC 10.

protocol—including the deafening procedure and the implantation of eye coils, electrodes, cochlear devices—nor does it consider the species of animals being used, and thus does not represent a reasonable or good faith effort by the PI to identify less invasive methods.

Had the IACUC compelled the PI to adequately investigate alternatives to the use of animals, as the *Guide* requires, it would have been discovered that less invasive methods and entirely non-animal alternatives exist for investigating the neural mechanisms of sound localization.

For example, instead of steel eye coil implants, many researchers use a non-invasive infrared method for tracking eye movement in cats and other animals that obviates the need to perform any surgery and "excels with even higher precision than the other methods." [36,37,38] This refinement allows experiments to avoid causing animals any pain and distress related to the initial implant surgery, coil replacement surgery (which is common because the hardware is fragile) and potential chronic eye irritation, pain, and injury.

In terms of entirely non-animal research methods available, imaging techniques such as positron emission tomography (PET)[39] and near-infrared spectroscopy (NIRS)[40] are being used in sound localization and hearing studies using human volunteers at world-class institutions across the world. Additionally, despite the PI's statement to the contrary in the protocol, single neuron recording in human volunteers is possible, including in the auditory cortex, using intracranial electroenecephalography (iEEG).[41,42,43,44] Human-based recording techniques not only eliminate the use of animals and the obstacle of interspecies extrapolation, but provide rich data about the intricacies of the functional organization of the human auditory cortex, many of which could not have been ascertained using animals.

Due to the inadequacy of the alternatives search and the IACUCs acceptance of this impoverished search, widely available methods to refine and replace animal use were not considered as is required by the *Guide*.

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[36] Körding KP, Kayser C, Betsch BY, König P. Non-contact eye-tracking on cats. J Neurosci Methods. 2001 Sep 30;110(1-2):103-11.

[37] Girard P and Koenig-Robert R.. Ultra-rapid categorization of fourier-spectrum equalized natural images:macaques and humans perform similarly. PLoS One 6:e16453 (2011).

[38] Ben-Simon A, Ben-Shahar O, Segev R. Measuring and tracking eye movements of a behaving archer fish by real-time stereo vision. J Neurosci Methods. 2009 Nov 15;184(2):235-43.

[39] Peterson, Pisoni, and Miyamoto. 2010. Cochlear implants and spoken language processing abilities: Review and assessment of the literature. Restor Neurol Neurosci. 28(2):237-250.

[40] Sevy, Bortfeld, Huppert, Beauchamp, Tonini, and Oghalai. 2010. Neuroimaging with Near-Infrared Spectroscopy Demonstrates Speech-Evoked Activity in the Auditory Cortex of Deaf Children Following Cochlear Implantation. Hear Res. 270(1-2):39-47.

[41] Bitterman Y, *et al.* Ultra-fine frequency tuning revealed in single neurons of human auditory cortex. Nature. 2008 Jan 10;451(7175):197-201.

[42]Howard MA, *et al.* A chronic microelectrode investigation of the tonotopic organization of human auditory cortex. Brain Res. 1996 Jun 17;724(2):260-4.

[43] Jacobs J, *et al.* Brain oscillations control timing of single-neuron activity in humans. J Neurosci. 2007 Apr 4;27(14):3839-44.

[44] Palmer AR, Summerfield AQ. Microelectrode and neuroimaging studies of central auditory function. Br Med Bull. 2002;63:95-105.

Request for Action

In connection with the aforementioned NIH-funded activities, UW-Madison has repeatedly violated the standards set forth in the *Guide* as outlined above and has also failed to report these flagrant noncompliances to OLAW, in violation of its Assurance and PHS Policy on Humane Care and Use of Laboratory Animals Section IV.F.3. In light of UW-Madison's misconduct, PETA respectfully asks that, at a minimum, you terminate any ongoing NIH support of the experiments in question. Given the scope of UW-Madison's violations, the fact that they directly and seriously impacted the welfare of animals, and UW-Madison's extensive history of noncompliance, it may even be warranted to withdraw UW-Madison's OLAW Assurance entirely, in accordance with PHS Policy Section IV.A.

Furthermore, as you are aware, section 4.1.1.5 of the NIHGPS states, "Charges to NIH grant awards for the conduct of live vertebrate animal activities during periods of time that the terms and conditions of the grant award are not upheld are not allowable." NIH has further elaborated to PETA in a letter that it, "does not permit charges to grant awards for the conduct of animal studies during periods of non-compliance with animal welfare regulations." The NIHGPS further states that, "[i]n cases where charges have been made for unauthorized animal activities, appropriate adjustments must be made to the grant to remove those charges." NIH Notice NOT-OD-10-081 re-affirms this policy, and states that "[s]pecific situations under which charges are not allowable" include the "conduct of animal activities in the absence of a valid IACUC approval of the activity," which is exactly what occurred at UW-Madison.

Because UW-Madison's conduct is in direct violation of the NIHGPS, as further affirmed by NIH Notice NOT-OD-10-081, PETA also urges the NIH to seek repayment of previous funds awarded to UW-Madison for this experiment during the noncompliant period.

Conclusion

The documents obtained by PETA demonstrate a pattern of carelessness towards animals and disregard for the *Guide for the Care and Use of Laboratory Animals* that resulted in enormous suffering for Double Trouble and other cats used in this highly invasive experiment. In light of the seriousness of these allegations and the fact that the NIH is still providing funding for this project, we urge you to swiftly investigate this complaint and expedite its resolution.

I look forward to hearing from you. I can be reached at Telephone # or JeremyB@peta.org. Thank you for your time and consideration.

Sincerely,

Jeremy Beckham
Research Project Manager
Laboratory Investigations Department

Exhibit A

UW-Madison
Treatment and Progress Records for Double Trouble

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# L005 / G07 SPECIES: cat SEX: F

Date	~~Time~~ Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
8/23/06	14:15	0.73 mL Dual-Pen SQ [redacted]
	14:45	0.12 mL Buprenorphine SQ, per [redacted] DVM
8/24/06	16:30	0.74 Dual-Pen SQ
MGH		I did not give painkiller today, per [redacted], DVM
		He looked at 607 & thought she didn't need it.
8/25/2006	11:30	0.76 Dual-Pen SQ [redacted]
8/26/2006	13:15	0.76 mL Dual-Pen, SQ [redacted]
8/27/2006	11:45	0.76 mL Dual-Pen, SQ [redacted]
8/28/2006	15:15	0.76mL Dual-Pen, SQ [redacted]
8/29/2006	12:00	0.76 mL Dual-Pen, SQ [redacted]
		This is the last day of penicillin treatment,
		per [redacted] DVM
10/11/06		PHYSICAL EXAM: WNL [redacted]
4/18/07		PHYSICAL EXAM: (S) BAR (O) [redacted]
		(A) GOOD CONDITION (P) NORMAL [redacted]
		MONITORING NAILS TRIMMED. [redacted]
5-09-07	900	APPEARS NORMAL. BARH. [redacted]
6-14-07	800	APPEARS NORMAL. BARH. [redacted] NAILS TRIMMED.
7-16-07	810	APPEARS NORMAL. BARH. [redacted]
8-13-07	820	APPEARS NORMAL. BARH [redacted]
9-21-07	1310	VOMIT IN CAGE - YELLOW, FOAMY. CAT NOT IN CAGE TO ASSESS. [redacted]
9.28.07	11:45am	No vomit was reported, appears normal. [redacted]
11-27-07	715	APPEARS NORMAL. BAR [redacted]
12.24.07	9:20	Appears normal BAR [redacted] J [redacted] DVM
01.23.08	1:35	Appears normal BAR [redacted]
2/12/08	815	NAILS TRIMMED APPEARS NORMAL, ACTIVE. BAR [redacted]
3/24/08	1015	SMALL AMOUNT OF PURULENT DISCHARGE AROUND HEADCAP.
		ACTIVE, BAR. [redacted]
3/25/08	1030 A	S/O: Small amount of purulent discharge on L side headcap. BAR, trend
		of mild weight gain
		A: WNL P: No action indicated at this time. [redacted], DVM

S- 0143

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# DOUBLE TROUBLE LO05/G07 SPECIES: FELINE SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
4/28/08	14:30	Trimmed nails. [redacted]
6/9/08	1255	3mls blood drawn from L cephalic + jugular for pre-surgical cbc + chem. [redacted] DVM.
6/10/08	8:15	S: Review of bloodwork from 6/9: CBC WNL. Chemistry WNL, but BUN levels higher end of normal.
		A: Healthy for procedure, but recommend monitoring of BUN + Cr throughout.
		P: Monitor kidney values frequently (~2X/wk) after procedure. [redacted] DVM
6-10-08	1200	100 mg cephazolin sub Q for prophalaxtic per [redacted] [redacted] DVM
6-11-08	7pm	cephazolin 100mg Buprenorphine .015mg SQ cat looks good. Has eaten 1 can food. [redacted]
6-12-8	8am	urine in pan. No apparent response to sound. When roused from sleep alert, reactive, vocalizing.
wgt 3kg		T=99.9 - leave heat lamp on for morning. Cat able to move to other side if too warm. Appeared to have eaten gravy. Buprenorphine 0.1ml SQ [redacted] DVM
	9:00	125 mg cephalexin in soft food. Cat eating well
	3:30	provided lab staff [redacted] with 1 bottle topical Nolvadine® oint. to be used around exit site of cutaneous catheters at time of bandage change. [redacted] DVM
	5:45	.015 mg SQ of Buprenorphine [redacted] 125mg cephalexin gel dressing Δ'd x2
6/13/08	730	0.1 ml BUPRENORPHINE GIVEN SQ.
	8:00 S/O	Alert, reactive, resting quietly. T=101.1 Incisions appear ok. Unable to safely (instrumentation) put in cat bag to draw post-op blood. Will attempt when lab staff here. [redacted] DVM
	9:30	125 mg cephalexin in water [redacted] and dressing Δ

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# DOUBLE TROUBLE L005/607 SPECIES: FELINE SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
6-13-8	11 am	(cont) [redacted] assisted with restraining cat for blood draw RF cephal. 1 ml [redacted] CREAT, elevated. [redacted] DVM
6-13	5pm	125 mg cephalexin in food [redacted] .015 mg buprenorphine S.Q. [redacted]
6-14-08	8:15a	125mg Cephalexin given PO in food. Alert, responsive, resting quietly. Got up immediately started eating [redacted] food with medications. [redacted]
	4:35p	125 mg Cephalexin given PO in canned cat food. [redacted]
6-15-08	8:30A	125mg Cephalexin given PO in canned wet food. BAR, active. [redacted] watery feces present in cage. [redacted]
	8:10P	125 mg Cephalexin given PO in canned cat food. Cat had lower jaw caught on bandaging material around chest had to free from it. Watery feces ? present in cage. Possibly from this a.m. cleaned up after observation [redacted]
6-16-08		S/O - Alert reactive, musician healing well. Moving well. Some slight ataxia when moving around cage. Lower canine caught on string f/ chest bandage - freed easily. Soft stool noted in cage - is on BID Cephalexin. Monitor. [redacted] A/P - Doing well. Make sure loose strings trimmed at all times f/ chest bandage. Monitor stool - suspect f/ antibiotics. [redacted] DVM
6-16-08	8:9am	125 mg cephalexin in food [redacted]
	1:30	Lab contacted, reported results of blood draw - WNL. Entered into record. [redacted] DVM
	2pm	125 mg cephalexin [redacted]
6/17/08	8:45	Nonformed feces in pan. Animal appears to be eating and drinking well. BAR [redacted]
	9:37	BAR, drinking when observed, incisions ok, jacket not being bothered. Moderate amount SE/D in/near litter box. A: suspect diarrhea related to current antibiotics. P: monitor for persistency. [redacted] DVM.

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble LOOS/40 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
6-17-08	9:40	125 mg cephalexin in food [redacted]
	6:30pm	125 mg cephalexin in food [redacted]
6/18/08		Nonformed feces in pan. Animal appears [redacted]
		to be eating and drinking well. QAR- [redacted]
	9:40	125 mg cephalexin in food [redacted]
	4:30	125 mg cephalexin in food [redacted]
6/19/08	8:10	Nonformed feces in pan, small amount.
		Normal urine. Animal is active, BAR. [redacted]
	8:10	Incision lines dry + intact, no swelling or
		redness - healing well. [redacted]
	15:45	Postpone follow up bloodwork (BUN+Cr) to
		6/20. [redacted], DVM.
6/20/08	9:15	No feces in pan. Animal slightly ataxic
		and dehydrated. Appears to have consumed
		1/2 can of food. Will check with DVM
		for plan. [redacted]
	13:40	~2 ml blood drawn from right cephalic.
		Animal is eating canned food that was
		offered by lab. Moving around well,
		climbing on cage. BAR. [redacted]
6/21/08	8:00 A	lethargic, still eating canned food and
		attempting to eat dry food but seems to be
		having some trouble. no feces noted [redacted]
6/22/08	9:15 A	slightly less lethargic than yesterday, has not
		eaten much, feels thin [redacted]
6-23-8	8:45	No urine/stool in pan. Responsive, alert, normal ambulation.
		Bladder - palpable - WNL. Colon - palpate empty.
		Appears to be eating (dried food on bottom of cage
		Lab staff weighs daily - WNL for this cat.
		Recommend close monitoring of eating/stool. [redacted] DVM
6/23/08	15:20	NO urine or feces in pan, was taken to lab

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble 607 SPECIES: feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
6/23/08	cont..	for most of morning. Observed animal
		eating dry cat chow. Active and BAR. [redacted]
6/24/08	8:20	No urine or feces in pan. Observed animal
		eating dry food when offered. Appears
		active, BAR. [redacted]
	2:30	Contacted by [redacted] in lab.
		Removed surgical staples (clear 2) @ forearm & electrode
		had not healed by 1° intention. Rec. apply topical AD. Keep
		covered. Upon exam - BAR. ½" wound dorsal @ SubQ
		around electrode.
		Plan: sedate, examination, clean, flush edges & close. [redacted] DVM
6-25-08	S/O	No Stool/urine. Alert, reactive. 2F out of sweater. NPO overnight
	10 am A/P	Sedate: Dormitor 0.08 ml / Ketamine 0.15 ml IM [redacted]
		Clean debride opening, amend @ heals SQ - 2 sutures dorsally
		3 sutures ventrally. * Remove suture 10-14 days. *
		Vomited small amt. food, hair & string. Palpated abd. - bladder
		full - easily expressed. ~2" soft stool in colon. Small amt.
		extruded hard string. [redacted] present, cleaned free strings
		and taped edges of sweater. 150 ml SQ 0.9% Na Cl
	10:35 am	0.05 ml Antisedan IM. Sternal and ambulating within 5 min.
		Returned to cage 10:45. Started eating. [redacted] DVM
	4:30	Alert, reactive. Bandage/sweater in place. Eating @ [redacted] DVM
6-26-8		Fed gone. NS feces/urine. Bladder full.
		Alert, reactive. Check w/ Jen 6:45 to feed after more food
		this week. [redacted] DVM.
	pm	via email - apparently when she goes to bed - passes
		stool there. Rec. that is entered into medical record
		+/- advise vet staff. [redacted] DVM.
6-27-8	8:30	tooth stuck on sweater/string. BAR. Sm. amt. urine
		no feces. [redacted] DVM
6/28/08	955	NO URINE OR FECES IN LITTLER PAN. APPEARS TO BE

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# DOUBLE TROUBLE G07 SPECIES: FELINE SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
6/28/08		EATING AND DRINKING NORMALLY. NO VOMIT PRESENT.
		INCISIONS HEALING. GREAT ATTITUDE, ACTIVE, BAR [redacted]
6/29/08	1600	STILL NO URINE OF FECES IN CAGE PAN. BLADDER FULL
		ON PALPATION, ABLE TO MANUALLY EXPRESS SOME URINE,
		APPEARED NORMAL. COULD NOT PALPATE ANY FECES, BUT
		DID NOT SEEM PAINFUL. NO VOMIT IN CAGE. EATING
		AND DRINKING NORMALLY. ACTIVE, BAR. [redacted]
6-30-08	8:00 am	No urine/feces. Did she go to lab this weekend?
		Sutures under bandage look good → lab to apply Nolvadent @ bandage.
		Colon palpates full. Bladder soft.
		BAR. [illegible]. Eating. [redacted]
6/30/08		Cat has defecated in the lab litter box
		on the following dates - 6/25, 6/27, 6/28,
		6/30. Will be recorded in this log in the future.
		Original recordings can be found in Animal Husbandry
		activity log in the cats' holding room.
7/1/08		Used litter box downstairs.
7-14-8		Am visit ✓. Removed 5 cuticular sutures fr/around Ⓛ dorsal
		electrode exit. No feces in pan, urine recorded in lab book.
		Small stool palpated. Ⓛ front exit pouch open, [illegible] lead
		exposed. Repositioned & closed [illegible] [redacted] DVM
		Plan ✓ w/ lab staff re feces.
		Note: re recording feces as noted 6-30-08. Will only need to be
		recorded in animal housing log in animal room [redacted]
7/31/08	8:25am	NO urine or feces noted by ART; readily eats when
		given cat chow, BAR, Active ([redacted]).
	3:30pm	Normal urine & feces in box ([redacted])
8/5/08	8:40AM	No feces in box this am; urine normal - in lab today [redacted]
8/13/08	830 A	S/O: Reported for mild tremors, not as social. Animal is resting,
		mild twitching of ears + head, slightly more prominent

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20100921 - 000285

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble 607 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
8/13/08	cont	when noise made. Slight head tilt to R. NO evidence of nystagmus. Interested in food, no feces in box.
		A: mild neurologic signs, possibly related to cochlear implants.
		P: Recheck later this AM with [redacted], contact lab to see what if procedure done yesterday [redacted] DVM
	915	S/O: Lab replaced battery on 8/12 for cochlear implant. Volume turned down + twitching stopped.
		A: suspect over stimulation of auditory nerves.
		P: Lab will adjust. No action indicated unless behavior returns. [redacted] DVM
8/15/08	800A	S/O: Reported for pacing. Animal is pacing in cage, slightly unusual since animal usually resting or quiet. Eating ate food. Normal feces + urine in pan. Stops to be pet.
		A: mild pacing, possibly agitated, unknown cause.
		P: Recheck this PM. Notify lab. [redacted] DVM
	1510	S/O: Resting, woke up, normal locomotion + attitude, ate a piece of food offered.
		A: Appears WNL, unable to contact lab this AM.
		P: NAI unless report continues. [redacted] DVM
8/25/08	850am	Weighed; trimmed nails [redacted]
9/19/8	1:40	Lab staff reported asymetry to face ⇒ R eye/eyelid. PLR - WNL. No ocular discharge, conjunctival swelling or erythema. No facial soft tissue swelling. No blepharospasm. Lab staff thought may have related to electrical shock.
		[redacted] 9-22-08 [redacted] DVM.
10/1/08	8:20	Wire loose from Right side of jacket - playing with cord overnight / this a.m.? small plastic piece found on cage floor; wire placed back into jacket and lab was contacted [redacted] BAR, Active [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
10-22-8		Small draining wound dorsal cranial, skin; over where
		surgical implant is (R) side
		BAR: Acting normally per lab staff.
	A/P	(R) ? possible reac. to implant / infection
		Plan - clean as needed, daily at first. Apply topical Abx's
		Monitor progress. [redacted] Dvm
	5:20	Triple antibiotic ointment applied to head
10/23/08	835AM	Cranial wound showed, small hole - draining fand
		w/ mod. tissue. swelling around. BAR, Active, normal
		feces in urine [redacted]
10/24/08	835am	~~Patient~~ ① Cranial head wound open, moist w/ bloody
		purulent discharge, moderate swelling. Sample
		taken of discharg for culture. T=100.5°F. BAR,
		Active, normal urine & feces in box [redacted]
10/24/08	16:20	Per [redacted] vum, gave 62.5 mg clavamox (1.0ml) PO
		BID. Will possibly recheck when final culture
		comes back. [redacted]
		Gave 62.5mg (1.0ml) Clavamox PO in small
		scoop of wet cat food. Per [redacted] DVM, flushed
		wound cranial head with Chlorhexidine
		solution & small amount of discharge expelled. [redacted]
10/25/08	8:25 A.	gave 62.5 mg (1.0ml) clavamox po in small amount
		of cat food began eating immediately, minor swelling
		and discharge at head wound, BAR Active. [redacted]
	17:30	gave 62.5 (1.0mL) clavamox PO in small amt
		of wet food. BAR active [redacted]
10/26/08	8:20	gave 62.5 (1.0mL) clavamox PO in small amt
		cat food. Began eating immediately. was resting
		quietly upon entering room BAR [redacted]
	17:20	wound red and slightly moist. gave 62.5 (1.0mL)
		Clavamox PO in cat food. BAR [redacted]

① Entry error [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# "Double Trouble" G07 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
10/27/08	830	Gave 1.0ml Clavamox (62.5mg) in sm amt wet
		cat food - ate readily. BAR, Active, swelling around
		cranial wound increased. T = 102.2°F. [redacted]
	8:45	Purulent and bloody discharge expelled
		from cranial wound, swelling around wound
		opening increased. Soft feces in litter pan,
		normal urine. Animal appears slightly
		depressed today. QAR. Obtained weight
		and trimmed nails. [redacted]
	9:10	Flush head wound w/ diluted betadine solution. Cat
		did not seem irritated or painful by flushing.
		Plan - Assessment - per lab - reactive inflamm/infection have
		been anecdotally reported w/ these cranial electrodes.
		Plan - continue 1x day flush - diluted betadine soln.
		oral med BID - pending culture results.
		Reassess daily. [redacted]
10/27/08	16:15	Gave 62.5mg (1.0ml) Clavamox PO with small
		amount of canned food. BAR. [redacted]
10-28-8	8:30	Gave 62.5 mg (1ml) Clavamox PO w/ food [redacted]
		Semi-annual PE - T = 101 °F HR - 180 RR - WNL. QAR.
		Abd. palpation - bowels slightly thickening. Soft stool.
		Flushed wound w/ diluted povidone iodine (6cc). <1/2cc
		mucopurulent discharge.
		A/P abscess 2° to chronic instrumentation, appears to be
		healing. G.I. upset 2° to ab-administration.
		Continue on study per protocol. Culture pending [redacted]
		Gave 1.0 ml (62.5 mg) Clavamox PO with small
10/28/08	16:05	amount of food [redacted]
10/29/08	8:35	TEMP. = 101.1°F. Flushed with dilute Betadine for
		cranial wound. Expelled purulent and bloody
		discharge from wound. Animal is active + BAR. [redacted]

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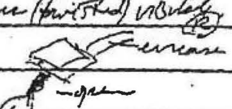
TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble G07 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
10-29-08	805AM	Gave 1.0ml Clavamox (62.5mg) in sm amt (~1T)
		of wet food - ate readily. Stool soft. BAR, Active [redacted]
	10:20	Culture result entered. Arcanobacterium pyogenes
		This seems to be responding to oral. But treat [illegible] u/kg
		IM/SQ BID for 3-21 days, will daily reassessment & update
		in plan. [redacted] Dm
	15:00	Gave 0.07ml Pen G (300000 u/ml) SQ. BAR, Active [redacted]
		Gave + 0.15 ml SQ = total .22 ml dose
		Dose miscalculation - total dose 0.22 ml. [redacted] Dm
10/30/08	825	Gave 0.22ml Pen G (300000 u/ml) SQ. BAR,
		Active, small amt soft stool in pan; flushed head
		wound with dilute betadine, moderate amt of
		viscous purulent discharge. [redacted]
10/30/08	15:30	Gave 0.22ml Pen G (300,000 U/ml) SQ. Active &
		BAR [redacted]
10/31/08	9:00	Gave 0.22ml Pen G (300,000 u/ml) SQ. QAR; flushed
		head wound w/ dilute Betadine (1%), sm amt of thick
		purulent discharge, moderate thickening of tissue
		around wound. [redacted]
	Note:	During AAALAC visit techs did all tx. and consultation w/ vet via
		email & phone. Vet communication w/ research staff
		regarding sedating and placing drain tube: [redacted] ak'd 10-31-8 [redacted] Dm
	AM am	Sedated Ketamine/domitor - See anesthesia record.
		Careful dissection of fistulated area top of head revealed electrode
		connector (?) with small piece loose flaps silicone &
		tubing running laterally coated & intact. Tubing running
		medially was not encased with multiple wire fibers (twisted) visible
		"Connector" was moveable w/ moveable sections 
		mosquito forceps could be passed ventrally
		under connector but no "large" fistulous tract could be
		identified w/out debriding & opening subQ tissues. Skin over

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20100921 - 000277

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# DT 607 SPECIES: Felis SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
10-31-8	(cont),	connector (original incision site) fractured. Placed [redacted] surgical sub Q drain difficult w/out full anesthesia & careful dissection around electrodes.
		[redacted] obtained instrumentation. Available for consult.
		Two simple interrupted sutures placed – one cranial/one caudal with central section of existing drainage left open.
		Plan is to arrange transport to CVM for radiograph to assess placement of electrode and anchor screw & underlying bone.
		607 given Antisedan IM, monitored until sternal & ambulatory and returned to cage w/ littermates. ~12:30 pm. [redacted] DVM
	Water	AALAC exit interview. Cage staff [redacted] until continual ½ hr check, LAR staff reported small amt. urination 12:40 pm.
	15:00	BAR. Offered small amt dry food w/ approx 1/4 D. [redacted] DVM
		Plan, as above, plus continue BID Penicillin inj SQ/IM. [redacted] DVM
	15:15	0.22 cc Pen G - (use 20g needle) SC. [redacted] DVM
		—NO FLUSH WIPED OVER WEEKEND (surgical wound). [redacted] DVM
11-01-08	750	0.22 mL PEN G (300,000 iu/mL) GIVEN SQ. VERY SMALL AMOUNT OF PURULENT DISCHARGE FROM WOUND ON HEAD. URINE, BUT NO STOOL IN CAGE. ACTIVE, BAR. ALL FOOD EATEN. [redacted]
	1805	0.22mL PEN G (300,000 iu/mL) GIVEN SQ. [redacted]
11-02-08	810	0.22mL PENG (300,000iu/mL) GIVEN SQ. WOUND ON HEAD SCABBED, NO DISCHARGE BUT SLIGHTLY SWOLLEN. VERY ACTIVE, BAR [redacted]
	1715	0.22mL PENG (300,000iu/mL) GIVEN SQ. [redacted]
11-3-08	855 Am	BAR, SH soft Stool in pen. Pen-G (300,000 iu/m) 0.22ml given SQ. Approx 2cm lump on top of head associated with open wound and Sx incision, when pressure applied mod amount of thick yellow/red purulent discharge. Per [redacted], flushed with dilute betadine [redacted]
11/3/08	1	
11/3/08	10:00	Gave 0.22ml Pen-G (300,000 iu/ml) SQ. [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# DT GOTT SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11/4/08	B:35am	BAR, Active, Gave 0.22 ml Pen G (300,000iu/ml) SQ wound on top of head scabbed over, soaked with dilute betadine & then flushed with the same, small amount of purulent discharge. [redacted]
	10:30	Plan: continue BID short acting penicillin until wound healed (had not been flushed over weekend). Cont. SQ flush. Arrangements in process to transport 11-5-8 for scheduled skull/cochlear implant. (Ket-Dex). [redacted] Dex
	14:40	Gave 0.22 ml Pen G (300,000iu/ml) SQ, BAR, Active [redacted]
11/5/08	BID	BAR, Active. Gave 0.22 ml Pen G (300,000iu/ml) SQ. Cranial wound cleaned with betadine and flushed with dilute betadine, sm amt purulent discharge [redacted]
	8:50	Transport LAR van #0 CV/M for radiographs.
	8:56	Dantin 0.08 ml Ketamine 0.2 ml — IM.
	9:20	Return transport to [redacted]
	9:30	Return to cage. Awake
	9:32	Antisedan 0.08 ml IM
	9:35	Standing & eating A/D offered.
	12:20	BAR, slightly groggy, opened cage to full size, eating remainder of A/D wet food, urinating and cage [redacted]
	2:25	BAR, Active, ate all of wet food, Gave 0.22ml Pen G (300,000iu/ml) [redacted]
11-6-8	BID	flushed wound top of head w/2 ml dilute betadine solution 1.5cc discharge expressed from lateral pocket. Gave 0.22ml Pen-G (300,000iu/ml) [redacted] Dex Plan Cont. BID penicillin until no discharge + daily flush
	3:15pm	Gave 0.22ml Pen G (300000iu/ml) SQ, QAR [redacted]
11/7/08	8:30	Gave 0.22ml Pen G (300000, iu/ml) SQ. Flushed cranial opening with dilute betadine and expelled purulent discharge. Sutures intact at wound site. Animal is active. BAR. [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (G07) SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11/7/08	15:50	Gave 0.22ml Pen G (300,000 iu/ml) IM. BAR. [redacted]
11/8/08	750A	wound on head between As & AD is clean, dry, & scabbed - sutures remain intact - Flushed/soaked with dilute betadine solution - no discharge to expell; Administered 0.22ml (300,000 iu/ml) Pen G SQ; BAR; Active [redacted]
	10:30	Removed scab. flushed w/ small amt dilute betadine BAR [redacted]
	1:15pm	Administered 0.22ml (300,000 iu/ml) SQ; BAR; Active [redacted]
11/9/08	830A	wound on head is clean, dry & scabbed - removed scab & flushed abscess w/ a small amt of dilute betadine Solu. small amt purulent discharge expelled; Administered 0.22ml (300,000 iu/ml) SQ; BAR [redacted]
	4:00pm	wound on head slightly moist but clean with no active discharge; Administered 0.22ml (300,000 iu/ml) SQ; BAR; Active [redacted]
11/10/08	8:35AM	BAR, Active, Soaked head wound/fistula with dilute betadine, scab removed, flushed with dilute betadine, small amount of purulent discharge evacuated; Gave 0.22 ml Peni-G (300,000 iu/ml) SQ. Normal feces/urine [redacted]
	4:15pm	QAR, resting, Gave 0.22ml Pen G (300,000 iu/ml) IM. [redacted]
11/11/08	8:18AM	BAR, Active. Small scab removed from cranial wound, small amount of yellowish cloudy discharge expelled, flushed wound with dilute betadine. Gave 0.22 ml Pen G ~~(0.22ml)~~ SQ (300000 iu/ml). Normal feces/urine [redacted]
	3:45pm	Gave 0.22ml Pen G (300000 iu/ml) SQ, BAR, Active [redacted]
11/12/08	New Tx	Plan: Per [redacted] discontinue Betadine flushing of Cranial wound after this morning, switch to oral meds, Give 1.0ml Amoxicillin (50mg/ml) BID x 14 days [redacted]
	8:45AM	Cranial wound flushed with Betadine dilute solution, small amount of yellow purulent discharge. [redacted]
	9:15AM	Gave 1.0ml Amoxicillin (50mg/ml) in 1T wet food - gave to Lab ([redacted]) to give this morning [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble 607 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11-12-08	340pm	Gave 1.0ml Amoxicillin (50mg/ml) in 1 Tablespoon wet food, ate it readily, BAR, Active [redacted]
11-13-08	8:10AM	Gave 1.0ml Amoxicillin (50mg/ml) in 1 Tablespoon in wet food, ate readily, Removed scab off cranial head wound - Small amount yellowish purulent discharge evacuated, normal stool & urine [redacted]
	16:05	Gave 1.0ml Amoxicillin (50mg/ml) in 1 Tablespoon wet food, ate readily, BAR, Active [redacted]
11-14-08	820	Gave 1.0ml Amoxicillin (50mg/ml) in 1 Tablespoon wet food, ate readily, BAR, Active [redacted]
		Note: wound continues to heal. Switched to oral amoxicillin, BID 15 days. CC w/DI & lab staff regarding plan for cat. Fistulous tract suspected. If continues, will need to carefully place surgical drain under anesthesia. Daily monitoring by lab/vet staff. [redacted] DVM
11/14/08	14:15	Gave 1ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. [redacted]
11/15/08	8:15	Gave 1.0ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. Normal urine, no feces. Animal is active and BAR. [redacted]
11/15/08	17:10	Gave 1.0ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. BAR. [redacted]
11/16/08	8:55	Gave 1.0ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. Small amount of purulent discharge expelled from cranial wound. Active and BAR. [redacted]
11/16/08	16:25	Gave 1.0ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. BAR. [redacted]
11/17/08	8:15	Gave 1.0ml Amoxicillin (50mg/ml) PO with one tablespoon of wet food. [redacted]
11/17/08	14:50	Gave 1.0ml Amoxicillin (50mg/ml) Po w/ one tablespoon of wet food. [redacted]

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TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble G07 SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11/18/08	8:40	BAR, Active Gave 1.0ml Amoxicillin (50mg/ml) in small amount of wet food, PO. Soft feces in litter pan. Cranial wound soaked ½ large scab removed - sm amt purulent discharge evacuated. [redacted]
	3:45pm	Brought to lab - Gave 1.0ml Amoxicillin (50mg/ml) in small amt of wet food to be given P.O. [redacted]
11/19/08	7:45	Gave 1.0ml Amoxicillin (50mg/ml) in one tablespoon of wetfood. No feces or urine in litter pan, will check later in the day. Cranial wound scabbed and dry. Animal is active and BAR. [redacted]
	9:30am	Lab staff called. Exudate from collected site on ① top of head.
	2:55pm	Mucopurulent discharge ②. Oral Amoxicillin 50mg/ml in TBS of wetfood. Had been in lab all day. Plan continue oral meds. OK w/ PI regarding planned testing. [redacted]
11/20/08	8:10am	BAR, Active Gave 1.0ml Amoxicillin (50mg/ml) in one tablespoon wet cat food. ⑧ Cranial wound - scab removed & flushed with dilute betadine - mod. amount of thick yellow purulent discharge evacuated. ⑫ cranial wound soaked w/ dilute betadine, sm amt purulent/bloody discharge. no feces in pan, normal urine [redacted]
	3:15pm	BAR, Active, Gave 1.0ml Amoxicillin (50mg/ml) in one tablespoon wet cat food. [redacted]
11/21/08	8:00am	BAR: Active. Gave 1.0ml Amoxicillin (50mg/ml) in one tablespoon wet cat food; Both Rt & Left Cranial wounds flushed with betadine dilute solution; Rt wound large amount of thick yellow purulent discharge evacuated, Lt wound moderate amount of thick bloody purulent discharge evacuated, NO feces in box. [redacted]
	3:30pm	Amoxicillin 1.0ml (50mg) in sm. amt wet food to be given PO by lab this afternoon - Animal was sedated by lab

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S- 0129

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (G07) SPECIES: feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11-21-08	cont.	with Ketamine - still groggy - will wait until sternal & ambulatory
		to place treatment on cage [redacted]
11-22-08	8:15 A	no discharge or scabbing observed. slight
		swelling and redness on both right and left
		cranial wounds. Soaked wounds and flushed
		with dilute betadine solution. Gave 1 mL (50mg)
		Amoxicillin PO in small amount of wet cat food
		BAR Active [redacted]
	7:00 PM	gave 1 mL (50mg) Amoxicillin PO in small amt
		of wet cat food BAR [redacted]
11/23/08	8:15 A.	scab on right side cranial wound. Soaked and
		removed scab. Flushed both right and left
		cranial wounds with dilute betadine. Gave
		1 mL (50mg) Amoxicillin PO in small amt of
		wet cat food BAR Active [redacted]
	6:30 P	gave 1 mL (50mg) Amoxicillin PO in small amount
		of wet cat food, ate readily. BAR [redacted]
11/24/08	8:36	Gave 1.0ml Amoxicillin (50mg) PO with one
		tablespoon of wet food Normal feces and
		urine. (R) cranial wound scabbed and
		dry. (L) cranial wound closed with
		swelling, not able to expel discharge. Active
		and BAR. [redacted]
		Per MH, do not flush wounds or clean for
		today. [redacted]
	15:10	Gave 1.0ml Amoxicillin (50mg) PO with one
		tablespoon of wet food. Active and BAR. [redacted]
		Plan: continue BID Amoxicillin. Discuss w/ P.I. prognosis
		1 day/short term use. [redacted]
		As per [redacted], flush wounds and clean
		as needed [redacted]

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (GD7) SPECIES: Feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
11/25/08	8:15	Gave 1.0ml Amoxicillin (50mg) PO with one tablespoon of food. Flushed both Left and Right cranial wounds with dilute betadine. Bloody, purulent discharge from D cranial wound. Active + BAR. [redacted]
	16:00	Gave 1.0ml Amoxicillin (50mg) PO with one tablespoon of wet food. Active + BAR. [redacted]
11/26/08	8:35	TEMP. = 101.0°F. Gave 1.0ml Amoxicillin (50mg) PO with one tablespoon of wet food. D cranial wound mostly closed with tiny amount of purulent, red discharge. D cranial wound open, flushed with dilute betadine. Skin surrounding D cranial wound appears irritated, redness apparent. Expelled purulent discharge. Animal is eating and drinking normally. Active and BAR. [redacted] CONTINUE AMOXICILLIN BID. FLUSH/CLEAN LESIONS WITH SALINE SID + APPLY TRIPLE ANTIBIOTIC OINTMENT SID x 5 DAYS, THEN REASSES. [redacted] DVM
	14:30	Gave 1.0ml Amoxicillin (50mg/ml) PO with small amount of wet food, ate readily. BAR, resting in cage. [redacted]
11/27/08	830	Left cranial wound - enlarged bump ~ 1½cm, when palpated - large amount of purulent & bloody discharge evacuated. Right cranial wound - small amount purulent discharge. Flushed both with sterile saline. applied TAD to both - seems to be helping prevent scab formation. BAR, Active, Gave 1.0ml Amoxicillin (50mg/ml) PO, in sm. amt wet food - ate readily [redacted]
	510pm	Gave 1.0ml Amoxicillin (50mg/ml) in small amount

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S- 0127

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (G07) SPECIES: feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P, System and sign each entry)
11/27/08	Cont.	wet catfood - ate readily; Small amount of crusting around both cranial wounds, BAR, Active [redacted]
11/28/08	845AM	Gave 1.0ml Amoxicillin (50mg/ml) PO in small amount catfood (wet) - ate it all immediately. Cranial head wounds and top of head med. amount of red/yellow crusting - Chlorhexidine Scrub & Sterile saline used to clean top of head from debris, both open wounds (L) & (R) flushed with Sterile saline. Small amount yellow purulent discharge evacuated from (R) side, mod amt yellow purulent blood discharge from (L) side, Small ~ 1/8cm purple bruise on medial side of (L) wound, (L) periorbital tissue near wound swelling and redness decreased. BAR, Active, normal stool/urine [redacted]
	1515	50mg AMOXICILLIN GIVEN PO MIXED WITH CANNED FOOD. [redacted]
11/29/08	9:10AM	Gave 1.0ml Amoxicillin (50mg/ml) PO mixed with canned food; R & L cranial wounds and yellow/red crusting cleaned topically w/ Chlorhexidine Scrub Both wounds flushed with sterile saline and TAO applied topically [redacted] BAR, Active [redacted]
	445pm	Gave 1.0ml Amoxicillin (50mg/ml) PO mixed w/ wet food, BAR, Active [redacted]
11/30/08	9AM	Gave 1.0ml Amoxicillin (50mg) PO in wet food, cleaned top of head - large amount of drainage from (L) side - yellow/red crusting, flushed both wounds w/ sterile saline. BAR, Active. normal stool/urine [redacted]
	325pm	Gave 1.0ml Amoxicillin (50mg) PO in wet food, BAR, Active [redacted]
12/1/08	9:00	Gave 1.0ml Amoxicillin (50mg) PO in wet food. Cleaned and flushed wounds with sterile saline. Applied triple antibiotic ointment to both wounds. Mild discharge from left cranial wound, decreased redness around wound [redacted]

R0132

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (607 SPECIES: feline SEX: F

Date	Problem#	Symptoms, Diagnosis, Treatment (use S.O.A.P. System and sign each entry)
12/1/08	15:40	Gave 1.0 ml Amoxicillin (50mg) PO with wet food. [redacted]
12/2/08	8:30	cleaned cranial wounds, expelled purulent discharge from left cranial wound. Attempted to flush cranial wound with sterile saline, animal was not cooperating, small amount of flush did go into wound. Cleaned head and applied TAO to both wounds. Gave 1.0ml Amoxicillin (50mg) PO with small amount of wet food. Animal has good attitude. Active and BAR. [redacted]
	2:30pm	Gave 1.0ml Amoxicillin (50mg/ml) PO with small amt of wet food, top of head wounds dry - applied additional TAO, BAR, Active [redacted]
12/3/08	8:15am	Cleaned cranial wounds topically with chlorhexidine scrub & sterile water, removed scabbing & crust, expelled mod amt of bloody purulent discharge from left side. Attempted flushed of wounds with sterile saline - animal not cooperating, TAO applied topically to both wounds. Increased crusting & discharge from left side of head cap. Gave 1.0ml Amoxicillin PO (50mg) in small amt wet food. alert/acute, BAR, Active, Normal urine, no feces in box. [redacted]
①	9:00am	Weighed [redacted]
~~12/4/08~~ ①	12/3/08	3:30pm: Gave 1.0 ml Amoxicillin PO in small amt of wet food (50mg given). BAR, Active [redacted]
12/4/08	8:10am	Cleaned cranial wounds topically with chlorhexidine scrub & flushed with Sterile Saline, mod. amt of purulent discharge from (L) side. Rt side - no discharge. Applied TAO to both wounds. Gave 1.0ml Amoxicillin PO (50mg) in sm. amt wet cat food. BAR, Active [redacted]

① During dose [redacted]

R0131

TREATMENT AND PROGRESS RECORD

Laboratory Animal Resources
University of Wisconsin Medical School
Madison, WI 53792

ID# Double Trouble (G07) SPECIES: feline SEX: F

Date	Problem#	Symptoms, Diagnosis; Treatment (use S.O.A.P. System and sign each entry)
12/4/08	3:20pm	Gave Amoxicillin (50mg) 1.0ml PO in small amt wet cat food; BAR, Active. [redacted]
12/5/08	8:20	Cleaned cranial head of scabs. Expelled purulent discharge from left cranial wound and flushed with small amount of sterile saline. Redness increased around left cranial wound. Applied TAD to cranial wounds. Gave 1.0ml Amoxicillin (50mg) PO with wet food. Active and BAR. [redacted] More sensitive to touch. [redacted]
12-5-8	Note	2-28 Met with [redacted] / [redacted] regarding prognosis. Implant not working. Infection not responding to tx. Per. [redacted] terminal. [redacted] will schedule for cardiac infusion by next week latest.

Final Disposition (Please answer all questions below)

FOR INVESTIGATOR USE

What was the animal euthanized with (include dosage and route of administration)? 80 mg)
Sodium pentobarbital (5 cc, i.v.) following ketamine (~~xxx~~ to induce sedation.

Reason for Euthanasia:

- ☐ End of Study Per Protocol
- ☒ Other (please explain): her cochlear implant was no longer working and she developed a chronic infection

Was the animal submitted for Necropsy? No (Veterinary staff will attach to record)

Date 12/5/08 Sign [redacted]

FOR VETSTAFF USE

☐ ____ Check and initial when necropsy is completed and attached

S- 0126

Exhibit B

UW-Madison IACUC-Approved Protocol
"Behavioral and Physiological Studies of Sound Localization"

CONFIDENTIAL

emailed
OCT 21 2008

Revision 11/99
Protocol Code: [redacted]

Dog 7.28.08
RESEARCH ANIMAL RESOURCES CENTER
Renewal due: 11.30.08

UNIVERSITY OF WISCONSIN - MADISON ANIMAL CARE AND USE PROTOCOL REVIEW FORM

Forms should be typed or in computer-printed format. IBM & Macintosh word processing diskettes are available through Research Animal Resources Center (RARC) or the form can be downloaded via the RARC homepage: http://rarc.wisc.edu/

Return Completed Forms to RARC, 396 Enzyme Institute, 1710 University Ave, Madison, WI 53705

Office Use Only
Committee Action: _____

Veterinarian Signature _____ Date: _____

Chairperson Signature: _____ Date: _____

Type of Protocol Procedure: _____

Type of Surgical Procedure: _____

_X_ Survival Surgery _X_ Non-Survival Surgery ____ Rodent Surgery ____ Non-rodent Surgery

_X_ Multiple Major Survival Surgery ____ Exercise Exemption ____ Paralytic Agents ____ Restraint

____ Critical Veterinary Care _X_ Fluid/Food Restrictions ____ Nonstandard Housing ____ Nonstandard Husbandry

____ Occupational Health/Personnel Safety ____ Class B Dog/Cat ____ Biohazards ____ Radiation

____ Enrichment Exemption

NOTE: ALL PROTOCOLS ARE VALID FOR THREE (3) YEARS FROM DATE OF APPROVAL.

1. Principal Investigator/Project Director: [redacted] C
   Telephone Numbers: Office: [redacted] Lab: [redacted] Animal Emergency: [redacted]
   Home: [redacted] Fax: [redacted] E-mail Address: [redacted]

If Investigator is Unavailable for Emergency

Alternate to Contact With Authority to Act in Investigator's Absence: [redacted] C
Alternate Office Phone [redacted] Alternate Emergency Phone: [redacted]

2. University Department (of PI): *Physiology* Office Address: [redacted]
   Unit & Division Number (UDDS): *A 53 5400*

3. Is this protocol a *RENEWAL* application? (Circle appropriate category)

   If Renewal or Amendment application, please give current protocol code: *A 53 5400* [redacted]

4. Is this protocol for: *RESEARCH*; (indicate research type) *BIOMEDICAL*; *BEHAVIORAL*;
   Circle all that apply.

5. *Title: Behavioral and physiological studies of sound localization*

6. Classification of Research Animal Use (See attached schedule; circle highest category applicable) ~~1 2~~ 3 ~~4 5~~ D

7. Will ANY surgery be performed on any of the animals? *YES* If yes, you must fill out questions 24-30.

R0604

Will you be working with wild-caught animals? *NO* If yes, you must fill out questions 31-34.
Will you be using non-human primates? *NO* If yes, you must fill out questions 35 & 36.

8. Except for surgery (see question 25) will any procedures (e.g. blood collection, injections, euthanasia, etc.) be conducted in labs or will animals be housed outside of their normal animal housing areas? *YES* If YES, indicate building and room numbers and anticipated length of time away from normal housing area(s): *For the chronic experiments, the cats are brought to room [redacted] for the 2-4 hours needed for behavioral training or recording on each day. In cases when there is a minor procedure needed where the cats need to be lightly anesthetized for 5-10 minutes (e.g. repairing the dental acrylic head cap or to remove a broken eye coil) with ketamine hydrochloride (15-20 mg/kg) and acepromazine (.2 mg/kg) and atropine methyl nitrate (0.15 mg/kg) or atropine sulphate (0.04 mg/kg), the repair is done in room [redacted]. The vet staff will be notified when these minor procedures will be done. All major surgeries are done in the surgical suite ([redacted]), see question 25. For the acute experiments, the surgery and recordings are done in room [redacted] and the cats are kept there for the duration of the experiment. For each cat involved in chronic experiments, one trip to the [redacted] for an MRI will be made. The animal care staff of Laboratory Animal Resources of the Medical School will provide transportation for moving the animals outside of the [redacted] Lab staff will be responsible for all other animal transports within [redacted] following LAR procedures for transport within hallways with public access ([redacted]).*

For the new cochlear implant studies, we will need to confirm how far up the cochlea the implant is. One to two weeks after cochlear implantation we will obtain an x-ray to verify the position of the implants on the two sides. Cats will be transported to Room [redacted] or Room [redacted] at [redacted] contact person [redacted]) by animal care staff of Laboratory Animal Resources of the Medical School. Cats will be anesthetized for the procedure with ketamine hydrochloride and acepromazine with atropine, and monitored as previously described for the MRI procedure.

9. NOTE: NUMBERS OF ANIMALS REFERS TO THE TOTAL NUMBER OF ANIMALS THAT ARE ANTICIPATED TO BE USED DURING THE ENTIRE THREE YEAR LIFE OF THIS PROTOCOL.

a) Species of Animal:	Total Number For 3 yr.:	Source of Animals (e.g. commercial, U.W. breeding colony, or list other):
(#1) *Cats*	*30*	Class A dealer [redacted] *through UW Animal Care, or other investigators*

b) Will any of the animals be obtained from Class B dealers? (dogs & cats only) *NO*

c) Have any of the animals from above been part of any other protocols. *YES* If YES, how have you determined that the previous use will not compromise the animal's health and proposed current research.
Animals received from other investigators will be experimentally naïve or the prior use will not compromise my research or the health of the animals.

10. Building(s) or facility where the animals will be housed (normal housing).
Animal Care Facility in [redacted]

11. a) Outline the specific scientific goal(s) and significance of this research in straight-forward, non-medical, non-technical language that would be understandable to a layperson.

The overall aim of this project is to understand the neural mechanisms of binaural interaction, in particular in relation to sound localization. We are trying to dissect the physiological and anatomical characteristics of the neural circuitry that encodes the cues used for this important sensation. These results will help us to understand how the brain integrates auditory information from the two ears. This circuitry plays a major role in our ability to detect signals in the presence of background noise, which is the major symptom of elderly people with hearing loss. Understanding the neural mechanisms will help in the design of hearing aids and therapy while the cochlear implant project will improve our understanding of cochlear implants in human patients.

11. b) Provide a justification for the use of animals for this research. Indicate why it is imperative to use animals for this research and explain why alternatives such as computer simulation or in vitro systems are not possible.

We have considered alternatives to the use of live animals in our studies. In fact one of our goals is to produce a computer model of the neural circuitry involved in this important sense. Models do help us to refine our questions, which in turn reduces the number of animals needed. However, in order to generate realistic computer models and to test their adequacy requires physiological data that can only be obtained from recordings made in animals. We simply do not understand the mechanisms of the brain well enough to simulate it with computer models yet.

Will you be working with wild-caught animals? *NO* If yes, you must fill out questions 31-34.
Will you be using non-human primates? *NO* If yes, you must fill out questions 35 & 36.

8. Except for surgery (see question 25) will any procedures (e.g. blood collection, injections, euthanasia, etc.) be conducted in labs or will animals be housed outside of their normal animal housing areas? *YES* If YES, indicate building and room numbers and anticipated length of time away from normal housing area(s): *For the chronic experiments, the cats are brought to room [redacted] for the 2-4 hours needed for behavioral training or recording on each day. In cases when there is a minor procedure needed where the cats need to be lightly anesthetized for 5-10 minutes (e.g. repairing the dental acrylic head cap or to remove a broken eye coil) with ketamine hydrochloride (15-20 mg/kg) and acepromazine (.2 mg/kg) and atropine methyl nitrate (0.15 mg/kg) or atropine sulphate (0.04 mg/kg), the repair is done in room [redacted] The vet staff will be notified when these minor procedures will be done. All major surgeries are done in the surgical suite [redacted] see question 25. For the acute experiments, the surgery and recordings are done in room [redacted] and the cats are kept there for the duration of the experiment. For each cat involved in chronic experiments, one trip to the [redacted] for an MRI will be made. The animal care staff of Laboratory Animal Resources of the Medical School will provide transportation for moving the animals outside of the [redacted] Lab staff will be responsible for all other animal transports within [redacted] following LAR procedures for transport within hallways with public access (i.e. carrying a cell phone with the number for emergencies at all times).*

For the new cochlear implant studies, we will need to confirm how far up the cochlea the implant is. One to two weeks after cochlear implantation we will obtain an x-ray to verify the position of the implants on the two sides. Cats will be transported to Room [redacted] or Room [redacted] at [redacted] contact person [redacted] by animal care staff of Laboratory Animal Resources of the Medical School. Cats will be anesthetized for the procedure with ketamine hydrochloride and acepromazine with atropine, and monitored as previously described for the MRI procedure.

9. NOTE: NUMBERS OF ANIMALS REFERS TO THE TOTAL NUMBER OF ANIMALS THAT ARE ANTICIPATED TO BE USED DURING THE ENTIRE THREE YEAR LIFE OF THIS PROTOCOL.

a) Species of Animal:	Total Number For 3 yr.:	Source of Animals (e.g. commercial, U.W. breeding colony, or list other):
(#1) *Cats*	*30*	Class A dealer, Class A dealer, *through UW Animal Care, or other investigators*

b) Will any of the animals be obtained from Class B dealers? (dogs & cats only) *NO*

c) Have any of the animals from above been part of any other protocols. *YES* If YES, how have you determined that the previous use will not compromise the animal's health and proposed current research.
Animals received from other investigators will be experimentally naïve or the prior use will not compromise my research or the health of the animals.

10. Building(s) or facility where the animals will be housed (normal housing).
Animal Care Facility in [redacted]

11. a) Outline the specific scientific goal(s) and significance of this research in straight-forward, non-medical, non-technical language that would be understandable to a layperson.

The overall aim of this project is to understand the neural mechanisms of binaural interaction, in particular in relation to sound localization. We are trying to dissect the physiological and anatomical characteristics of the neural circuitry that encodes the cues used for this important sensation. These results will help us to understand how the brain integrates auditory information from the two ears. This circuitry plays a major role in our ability to detect signals in the presence of background noise, which is the major symptom of elderly people with hearing loss. Understanding the neural mechanisms will help in the design of hearing aids and therapy while the cochlear implant project will improve our understanding of cochlear implants in human patients..

11. b) Provide a justification for the use of animals for this research. Indicate why it is imperative to use animals for this research and explain why alternatives such as computer simulation or in vitro systems are not possible.

We have considered alternatives to the use of live animals in our studies. In fact one of our goals is to produce a computer model of the neural circuitry involved in this important sense. Models do help us to refine our questions, which in turn reduces the number of animals needed. However, in order to generate realistic computer models and to test their adequacy requires physiological data that can only be obtained from recordings made in animals. We simply do not understand the mechanisms of the brain well enough to simulate it with computer models yet.

c) Provide justification for why you have chosen the species cited in 9-A for your work.

We use cats (about 15/year, of which 10 are for acute[though we have not done any acute experiments in several years so we anticipate the number needed to drop] and 5 are for chronic experiments)) for the following reasons: the physiological, anatomical and psychophysical characteristics of their auditory system are very similar to those of humans and higher primates (which makes it likely that our results are also applicable to humans), their auditory system has been extensively studied by others such that most of our understanding of auditory physiology derives from studies in the cat, the relevant parts of their brain are relatively easily accessible, and they are not endangered or in short supply. . The other animal species that have been extensively used in studies of sound localization are guinea pigs, gerbils, chinchillas, and barn owls. The rodents are not good models for studies of localization because the behavioral evidence indicates that their localization acuity is considerably less than that of predators like the cat, barn owl or human. For prey, they need only determine the general direction of a sound source, not its precise location. Barn owls are not good models since they are so highly specialized (they are the only animals known to phase lock to frequencies above 3-4 kHz and they do not move their eyes) that results may not apply generally to other animals.

12. Explain how the number of animals required was determined and justify that need. Include control animals in this discussion.

We have been studying the auditory system for over 30 years here at UW-Madison and the number of animals requested represents an average taken over a number of years. We make an extensive effort to gather as much data from each animal as possible; the acute experiments may run continuously for up to 3-5 days while the chronic cats are kept for many months, even years. Ultimately, the number of cats needed is determined by the scientific aims of the experiment and is governed by many different considerations: a large number of neurons need to be sampled in order to gain statistical viability and to meet the demands of critical reviewers for our manuscripts, there are practical limits to the number of neurons we can study in each cat as it takes an hour or more to characterize each neuron, and every experiment does not work for many different reasons. It is not possible to state how many neurons are required to reach statistical viability since that depends upon the questions that we are addressing and the differences we see between different neurons in any given experiment. For example, if all the neurons consistently show a strong effect, then relatively few neurons are needed to reach significance and therefore few animals are needed; but if there is considerable variability between the neurons then many more cells are required to demonstrate the presence or absence of an effect, i.e. in a t-test. Thus, it is not only more practical but also more realistic to justify the number of animals based upon past experience. While we have averaged about 30 cats/year, this number is quite variable, depending upon the experiments that are being done at any given moment. This number allows us to collect enough data to keep up a productive publication record that ensures our constant funding from NIH over these 30 years. We anticipate reducing the number of acute experiments in the coming years so our estimate of animals needed is lower. We endeavor to use as few animals as possible; and the chronic experiments allow us to use the same animal many times.

13. Indicate any current or pending funding for this project:
Funding Source (1): *N.I.H.* Grant Number (1): [redacted]
Title of Grant (1): [redacted]

Funding Source (2): *N.I.H.* Grant Number (2):
Title of Grant (2):

14. Identify the person(s) or unit responsible for **daily** animal care:
Animal Care Facility

15. Personnel working with animals: Everyone must take the "Responsible Use and Care of Laboratory Animals" exam or course. Protocols cannot be processed until PI and all personnel are certified. For information, call RARC 262-1238.

All of the personnel have been certified by the guidelines of the ACUC of the University of Wisconsin-Madison. New personnel in the lab are trained by first observing surgeries, the daily handling of animals, and husbandry. After they have watched a number of surgeries and the daily procedures, they are slowly initiated into doing the tasks under careful guidance of the PI and other experienced personnel. [redacted] and [redacted] will be responsible for training and supervision of the new students while they are learning the surgery. All chronic sterile surgeries are performed in a dedicated surgical suite ([redacted]).

Last name, first name	Phone Number	Type and length of training/experience for animal use
[redacted]	[redacted]	[redacted]

[redacted] and [redacted] will be coming to Madison in February, 2009 to help us do the cochlear implant surgery. They will be here for less than 30 days (2-3 days during the surgery and 2 days when the implants are turned on). They will be doing the surgery under the direct and constant supervision of [redacted] and [redacted]

16. a) Give a brief summary of the methods and sources you use to keep current with pertinent information in your field in order to assure that alternatives to the use of animals have been considered, this work is not duplicating existing knowledge, and that the procedures are the least stressful to animals. If electronic databases are utilized, include sources, date of search, years covered by the search, and key words and/or search strategy used. This information for electronic databases is required by USDA Policy #12. Full text of USDA Policy #12 may be viewed at: http://www/aphis.usda.gov/ac/policy12.html.

Since my laboratory is interested in the way in which the brain processes information, there is no real alternative to using animals for the physiological experiments. Recordings from human subjects are not feasible since the information we need requires invasive recordings and we don't know enough to gain understanding with computer models. We maximize the use of animals by doing our psychophysical experiments on the animals themselves rather than trying to extrapolate from human psychophysics to physiological responses in animals. I routinely monitor on a daily basis a wide variety of sources for information in the literature related to the problems of interest. I have personal subscriptions to the two journals that are most relevant to my field: Journal of Neurophysiology and Journal of Neuroscience. I also regularly read Hearing Research, Science, Journal of Physiology (London), Nature, Journal of Comparative Neurology and Trends in Neuroscience, through subscriptions and copies available on the internet. In addition I regularly use the facilities of the Medical School Library to search using ProCite, an electronic database and to consult Index Medicus, Biological Abstracts, and papers published in book chapters or other journals. In the most recent search in May, 2008, I used ProCite to search Medline for all years since 1966 under the following keywords: animal welfare, alternatives to animal testing, auditory attention, superior olive, inferior colliculus, precedence effect, Franssen effect, and sound localization. I attend national and international meetings in which the most recent findings in the field are reported, and I communicate with a world-wide network of auditory physiologists over E-mail to keep up with the latest developments.

We are always mindful of trying to limit the stress to our cats since our experiments cannot succeed if the animal is under stress and not cooperative. The potentially stressful and invasive procedures that we employ include: 1) food deprivation for operant conditioning, 2) head restraint, 3) eye coil implantation for measuring eye movements, 4) microelectrode insertion into the brain, and 5) denervation of the ear muscles. For each of these procedures we have endeavored to minimize the stress. 1) Following a PubMed search on March 5, 2005 using the key words "cat" "sound localization" we found a reference (Malhotra et al., J. Neurophysiol., 92: 1625, 2004) in which cats were not food deprived but rather worked for canned cat food while having dry cat food ad lib following the training. We are presently piloting experimental procedures to see if we can implement this procedure so that the cats are not food deprived when they are in their cages. 2) Restraining the head of the cat is clearly stressful as the cats will usually struggle against the restraint until they learn that they cannot move their heads. To alleviate this stress, as well as for sound scientific reasons, we have now moved to an unrestrained head preparation for most of our experiments. This has made the cats much more relaxed and interestingly their localization accuracy has improved markedly. However, for some of our procedures it is still necessary to have the head restrained so this part of the protocol has been retained. 3) The use of the scleral search coil is a standard technique used by all of the eminent labs studying oculomotor function in cats or primates. However, it is invasive and requires surgery to implant and often the coils have to be replaced because of breakage after the many months of use. Therefore alternative methods could be advantageous. However the other common method relies on visual reflective techniques that are not adaptable to situations where the head is not restrained. For unrestrained heads the other common technique is electro-oculograms but these are very much inferior to the search coil technique that we use in terms of accuracy of measurement and it is no better in terms of surgical requirement. 4) There are not many alternatives to insertion of microelectrodes given that our experiments require recording from neurons in the brain. We are piloting experiments to use a multichannel microelectrode probe to allow us to record from many neurons simultaneously which will decrease the amount of time that we need to record from each animal. 5) The denervation of the ear muscles is not stressful, but does require a surgical procedure. Before resorting to this we tried anesthetizing the ear muscles but this procedure was very stressful to the cats, who do not like to have their ears touched. We decided that denervation was much less stressful than the alternatives. The last PubMed search was done on May, 2008 and covered the last 10 years (1998-2008)..

In summary the proposed work does not duplicate existing knowledge, alternatives to the use of animals do not exist and the

[redacted]

An undergraduate hourly worker, no previous animal experience will be trained by [redacted] has already been added to the protocol
An undergraduate hourly worker, no previous animal experience will be trained by [redacted], has already been added to the protocol
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Drs. [redacted] and [redacted] of the [redacted] will be coming to Madison in February, 2009 to help us do the cochlear implant surgery. They will be here for less than 30 days (2-3 days during the surgery and 2 days when the implants are turned on). They will be doing the surgery under the direct and constant supervision of Dr. [redacted] and [redacted]

16. a) Give a brief summary of the methods and sources you use to keep current with pertinent information in your field in order to assure that alternatives to the use of animals have been considered, this work is not duplicating existing knowledge, and that the procedures are the least stressful to animals. If electronic databases are utilized, include sources, date of search, years covered by the search, and key words and/or search strategy used. This information for electronic databases is required by USDA Policy #12. Full text of USDA Policy #12 may be viewed at: http://www/aphis.usda.gov/ac/policy12.html.

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In summary the proposed work does not duplicate existing knowledge, alternatives to the use of animals do not exist and the

proposed procedures are designed to minimize the stress to the animals.

b) Radiation or Biohazard Material Usage In Animals: mark all that apply, indicate specific material used, and show status (approved or pending) of Biological Safety (OBS-2) and/or Radiation Safety (99A) protocols.

Category	Specific Material(s)

_____ Recombinant DNA
_____ Genetically Altered Materials
_____ Infectious Agents
  _____ Bacteria
  _____ Virus
  _____ Prion
_____ Carcinogen or Mutagen
_____ Toxic Agent
Status of OBS-2: (circle one) APPROVED PENDING
_____ Radioactive Material
Status of 99-A: (circle one) APPROVED PENDING

Occupational Health & Safety: If you are using any agent that could be hazardous to humans or animals please provide any special precautions that should be followed by your lab personnel, animal caretakers, veterinarians, maintenance and/or sanitation personnel, or anyone else entering the areas where experiments are conducted or animals are housed. Include any special practices required for handling of any animal or experimental waste, animal carcasses, and cages and caging materials. Consider such requirements as: masks or respirators, eye protection, lab coats, gloves, and disposal methods.

Lab coats or dedicated uniforms are needed to enter the cat holding rooms.
The primary health and safety concern is an occasional obstreperous cat who scratches or bites one of our lab personnel. This is rare since we handle the cats daily and feed them when we bring them to the lab, but it has happened when the cat is acclimating to the lab or during a medical procedure. All lab personnel are aware of the potential dangers and in the case of bites or bad scratches we follow the LAR policy #206: the wound is cleansed immediately for 15 minutes and the incident is reported immediately to the LAR and veterinary staff. The person will go to the UW Health Service or Hospital emergency room for immediate treatment. The LAR and veterinary staff will monitor the animal for 10 days following the incident. We typically do not wear masks, gloves or lab coats when handling the cats since they are usually very friendly and receptive to us.
The only hazardous material that we use is 10% formalin on the rare occasions when we perfuse an animal when we want to keep the brain for histological purposes and need the results quickly. If there is no need for a quick perfusion or if there is no special histological stain, then we simply immerse the head and brain in formalin after the animal has been sacrificed. Perfusions are done under a hood with gloves, eye protection and lab coats.

You must address questions 17 separately for each species.

17. Experimental Protocol

a) In this section describe your experimental protocols, outside of normal husbandry, to be performed on the animals. **This response should provide the committee with a clear understanding of what specifically happens sequentially to each animal or group of animals and over what time period.** It is not necessary to repeat the surgical description that is provided in question 28, but the timing of the surgery within the experiment should be indicated. Be sure to include: all drugs given, including dosage range, routes and frequency of administration; nutritional intervention; social or environmental manipulation; method and amount of biological samples taken; methods of antibody production; use of radioactive materials, blood or other fluid sampling including method and amount, etc. Specify the expected sequence, frequency and duration of these procedures. **If this protocol is to cover an animal colony, use this section to detail breeding procedures/methods.** (Append additional page(s) if necessary)

Cats: Two different experimental preparations are used in the cat experiments: acute and chronic. Acute experiments are ones in which the cat is deeply barbiturate-anesthetized during the entire course of the experiment, which normally runs from 24 to as long as 120 hours, and is sacrificed at the conclusion. We will monitor the cat's anesthetic state about every hour (see #19 below) with a paw pinch and by monitoring the EKG signal for EMG activity. In addition we have a Pulse Oximeter machine (SurgiVet V3395) that makes it possible for us to monitor the heart and respiration rate. No additional care, outside of normal husbandry is needed for these cats.

In some chronic experiments, the aim is to train cats to localize sound sources using standard operant training techniques and then to record from neurons in the brain of the cats while they are actively performing the sound localization task. The

During this period no other procedures are done except to monitor the weight daily. At the end of this acclimation period we will consult with the vets to agree on the working weight, and the 'action weight' which is 85% of the working weight. If the cat's weight falls below the action weight, we will consult with the vets and monitor the cat's feeding. If necessary, deprivation is suspended until the weight returns to above the action weight. This regimen is maintained during the experimental phase while physiological recordings are done. Training sessions last 1-4 hours for 5 or 6 days/week and the cats are allowed an average of 75 (range of 60-120 depending upon the cat) gms (or ~175 cc or a range of 140-280 cc))/day of food over the weekend.. It is important to emphasize that in order for this project to succeed, the cats must be motivated and eager to perform. Thus, we take all possible steps to ensure that the cats are contented.

We have experimented on some of our animals with mixed success to see if we can reward the cat with highly desirable food (canned cat food possibly laced with tuna or other 'goodies') so that the cat would receive ad lib dry cat food at the end of the day. In this way the cat would not be food deprived, but rather food regulated, during the day. This regime has apparently been successfully used by another lab in [redacted], with whom we are consulting. If this is successful, then we should not have trouble with cat's falling below the action weight.

Once the head post and eye/head/pinna coils are implanted, we perform routine cleaning of the wound edge around the dental acrylic head cap of the cats once a week (or more if needed). Those with chambers or with the large craniotomies have their wound edge and chamber cleaned two or three times a week. To clean the wound edge we remove the scabs gently with Q tips. If there is some drainage, we may cleanse with hydrogen peroxide, rinse with tap water, and apply some triple antibiotic ointment if needed. The ointment is applied once per day for 5 days when needed. To clean the chamber, we fill the it with a solution of (24cc's gentamax (100mg/ml) in 1000ml 0.9% NaCl), let it soak several minutes, then suction out the fluid. We reseal the chamber with the wax.

In the grant [redacted] we will be studying selective auditory attention. This project is being done in collaboration with [redacted]. The basic protocol will be identical to those outlined above for the chronic experiments except that the recordings will be made from the primary auditory cortex rather than from the inferior colliculus. The cats will recover from the surgical procedure to insert the electrodes which will be done under isoflurane anesthesia. In our initial pilot experiments we will do this on untrained animals just to debug the electrode recording system. The behavioral task that the animals will be trained to do will also be different: cats will be working on a frequency discrimination task in which they are cued to attend to one side or the other using operant training as in our sound localization experiments. Our interest in these experiments is to probe the effect of behavioral attention on the responses of cortical cells. We plan to train the cats to attend to one of two speakers and to respond with an eye movement when there is a change in frequency in a train of tone bursts. We hypothesize that cortical cells will show increased sensitivity when the cat is attending to the speaker. The basic animal protocol is otherwise identical to our localization experiments.

In some experiments we need to measure the head related transfer function (HRTF) of the cats to determine the acoustic input to the ears. This will be done before euthanizing the cats. Cats are deeply anesthetized initially with ketamine (15-20 mg/kg) and acepromazine (.2 mg/kg) (see #27). Anesthesia is maintained with sodium pentobarbital administered IV through a venous cannula (1.14mm ID, 1.57mm OD) implanted into the femoral vein. The sodium pentobarbital is diluted (50:50) with saline and given as needed to maintain an areflexic state (usually about 10-20 mg/kg initially and 3-5 mg/kg/hr in the steady state). A tracheal cannula (3.0-4.5mm OD) will be installed to facilitate the animal's breathing. The cats are usually able to breathe naturally on their own, but we do have a ventilator on hand if breathing becomes difficult. Generally, the acoustical noise made by the ventilator interferes with the acoustic response properties of the auditory neurons we are trying to study, so a ventilator is used only when necessary. The acoustical responses in the ears of the cats and kittens to free-field sounds will be recorded by placing a small probe-tube microphone (3.1 mm in length, 1.5mm in diameter) into the ear canal via a small slit in the posterior portion of the pinnae.

Deafening procedures

In a new series of experiments currently being proposed, cats will undergo surgery in preparation for training of the chronic experiments as described above. They will recover and be trained for psychophysical experiments as normal, but once they have been fully trained, they will undergo a deafening procedure using one of the following procedures: 1) subcutaneous (SQ) or intramuscular (IM) injections of neomycin sulfate (~50 mg/kg), 2) a one-time combination of subcutaneously injected kanamycin (300-400 mg/kg) along with intravenous ethacrynic acid (10-25 mg/kg), or 3) direct injection of neomycin sulfate (2.5 ml of a 10 mg/ml solution) into scala tympani of the cochlea of both ears through the round window. The first two procedures will be done prior to the cochlear implant surgery while the third procedure will be done on the day of implanting the electrodes when the round window is exposed surgically. In the cases of procedures 1 and 2, the veterinary staff will be consulted before the antibiotic treatment is begun. For procedure #1, the injections and/or IV will be repeated daily for up to 21 days, and will be stopped once effective deafening of the cat has been determined. Careful restraint practice will be utilized for the regular IM injections. The cats will be restrained within nylon, zippered bags before the injection is administered for the safety of the staff. Injection sites will also be rotated in an effort to minimize the cat's discomfort. The level of deafness will be determined using both measurement of the auditory brainstem response

electrophysiological recording is done by inserting fine tipped microelectrodes into the brain through an intact dura. Since the training regimen usually takes several months and the recording phase also extends over many months, some of these cats have been in the lab for over a year. Because the dura hardens as soon as the skull is opened and this hardening limits the number of days we can penetrate it with the fine microelectrodes, multiple surgeries are required for this experiment. Typically, we begin by simply bringing a cat [redacted] into the lab and feeding it in the lab once a day for a couple weeks so it becomes accustomed to the personnel and to getting fed out of its own cage. Prior to surgery each cat will have an MRI to determine if the inferior colliculus is accessible to recording, or, if in that particular cat, it is covered by the bony tentorium. Cats will be transported to the [redacted] for the MRI scan by animal care staff of Laboratory Animal Resources of the Medical School.

For all of the surgical procedures described below, cats will be initially anesthetized for the procedure with intramuscular injection of ketamine hydrochloride (15-20 mg/kg) and acepromazine (.2 mg/kg), and given atropine methyl nitrate (0.15 mg/kg) or atropine sulphate (0.04 mg/kg), to suppress mucus. For minor procedures that only require anesthesia for less than 30 minutes this ketamine/acepromazine/atropine mixture will suffice. If the surgical procedure is longer than about 30 minutes (e.g. for the surgeries to implant head restraint device, eye or ear coils, or recording chambers), we insert an endotracheal tube to administer gas (isoflurane) anesthesia. The dose of isoflurane will be adjusted from about 2 to 3% as needed. Recently we have had some problems with a few cats that appear to be hypersensitive to the ketamine/acepromazine cocktail. On the advice of RARC vets we have used Midazolam (0.066- 0.22 mg/kg) and Oxymorphone (0.1-0.4 mg/kg) to induce the cats in order to do the MRI or to insert the endotracheal tube for gas anesthesia. As oxymorphone has become more difficult to obtain, after consultation with the vet staff we have been pre-medicating about 1/2 hour before induction with 0.1 ml (.03mg/ml) buprinorphine, IM, SQ, or sublingually. Induction can be achieved with an intramuscular injection of ketamine 5 mg/kg (100mg/ml) and medetomedine .25 mcg/kg (1mg/ml) mixed together. During the surgical procedure, we will observe and record heart rate, respiration rate, and body temperature about every 15 minutes from the time of induction until the animal has recovered from anesthesia and returned to its cage.

The entire MRI procedure takes about 30 minutes so it is usually done under ketamine/acepromazine/atropine. If the MRI indicates that the inferior colliculus is covered by the bony tentorium, we will not use that animal. If the inferior colliculus is accessible to recording, we will proceed with the following surgery several weeks later. We implant the head holder and eye coils needed for the experiment on its skull under deep anesthesia and aseptic conditions. Several months of daily training follows. When the cat is adequately trained, another surgical procedure is done to open the skull to allow microelectrode penetrations. It is not possible to specify exactly what constitutes an adequately trained cat since it depends very much on the type of experiment we are doing. Lately we have actually done a considerable amount of animal psychophysics once the cat is trained so this is very much a judgement call. This is then the beginning of the experimental phase during which we will have daily recording sessions with the cat while it is awake and behaving for as long as the dura is not too hard, usually 4 months or so. For some experiments we are interested in the role of the external ears in localization and we have a last surgical procedure in which the nerve supply to the external ear muscles are cut to prevent the ears from moving. In addition, on occasion we have to replace a broken eye or ear coil with another surgical procedure. As an example, from 1997 to 2004 years, we used 14 cats in the chronic experiments, 7 of which are still alive and running experiments. For these 14 cats we have averaged 109 days (minimum of 21 days) between eye coil replacements in the cases where a replacement was required. This does not accurately estimate the failure rate of coils since it does not include cats who were euthanized with no coil problem or those still running. Nonetheless it does provide a rough estimate of the expected time between coil replacement surgeries. If in any given cat this rate of coil replacement becomes more than double, we will consult with the vets to troubleshoot the problem.

For the initial surgery, the cats are implanted with a number of devices under anesthesia (see below) and then are allowed to recover from the surgery and trained on a behavioral task for a period of several weeks to months. The cats are trained by depriving them of food in their home cages and having them work for a food reward. The cats will be trained under computer control to look at a sound source presented in front of the animal. Initially the acoustic stimulus will be coupled with a visual one in order to train them to look at the source. With time the visual stimulus will gradually be removed. Upon completion of a successful trial the cat will be given a small amount of cat food in the form of a soft paste. Each day the cat will be expected to make 200-500 successful trials, but the number of successful trials varies considerably depending upon the difficulty of the task, the degree of training of the animal, and our goals in any given experiment.

The cats' weight and food consumption during the training sessions are monitored and recorded daily during weekdays to ensure that they are getting enough nutrition. To ensure that the cats are not overdeprived, we will establish a 'working weight' for each cat and make sure that their weight is at least 85% of this working weight. Since the cats usually arrive into LAR a bit overweight, we will establish each cat's working weight in consultation with the veterinarians after an initial period of acclimation to the lab. Usually this entails a few weeks in which we restrict the cat's access to food to the period when it is brought down to the lab. The cat will be provided with food ad lib during this period and we will monitor the amount of food that each cat eats to determine how much it needs to maintain a given weight. This also provides an opportunity for the cat to acclimate to handling and being put into the bag.

(ABR) or by psychophysical determination. The ABR recording will be performed once before the surgery to gather baseline data on the hearing cat, and again 10 days into the deafening procedure to test for any residual hearing. If there is residual hearing, neomycin injections will continue and another ABR recording will be performed after 4-5 days. During ABR recordings, cats will be anesthetized with Acepromazine (.2 mg/kg SC) and Ketamine (15-20 mg/kg IM). The cat's head and back between the scapula may need to be shaved to provide better electrode access. Three sterile #27 gauge needles will be inserted subcutaneously at the base of the neck, ear, and in the torso for recording purposes. Vital signs will be monitored every 15 minutes for the duration, as we do for all other minor procedures in the lab. This procedure typically takes about an hour, and will be performed in room 290 MSC. Additional doses may be, but are not likely to be, necessary if effective deafening has not yet been achieved. Due to the fact that both neomycin and kanamycin are nephrotoxic, renal function will be monitored with regular blood creatinine and/or blood urea nitrogen (BUN) tests every other day after the treatment has begun and continued as needed based on clinical observation and upon consultation with the veterinary staff. Urine test strips may also be used in conjunction with or instead of the blood tests upon consultation with the veterinary staff. In addition we will assess renal function and overall health of the cats selected for these experiments by doing blood work (chemical profile and CBC) prior to surgery. The use of either neomycin or kanamycin is a standard deafening procedure in the literature, and there are several sources that report using either neomycin(Beitel et al., J. Neurophysiol., 83: 2145, 2000; Klinke et al., Science, 285: 1729, 1999; Miller et al., Hearing Res., 92: 85, 1995) or kanamycin (Beitel, et al., 2000) that perform chronic experiments and do not note any problems with kidney failure. One paper notes specifically that up to three days after a deafening procedure using kanamycin with ethacrynic acid there were normal BUN and creatinine levels in the cat (Xu et al., Hearing Res., 70(2): 205, 1993). Along with the use of ethacrynic acid, there is also the risk of water and electrolyte imbalances, and so regular serum chemistries will be used to monitor the cat. During the period of antibiotic treatment the cats will be carefully monitored by 1) daily clinical monitoring of behavior and overall health and 2) measurement of BUN and other blood parameters by the veterinary staff at least 3 times per week during the first 2 weeks of neomycin administration, with additional measuring to be performed as needed based on test results and clinical appearance. The level of deafness will be determined using measurement of the auditory brainstem response (ABR). If the deafening procedure with antibiotics proves to be problematic, we will explore other well-known methods for deafening, such as noise exposure.

Although the literature reveals the use of systemically injected ototoxic antibiotics, [redacted] has presented the idea of topical application of the antibiotics as a lower-dose alternative. The third procedure mentioned above represents this alternative to systemic neomycin applications by direct injection into the cochlea done at the time of implant The cochlear implant study is currently a pilot study, and if it is later discovered that topical application does not cause any problems that would be inherent in the proposed experiment, we are open to using a protocol suggested by the veterinary staff for future cats. We are also open to consultation with veterinary surgeons such as [redacted] or [redacted] before conducting the surgery.

After the deafening procedure has been completed, the cat will then undergo a surgery to implant bilateral cochlear implants. Cochlear implants, which will be cat-sized electrode arrays, will be inserted into both ears (see Question 28 for full surgery description). Care will be taken to achieve the same insertion depth on both sides, and the function of the stimulating electrodes will be determined before the end of the surgery. The cat will then undergo the standard recovery process before it is retrained behaviorally, also according to standard procedure. The cat's electrodes will be connected to a signal processor and stimulated in a manner analogous to that of human cochlear implant wearers during the new behavioral experiments. Measures will be taken to ensure that levels of stimulation are within the normal hearing range and are not causing the cat visible discomfort. Later, the cat will also undergo the surgery to allow for neuronal recording. Multiple surgeries are necessary for this experiment since we will need one surgery to implant the eye coils needed for training, another to implant the cochlear implant after it is trained, and a third to implant the recording chamber for neuronal recordings. There will be at least several months between the initial implantation of the eye coils and the cochlear implant surgery. Then, there will be at least several more months between the cochlear implant surgery and the implantation of the recording chamber.

b) Do any animals undergo any type of restraint beyond normal housing methods? *YES* If YES, indicate method, length of restraint, and justification for such restraint. If the design of the study requires continuous restraint for longer than 12 hours without the opportunity for exercise, be sure the justification addresses need for such an extended period and include the maximum length of time the animals will be restrained. Include any plans for providing additional enrichment and any steps taken to avoid physical discomfort during the restraint. (See Campus Policy on Non-human Primate Chairing if applicable - available on the web at: www.rarc.wisc.edu

In some of the chronic cat experiments, the cats are restrained by holding their heads in a fixed position by clamping on the head holder that has been implanted on the skull. For all chronic experiments the cat is placed into a nylon bag to restrain its movements during training and recording. The cat can freely moves its limbs in the bag but cannot ambulate. The head holder and head restraint is needed for those experiments in which we need to maintain the head in a known position since the position of the head relative to the acoustic stimulus is the primary variable in the acoustic cues for sound localization. A typical training or recording session will last from 1.5 to 3 hours, after which they are allowed to run free in the lab. They are never restrained over 4 hours. If the animal shows any discomfort at any time during the training or recording period, the experiment is immediately

terminated for the day since success of the training and recording depends on a cooperative animal. Therefore we go to great lengths to ensure that the cats are not in discomfort.

c) Are any animals subjected to fluid or food restriction? *YES* If YES, discuss level of restriction, expected consequences, and justification for such restrictions

For the chronic cat preparation, we control access of food to the animals in their home cages for the period that they are being trained and tested and feed them only during the behavioral training and testing in the lab. This is necessary to motivate them to do the behavior. If they are not food deprived then they will not perform the behavioral task. As described above in section 17a, the cats' weight and amount of food consumption during the training sessions are monitored daily to ensure that they are getting enough nutrition. Typically during daily training, the cats earn enough food rewards to satiate themselves. Training sessions last 1-4 hours for 5 or 6 days/week and the cats are allowed 100 gms/day of food over the weekend. If the body weight falls below 85% of the established normal 'working weight', controlled-access feeding is suspended until the weight is regained. Weight is recorded for each training and recording session. We consult regularly with the lab animal veterinarians regarding the food deprivation of each animal and ensure that their weight has not fallen below the 'action weight'. We have had cats maintained in excellent health under these controlled food access conditions for up to 3 years with no untoward effects. It is also not uncommon for us to have a hiatus in the training or testing of any given cat for weeks or even months. During the hiatus, cats are given food ad lib.

d) Will any animals require nonstandard husbandry exemption (e.g. exercise exemption, extended cage cleaning periods, etc.)
NO If YES, indicated nonstandard husbandry required and justification for this practice.

18. For animals experiencing more than momentary or slight pain or discomfort as a result of your procedure(s), describe what you will do to relieve this discomfort and assure that no animal experiences undue pain or distress during the course of your research. Include drugs, dosages, nursing care, mechanical devices, humane euthanasia, etc. If you do not believe animals will experience any momentary or slight pain, provide explanation for that belief.

See item 29b for post-anesthetic procedures

19. Describe how frequently and how you will monitor your animals to insure they are not experiencing pain or discomfort from your procedures or from unanticipated illness or injury. Include criteria when euthanasia would be utilized for dealing with the unanticipated illness or injury not necessarily directly related to your research.

Acute: while we would like to monitor our acute cats every 10-15 minutes, this is unduly disruptive to the experiments. These cats are placed in a sound proof room so that they hear only the sounds that we deliver to them in our acoustic experiments. This is an essential ingredient of our experiments. Recording from single neurons is a delicate procedure, which can easily be disrupted if the animal is touched or even by walking into the chamber and recordings can last more than one hour. Therefore, we check the animal about every hour between recordings by areflexia to a painful paw pinch. Most cats under deep barbiturate anesthesia only need additional doses every 4 hours or more, though there is considerable variability in this time from cat to cat. To allow us to monitor the cats from outside the chamber, we will attach EKG leads and monitor and document the signal for movement EMGs or changes in heart rate using a pulse oximeter..

Chronic: since these cats are awake and behaving during the training and experiments, they quickly let us know if there is any discomfort or pain. In order for these experiments to succeed, it is essential that the cats are happy so much of our efforts are directed towards assuring that they are not in discomfort.

If there is an unanticipated illness or injury the lab animal veterinarian is contacted for treatment.

20. If experiments could induce chronic disease, tumors or radiation sickness, describe the specific criteria for termination of the affected animals. This description should be detailed enough so as to indicate such things as tumor size, specific animal characteristics or behaviors, weight loss criteria, clinical signs, etc.
NA

21. Describe the methods of euthanasia used, including drugs, dosage, and any sedation and provide necessary justification as necessary. Euthanasia methods must follow the Report of the American Veterinary Medical Association (AVMA) Panel on Euthanasia (2000).
NOTE: Even if euthanasia of animals is not part of this project, complete this section to provide direction in cases of unanticipated illness or injury.

All cats are euthanised by an IV overdose (100-150 mg/kg bw) of sodium pentobarbital. Death is confirmed by the cessation of respiration followed by cessation of the heart beat.

22. If the animals are not euthanized at the end of the study, what will happen to them?

Animals may be transferred to other investigators with the approval of a veterinarian.

23. Will any animal products be used for human consumption? *NO*
If YES, list any drugs to be given to the animals, and their withdrawal times before consumption:

I plan to follow the provisions for the care, use and treatment of animals found in the NIH "Guide for the Care and Use of Laboratory Animals," or the "Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching". I assure that these procedures do not unnecessarily duplicate previous experiments.

Signature of PRINCIPAL INVESTIGATOR/PROJECT DIRECTOR:_____

Attending Veterinarian:_____
(SVM ONLY)

INVESTIGATOR SIGNATURE:

To the best of my knowledge, I certify that the information provided in this Animal Care and Use Protocol is complete and accurate. I understand that approval must be renewed annually, that every third year the ACUC must perform a new review of my protocol, and that I might be required to complete a newer version of the Animal Care and Use Protocol and provide additional information at the time of the triennial review.

I also understand that ACUC approval must be obtained by an amendment to this protocol before I:

- Use additional animal species, increase the number of animals used, or increase the number of procedures performed on individual animals;
- Change procedures in any way that might be considered a significant departure from the written protocol;
- Perform additional procedures not described in this Animal Care and Use Protocol;
- Allow other investigators to use these animals on other protocols, or use these animals on another of my ACUC-approved protocols.

I further certify that:

- No personnel will perform any animal procedures until they have been approved by the ACUC via RARC. When new or additional personnel become involved in these studies, I will submit their qualifications, training, and experience to the ACUC and seek ACUC approval before they are involved in animal studies;
- I will ensure that all personnel are enrolled in an institutional Occupational Health and Safety Program prior to their contact with animals, or have declined in writing to participate, if allowed by local policy;
- I will provide my after-hours telephone numbers to the animal care staff in case of emergency.

I plan to follow the provisions for the care, use and treatment of animals found in the NIH "Guide for the Care and Use of Laboratory Animals," or the "Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching". I assure that these procedures do not unnecessarily duplicate previous experiments.

Signature of PRINCIPAL INVESTIGATOR/PROJECT DIRECTOR: [redacted] 11/24/08

Attending Veterinarian (SVM ONLY) _____

QUESTIONS FOR PROJECTS INVOLVING SURGICAL PROCEDURES

24. Which of the persons listed on question 15 will be performing surgery?:

Last name, first	Type and length of SURGICAL training/experience
[redacted]	[redacted]

25. Where will surgery be performed? Room(s): *Acute* [redacted] *Sterile* [redacted] Building: [redacted]

26. How many animals listed in question nine will undergo surgery? *All*

27. Describe anesthetic method used, including all drugs, dosages, routes of administration and supplementation schedules. Include how anesthesia level is monitored.

Acute: Animals are induced with ketamine hydrochloride (15-20 mg/kg) and acepromazine (.2 mg/kg) and atropine methyl nitrate (0.15 mg) given intramuscularl. An intravenous cannula is then inserted to administer additional doses (3-5 mg/kg) of sodium pentobarbital (diluted 50:50 with saline) as needed to maintain areflexia. The heart rate and respiration rate are monitored about every 15 minutes throughout the initial stages of the surgery that typically lasts several hours and then at 30-60 minute intervals during recording. These physiological measure will all be documented. It is not possible to monitor at higher rates during recording since the animal has to be isolated in the sound proof room and entering the room will usually cause loss of recording contact with the single neurons (see #19 above). Temperature is continuously monitored and regulated with a water-circulating heating pad. Intravenous lactated Ringer's (10 ml/kg/hr), with additional dextrose (5% in saline) for long term (2 day) experiments, is given via an IV drip to prevent dehydration. Rarely if a cat goes into respiratory distress, we will give Dopram (1.5 mg/kg, single injection IV) with possible repeated injections at 5 min intervals if needed. Typically these experiments will last 36-48 hours though on rare occasions they have been known to last as long as 80 hours. Because of the extensive surgery required in these acute experiments, it is not feasible to do them aseptically though we endeavor to keep the wounds as clean as possible. For these acute experiments the animals are kept areflexic with supplemental pentobarbital injections as long as the experiment lasts and euthanized at the end.

Chronic: cats are induced with ketamine (15-20 mg/kg) and acepromazine (.2 mg/kg) and atropine as above, or Midazolam (0.066- 0.22 mg/kg) and Oxymorphone (0.1-0.4 mg/kg), and a venous cannula inserted for administration of fluids. An endotracheal tube is then inserted to enable gas anesthesia (isoflurane and oxygen) to be used for the duration of the surgery, which lasts about 4-7 hours. Expired CO_2 is monitored throughout the surgery and fedback through an audio signal so that we have a continuous monitor of the animal's breathing pattern. We will also monitor heart rate, respiration rate and temperature about every 15 minutes during the course of the surgery as well as during the recovery phase after the anesthesia has been discontinued, until the animal has recovered from anesthesia and returned to its cage. These chronic surgeries are never longer than 12 hours.

a) Are any paralytic agents being used *NO* If YES, indicate agent, justification for use, and any special monitoring techniques used to assess animal condition while under paralysis.

28. **Surgical Procedures** (see relevant Campus policies at www.rarc.wisc.edu or Medical School policies at www.acu.medsch.wisc.edu/Policies/policies

a) Describe the surgical procedure(s), including a narrative description(s) giving: reason for the surgery, incision site(s), tissue isolation methods, wound closure, and an estimate of time required to complete the surgery. Note: aseptic procedures must be used for all survival surgery. (Append additional page if necessary)

Acute surgery: There are several reasons that surgery is necessary: first, the experiments require that the animal be kept from moving because the acoustic stimuli have to be delivered to the tympanic membrane in a precise manner and because we would like to record from a single neuron in the brain for long periods of time and secondly, we gain access to the brain by drilling holes in the skull.

Cats are deeply anesthetized initially with ketamine and acepromazine (see #27) and then with sodium pentobarbital administered IV. A venous cannula for infusion of additional anesthetic and a tracheal cannula are installed. In addition we maintain a drip of lactated Ringer's with dextrose via the IV line to keep the animal properly hydrated and osmotically balanced. The head is clamped in a head-holder and in most cases the external ears are removed by incisions made around the ears and deep dissection of the muscles and connective tissue. A midline incision is made on the skull and the muscles and tissue overlying the skull are removed or retracted. The bone overlying the area of interest is removed and the cerebral cortex or cerebellum are aspirated to

expose the inferior colliculus, or other area under study. In some cases a ventral approach is used to expose the ventral aspect of the brainstem; in these cases the esophagus is ligated and the basiocciptal bone along the ventral surface of the brainstem is exposed. The surgery usually takes about 4-6 hours to complete; it is followed by electrophysiological recording usually for 1-2 days, and occasionally longer. The cat is kept deeply anesthetized throughout this period with additional doses of IV pentobarbital. The core body temperature is continuously monitored with a rectal thermometer and maintained at 39° C with a water-circulating heating pad. I am consulting with the Veterinary staff on additional antibiotics, nutrients, vitamins, amino acids and anti-inflammatory agents that can be administered for these prolonged nonsurvival cases.

For these acute terminal experiments the surgeries are not done aseptically. One of the main reasons for doing these experiments acutely with anesthetized animals (as opposed to a chronic preparation in an awake animal) is that we want to deliver the acoustic stimulus as close as possible to the ear drum to avoid the complicated acoustic filtering properties of the external ear and ear canal. To do this we remove the external ear and cut the ear canal close to the eardrum so that we can fit a tube that is connected to a speaker along with a probe tube to calibrate the whole system acoustically. This means that a considerable amount of hardware has to be placed right outside the eardrum. Removing the external ear requires opening a rather large surgical field so there is an open wound around the ear canal that cannot be sealed due to hardware considerations. We "keep the field as clean as possible" by closing the open wound as much as possible with wound clips but unfortunately it is not possible to close it completely. We also cauterize all blood vessels and wound edges to minimize bleeding.

In practice this is not really an issue any more for two reasons: first we are phasing out these acute experiments. We have not done one of these in the lab in the last 4 years or so. Second, very rarely do the experiments last more than about 30-40 hours. On exceptional cases they have lasted for 5 days but we've only had one such cat in 30 years! We will alert the RARC vets in case we decide to do one of these acute surgeries again.

Chronic surgery: Multiple surgeries are needed for these experiments, all of which are done aseptically. During the initial surgery, a head holding device is implanted on the skull and eye and ear coils are implanted around the eyeball and subcutaneously on the ear. During the second surgery a hole is drilled in the head to provide access to the dura and brain for the microelectrode. Sometimes a third surgery is needed to cauterize the nerves innervating the muscles of the ear in order to prevent ear movements. For some of the experiments, the cats will be bilaterally implanted with cochlear implants in order to study binaural processing in a deaf model with restored hearing.

The initial procedure is done under sterile conditions with the cat deeply anesthetized with isofloraneloxygen (above) to install a device for holding the head and coils of wire around the eyeball; on the head and in the external ears to monitor eye, ear and head movements. The eye coil is placed by cutting the conjunctiva around the eyeball, placing the coil around the globe, and leading the ends of the wire subcutaneously up to the top of the head. This procedure has been previously used in this laboratory ([redacted]). Likewise the coils on the ears are placed subcutaneously behind the ear by an incision behind the ear which is enlarged by blunt dissection so that a small coil of three turns of Teflon coated stainless steel wire can be placed underneath the skin. The ends of the wire are also led subcutaneously to the top of the head where each wire is soldered to a connector. Each of the connectors is embedded in the dental acrylic base. The use of a coil for measuring eye movements has become standard for neurophysiological studies of the oculomotor sytem. Virtually all investigators are using this technique. It has very significant advantages over the electrooculogram in accuracy, stability and resistance to artifacts.

The head holder is a stainless steel post with an anchoring base that is attached to the skull by embedding the base in dental acrylic, which is in turn fastened to the skull by 10-15 small titanium screws obtained from a neurosurgical supply house. An incision is made along the midline of the dorsal aspect of the skull and the overlying tissue and muscle attachments are carefully dissected free to expose an area of skull about 4 cm diameter. Since we are interested in preserving as much as possible the natural ear movements of the cat, we try to disturb the muscle attachments as little as possible. The titanium screws are screwed into the bone, after which they are embedded in layers of dental acrylic. The entire surgery for implanting the eye and pinna coils and head holder takes about 6 hours. Our past experience shows that the behavioral training is well-tolerated by the cats. Some of them will readily jump into the restraint bag when brought into the lab in the morning, apparently eager to begin testing.

The second surgical procedure comes several months later after the cat has been throughly trained. A recording chamber for electrophysiological recordings of single cells in the brain will be implanted on the skull under sterile conditions. After retracting the skin over the skull, a hole of 1.27 cm diameter will be made in the skull over the superior or inferior (depending upon the experiemental goals) colliculus with a trephine, taking care not to injure the overlying dura. A specially-made, plexiglass recording chamber which holds the microdrive and microelectrode is glued over the hole with dentral acrylic. This surgical procedure takes about 2 hours. The recordings are then done on a daily basis over 2-4 months.

In some cats some additional procedures are necessary. To study the effect of eliminating pinna movements during sound

localization, we cut the nerves innervating the ear muscles after the animal has been thoroughly studied under normal conditions. The ear muscles are de-efferented by ligating the the temporal and auriculoposterior branches of the facial nerve that innervate the pinna muscles. This procedure will also be done aseptically under isoflurane anesthesia. The relevant branches of the facial nerve that innervate the pinna will be identified by electrial stimulation. In some cats we will de-efferent the pinnae after behavioral training but before physiological recordings to study the behavioral effects of de-efferentation, in other cats we will de-efferent the pinnae after recording from a sample of cells to study the physiological effects of de-efferentation

For the surgery to implant cochlear implants, the anesthesia administered for the surgery will remain the same as outlined above for the chronic surgeries. A tracheal cannula may be inserted to allow for easier airway access. The skin and muscles overlying the back and top of the skull will be reflected, and the ear canals will be transected. If the deafening procedure using local injection of antibiotic into the cochlea is being used (procedure #3 in item 17 above), that procedure will be done at this time since this procedure is very invasive, and we want to minimize surgical exposure of the cochlea. The tympanic bullae will be opened to allow access to the round window. The round window will be carefully incised, the oval window opened by removal of the stapes footplate, and the perilymph gently aspirated. A polyurethane cannula will be inserted into the scala tympani through the round window opening, and the ototoxic aminoglycoside, neomycin sulfate (2.5 ml of a 10 mg/ml solution in normal saline; Sigma) will be slowly perfused through the cochlea over a 5 min period, with gentle aspiration at the oval window. The intracochlear electrodes, which will be cat-sized electrode arrays, one in each ear, will be surgically implanted into each cochlea through the exposed round window. Care will be taken to achieve the same insertion depth on both sides, and the function of the stimulating electrodes will be determined before the end of the surgery. The wires will be pre-soaked in a dilute Ampicillin solution (50 mg/ml) and the subcutaneous tissue will also be irrigated with this dilute Ampicillin solution. The lead wires from the implant will be tied to small fixation screws embedded in the bone of the bulla and led to a connector at the base of the back of the neck. The transmitter assembly will be held n a backpack worn by the cat. This system has been designed by [redacted] *and they are helping us with the necessary hardware and surgical technique. Stay sutures will be used to fix the receiver package and prevent it from migrating. The sutures will be inserted through small holes drilled into the skull using a fine burr and then tied around the package. The lead wire will also be held in place with sutures. To prevent middle ear infection, an autograft of connective tissue will seal the electrode entry point into the cochlea.*

b) Detail the aseptic procedures used for this survival surgery including incision site preparation, instrument sterilization, and clothing worn.

Sterile surgeries are performed in room [redacted] *which is provided by the UW Animal Care facility. All instruments are sterilized in an autoclave or by submersion in an antiseptic solution of cetylcide II. We usually have at least three people involved in every surgery: two wear sterile gloves and gowns and the third maintains watch on the animal's condition and takes notes. Everyone wears a face mask and cap. The incision site is scrubbed with betadyne followed by alcohol.*

29. Will the animals be allowed to recover from surgery? YES

If YES, describe the post-anesthetic and post-surgical monitoring and care procedures, including all drugs and dosages, how body temperature will be maintained during recovery, who will do the monitoring, frequency/duration of monitoring, the parameters which will be evaluated, and method of maintaining written records of these examinations. Describe measures designed to alleviate post-operative discomfort. (For Medical School: see analgesic policy at: www.acu.medsch.wisc.edu/Policies/policies).

Immediately after the surgery is completed, the anesthetic inhalant is turned off and O_2 is given. Within a few minutes, the cat will naturally start coughing as it detects the irritation of the endotracheal tube, at which point we remove it. The cat is then closely monitored usually by cradling in our hands immediately until it is sternally recumbent and then placed under a heat lamp during the overnight postsurgical period. If the cat is not fully recovered, we monitor heart rate and respiration every 15 minutes or so and turn it from side to side to prevent the buildup of fluid in the lungs. The cat is considered to be fully recovered when it is able to get up and move around unassisted. Cefazolin (30 mg/kg, IM or SQ) is administered at the initiation of surgery and continued three times per day until the cat can take oral antibiotics. At that point the oral antibiotic Orbax (5-7 mg/kg) or Cephalexin (35 mg/kg BID) is given daily for 1 week. If it appears that the cat will require more than one week of antibiotic, we will consult the veterinarians. During surgery we administer fluids (lactated Ringer's) to prevent dehydration, topical analgesics (Lidocaine) to the wound margins, Bacitracin ophthalmic ointment to the eyes, eye drops (artificial tears) are applied every 15 minutes as a protectant from time of intubation until eye coil placement is complete, and ketoprofen (2 mg/kg s.q.), buprenorphine (0.005 - .01 mg/kg SQ), or non-steroidal anti-inflammatory drug such as carprofen (1-2 mg/kg) for pain killing about 30 minutes before the gas anesthesia is turned off. These chronic cats may experience some pain (as assessed by criteria that are used in human infants, such as body weight, respiration, mobility, and vocalizations) *during the recovery period in which case additional ketoprofen (1-2 mg/kg s.q daily) or buprenorphine (0.005 – 0.01 mg/kg SQ) or other analgesic as recommended by an RARC veterinarian is given for up to 2-5 days post-surgery. The veterinarians will be consulted in cases where additional analgesics beyond the initial 2 day period are thought to be needed. Careful post-op monitoring by the vet staff will be performed with possible post-op blood work. The veterinarian staff will be informed before the chronic surgeries are performed so that postoperative assessment of analgesia can be monitored.*

30. Will any animal(s) be allowed to recover from more than one major operative procedure? *YES*
(A major operative procedure is defined as any surgical intervention that penetrates and exposes a body cavity or any procedure which produces permanent impairment of physical or physiological functions.)

a) If YES, provide scientific justification for performing these procedures and list the species and number of animals:
The multiple surgical procedures are necessary in the chronic cats for five reasons. First, the duration of the final electrophysiological recordings are limited by the hardening of the dura that occurs once the skull is exposed, which makes it too tough to penetrate with a microelectrode. Since cats must be trained for many months before recordings can begin, it is necessary to complete the training before opening up the skull. In order to train the animals, we must implant the eye coils and head holder so a surgical procedure is needed. If both procedures were done at once, the dura would be too hard before we had completed the training. Thus, one surgery consists of implanting the eye and ear coils and the head restraint device, and a second surgery is needed to implant the recording chamber. Second, in some animals we have had trouble with breakage of the eye coils which requires that they be removed and reimplanted. These cats will then undergo additional sugeries. Third, in some of the pinna de-efferentation experiments we will study the behavioral effects of de-efferentation, which requires the initial surgery to implant the eye coils and head restraint so that the animal can be trained. Only after training is complete, which may require up to several months of time, can the pinnae be immobilized in a second surgical procedure. Four, we will also compare the responses of cells in a normal cat with the same population of cells in the same cat with the pinnae immobilized. These cats will require a third surgery to de-efferent the pinnae following a few months of physiological recording. Fifth, the cats with cochlear implants must undergo surgery in order to be trained behaviorally before they are deafened and given cochlear implants. In order to be trained behaviorally, they must first undergo the standard first surgery of head holder and eye/pinna coil implantation. After deafening, they must then undergo the cochlear implant surgery. We endeavor to keep each of the surgical procedures as short as possible. Not all cats will be subjects in all experiments. All of them will undergo the first (head holder and eye and pinna coil implantation) and last procedures (recording chamber implantation). By using the cats for several experiments we are able to minimize the use of animals. We plan to use 3-5 cats/year in this chronic preparation. The other cats used for the acute experiments do not recover from the anesthesia.

b) What is minimum length of time between the operative procedures?

We anticipate that no more than 3 eye coil implantations per eye will be required within a given 12 month period. If additional replacements are needed, the veterinarian staff will be consulted. In the case of replacement or repair of the head implants, we will consult with the vets if the head cap must be replaced or repaired more than once to make sure that the animal's condition is suitable for repair or replacement.

QUESTIONS FOR PROJECTS USING WILD-CAUGHT ANIMALS

It is the responsibility of the PI to obtain all necessary state and federal permits for work with wild animals.

31. Do you capture wild animals or do experimental manipulations (or procedures) on animals in the wild?
NO

32. If you capture wild animals, describe how they will be trapped, what types of traps will be used, and how often traps will be checked.

33. a) Describe quarantine procedures and precautions to prevent exposure of humans and other animals to zoonotic diseases.
b) If animals will be release back to the wild, justify why this release will not result in disease exposure to wild population

34. If wild animals will be anesthetized and released to the wild, describe anesthetic doses, method of administering and procedures for assuring that animals are sufficiently recovered from anesthetic to be released.

QUESTIONS FOR PROJECTS USING NON-HUMAN PRIMATES

35.a) If non-human primates used in your study must be housed individually due to scientific consideration, provide that scientific rational.

b) Provide scientific rationale for any restrictions to environmental enrichment. Include the specific restriction(s) such as: puzzle feeders, cage perch, wooden chew sticks, food treats (bananas, carrots, oranges, other fruit or vegetables), etc.

Exhibit C

January 24, 2012 USDA-APHIS Inspection Report for University of California, San Francisco

USDA

United States Department of Agriculture
Animal and Plant Health Inspection Service

FORM
321215341307 17 (ISO_ID

Inspection Report

UNIVERSITY OF CALIFORNIA, SAN FRANCISCO

Customer ID: **9199**
Certificate: **93-R-0440**
Site: 001
THE UNIVERSITY OF CALIFORNIA, SAN FRANCISCO

UCSF BOX 0547

Type: ROUTINE INSPECTION
Date: Jan-24-2012

SAN FRANCISCO, CA 94143

2.31 (d) (1) REPEAT

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC).

2.31(d) IACUC review of activities involving animals. (1) In order to approve proposed activities,.. the IACUC shall determine that the proposed activities or significant changes in ongoing activities meet the following requirements:
(i) Procedures involving animals will avoid or minimize discomfort, distress, and pain to the animals;

The IACUC-approved protocol " Incubation Periods of Prions and other Neurodegenerative Diseases" states that voles are to be monitored for neurologic signs following intra-cerebral inoculation with prion infected tissue "Twice weekly until the first sign, then daily Monday through Friday". According to the protocol the animals should be euthanized either "...at the earliest point possible in the clinical progression of the disease, to ensure that no animal ever suffers or is in distress" or ".. following the presence of two neurological signs.." According to this protocol, an animal developing one neurologic sign on a Friday could develop additional neurologic signs over the weekend and experience unnecessary discomfort, distress, and pain prior to dying of prion disease or being humanely euthanized on Monday. Incubation period data for eight animals placed on this study showed that three animals were found dead from prion disease on a Monday, with no recorded observed neurological signs.

The IACUC should ensure that animal activities include provisions that avoid / minimize pain and distress by providing for adequate monitoring of animals as necessary to prevent discomfort, distress, and pain.

This is a REPEAT citation. There was a previous citation under this regulation on Jan 5, 2011, and it was to be corrected by April 3, 2011.

2.31 (e) (2)

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC).

2.31(e) A proposal to conduct an activity involving animals, or to make a significant change in an ongoing activity involving animals, must contain the following:..
(2) A rationale for involving animals, and for the appropriateness of the species and numbers of animals to be used:

Prepared By: PAMELA L SMITH, D.V.M. USDA, APHIS, Animal Care **Date:** Feb-01-2012
Title: VETERINARY MEDICAL OFFICER Inspector 6036

Received By: (b)(6),(b)(7)(c) **Date:** Feb-16-2012
Title:

Inspection Report

***In the IACUC-approved Amblyopia and Strabismus study protocol, the rationale for the numbers of animals to be used in the study states " In my field, the number of animals needed for research is not guided by statistical criteria or tests... Because the success rate of experiments is unpredictable, given the innate nature of scientific investigation, and the yield of data from any given animal is variable in quantity and quality, it is hard to predict the number of animals required for our research projects. The number of 15 monkeys is the most accurate projection that I can provide of expected animal use over the next 3 years. This figure is also based on our experience with the requirements for manuscripts reporting scientific results in per-reviewed publications in my field. We utilize the minimum number of animals required to meet my scientific field's publication standard. "

Stating that the number of animals to be used cannot be determined using statistical criteria or tests, because of the nature of your field or research, is not an appropriate explanation of how the numbers requested were determined unless an acceptable alternative explanation is provided. Citing requirements for publication of manuscripts is also not an adequate explanation for the need to use a specific number of animals for experimentation. The investigator is responsible for providing the IACUC committee with a rationale for animal use that includes an explanation for the number of animals that is based on specific goals, statistical thresholds, or some other more concrete criteria which reflect the purpose of the investigation, beyond manuscript publication.

*** In the IACUC-approved Incubation Periods of Prions and other Neurodegenerative Diseases protocol, the description of the criteria for the number of animals needed states "We perform on average 44 samples per year in hamsters... and 25 samples in voles." But a chart above this description showing the number of animals acquired for this protocol lists 1056 hamsters and 750 voles. There is no explanation for these numbers, and it remains unclear how many animals the investigator proposes to use for this research under this protocol.

*** In the IACUC-approved Rabbit Model of Bacterial Endocarditis protocol under Category D there is contradictory and inadequate information about the number of rabbits to be used. Under the description of statistical tests/rationales used to determine the number of animals needed, it describes the use of 100 rabbits per year for three years for a total 300 rabbits, and then later states that under category D the total is 400 rabbits , with no clear explanation as to how the extra 100 rabbits came into the equation. There is also no explanation as to how either the 100 rabbits per year nor the 300/400 rabbits total will be used to meet their stated statistical goals.

*** In the Cellular Mechanisms of Vascular Injury protocol, the rationale for the number of animals calls for two different injury models, and under each injury model there are three treatment modalities. For some unexplained reason the protocol states that each treatment modality will have its own "control - no treatment " rabbit. It is not explained why each treatment modality requires a control animal rather than having a control under each injury model, alongside the three treatment modalities for each injury model. Having a control for each treatment modality adds an additional 100 rabbits to the protocol over having one control per injury model.

Prepared By: PAMELA L SMITH, D V M. USDA, APHIS, Animal Care **Date:** Feb-01-2012

Title: VETERINARY MEDICAL OFFICER Inspector 6038

Received By: (b)(6),(b)(7)(c) **Date:** Feb-16-2012

Title:

Inspection Report

An appropriate rationale for the number of animals the investigator proposes to use is necessary to ensure that the minimum number of animals is used in an activity involving animals. Errors or lack of information in a proposed activity could result in animal use that was not approved with a clear or accurate understanding of the need for this animal use, which could jeopardize animal welfare.

The IACUC should ensure that each activity using animals contains an appropriate rationale for the numbers of animals to be used in that activity.

To be corrected by May 1, 2012.

2.31 (e) (3) REPEAT

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC).

2.31 (e) A proposal to conduct an activity involving animals, or to make a significant change in an ongoing activity involving animals, must contain the following:...
(3) A complete description of the proposed use of the animals.

***The protocol "Incubation Periods of Prions and other Neurodegenerative Diseases" was approved with an incomplete description of when animals were to be monitored for neurological signs. According to the stated monitoring parameters in the protocol, following prion inoculation the animals are to be monitored " twice weekly until the first sign..." However, the protocol does not specify when the monitoring should take place during the twice a week monitoring period. As written, monitoring could take place on consecutive days leaving five days when the animals would not be assessed for neurological signs. The protocol should be more specific as to when the animals are monitored for neurological signs in order to ensure that they do not wait past study end points for euthanasia. The protocol also contains conflicting statements as to when the animals are to be euthanized. One section of the protocol states that "Each animal is humanely euthanized at the earliest point possible in the clinical progression of the disease, to ensure that no animal ever suffers or is in distress." Elsewhere in the protocol stated endpoints are "...5 days past onset of clinical signs in hamsters" and that voles "...are euthanized following the presence of two neurological signs..." The protocol also states that the animals are still able to eat, drink, and move around the cage, yet some of the neurological signs that animals are monitored for include "Can't get up", "Paralysis", and "Convulsion". The protocol does not differentiate between the animals with severe neurological signs and those with mild signs with regards to euthanasia of infected animals. Presumably, if a vole was only having convulsions it would not be euthanized because it was only showing one neurological sign.

It is the responsibility of the IACUC to ensure that the investigator provides a complete description of proposed activities that involve the use of animals in order that those activities may be adequately reviewed and determined to be in accordance with the Animal Welfare Act.

This is a REPEAT citation. There was a previous citation under this regulation on Jan 5, 2011, and it was corrected by the time of the exit interview for that inspection.

An exit interview was conducted with facility representatives.

Prepared By: PAMELA L SMITH, D V M. USDA, APHIS, Animal Care **Date:** Feb-01-2012

Title: VETERINARY MEDICAL OFFICER Inspector 6036

Received By: (b)(6),(b)(7)(c) **Date:** Feb-16-2012

Title:

Exhibit D

UW-Madison Procedure Log for Double Trouble
April 24, 2008

IACUC Protocol # [redacted] PI [redacted]
Animal Identification (name or #) G07 Date 4/24/08
Procedures ABR

ID#/ Species G07
Body weight 2.88
Age ____ Sex F
Health status/preoperative physical exam:

Fasting period ____ (hours)

Induction and Summary of Drugs/Fluids:

Time	Drug/Fluid	Purpose	Dosage: mgs/vol	Route	Comments	Initial
1105	ketamine		0.60mL 60mg	IM		[redacted]
1105	acepromazine		0.06mL 0.6mg	IM		

Maintenance

Gas Anesthesia ☐ yes → ☐ Isoflurane Start Time: 1105
☒ no
Respiration ☒ spontaneous Endotracheal tube size: ____
☐ controlled ventilation

Vital Signs Monitoring: (Interval is every ____ min during surgery, ____ min during recording.)
Observations should be recorded from the time that the animal is stable after induction until the animal is terminated or recovered and placed back in their cage. Recovery is defined as when the following conditions are met: 1)the animal is physiologically stable, 2) analgesia is assured, and 3) the animal can lift it's head and remain sternal.

% Gas = anesthetic vaporizer setting, HR = heart rate, RR = respiratory rate, Temp = body temperature, O^2 Sat = oxygen saturation. Optional monitored parameters: muscle tone, capillary refill time, urine output.

Time	% Gas	HR	RR	Temp	O^2 Sat	Other (reflexes for acute exp)	Surgical Events / Comments / Additional Drugs	Initial
1:20		182		96	100%			[redacted]
~~1:35~~ 1145		224		99.5	90%	turned	anesthesia too light? Kicking	[redacted]
1:55		192		99.3	94%	probes on measurements turned. cath sternal	waking up - no more	[redacted]
2:10							5:00 returned to cage	[redacted]

Summary:

~~Surgery~~ start time: 1105 ~~Surgery~~ end time: 1145
Surgeon: [redacted]

Exhibit E

UW-Madison Procedure Log for Double Trouble
November 21, 2008

IACUC Protocol # _____ PI [redacted]
Animal Identification (name or #) 607 Double Trouble Date 11-21-08
Procedures EABR

ID#/Species 607
Body weight 7.22kg
Age _____ Sex F
Health status/preoperative physical exam:

Fasting period 7-24 (hours)

Induction and Summary of Drugs/Fluids:

Time	Drug/Fluid	Purpose	Dosage: mgs/vol	Route	Comments	Initial
11:00	Ketamine	Preanesthetic	100 mg/ml	IM	.8mL 80mg	[redacted]
	Acepromazine	Preanesthetic	10 mg/ml	IM	.06 mL	
	Ketamine		100 mg/mL	IM	.4mL 40mg	[redacted]

Maintenance
Gas Anesthesia ☐ yes → ☐ Isoflurane Start Time: 11:00
☒ no
Respiration ☐ spontaneous Endotracheal tube size: _____
☐ controlled ventilation

Vital Signs Monitoring: (Interval is every ____ min during surgery, _____ min during recording.)
Observations should be recorded from the time that the animal is stable after induction until the animal is terminated or recovered and placed back in their cage. Recovery is defined as when the following conditions are met: 1)the animal is physiologically stable, 2) analgesia is assured, and 3) the animal can lift it's head and remain sternal.

% Gas = anesthetic vaporizer setting, HR = heart rate, RR = respiratory rate, Temp = body temperature, O^2 Sat = oxygen saturation. Optional monitored parameters: muscle tone, capillary refill time, urine output.

Time	% Gas	HR	RR	Temp	O^2 Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
11:00							Triple antibiotic inj	[redacted]
11:05							Thermometer inserted	[redacted]
11:05		174		101.5	89			

Summary:

Surgery start time: 11:00 Surgery end time: 1:15
Surgeon: [redacted]

Date 11-21-08 ID# 807

Time	% Gas	HR	RR	Temp	O² Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
11:10							Shaved. Put in chamber	[redacted]
11:15		217		105	80		3 electrodes inserted	[redacted]
11:20		1					On heating pad, head fixed	[redacted]
12:00		135		101	90		Checked anesthesia. Was light	[redacted]
12:05							Additional Ketamine 40mg injection	[redacted]
12:15		140		100	90			[redacted]
12:30		140		100	90			[redacted]
12:45		140		99.5	92			[redacted]
1:00		135		99.5	92			[redacted]
1:15		200		99	90		Cat waking up	[redacted]

R0151

Exhibit F

UW-Madison Procedure Log for "Cat 33"
December 17, 2008

IACUC Protocol # _____ PI [redacted]

Animal Identification (name or #) cat 33 Date 12/17/08

Procedures _____

ID#/ Species _____

Body weight 4.09Kg (8.89lb)

Age _____ Sex F/S

Health status/preoperative physical exam:

T= 99.7°F RR = 20 bpm HR = 128bpm

Fasting period 7 1/2 hrs (hours)

Induction and Summary of Drugs/Fluids:

Time	Drug/Fluid	Purpose	Dosage: mgs/vol	Route	Comments	Initial
9:09Am	Buprenorphine		0.1 ml	Buccal		[redacted]
9:13Am	Domitor		0.1ml	IM		
9:13Am	Ketamine		0.23ml	IM		

Maintenance

Gas Anesthesia ☑ yes → ☑ Isoflurane Start Time: 9:40

☐ no

Respiration ☑ spontaneous Endotracheal tube size: 3.5

☐ controlled ventilation

Vital Signs Monitoring: (Interval is every _____ min during surgery, _____ min during recording.)

Observations should be recorded from the time that the animal is stable after induction until the animal is terminated or recovered and placed back in their cage. Recovery is defined as when the following conditions are met: 1)the animal is physiologically stable, 2) analgesia is assured, and 3) the animal can lift it's head and remain sternal.

% Gas = anesthetic vaporizer setting, HR = heart rate, RR = respiratory rate, Temp = body temperature, O^2 Sat = oxygen saturation. Optional monitored parameters: muscle tone, capillary refill time, urine output.

Time	% Gas	HR	RR	Temp	O^2 Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
9:13							preanesthesia	[redacted]
9:30						Sodium Chloride	insert I.V.	24 ga
9:40	3% O2 .5				100ml/hr fluid rate	capillary refill	intubated 3.5	

Summary:

Surgery start time: 9:13 Surgery end time: _____

Surgeon: _____

[redacted]

[redacted]

Date 12-17-08 ID# cat 33

Time	% Gas	HR.	RR	Temp	O² Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
9:45	1.5%	116			98	1 drop each eye	artificial tears cephazolin 1cc	[redacted]
9:50	1.5	118	16	97°	98		shaved head	[redacted]
10:05	1.5	103		99.5	98		positioned cat 2 drop AK dilate	[redacted]
10:15					8ml/hr fluid	preparing instruments	scrubbed head art tears	[redacted]
10:20	1.5	61	17	99	94		started dissection (L) eye for coil	[redacted]
10:30	1%	60	38	98.6	90	.9 l/min	art tears	[redacted]
10:45	1%	190	21	98.4	72		Dissecting left eye Coil placed in left eye	[redacted]
							18 mm coil in left eye 3 loops	[redacted]
11:00	1%	84	22	98.4	96		Suturing left eye	
11:10						.9 l/min O2	3 sutures in left eye Nexaband on sutures	[redacted]
11:15	1%	99	22	98.4	84		Art. Tears Opthalmic ointment	[redacted]
11:30	1.5%	98	23	98.6	88		in left eye Begin dissecting R. Eye	[redacted]
11:35							Coil placed in right eye 18mm	[redacted]
11:45	1.5%	103	23	98.8	93		Suturing R. Eye	[redacted]
12:00	1.5%	102	22	99.0	91		Adhesive suture in R. Eye Ointment added to R. Eye	[redacted]
12:15	1.5	112	20	99.1	92		started dissecting L ear L coil in 10mm 6 loop	[redacted]
12:30	1.75%	113	23	99.3	94		sutures L ear Nexaband (R) ear insert R coil 10mm 6 loop	
12:45	2.%		21					[redacted]
1:00			24				Incision on top of skull Cleaning bone	[redacted]
1:15	2	164	24	99.5	89		Drilling bone Putting in screws	[redacted]
1:30	2	119	21	~~98~~ 99.5	96			[redacted]
1:45	2	128	21	99.5	77		More Screws	
2:00	2	122	20	99.5	86		13 screws total	[redacted]
2:15							Applying dental cement	[redacted]
2:30	2	118	21	99.3	92		/	[redacted]

Date 12-17-08 ID# Cat 33

Time	% Gas	HR	RR	Temp	O² Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
2:45	2	120	21	99.5	94		Running wires through scalp to implant	[redacted]
3:00	2	121	21	99.5	94			
3:15	2-1.5	120	20	99.5	95	Rear 57.4 Lear 52.5	Soldering wires to plug. Shrink tubing	
3:30	1.5	113	22	99.3	89			
3:45	1.5	115	21	99.3	89	HB. 1 Rage		
4:00	1.5	118	23	99.3	92			
4:15	1.5	119	22	99.3	93			
4:20	1.5	180	11	99.5	72	Left eye: 1/4 2/3	Cat stopped breathing, cleared trachea of fluid	
4:35	1.5						Isoflorane removed to clear trachea, cat woke up	
4:35	1.5	110	18			manual ventilation	briefly. Resumed breathing	
4:45	1.5	107	10	99.7	95		cementing connector	[redacted]
5:00	1.5	107	9	99.7	91			
5:15		118	9	99.8	86		cementing head cap	[redacted]
5:30	1.5	116	16	99.5	91			
5:45	1.5	116	21	99.5	87		cement fixing rest of skull	[redacted]
6:00	1.5		25				1cc cephazolin	[redacted]
6:15	1.5	102	20	99.7	82		.04 mL Ketoprofen	[redacted]
6:30	1.0%						suturing wounds	[redacted]
6:45	1	119	36	99.7	88		One stitch on right cheek.	[redacted]
	Placed in room with heat lamp. Fed one can wet food. Is awake and moving around						Three stitches behind head part, one stitch in front Rectal thermometer out I.V. out .66mL saline total Heating pad off extubated	[redacted]
6:45				.1 mL	Buprenophine		cat walking	[redacted]
							Cat sternal cat walking returned to cage	[redacted]

Exhibit G

UW-Madison Procedure Log for Double Trouble
June 11, 2008

IACUC Protocol # _____ PI [redacted]

Animal Identification (name or #) 607 Date 6-11-08

Procedures Bilateral cochlear implants

ID#/ Species Cat

Body weight 3Kg

Age 8 Sex M

Health status/preoperative physical exam: good

Fasting period 12 hrs (hours)

Induction and Summary of Drugs/Fluids:

Time	Drug/Fluid	Purpose	Dosage: mgs/vol	Route	Comments	Initial
10:00			.54mg atropine .6cc Midazolam .6mg Butorphanol			[redacted]
10:00	5% iso 1% O2		& mask			
10:05	KA 0.6cc/6mg Lidocaine				intubate-not in	
10:07	5% iso					
10:10	1cc #/ 2mg ~~Midazolam~~ lidocaine				intubate	
10:12	5% iso					
10:14	cefazolin		100mg cefazolin			

Maintenance

Gas Anesthesia ☒ yes → ☒ Isoflurane Start Time: 10:07

☐ no

Respiration ☐ spontaneous Endotracheal tube size: 3.5

☐ controlled ventilation

Vital Signs Monitoring: (Interval is every ____ min during surgery, ____ min during recording.)

Observations should be recorded from the time that the animal is stable after induction until the animal is terminated or recovered and placed back in their cage. Recovery is defined as when the following conditions are met: 1)the animal is physiologically stable, 2) analgesia is assured, and 3) the animal can lift it's head and remain sternal.

% Gas = anesthetic vaporizer setting, HR = heart rate, RR = respiratory rate, Temp = body temperature, O^2 Sat = oxygen saturation. Optional monitored parameters: muscle tone, capillary refill time, urine output.

Time	% Gas	HR	RR	Temp	O^2 Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
10:15						Ketadrol + alcohol prep for I.V		[redacted]
							Lubes ophthalmic ointment	
10:30	1.5	170 start cut	10		98 start to climb	10ml/hr	I.V in	

Summary:

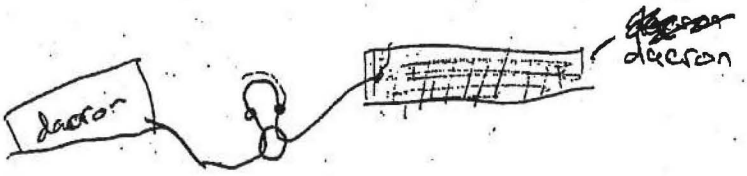
Surgery start time: 10:15 Surgery end time: [redacted]

Surgeon: [redacted]

R0156

Date 6-11-08 ID# 607

Time	% Gas	HR	RR	Temp	O² Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
	2						shaved throughout below ear except head and [redacted]	
10:40	2						Isoflurane turned up to 2	
							Isoflurane up to 2.5	
10:45	2.5	188	16	94.6	100		covered cat	[redacted]
11:00	2	196	5	93.2	97		draping cat	[redacted]
11:10							Initial Incision directly below	[redacted]
11:15	2	178	9	92.7	97		ear running rostral-caudal	[redacted]
11:20	1.75						Bulla uncovered	[redacted]
11:30	4	190	21				Anesthetic mask came off, animal	[redacted]
11:35	3	161	5	93.2	97		showed signs of waking	[redacted]
							Hole drilled in bulla, microscope used	[redacted]
							at this point. Bulla is cleaned.	[redacted]
11:45	2	140	4	93.2	96		Round window being uncovered.	[redacted]
							Normal Round window & middle ear	[redacted]
11:50							Drilling hole through cortical bone	[redacted]
							Cat gut run through hole drilled	[redacted]
							in cortical bone. Also run through	[redacted]
12:00	2	135	5	93.7	97		Amoxicilin goes in bulla and around wound	
	1 3/4						25# needle & 1 mL syringe used to	[redacted]
							Round window opened, perilymph	[redacted]
							Sucked out superficially, neomycin injected into ~~cochlea~~	[redacted]
							~100 mL injected. 4 times. Muscle	[redacted]
							tissue removed for preservation and	[redacted]
				Aspiration following injection	again neomycin		preparation for packing in around round window	[redacted]
							Electrode array soaked in antibiotic	[redacted]
12:15	1 3/4	138	5	94.1	96		Electrode array guided through	[redacted]



R0154

Date 6-11-08 ID# 607

Time	% Gas	HR	RR	Temp	O² Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
12:15							Cochlea. Cat gut cut after HF	
	All electrodes inserted into scala tympani. No evidence of traumate cochlear						dacron run through. Dacron tied off in double knot. Dacron ends cut off. Crushed muscle packed	
							into round window. Gently aspirated. Fine lead wire guided	
							into bulla. Loctite used to secure	
12:30	1 3/4	139	7	94.5	92		dacron knot. Blue dental cement	
							in syringe, uses syringe to fill	
							~ 1/3 of bulla away from round	
							window. Bulla not needed to be	
							dried out significantly. Not necessary	
							for dental cement to harden properly	
12:40							Begun suturing muscles.	
							Internal sutures completed, antibiotic to	
12:45	1 3/4	145	10	94.8	96		applied. ~~External sutures~~	
							Incision made to right of left	
							ear and plication through to	
							surgical incision. Lead pulled from	
							surgical incision under skin to	
							emerge from incision to right of	
							ear. External sutures in place.	
1:00	1 3/4	149	11	94.8	95		~~Cat flipped, other side begun~~	
							~~Incision made below ear~~	
	3 sutures made in top incision						Skull exposed. Hand drill	
							used to drill. Clamps attached	
							to lead wire, titanium screw	
							through clamps screwed into	
							hole in skull. Second hole drilled,	
							clamp attached, screwed in.	
							Lead moved to back of incision,	
							suturing begun.	
1:15	1 3/4	138	9	95.2	98		Incision made above left shoulder	
							lead run under skin ~~from~~ end cut	
							of this incision.	
1:30								

R0155

Date 6-11-08 ID# 607

Time	% Gas	HR	RR	Temp	O^2 Sat	Other (reflexes for acute exp)	Surgical Events/ Comments/ Additional Drugs	Initial
[redacted] 1:45	1 3/4	141	10	94.5	95		IV reinserted, cat flipped, covers, gloves, gown changed, cat rescrubbed	
[redacted] 2:00	2	152	7	93.9	93		First incision made below ear. Searching for bulla.	
							Bulla uncovered. Moving tissue out of the way to begin drilling.	
[redacted] 2:05							Bulla opened. Bulla is clean. Round window exposed	
[redacted] 2:15	2	144	13	94.3	96	28 mL infused	Holes drilled in bulla, cat gut run through, tied to dacron	
[redacted] 2:30	2	167	11	94.6	98		Dacron tied off. Injecting neomycin. Cat gut suture	
	1.75						cut. preparing to insert electrodes.	
[redacted] 2:45	1.75	138	4	95.2	97		Electrodes in. Round window is clean	
							Loctite applied to dacron knot. Filling bulla with dental cement	
[redacted] 3:00	1.75	133	6	95.7	98		Layer one sutured (4 sutures)	
[redacted] 3:05	2						Skin stapled closed. Incision made behind ear	
3:20	2	139	6	96.1			Lead run under skin to incision. Clamps attached to leads	[redacted]
							Administered Buprenorphine .015mg	[redacted]
-	-						Cephazolin 100mg	
							drill titanium screw in attach clip anchor;	[redacted]
3:30	2	141	9	96.4	94		lead wires down back.	[redacted]
						wound clips applied.	sequester ā amoutile clean wound	[redacted]
3:45	2	137	11	96.6	92	putting leads in connectors		[redacted]
3:50	1.5					applying shrink tubing. Shrink tube		
3:55	1.0					wounds sprayed ō opsite		
4:05				Probes being removed			endo trach. removed	[redacted]
4:09	0						Fluids total 66 mls 0.9% Saline ō total for procedure	

4:15 extubated heat on
4:30 cat turned [redacted]
4:35 cat sternal
5:00 cat returned to cage ō heat lamp fed [redacted]

R0153

Wolff, Axel (NIH/OD) [E]

From: Jeremy Beckham [JeremyB@peta.org]
Sent: Wednesday, September 12, 2012 10:08 AM
To: Wolff, Axel (NIH/OD) [E]; Battey, James (NIH/NIDCD) [E]; Collins, Francis (NIH/OD) [E]; NIDCDInfo
Subject: Urgent Complaint from PETA re UW-Madison
Attachments: Sep 12 2012 NIH OLAW Complaint from PETA re UW-Madison.pdf

Importance: High

September 12, 2012

Axel V. Wolff, M.S., D.V.M., Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
RKL 1, Suite 360, MSC 7982
6705 Rockledge Dr.
Bethesda, MD 20892-7982

James F. Battey, Jr., M.D., Ph.D., Director
National Institute on Deafness and Other Communication Disorders
31 Center Drive, MSC 2320
Bethesda, MD 20892-2320

Francis S. Collins, M.D., Ph.D., Director
National Institutes of Health
9000 Rockville Pike
Bethesda, MD 20892

Via e-mail: wolffa@od.nih.gov; batteyj@nidcd.nih.gov; francis.collins@nih.hhs.gov

Dear Dr. Wolff, Dr. Battey, and Dr. Collins:

Attached please find a complaint from PETA regarding apparent violations of the Public Health Service's *Guide to the Care and Use of Laboratory Animals,* U.S. Government Principles, and the NIH Grants Policy Statement. These violations are related to the use of cats in sound localization experiments as part of an NIH-funded project at UW-Madison.

Please contact me should you have any questions. Thank you for your attention to this urgent matter.

Sincerely,

Jeremy Beckham
Research Project Manager
Laboratory Investigations Department
People for the Ethical Treatment of Animals (PETA)
Telephone # JeremyB@peta.org