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Office of Research Infrastructure Programs

Rigor and Reproducibility of Animal Studies: Extrinsic Factors Workshop

Session 3. Large Animals (Nonhuman Primates/Swine)

September 30, 2022
Virtual Meeting

Final Report

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Executive Summary

The Extrinsic Factors Workshop was held in three sessions to better understand extrinsic factors and their effects on biomedical research. Session 3 was focused on extrinsic factors in the use of large animals, particularly nonhuman primates (NHPs) and swine, for biomedical research. Drs. Joyce Cohen and Kiho Lee served as the session co-chairs. Discussions in Session 3 addressed the use of precision medicine in NHP research; considerations for husbandry of swine; construction, housing, and caging factors; and equipment and technology factors. The speakers identified various extrinsic factors for consideration in research, including lifetime exposures, stress, social structure, sex and reproduction, diet, early biobehavioral organization, density, housing, enrichment, temperature, humidity, lighting, the microbiome, breed, disease status, and litter size. The participants also discussed monitoring and reporting extrinsic factors in research using large animals. They noted that these factors are important to monitor, but investigators often are discouraged from including extensive methodology sections in publications. Additionally, it was noted that many investigators are hesitant to modify their established systems. The participants discussed the need to maintain a balance between controlling extrinsic factors while ensuring that experiments remain generalizable and translatable. The need for additional funding to understand extrinsic factors was emphasized. Other considerations include the requirements for increased throughput and sample size. Investigators must remain flexible as technical and scientific opportunities present new considerations in biomedical research.

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Kiho Lee, Ph.D., University of Missouri

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Timothy Allen, Ph.D., Florida International University
Kelly Ethun, D.V.M., Ph.D., DACLAM, Emory National Primate Research Center
Kathleen Grant, Ph.D., Oregon National Primate Research Center
Shannan Hall-Ursone, D.V.M., Southwest National Primate Research Center
Erin Kinnally, Ph.D., California National Primate Research Center (CNPRC)
Gota Morota, Ph.D., Virginia Polytechnic Institute and State University
Linda Saif, Ph.D., The Ohio State University
Gregory Timmel, D.V.M., M.S., DACLAM, CNPRC
Christopher Tuggle, Ph.D., Iowa State University
Kristin Whitworth, Ph.D., National Swine Resource and Research Center

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Workshop Report

Opening Remarks

James Fox, D.V.M., M.S., DACLAM, Massachusetts Institute of Technology
Xiang-Ning Li, M.D., Ph.D., Office of Research Infrastructure Programs (ORIP), Division of Program Coordination, Planning, and Strategic Initiatives (DPCPSI), Office of the Director (OD), National Institutes of Health (NIH)
Guanghu (Jeff) Wang, Ph.D., M.B.A., ORIP, DPCPSI, OD, NIH

Dr. Xiang-Ning Li welcomed the attendees to Session 3 of the workshop. Dr. Li reminded the participants of NIH's dedication to rigor and reproducibility, which was emphasized by Dr. Robert W. Eisinger, Acting Director, DPCPSI, during Session 1. In 2021, the NIH Advisory Committee to the Director Working Group on Enhancing Rigor, Transparency, and Translatability in Animal Research recommended that the NIH encourage and support work to better understand, monitor, record, and report important extrinsic factors related to animal care that might affect research results.

Dr. Li also reminded the participants that ORIP has long devoted efforts to enhancing rigor and reproducibility, which was emphasized by Dr. Franziska Grieder, Director, ORIP, during Session 1. ORIP has supported this effort through scientific research workshops (e.g., Zebrafish and Other Fish Models: Extrinsic Environmental Factors for Rigorous Experiments and Reproducible Results; Validation of Animal Models and Tools for Biomedical Research) and publications of future funding opportunity announcements (e.g., NOT-OD-22-039). This workshop is one of several steps toward fulfilling ORIP's Strategic Plan by addressing the important endeavor of enhancing animal study rigor and reproducibility in NIH-supported research. The Extrinsic Factors Workshop seeks to better understand extrinsic factors and their effects on biomedical research. ORIP is modifying its infrastructure programs to address reproducibility in animal studies.

Dr. Jeff Wang, Workshop Coordinator, also welcomed the attendees. He provided examples of extrinsic factors related to animal research, which include temperature, humidity, noise, and lighting. Housing conditions—such as size and material of enclosure, number of animals per enclosure, bedding material and thickness, and cleanliness and cleaning schedules—also must be considered. Dr. Wang emphasized that the effects of extrinsic factors can be highly complex and often include multiple interactions. This issue has been understudied and under-documented. The goal of the workshop is to discuss the current status, needs, and strategies related to management, monitoring, and reporting of extrinsic factors to enhance the reproducibility and rigor of animal research. The focus is on the most widely and commonly used animal models, relevant extrinsic physical factors, and modern technologies. Dr. Wang expressed appreciation to the organizing committee members, speakers, and participants for their engagement.

Dr. James Fox, Workshop Chairperson, briefly highlighted Sessions 1 and 2 of the workshop. He emphasized that the topic of extrinsic factors is highly relevant to biomedical research, both for investigators and vivarium staff members. Dr. Fox also introduced Drs. Joyce Cohen and Kiho Lee, Session 3 Co-Chairs.

Keynote Presentation: Precision Medicine—Targeting Individual Differences to Increase Rigor and Translatability

Erin Kinnally, Ph.D., California National Primate Research Center (CNPRC)

Dr. Erin Kinnally presented on rigor and reproducibility in the context of precision medicine. She explained that CPNRC's BioBehavioral Assessment (BBA) Program is focused on addressing the key questions of who gets sick and why. Dr. Kinnally briefly described precision medicine, which is based on the concept that humans differ in disease susceptibility, and treatments ideally will leverage these

individual differences to improve outcomes. Genetics, lifetime exposures, and biobehavioral organization might play a role. Biomedical models for disease should address these factors for optimal translatability.

The BBA Program has assessed 5,600 infant rhesus macaques to understand individual differences in temperament, stress, reactivity, cognition, and social motivation. Recently, the program was embedded into CPNRC's operating base grant. The program's assessments since have expanded to incorporate a lifespan approach; the team is developing an aging BBA for macaques older than 15 years of age. To date, 128 aging macaques have been assessed, and 50 aging animals will be assessed annually in the future. Dr. Kinnally highlighted discoveries resulting from BBA models, which relate to airway hyper-responsiveness, social motivation, and depressive behavior. Future directions of the program include studying long COVID-19, wildfire exposure, and retinal disease.

Dr. Kinnally highlighted work with the aging BBA. The goals of this initiative are to characterize healthy biobehavioral aging in monkeys and to determine the factors that promote healthy biobehavioral aging (e.g., social connection, lifetime exposures, stress, diet, genetics/epigenetics, early biobehavioral organization). The overarching objective is to increase use of individual differences in nonhuman primate (NHP) disease models to increase rigor and reproducibility. All data are made available to investigators.

The aging BBA was based on five major domains of macaque aging: cognitive, motor, physiological/cellular, emotional, and social. Various tests were developed based on metrics that allow investigators to determine how the monkeys are aging across these domains. The tests include vision (e.g., near, distance), gait (e.g., speed, cadence, number of steps), decision making (e.g., reaction time, complexity, predictability), depressive behavior, and social cognition (e.g., social stimulus response, human imitation).

Dr. Kinnally noted the importance of increasing throughput through technology. Efforts in this area include coding videos, automating behavioral observation software, and recording through eye-tracking tablets. She noted that automation software increases capacity for capturing certain metrics that previously were inaccessible to researchers.

The BBA programs can increase the use of NHP models for aging. Dr. Kinnally noted that stakeholders in this effort include NHP model developers, the National Primate Research Centers (NPRCs), and individual investigators. Engagement efforts include sharing standardized protocols, data, and video training resources. Dr. Kinnally noted that individual monkeys can be studied at investigators' request.

Discussion

- In response to a question from Dr. Amy Ryan, Dr. Kinnally affirmed that the program is assessing macaques across different housing conditions and rearing histories. The animals' health and research history can be accessed.

- Dr. Sarah Carratt wondered about differences in biobehavior between macaques housed in U.S. versus European Union facilities. Dr. Kinnally responded that this question would be interesting to explore.

- Dr. Cohen asked how often BBA information is leveraged before animals are assigned to projects. She asked whether this process occurs for all assignments at the CNPRC or whether it is implemented only upon request. Dr. Kinnally explained that BBA infant data have been available for years through the internal health record system, which is available to CNPRC investigators. This resource is used frequently for behavioral health studies. Data are being made available on the public website.

- In response to a question from Dr. Fox, Dr. Kinnally spoke on whole-genome sequencing efforts at the program. She noted that genotyping efforts in this area have expanded over the years. Several efforts related to whole-genome sequencing are being pursued, and many tissue samples are available for analysis.

- Dr. Fox asked whether the program is engaging with other NPRCs to implement the study in New World monkeys. Dr. Kinnally stated that she recently became involved in a multi-NPRC effort to develop BBAs for marmosets.

Keynote Presentation: Husbandry of Biomedical Swine Models
Kristin Whitworth, Ph.D., National Swine Resource and Research Center (NSRRC)

Dr. Kristin Whitworth highlighted the NSRRC and management of extrinsic factors in animal research. She first listed the objectives of the NSRRC, which are to (1) ensure that biomedical investigators have enhanced access to critically needed swine models involving human health and disease and (2) serve as a central repository for materials (e.g., germline and somatic cells, gene probes, markers) and information.

The NSRRC has generated more than 1,300 cloned pigs, of which 685 were cloned by zygote injection. More than 90 distinct models have been developed, 13 of which were developed in 2021. The NSRRC distributes pigs, pig tissues, pig cell lines, and protocols and facilitates cloning and embryo transfers across the United States. Five housing facilities have been established at the University of Missouri; the NSRRC is a high-biosecurity xenobiology building that can house about 100 pigs. Overall, the entire campus has the capacity to house between 350 and 400 pigs.

NSRRC models support numerous areas, including such specialties as somatic cell genome editing, reproductive biology, xenotransplantation, immunology, neurobiology, and heart valve transplantation; conditions like Fanconi anemia, congenital heart defect, pulmonary fibrosis, seizure, cancer, Alzheimer's disease, influenza, cardiovascular disease, liver disease, and hepatitis; and bone, eye, and ear studies. Dr. Whitworth emphasized that the animals are used for multiple projects to maximize the use of tissues.

The NSRRC provides services to numerous organizations and institutions, including the University of Missouri, Oregon Health & Science University, University of Pittsburgh, Columbia University, and University of Louisville. The NSRRC also supports several NIH Institutes, Centers, and Offices (ICOs). Highly distributed pig models include enhanced green fluorescent protein pigs (e.g., retinal stem cells, spinal cord regeneration), Cre-inducible cancer models, and xenotransplantation models (e.g., *GGTA1*, *hCD55*). Additional models are in development. Dr. Whitworth noted that the NSRRC recently received C06 funding to expand the facility.

The *Guide for the Care and Use of Laboratory Animals* specifies the number of swine per pen, based on the animal's weight and pen size. For larger animals, particularly swine, it is important that the configuration of the space allow the animals to turn around and move freely. Food troughs and water devices should be provided in sufficient numbers to allow ready access for all animals. Direction on temperature and relative humidity also is provided in the *Guide*. General guidance for animal facilities relates to structural strength, water and power, and storage. Other standards relate to feeding and watering.

Dr. Whitworth spoke with NSRRC's Director of Animal Care and Quality Assurance, as well as investigators, to determine shortfalls and opportunities related to extrinsic factors. Suggestions included improving animal welfare (e.g., proactive approach, experienced veterinary and animal care staff, maintenance of low animal stress levels, best feeding practices), optimizing the *Guide* requirements (e.g., funding for density studies, optimal enrichment), reliable phenotyping and characterization (e.g. third-party validation, standard rubric), and infrastructure (e.g., video monitoring equipment,

image-based health assessments, bioinformatics). The group proposed establishing specialty centers around NIH ICOs, as well as a pig transport center. Investigators could travel to perform their work in the best location, similar to the NPRCs. Other ideas include collaborations between animal science departments and medical centers, as well as the addition of pig facilities and staff.

Dr. Whitworth also pointed out that some inconsistency might be beneficial in research; pigs are outbred with large litters, robust performance, and high fertility. Miniature pigs are inbred, with small litters and infertility. She shared a perspective that if a model provides a predicted phenotype regardless of its environment, it is a reliable and reproducible model. She also noted the need for extension of funding periods and reasonable requirements for documentation.

Discussion

- Dr. Joseph Newsome emphasized the need for expert training on swine management and reproductive physiology for non-animal science programs that use these models. Dr. Whitworth agreed but noted that hands-on experience is needed, and biosecurity restrictions can impose challenges for access.

Presentations: Construction, Housing, and Caging That Impact Rigor and Reproducibility in Studies Using Large Animals

Impacts of NHP Enclosure Design on Welfare and Rigor and Reproducibility

Gregory Timmel, D.V.M., M.S., DACLAM, CNPRC

Dr. Gregory Timmel presented examples of NHP enclosure designs and their effects on rigor and reproducibility. He also discussed recent research on innovative enclosure designs. He shared examples of large outdoor enclosures, corn crib–style enclosures, indoor housing, and indoor–outdoor housing. He noted that these designs can be applied for different types of experiments and breeding practices. Dr. Timmel highlighted examples of environmental enrichment and considerations for management. He pointed out that all NHP species should be considered in this context, not only macaques. Additionally, smaller species can be easier to house socially in smaller areas.

A study was performed at the Emory National Primate Research Center (ENPRC) to determine the effects on animal welfare (e.g., behavior, health, stress) of providing young male rhesus macaques access to larger outdoor spaces. The animals showed strong preference for the outdoor space, were more physically active and played with one another, and had diminished cortisol levels. No effects were observed on other behaviors, alopecia, body condition score, or diarrhea cases. Additionally, no consistent patterns of response on cognitive bias testing were observed.

Dr. Timmel underscored the importance of NHP studies to assess the effects of housing on animal health and behavior. He also briefly highlighted a study on the effects of psychosocial stress on immune response during acute simian immunodeficiency virus (SIV) infection in a pigtail macaque model. Another study examined social housing status and attentive response in rhesus macaques. Dr. Timmel noted a study that reported the effect of single housing on innate immune activation in pigtail macaques infected with SIV.

In summary, factors to consider in NHP research include social housing, enclosure features (e.g., size, location, complexity), indoors versus outdoors, substrates, secondary enclosure schedules, natural versus artificial lighting, light cycle, behavioral enrichment, individual animal effect, and phenotype. He emphasized that much still is unknown, and more work in this area is needed.

Automated Feeding Stations Provide a Rigorous Tool for Enhancing Outcome Accuracy on Social Determinants of Nutritional Health and Breeding Colony Management in Monkeys
Kelly Ethun, D.V.M., Ph.D., DACLAM, ENPRC

Dr. Kelly Ethun discussed the use of automated feeding stations as a tool to study social determinants of nutritional health and novel colony management strategies. She explained that automated feeders have been used at the ENPRC for neurobehavioral–nutritional health translational studies (e.g., stress-induced eating) and for resource-related research. These studies would have been impossible without a noninvasive, automated method to accurately monitor and control the dietary environment of socially housed monkeys on an individual basis.

ENPRC's automated feeding systems have been used as a basis for high-throughput designs for use in outdoor corral-based feeding studies. This work was conducted in partnership with Research Diets, Inc. Improvements include a stainless-steel hopper, waterproof frame, Power over Ethernet computer and Category 6 cable, and a highly sensitive dispensing system that measures food pellets on a weighting platform with high accuracy. Dr. Ethun shared a video demonstrating the system's principle of operation.

The ENPRC field station houses more than 2,300 animals, primarily in large outdoor enclosures. The Center has purchased 32 commercial BioDAQ feeders to outfit eight compounds, which house about 750 animals. The feeders are a valuable resource to the Center and provide a modern alternative to conventional bin-feeding practices. Disadvantages of the bin hoppers include food waste, potential contamination, availability to vermin and other pests, and lack of accurate consumption monitoring. Animals fed from automated feeders, in contrast, waste less food, and intake can be quantified accurately.

Dr. Ethun noted that efficient social health surveillance methodologies can be employed to identify groups at risk for social instability before the onset of fighting. Potential consequences of unresolved conflicts include increased wounding and stress. Social stress resulting from group instability can lead to decreased reproductive success, decreased animal availability, increased variability in neurobehavioral and immunological processes, and decreased reproducibility. This new approach can enable automated and real-time monitoring. Dr. Ethun shared a case study of intrafamily aggression and daily caloric intake.

The Feeding Interaction Network (FIN) Project, funded by ORIP, involves the development of advanced computational approaches using temporal proximity feeding interaction data to enhance social health surveillance of rhesus breeding groups. FIN network analyses can be used to examine co-feeding patterns among group members and identify subgroups in feeding communities that are associated with changes in kinship and dominance structures. Management can use timely information to increase observations and develop strategies to intervene prior to escalation of aggression. New computational methods can be used to develop and validate community detection algorithms and machine learning models.

In summary, feeding behavior of socially housed monkeys provides useful information about nutritional health and the influence of socioenvironmental factors. Automated feeding data provide information about individual variability in caloric intake, feeding duration and timing, diet preference, and partner choice. Studies investigating feeding behavior of group-housed rhesus monkeys require high-throughput and modern equipment to ensure rigor and reproducibility. Modern primate research facilities need rigorous monitoring systems to optimize NHP well-being and foster the conduct of high-quality science.

Biocontainment Protocols for Improving Survivability of Severe Combined Immunodeficient (SCID) Pigs
Christopher Tuggle, Ph.D., Iowa State University

Dr. Christopher Tuggle discussed protocols for improving survivability in studies using SCID pigs. He explained that SCID pigs cannot reject xenografts of human cells. SCID pigs are used in cancer xenograft

studies, stem cell xenograft studies, and cell–drug interaction testing. Researchers are interested in exploring whether the SCID pig can be developed as an alternative biomedical model to rodents in such xenograft studies as well as in humanization (creation of a human immune system in the SCID pig). Attributes of a good animal model include accurate modeling of human disease or condition and routine availability for low cost, with minimal requirements for client husbandry expertise.

Dr. Tuggle has demonstrated that an ovarian cancer cell line can survive in a SCID pig model. In this work, the SCID pig model has allowed the development of a tumor similar to that in human patients. The SCID pig might be better than the SCID mouse for modeling certain markers. Human skin transplants also have been shown to survive on the SCID pig in a proof-of-concept study. Additionally, injection of human hematopoietic stem cells demonstrates substantial humanization at the neonatal stage.

In an ORIP-funded project, investigators developed three positive-pressure "bubbles" for SCID pigs. High-efficiency particulate air (HEPA)–filtered air flows out. Water is sterile filtered, ultraviolet treated, and acidified. Personnel wear Tyvek, double gloves, a hair net, and a surgical mask. The three bubbles are designated for entry, production, and fee for service. Husbandry initially was performed through snatch-farrow carrier gilts. This approach provides colostrum and a complex microbiota and is as close to natural as possible. However, reproduction is inefficient, and the system requires a large, high-risk investment. Biosecurity risks also are of concern.

The team now is exploring options for cesarean section to create gnotobiotic pigs that initially are raised in isolators and later are transferred to biocontainment. This approach decreases exposure of piglets to outside air during their most susceptible stage and is performed in a batch process. The approach also allows manipulation of different colostrum sources and microbiota types, which could help researchers move toward standardization. Disadvantages include pig fragility and the need to move to the bubble at 4–6 weeks for most projects. Additionally, large-animal expertise is needed.

Survivability was greatest for the snatch-farrow approach, although this approach—unlike the cesarean section—was used only in non-cloned pigs. Testing in this area is in progress. Dr. Tuggle explained that the major cause of death was early euthanasia due to poor health, generally resulting from sepsis. The source of bacteria is unknown, and more investigation in this area is needed. Treatments are provided based on clinical signs. Gut microbiome sequencing revealed that the number of bacteria is low, compared to process controls. As expected, the bacterial communities are not highly diverse. *Lactococcus* is the most abundant operational taxonomic unit across all samples, and *Enterococcus* is observed at later time points.

In summary, the combination of SCID and gnotobiotic condition might be uniquely difficult to raise. Widespread sepsis is caused by poor neonatal gut health, leading to lack of gut closure and sepsis by opportunistic bacteria. Cloned SCID pigs also might be uniquely susceptible. Complex microbiota and porcine colostrum might be required for reproducible husbandry of SCID pigs. Future plans include focusing on early gut health and gut closure, comparing the outcome of defined versus complex microbial populations, investigating immunoglobulin source and delivery, and comparing cloned and non-cloned pigs.

Gnotobiotic Pigs Infrastructure and Biological Model
Linda Saif, Ph.D., The Ohio State University

Dr. Linda Saif presented on infrastructure and biological models for gnotobiotic pigs. She first explained that germ-free pigs require a large-animal germ-free facility with temperature-controlled rooms, an air system with a central turbine unit and HEPA filtration, and positive-pressure isolators. Dr. Saif briefly outlined the layout of the facility, noting the inlet and outlet filters, housing isolator, isolator floor, and

enrichment toys. Extrinsic environmental factors include temperature, lighting, control of humidity, and space limitations. Extrinsic host factors include swine breed, disease status, litter size, and sterility.

Dr. Saif asserted that the pig has multiple advantages over other animal models (e.g., rodents, NHPs). Considerations include similarity to humans, ethical acceptance, high hygienic status, good animal compliance, sufficient sample material, good reproduction data, and established genetic modification. Maintenance costs of swine, however, remain a significant limitation. Dr. Saif noted that pigs can provide a dual model to study enteric viral infections and vaccines. Pigs are the only animal model that is susceptible to human rotavirus diarrhea and norovirus. Pigs are outbred and are anatomically, physiologically, and genetically similar to human infants. Additionally, extraneous enteropathogens, microbiota, and maternal antibodies are absent. For these reasons, the pig is a highly relevant model for mechanistic and translational research on human disease.

Germ-free piglets provide a unique model for studies of the microbiome, rotavirus infection, immunity, and vaccines. Goals and gaps include an approach for colonization with neonatal human fecal microbiota and probiotics, a model to understand the effects of the microbiome and probiotics on immunity and vaccines, and a model for evaluating why current rotavirus vaccines fail in field environments. Dr. Saif highlighted previous work demonstrating probiotic effects on rotavirus immunity and vaccines. Nutritional effects also have been demonstrated in pig models for vitamin A deficiency and protein malnutrition.

Gaps in the gnotobiotic pig model include lack of immunological tools and reagents, limited availability of gene expression microarray and proteome, limited knockout and genetically modified pigs, need for interagency partnerships, and need for federal support for germ-free large-animal facilities.

Discussion

- A participant asked whether the sex of experimenters could affect experimental outcomes in NHPs, similar to mice. Dr. Timmel replied that he was unaware of studies on this topic but that this could represent a potential variable.

- Dr. Cohen asked whether smaller automated feeders could be used in more conventional NHP facilities. Dr. Ethun responded that smaller feeders are commercially available.

- In response to a question from Dr. Lee, Dr. Saif explained that gnotobiotic pigs can be maintained in isolator bubbles for 8 weeks. Longer-term options are available if needed. The animals can be maintained in a calf isolator for 3 months.

Presentations: Equipment and Technology That Enhance Rigor and Reproducibility in Studies Using Large Animals

Markerless Motion-Capture Technology

Shannan Hall-Ursone, D.V.M., Southwest National Primate Research Center (SNPRC)

Dr. Shannan Hall-Ursone discussed the incorporation of gait and movement analysis technology to enhance clinical care and research outcomes. The specific aims of this project were to (1) adapt the Southwest Research Institute's markerless biomechanics technology to track baboons at Texas Biomedical Research Institute and (2) determine whether this technology could be used noninvasively to obtain information from animals that would electronically determine normal movement. The end goal was to use the analysis to identify abnormal movements using predetermined data and identify early signs of injury or disease. This information provides both clinical and research value.

The Human Performance Initiative was aimed at tracking joint angles and positions using monocular cameras, with no instrumenting of test subjects. The results of this study potentially could benefit two aspects of the 3Rs (i.e., replacement, reduction, and refinement) of animal research. If favorable results are obtained, reduction will be achieved, because the technology can aid in finding optimal candidates for study. Refinement can be achieved by using this technology to aid in early endpoints, as well as pain assessment. For clinical cases, understanding changes in the animal's movements that indicate pain will allow researchers to initiate treatment earlier, therefore increasing animal quality of life and welfare.

Dr. Hall-Ursone outlined the experimental setup, which included an enclosure with indoor and outdoor access, with cameras positioned in selected areas. Initial tasks included updating the neural network and annotation, identifying and labeling more than 300 images of baboons, designing and deploying a data-capture system for the baboon enclosure, adapting a human performance capture tool to fit the application, and capturing about 20,000 frames of data from seven baboons. Dr. Hall-Ursone shared a video recording using this approach.

Research applications of the technology include marmoset and baboon models for Parkinson's disease, a marmoset model for multiple sclerosis, and treadmill studies to characterize gait in baboons and marmosets. This project has fostered a strong partnership with the Southwest Research Institute, and grants currently are being pursued by two SNPRC scientists. Another investigator is determining how to modify the technology for use in marmosets. Dr. Hall-Ursone noted that a method for animal identification must be developed. Additionally, radiographic evaluation could help provide evidence for correlation between arthritis and movement.

Future tasks include using collected data to update baboon monitoring, correlating observed behavioral or physical changes with changes in posed data, and determining the technology's use in additional animal models and project applications.

Rigor and Reproducibility in Cognitive Behavior Without Social Isolation During Testing
Kathleen Grant, Ph.D., Oregon National Primate Research Center

Dr. Kathleen Grant spoke on the assessment of cognitive behavior in an animal model for voluntary alcohol self-administration. She explained that traditionally, cognitive behavior is assessed in NHPs in an isolated environment. She noted that lack of throughput is a limitation of this approach. She presented a new experimental setup, in which operant panels are embedded in quad housing cages for food and fluid intake and cognitive behavioral testing. Modifications include caging, electricity, cable management, and husbandry.

Advantages of the approach include precise measures of fluid and food intake; precise measures of timing; event-triggered chains of behavioral assessments; and lack of disruption due to relocation, which allows spontaneous sequencing of behaviors. Dr. Grant outlined the experimental model, which includes key timepoints in training and self-administration. Cognitive testing is performed via magnetic resonance imaging and hypothalamic-pituitary-adrenal axis function. Data can be analyzed and grouped via machine learning. She showed a representative video of individual differences in animal choices.

Dr. Grant shared data indicating that drinking behaviors can be predicted by baseline performance. Imaging studies indicate that the striatum, orbitofrontal and prefrontal cortices, and ventromedial prefrontal and superior temporal cortices play a role in determining performance. Performance can be improved through manipulation of the putamen with designer receptors exclusively activated by designer drugs (DREADDs).

Dr. Grant asserted that this approach is novel, efficient, replicable, predictive, sensitive, and longitudinal. Future directions include expansion into all housing environments. She concluded by emphasizing several

points: (1) cohort designs can test and reaffirm the rigor and reproducibility of scientific approaches, (2) allowing individual differences helps reveal predispositions to behavioral disorders mediated by neural circuitry, (3) throughput is essential for integration across data domains, and (4) a tissue and data repository procedure allows *ex vivo* synaptic recording and banking of brain areas and peripheral tissues.

Machine Learning–Enabled Pig Activity Monitoring
Gota Morota, Ph.D., Virginia Polytechnic Institute and State University

Dr. Gota Morota discussed recent work on machine learning and new technologies to perform monitoring in pigs. He first highlighted three components of genetics: phenotyping, genotyping and sequencing, and the phenotype–genotype relationship. The cost of genome sequencing has decreased substantially in recent years; as a result, phenotyping now is more expensive (i.e., in both money and labor) than genotyping.

Phenotyping has emerged as a major bottleneck in recent years. Phenotypes that are difficult to collect with current technologies include activity, behavior, social interaction, and repeated records. Real-time, continuous monitoring is critical in large animals (e.g., pigs). Available new systems include computer vision (e.g., videos, images) and wearable sensors.

New 3D depth-sensor cameras can be used to generate both color and dense-depth images, as well as other information. Computer vision systems for automated monitoring can record morphology (e.g., growth rate, body weight, body condition score) and activity (e.g., distance traveled; frequency of standing, sitting, or lying down; food and water intake).

Dr. Morota highlighted work focused on continuous monitoring of pig body weight from image data. He shared an example of a depth video. The images can be used to determine width, length, and height, which are used to calculate body volume; volume is highly correlated with body weight. Dr. Morota also discussed determination of pig activity, which involves tracking the same animal consistently across many frames and performing multi-object tracking. Trackers include sparse optical flow, multiple-instance learning, and channel and spatial reliability. Motion hotspots also can be detected.

Continuous monitoring of activity also can be achieved through the use of wearable sensors, which are attached to the animal's back with a harness. Dr. Morota briefly outlined the experimental setup. Data are annotated using Data Capture Lab software. Several behaviors—such as eating, lying down, walking, and standing—can be determined through machine learning performance comparison.

Dr. Morota concluded by outlining future directions for this work. He noted challenges, which include video data size and identification, battery duration, and the need for real-time monitoring. He suggested exploring ways in which computer vision and wearable sensors can be leveraged to help one another.

Automated Spatial and Nonspatial Memory Testing in Laboratory Pigs
Timothy Allen, Ph.D., Florida International University

Dr. Timothy Allen discussed his work on automating spatial and nonspatial memory testing in laboratory pigs. His laboratory is focused on the relationship between neurobiology and cognition. The group has used rodent models primarily, with a typical cross-species approach (i.e., moving directly between rodents and humans). He noted, however, that an additional animal model is needed to understand these systems. The group has developed pigs as a preclinical model for behavioral neuroscience.

The pig brain is about one-tenth the size of the human brain and is heavily gyrated, with long-distance networks and well-defined hippocampal formation. Previous studies suggest that pigs have strong spatial memory and can learn spatial tasks easily. To facilitate rigorous comparisons with rodent work,

Dr. Allen's group built a large, automated maze suitable for pigs. Tracking is performed in real time from a separate location. Dr. Allen shared representative videos and data from maze-based tracking experiments. He also highlighted the use of touchscreens to test cognition in pigs by assessing fixed and conditional associations. He shared a representative video of the animal performing the task. The generated data can be compared directly with results from experiments in humans.

The team also developed the "HogHat," which can be applied for neural recordings. Depth electrodes are implanted into the brain through the device for measurement of chronic pig neurophysiology. Additionally, spatial behaviors can be correlated to brain volumetrics through structural and diffusion-weighted neuroimaging.

Dr. Allen concluded by emphasizing that automated cognitive assessments can be performed reliably in pigs—with all the laboratory rigor available in rodents—using the automated maze and touchscreen setups. These setups allow rigorous comparisons with rodent and human tasks and facilitate pigs as a large-animal model for translational neuroscience. He emphasized that cognition is an important intrinsic factor, and laboratory tests will be needed to better understand this variable.

Discussion

- In response to a question from Dr. Matthew Jorgensen, Dr. Hall-Ursone clarified that the motion-capture system can identify and differentiate among individual animals.

- Dr. Cohen asked whether the motion-capture system can be used to capture abnormalities in animals prior to their use in experiments. Dr. Hall-Ursone commented that several efforts in this area are ongoing, and the technology could be applied for this use in the future.

- Dr. Ryan asked whether the animals in the cognition studies are separated from their pair-mates during study time or are singly housed during the entire experiment. Dr. Grant explained that the animals spend 2 hours per day with their partner and are separated during the rest of the period. She noted that the team is investigating other options that use radio-frequency identification technology, which could allow the pair-mates to stay together.

- Dr. Cohen asked Dr. Grant how the lack of relocation might affect study results. Dr. Grant responded that this approach is more efficient and highly replicable. She stated that she is interested in applying the tool to more complex housing environments.

- Dr. Grant remarked that low performers tend to be distracted by other factors in the room. She is interested in exploring attention deficit behaviors further. Mr. Alan Olzinski asked whether distracted animals might respond differently in isolation. Dr. Grant agreed that isolation likely would affect the results, but the team was interested in testing natural disposition.

- Dr. John Vanchiere wondered about the use of attention-deficit/hyperactivity disorder (ADHD) medications to help low performers. He noted that untreated ADHD might be related to future alcohol-use disorder. Dr. Grant was uncertain but noted that training of animals is likely to play a role in changing behavior. She noted that the field of medicine is moving toward specific circuitry manipulations and underscored the importance of more work in this area.

- Dr. Lee asked how many pigs can be tracked simultaneously. Dr. Morota replied that currently, the technology can accurately monitor as many as three pigs at once. Dr. Lee commented that for larger facilities, multiple cameras would be needed. In response to a follow-up question, Dr. Morota stated that the cost of the sensor is about $20–30, and the cost of the video camera is about $250–300.

- Dr. Lee asked about the size of the pig maze and touchscreen room. Dr. Allen noted that mazes are 17 feet long by 15 feet wide, and height is the greatest constraint. A hang-over version of the touch screen has been developed, so the devices can be moved as needed. He suggested that in the future, multiple pig touchscreen experiments could be performed simultaneously. The ideal weight limit is about 150 pounds; the group's heaviest pig in these experiments was about 220 pounds.

- A participant asked whether odor-based memories are addressed in repeated tests. Dr. Allen stated that the space is sanitized between animals, with a period of several hours between tests. The effects of odor cues cannot, however, be eliminated entirely.

- In response to a question from Dr. Lee, Dr. Allen stated that he has not observed sex differences in spatial memory. He noted that social dynamics play a role in results.

- Dr. Reginald Miller commented that the differences between adult and adolescent memory in smaller pigs should be considered. Dr. Allen noted that the tests can be used to determine changes in memory during development. The pig brain reaches its adult state by about 9–12 months. He noted that his team is collecting a data bank of pigs up to 6 years of age, and a substantial change in spatial performance has not been noted.

- Dr. John Hasenau asked Dr. Allen whether the HogHat studies can be performed in a social environment. Dr. Allen explained that the pig skull is thick and hard, which allows mounting of the device. Therefore, paired housing likely would be feasible, but he has not yet attempted to do so. Currently, the pigs are housed in neighboring pens.

Group Discussion and Summary

- Dr. Katherine Roe asked whether investigators who purchase animals through the NPRC system are provided a historical knowledge (e.g., rearing, housing, experimental history) of their animals. Dr. Kinnally responded that access to this information is dependent on various factors. She agreed that it would be a good idea to inquire about these factors and report them, if they are available. Dr. Lee added that these factors are challenging to obtain for pig research.

- Dr. Cohen asked the participants for their opinions on reporting of various extrinsic factors in publications. Dr. Grant commented that these factors (e.g., time since acquisition, age of acquisition, time in the experimental facility) are important to understand. She noted, however, that methods sections often are truncated in publications, and many journals discourage submission of supplemental materials. Medical records would be beneficial in experimental designs but likely would not be included in publications. Several participants commented that housing conditions are regularly reported. Dr. Whitworth noted that for pigs, the important factors are dependent on the experimental questions. Journals also have specific requirements.

- Dr. Fox inquired about addressing the history of NHPs imported from China. Dr. Cohen agreed on the importance and challenges of this issue. Dr. Saif noted that previous and current housing status is likely to affect the microbiome in NHPs. Dr. Fox added that this topic represents an important consideration that could be explored in a future workshop.

- Dr. Saif wondered whether animal models could be used to explore differences in susceptibility to COVID-19 and other diseases among socioeconomic groups. Dr. Cohen agreed that this topic would be interesting to explore, and NHPs have served as a valuable model for COVID-19 research. Dr. Grant added that Dr. Ethun's work on social status could provide opportunities for

further exploration in this area. Dr. Ethun noted that NHP social status has been used as a model for socioeconomic studies. She described studies examining changes in gene expression during rearrangement of social groups.

- Drs. Whitworth and Allen added that swine also establish a social hierarchy. Drs. Saif and Tuggle noted that field studies on this topic have been completed. Dr. Saif added that relevant variables (e.g., transport-induced stress) could be considered in this context.

- Several participants suggested that social rank be considered as an extrinsic factor. Dr. Allen cautioned that numerous factors might be present in these dynamics, and further investigation is needed. Dr. Ethun added that the overall effect of chronic stress might be more relevant. Dr. Grant noted that gestation in high-stress environments should be considered in the context of epigenetic changes. She added that protective effects have been reported.

- Dr. Fox asked about consideration of the Animal Research: Reporting of *In Vivo* Experiments (ARRIVE) 2.0 Essential 10 Checklist. Dr. Grant expressed support for the guidelines. She noted the need to address statistical requirements, particularly in the context of artificial intelligence. Dr. Ethun agreed on the importance of reporting extrinsic factors.

- Dr. Saif wondered about the importance of incorporating sex as a biological variable. Dr. Grant explained that in rhesus macaques, a spectrum of sexual maturity is present between 2–4 years of age. The brain continues to develop until 6–7 years of age. She noted that researchers often do not monitor these variables and therefore cannot report them. Dr. Saif also commented on the importance of reporting previous pregnancies and miscarriages in research.

- In response to a question from Dr. Fox, Dr. Saif underscored the importance of the microbiome on nutritional factors and the immune response. She noted that sterilization of pig diets is expensive. She added that she plans to refine and explore these dynamics in future studies. Dr. Cohen remarked that NHP researchers also are focused on characterizing the microbiome, including comparative studies of wild and captive animals. Most of these studies are focused on the microbiome in the context of HIV.

- Dr. Saif noted that companion animals tend to share microbiota with their owners. She wondered whether similar studies have been conducted in NHPs. Dr. Cohen was unaware of studies in this area. Dr. Fox remarked that Helping Hands: Monkey Helpers for the Disabled, Inc. might provide opportunities for study in this area.

- Dr. Roe asked about the feasibility of establishing a balance between controlling extrinsic factors while ensuring that experiments remain generalizable and translatable. Dr. Cohen agreed that every factor cannot be controlled fully, but thorough reporting can provide insight into which factors are most important. Dr. Grant added that many investigators are hesitant to modify their established systems. Dr. Timmel remarked that more research on these variables is needed. Such experiments are expensive to perform, and additional funding opportunities will be needed.

- Dr. Allen pointed out that considerations related to reproducibility necessitate both increased throughput and sample size; a balance between those two needs must be considered. Dr. Grant pointed out that NHP researchers have adapted their experimental designs over time to new advances in housing and enrichment. She noted the importance of ongoing flexibility in research.

Session Wrap-Up, Workshop Closing, and Adjournment

Dr. Li reminded the participants of NIH's dedication to rigor and reproducibility in biomedical and biobehavioral research, which was emphasized throughout the workshop. He briefly highlighted the previous session topics, co-chairs, and presenters. Dr. Li listed take-home messages from the workshop:

- Many variations exist among animal models, species, and strains of animals. Each component has specific characteristics, extrinsic factors, and needs for consideration.
- Researchers cannot standardize every extrinsic factor, because too many exist. More studies and discussions on this topic will be needed.
- Monitoring, recording, analyzing, and reporting will be needed.
- Current gaps include equipment for sensing, detecting, monitoring, real-time analyzing, and reporting. Other needs include newly designed fish tanks and uniform lighting in rodent cages.
- Reporting of extrinsic factors will help researchers increase transparency and will help other researchers manage those factors in their work.
- These efforts will entice more stakeholders to join the effort to improve rigor and reproducibility.

Dr. Li explained that the organizing committee will continue to meet after the workshop. Co-chairs of each group will develop a sub-report on each session; a summary of the overall workshop will be produced, and gaps might be suggested. ORIP's Division of Construction and Instruments (DCI) will analyze and identify gaps, inadequacies, or deficiencies in its infrastructure programs and will explore potential avenues to address gaps in monitoring, recording, and reporting. Dr. Li also noted that DCI manages construction and instrumentation programs.

In collaboration with other NIH ICOs, DCI will seek to promote awareness of, advocate support for, and work with other stakeholders to enhance rigor and reproducibility. Dr. Li emphasized that efforts from the scientific community will be needed to address these issues. He thanked the speakers, organizers, and participants for their engagement during the meeting.

Dr. Wang underscored ORIP's commitment to addressing the issue of rigor and reproducibility and emphasized that work in this area is ongoing. Dr. Wang adjourned the meeting.

Appendix A: Meeting Agenda

Session 3. Large Animals (Nonhuman Primates/Swine)
Virtual Meeting
September 30, 2022

12:00–12:10 p.m.	**Opening Remarks** *James Fox, D.V.M., M.S., DACLAM, Massachusetts Institute of Technology* *Xiang-Ning Li, M.D., Ph.D., Office of Research Infrastructure Programs (ORIP), Division of Program Coordination, Planning, and Strategic Initiatives (DPCPSI), Office of the Director (OD), National Institutes of Health (NIH)* *Guanghu (Jeff) Wang, Ph.D., M.B.A., ORIP, DPCPSI, OD, NIH*
12:10–12:40 p.m.	**Keynote Presentation: Precision Medicine—Targeting Individual Differences to Increase Rigor and Translatability** *Erin Kinnally, Ph.D., California National Primate Research Center (CNPRC)*
12:40–1:10 p.m.	**Keynote Presentation: Husbandry of Biomedical Swine Models** *Kristin Whitworth, Ph.D., National Swine Resource and Research Center*
1:10–2:30 p.m.	**Presentations: Construction, Housing, and Caging That Impact Rigor and Reproducibility in Studies Using Large Animals**
	Impacts of NHP Enclosure Design on Welfare and Rigor and Reproducibility *Gregory Timmel, D.V.M., M.S., DACLAM, CNPRC*
	Automated Feeding Stations Provide a Rigorous Tool for Enhancing Outcome Accuracy on Social Determinants of Nutritional Health and Breeding Colony Management in Monkeys *Kelly Ethun, D.V.M., Ph.D., DACLAM, Emory National Primate Research Center*
	Biocontainment Protocols for Improving Survivability of Severe Combined Immunodeficient Pigs *Christopher Tuggle, Ph.D., Iowa State University*
	Gnotobiotic Pigs Infrastructure and Biological Model *Linda Saif, Ph.D., The Ohio State University*
2:30–2:40 p.m.	**Break**
2:40–4:00 p.m.	**Presentations: Equipment and Technology That Enhance Rigor and Reproducibility in Studies Using Large Animals**
	Markerless Motion-Capture Technology *Shannan Hall-Ursone, D.V.M., Southwest National Primate Research Center*
	Rigor and Reproducibility in Cognitive Behavior Without Social Isolation During Testing *Kathleen Grant, Ph.D., Oregon National Primate Research Center*

	Machine Learning–Enabled Pig Activity Monitoring *Gota Morota, Ph.D., Virginia Polytechnic Institute and State University*
	Automated Spatial and Nonspatial Memory Testing in Laboratory Pigs *Timothy Allen, Ph.D., Florida International University*
4:00–4:30 p.m.	**Group Discussion and Summary**
4:30–4:50 p.m.	**Session Wrap-up**
4:50 p.m.	**Adjournment**

Appendix B: Participants List

Session 3. Large Animals (Nonhuman Primates/Swine)
Virtual Meeting
September 30, 2022

Leigh Allen, *Eunice Kennedy Shriver* National Institute of Child Health and Human Development (NICHD), National Institutes of Health (NIH)
Timothy Allen, Florida International University
Yoko Ambrosini, Washington State University
Amanda Armijo, Massachusetts Institute of Technology
Matthew Arnegard, Office of Research Infrastructure Programs (ORIP), Division of Program Coordination, Planning, and Strategic Initiatives (DPCPSI), Office of the Director (OD), NIH
Carmen Arsuaga, Nationwide Children's Hospital
Jill Ascher, Office of Scientific Resources, Office of Research Services, OD, NIH
Julia Bachman, National Institute of Neurological Disorders and Stroke (NINDS), NIH
Kate Baker, Tulane National Primate Research Center (TNPRC)
Ashley Barnes, ORIP, DPCPSI, OD, NIH
Doug Bartels, National Ferret Research and Resource Institute, The University of Iowa
Taylor Bennett, National Association for Biomedical Research
Skip Bohm, TNPRC
Francesca Bosetti, NINDS, NIH
Aleaya Bowie, Vanderbilt University Medical Center
Christopher Braunger, Washington National Primate Research Center
Jacqueline Brockhurst, Johns Hopkins University School of Medicine
Patricia Brown, Office of Laboratory Animal Welfare (OLAW), Office of Extramural Research (OER), OD, NIH
Monika Burns, Novartis
Ceara Byrne, Massachusetts Institute of Technology
Sarah Carratt, Seagen
Shreaya Chakroborty, National Institute on Aging (NIA), NIH
Anthony Chan, Center for Scientific Review (CSR), NIH
Susan Chandran, ORIP, DPCPSI, OD, NIH
Michael Chang, ORIP, DPCPSI, OD, NIH
Naomi Charalambakis, Federation of American Societies for Experimental Biology
Beatrice Chen, Brigham and Women's Hospital
Megan Clark, OLAW, OER, OD, NIH
Joyce Cohen, Emory University
Kristine Coleman, Oregon National Primate Research Center (ONPRC)
Ricki Colman, University of Wisconsin–Madison
Karina Concha, Florida Atlantic University
Meghan Connolly, Office of Scientific Resources, Office of Research Services, OD, NIH
Miguel Contreras, ORIP, DPCPSI, OD, NIH
Maria Crane, Emory National Primate Research Center (ENPRC)
Devon Crawford, NINDS, NIH
Joette Crews, Emory University
Christina Cruzen, University of Washington
John Dennis, U.S. Food and Drug Administration (FDA)
Cynthia Doane, The University of Arizona
Samantha Earlywine, Nationwide Children's Hospital
Catalina Echeverri, The Rockefeller University
Mark Eichelberg, American Physiological Society

John Engelhardt, The University of Iowa
Kelly Ethun, ENPRC
Ted Evans, Georgia Institute of Technology
Jeffrey Everitt, Duke University
Niora Fabian, Massachusetts Institute of Technology
Cameron Fili, FDA
Craig Fletcher, The University of North Carolina at Chapel Hill
James Fox, Massachusetts Institute of Technology
Olga Franco, Charles River Laboratories
Emily Franklin, Massachusetts Institute of Technology
Maria Fe Lanfranco Gallofre, NIA, NIH
Sarah Gillis-Smith, Massachusetts Institute of Technology
Neera Gopee, OLAW, OER, OD, NIH
Melanie Graham, University of Minnesota
Kathleen Grant, ONPRC
Travis Hagedorn, The University of Kansas Medical Center
Shannan Hall-Ursone, Southwest National Primate Research Center
Caitlin Haller, Nationwide Children's Hospital
Lisa Halliday, Biologic Resources Laboratory
Susan Harper, Inwood Animal Center
John Hasenau, Lab Animal Consultants
Renee Hernandez, GlaxoSmithKline
Nancy Hitt, NINDS, NIH
Tuan Hoang, Fluid Synchrony, LLC
Camila Hochman Mendez, Texas Heart Institute
Lydia Hopper, Johns Hopkins University
Charlotte Hotchkiss, University of Washington
Charlie Hsu, University of Washington
Denise Hsu, U.S. Military HIV Research Program
Maureen Humphrey-Shelton, U.S. Army Medical Research and Development Command
Eric Hutchinson, Johns Hopkins University
Naveena Janakiram, National Cancer Institute (NCI), NIH
Remi Jawando, Seagen
Walter Jeske, Loyola University Chicago
Alex Johnson, TNPRC
Crystal Johnson, Georgetown University
Lisa Jones-Engel, People for the Ethical Treatment of Animals
Matthew Jorgensen, Wake Forest University School of Medicine
Deepak Kaushal, Texas Biomedical Research Institute
Kylie Kavanagh, Wake Forest University School of Medicine
Roseann Kehoe, Rutgers, The State University of New Jersey
Greena Kim, Emory University
Erin Kinnally, California National Primate Research Center (CNPRC)
Madison Klanke, Turner Scientific
Kim Klukas, The Hormel Institute, University of Minnesota
Donna Kupniewski, Monell Chemical Senses Center
Kelsey Lambert, Wake Forest University School of Medicine
Malcolm Lane, University of Maryland Baltimore
Kiho Lee, University of Missouri
Karen Lencioni, California Institute of Technology
Jori Leszczynski, University of Colorado Denver and University of Colorado Anschutz Medical Campus
Denyse Levesque, ENPRC
Xiang-Ning Li, ORIP, DPCPSI, OD, NIH

Xin Li, New York University
Alex Lindquist, University of Colorado Boulder
Courtney Lunger, Massachusetts Institute of Technology
Alexander Mamishev, University of Washington
John Manker, Turner Scientific
Lindsay Marshall, The Humane Society of the United States
Drew Martin, ONPRC
Julie Mattison, NIA, NIH
Rachele McAndrew, Massachusetts Institute of Technology
Derek McLean, Office of AIDS Research, OD, NIH
Andres Mejia, Wisconsin National Primate Research Center
Ana Melero, University of Valencia
Istvan Merchenthaler, University of Maryland Baltimore
Reginald Miller, Mount Sinai School of Medicine
Oleg Mirochnitchenko, ORIP, DPCPSI, OD, NIH
DP Mohapatra, NINDS, NIH
Elizabeth Moore, Cornell University
Rafael Moreno Gómez-Toledano, Universidad de Alcalá
Gota Morota, Virginia Polytechnic Institute and State University
Joseph Mudd, Tulane University
Stephanie Murphy, ORIP, DPCPSI, OD, NIH
Judy Murray, Charles River Laboratories
India Napier, Massachusetts Institute of Technology
Joseph Newsome, University of Pittsburgh
John Norton, Duke University
Alan Olzinski, GlaxoSmithKline
Carly O'Malley, Charles River Laboratories
Allison Ostdiek, The University of Chicago
Rebecca Osthus, American Physiological Society
Missy Painter, Johns Hopkins University
Matt Parsons, Henry M. Jackson Foundation for the Advancement of Military Medicine
Kelly Pate, Massachusetts Institute of Technology
Norman Peterson, Seagen
Katy Phillip, The University of Arizona
Mahesh Pillai, The University of Toledo
Larisa Poluektova, University of Nebraska Medical Center
Ori Pomerantz, CNPRC
Carol Raymond, U.S. Army Institute of Surgical Research
Gregory Reinhard, University of Pennsylvania
Francisco Rendon-Gonzalez, Regeneron Pharmaceuticals
Katherine Roe, People for the Ethical Treatment of Animals
Amy Ryan, National Institute of Mental Health, NIH
Linda Saif, The Ohio State University
Melissa Sanchez, U.S. Army Institute of Surgical Research
Alfredo Sancho, Office of Intramural Research, OD, NIH
Rachel Sarabia Estrada, Mayo Clinic
Sarah Schlink, University of Missouri
Jenna Schmidt, University of Wisconsin–Madison
Caroline Schomer, The University of Texas at Austin
Diana Scorpio, Texas Biomedical Research Institute
Riti Sharan, Texas Biomedical Research Institute
Anuj Sharma, Office of Research Integrity, U.S. Department of Health and Human Services
Karlie Sharma, National Center for Advancing Translational Sciences, NIH

Vanessa Sherk, CSR, NIH
Bhupinder Singh, Rutgers, The State University of New Jersey
Anna Skorupski, University of Pittsburgh
Heather Smith, Office of Animal Care and Use, Office of Intramural Research, OD, NIH
Jeff Stanton, ONPRC
Xiaoping Sun, NIA, NIH
Elizabeth Sypek, NINDS, NIH
Debra Szczepanski, The University of Texas Southwestern Medical Center
Ginger Tansey, National Eye Institute, NIH
Nick Tataryn, Vanderbilt University Medical Center
Ei Terasawa, University of Wisconsin–Madison
Nicklaus Thompson, University of Washington
Biao Tian, ORIP, DPCPSI, OD, NIH
Gregory Timmel, CNPRC
Elizabeth Tobey, National Agricultural Library
Ferenc Toth, University of Minnesota
Drew Townsend, National Institute on Drug Abuse, NIH
Elise Trowel, Tufts University
Emily Trunnell, People for the Ethical Treatment of Animals
Jacquelyn Tubbs, OLAW, OD, OER, NIH
Christopher Tuggle, Iowa State University
Rebecca Turcios, The University of Chicago
Patricia Turner, Charles River Laboratories
George Umanah, NINDS, NIH
John Vanchiere, Louisiana State University Health Shreveport
Tyara Vazquez, The University of Toledo
Jean Verheyden, National Institute on Deafness and Other Communication Disorders, NIH
Jayalakshmi Viswanathan, NIA, NIH
Colby Vorland, Indiana University
Anil Wali, NCI, NIH
Jeanne Wallace, Vanderbilt University Medical Center
Michael Wallis, Johns Hopkins University
Guanghu (Jeff) Wang, ORIP, DPCPSI, OD, NIH
Erica Watson, GlaxoSmithKline
Rachel Weinberg, NINDS, NIH
Sylvia West, Emory University
Sarah Wheelan, National Human Genome Research Institute, NIH
Kristin Whitworth, National Swine Resource and Research Center
Sarah Woller, NINDS, NIH
Dan Xi, NCI, NIH
Jianhua Xu, National Institute of General Medical Sciences, NIH
Erin Yu, Vanderbilt University Medical Center
Amanda Ziegler, North Carolina State University
Sige Zou, ORIP, DPCPSI, OD, NIH