INSTITUTIONAL BIOSAFETY COMMITTEE MEETING
August 15, 2018
3 PM, Plant Biotechnology Bldg., Room 410

MEMBERS PRESENT: Chair, David White; Vice Chair, Elizabeth Fozo; Marc Caldwell, Tamara Chavez-Lindell, Lori Cole, Paul Dalhaimer, Brittany Isabell, Reza Hajimorad, George Dizikes, Jun Lin, Reggie Millwood, Deidra Mountain, Jae Park, Ling Zhao

Ex-Officio – Linda Hamilton, Scott Moser, Brian Ranger, Jessica Woofter

MEMBERS ABSENT: Doris D'Souza, Melissa Kennedy

OTHERS PRESENT: Bryce Burton

Opening:

The Chair called the meeting to order at 3:03 PM. The minutes of July 10, 2018, were reviewed and approved pending edits to Dr. Kerro Dego's requirements to add a clarification that IACUC protocols are active.

IBC Applications:

#IBC-06-292-2 (Guoxun Chen) Recombinant DNA & Human Derived Materials, III-D-3, 3-year rewrite
Dr. Chen's research investigates the role(s) of various metabolic proteins in mediating the roles of vitamin A in energy metabolism and the development of metabolic diseases, such as obesity and diabetes. To achieve efficient transfection of various mammalian cell lines (mouse, rat, and human), Dr. Chen is proposing the use of recombinant, replication-incompetent adenoviral vectors to deliver/express genes of interest. Briefly, the adenoviral system is based on a binary plasmid system in combination with HEK 293 packaging cells (which express the E1 gene region necessary for viral replication). The proposed containment level was set at BSL-2. The committee voted to approve the registration pending removal of references to the Jessie Harris Building; updating the location of the CCI 250 biosafety cabinet; correction of typographical errors and incorporation of annual update information in the technical summary; updating the spill response; clarifying PPE cleaning procedures; and completion of the Biohazardous Sharp Waste section. There was one abstention.

 # IBC-12-380-1 (Neal Stewart) Recombinant DNA, III-E-2-a, 3-year rewrite
Dr. Stewart's research involves functional analysis of several genes belonging to the ATP-binding (ABC) superfamily, including those related to herbicide resistance or bacterial pathogen resistance. Briefly, full-length cDNA of candidate genes are being cloned into plant expression vectors, which

will be used to transform susceptible biotypes of horseweed, tobacco, and/or Arabidopsis plants. Plants will be transformed using Agrobacterium-mediated transformations to deliver individual vector constructs. Similarly, RNA interference (RNAi) and genome editing (CRISPR/Cas9) constructs will be designed to knock-out or inhibit ABC candidate gene expression in resistant biotypes. Plants will be grown in growth chambers and greenhouse (with pollen screens, pollen bags, or flower removal). Any field releases will be under the authority of a USDA APHIS BRS permit/notification. The proposed containment level is BL-1/BL-1-P. The committee voted to approve the registration as written. There was one abstention.

#IBC-12-385-1 (Elias Fernandez) Recombinant DNA, III-E, 3-year rewrite

Dr. Fernandez's research covers the characterization of the biophysical properties of nuclear receptors for the development of more effective therapies. Entire genes or subdomains of the genes will be sub-cloned into pET vectors and overexpressed in *E. coli* BL21/DE3 cells. Protein will be isolated/purified by conventional means and used for biophysical assays (e.g. X-ray crystallography). The research will be conducted at BSL-1. The committee voted to approve the registration pending clarification of volumes of *E. coli* culture, decontamination procedures used for culture vessels; clarification of waste treatment/disposal; an updated spill response; updated facility information including room numbers and autoclave information; and confirmation that the 10 liters used is not stored in a single vessel.

#IBC-15-430-2 (Oudessa Kerro Dego) Recombinant DNA & Infectious Agents, III-D-1-a, 3-year rewrite (*resubmission*)

Dr. Kerro Dego's registration covers the testing and identification of known virulence factors of *Escherichia coli*, *Staphylococcus aureus*, *Mycoplasma bovis*, and *Streptococcus uberis* isolates from bovine mastitis using standard and special culture methods and PCR. The research objectives also include co-incubating *E. coli, S. aureus,* or *S. uberis* with mammary epithelial cell line (MAC-T cells) and evaluation of bacterial gene expression patterns by real-time PCR or RNA-Sequencing. Both methods will help evaluate the presence of zoonotic and foodborne pathogens in the dairy farm environment. Several other Risk Group 2 pathogens may be used as reference strains for these assays. Additionally, gene knockouts will be created in *S. aureus, E. coli, M. bovis,* and *S. uberis* via overlapping extension PCR/homologous recombination using temperature-sensitive shuttle vectors. The committee voted to approve the registration pending clarification that challenge studies will be contingent upon IACUC approval; the addition of the lab space located at ETREC Little River Dairy; clarification of the *S. aureus* strain to be used for mastitis challenges; and an approval provision indicating that *M. bovis* challenges will be at the discretion and approval of the IACUC and ETREC Little Dairy management. There was one abstention.

#IBC-15-432-2 (Ahmed Bettaieb) Recombinant DNA, Infectious Agents & Human Derived Materials, III-D-3-a, 3-year rewrite

Dr. Ahmed Bettaieb is investigating the regulatory roles of protein tyrosine phosphatases (e.g. Fas, nephrin, prolactin, etc.) in glucose metabolism homeostasis, energy expenditure, and pathological disease signaling. His research will include the use of human and nonhuman primate cells and tissues (adipose tissue) as well as 2nd and 3rd generation replication-deficient lentiviral vector systems. Briefly, pseudotyped lentiviral vectors will be used in human, mouse, rat, and/or nonhuman primate cell lines to 1) knock down expression of proteins of interest with commercially available shRNA constructs, and 2) overexpress a protein of interest from commercially available ORF clones. Component lentiviral plasmids will be maintained and propagated in *E. coli* K-12

strains. HEK293 cells are used for generation of recombinant lentiviral particles, which are subsequently transduced into target cell lines as described above (standard molecular protocols). All open vessel procedures involving mammalian cell lines and lentiviral vectors will be performed in a certified Class II biosafety cabinet using BSL-2 containment and precautions. The outlined safety precautions, waste segregation/treatment strategies, and emergency response procedures for lentiviral vectors and mammalian cells/tissues were deemed acceptable by the committee. Additionally, Dr. Bettaieb will be using transgenic knockout mice lacking protein tyrosine phosphatases and their interacting partners in adipose, liver, kidney, lungs, brain, muscle or pancreatic tissue for his studies (cre-lox system). All mice will be purchased or transferred and can be contained at BSL-1; therefore, this component is exempt from IBC approval per section III-F-8/Appendices C-VII and C-VIII. The committee voted to approve the registration pending an update to the spill response. There was one abstention.

#IBC-15-433-1 (Subimal Datta) Recombinant DNA, III-D-4-a, 3-year rewrite

Dr. Datta studies the cellular and molecular mechanisms involved in homeostatically regulated rapid eye movement (REM) sleep in a rat model. A combination of canine-associated virus (CAV) and adeno-associated virus (AAV) will be used to over-express designer muscarinic-like receptors in specific neural circuits to test whether pharmacological activation of these receptors initiates REM sleep. AAV will be delivered intracranially to the pedunculopontine tegmentum, and CAV will be delivered in the same fashion to the subcoeruleus nucleus. Viral vectors carrying the transgene of interest are purchased from the UNC-Chapel Hill Vector Core Facility (no onsite production). Containment was set at BSL-1/ABSL-1. The committee voted to approve the registration pending clarification of disinfection procedures for safety glasses; the addition of a statement that autoclave validations are performed regularly by UHS; and an approval provision that animal work would be contingent upon IACUC approval.

Old Business:

Administrative Report

i. Contingencies

Following up on July 10, 2018, IBC meeting, Dr. Shigetoshi Eda's registration (#06-276-2) was edited to include all ATCC numbers and organisms were listed separately; corrected the source and origin for *Mycobacterium* sp. (incl. *avium, kansasii, phlei, fortuitum*); provided clarifying statement about collection/source of swabs and blood; and included updates to the occupational health section, including the hepatitis B vaccine offer for those working with human blood and a routine TB skin test for those working with *M. bovis* (BCG). Dr. Tim Sparer's registration (#06-277-2) was edited and references to tumor modeling in animals and IACUC Protocol #1668 were removed. Dr. Barry Rouse's registration (#06-280-2) is still pending clarification of procedures included in other cited IACUC protocols; updates to the spill response plan; updates to the biosafety cabinet certification date; and updates to the autoclave validation dates. Dr. Jon Wall's registration (#09-342-2) was edited to include updates to the autoclave validation dates and spill response. Dr. Neal Stewart's registration (#12-382-1) was edited to include a clarifying statement that low-risk waste containers will be in a color other than red and will not

display the biohazard symbol. Dr. Josh Bembenek's registration (#12-392-1) was edited to include a clarification of assays mentioned in the technical summary and an update to autoclave validation dates.

ii. *Administrative Approvals*
Dr. Elizabeth Lennon's amendment (#16-436-2) was administratively approved by the Biosafety Officer to include the addition of human IgE from myeloma patients.

iii. *Administrative Terminations*
None.

iv. *Administrative Exemptions:*
None.

v. *Accidents, Injuries/Exposures:*
None.

vi. *Laboratory Report (Hamilton)*
None.

vii. *iMedRIS Update, Manual Reviews, & System Orientation (Woofter)*
None.

Charter Revision Update
Brian notified the committee he will have SOPs ready for review on the next agenda.

WLS and JHB Lab Moves to the New Mossman Bldg. Update
Brian updated the committee on the Mossman laboratory moves, including concerns related to lab design and directional airflow.

New Business:

Bob Emery Peer Review/Recommendation
Dr. Bob Emery from the University of Texas Health Science Center at Houston reviewed the University of Tennessee's compliance offices. Brian summarized his preliminary findings to the committee, which included recommendations for staffing and organizational management.

BIORAFT Discussion
Brian notified the committee that the Biosafety Office reviewed a demo of BIORAFT. Jessica Woofter sent the committee a link to view the demo.

The meeting was adjourned at 4:35 PM. The next meeting is tentatively scheduled for September 27, 2018, in the Plant Biotechnology Building, Room 410 at 1 PM.