

INSTITUTIONAL BIOSAFETY COMMITTEE MEETING

July 10, 2018

10 AM, Plant Biotechnology Bldg., Room 410

MEMBERS PRESENT: Chair, David White; Vice Chair, Elizabeth Fozo; Lori Cole, Reza Hajimorad, George Dizikes, Melissa Kennedy, Reggie Millwood, Deidra Mountain, Jae Park

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

MEMBERS ABSENT: Marc Caldwell, Tamara Chavez-Lindell, Doris D’Souza, Paul Dalhaimer, Brittany Isabell, Jun Lin, Ling Zhao

OTHERS PRESENT: Dr. Sarah Pruet

Opening:

The IBC Chair, Dr. David White, called the meeting to order at 10:08 AM. The minutes of May 16, 2018 were reviewed and approved as written.

IBC Applications:

#IBC-06-276-2 (Shigetoshi Eda) Infectious Agents and Human-derived Materials, 3-year rewrite

Dr. Shigetoshi Eda’s registration covers two novel detection technologies for the diagnosis of Johne’s disease (JD) caused by *Mycobacterium avium* subsp. *paratuberculosis* (MAP). Dr. Eda’s research also hopes to develop rapid diagnostics for other diseases such as Crohn’s disease, bovine tuberculosis, mastitis, malaria, and Lyme disease. This registration covers approximately 20 Risk Group 2 infectious agents for specificity and sensitivity controls for the detection assays in development. Containment was set at BSL-2. The committee voted to approve the registration as written.

IBC-06-277-2 (Tim Sparer) Recombinant DNA & Infectious Agents, III-D-2-a; 3-a; 4-b, 3-year rewrite

Dr. Tim Sparer’s registration covers various studies involving human and murine cytomegaloviruses (CMVs) and host immunity, particularly the role of viral chemokines in modulating the host immune response to CMV infection. Baculoviruses will be used to generate recombinant viral chemokines to be used in a variety of *in vitro* and *in vivo* assays to determine their effects on neutrophil function. Human CMVs will be propagated and titered for use in viral engulfment experiments. Murine CMVs (wild-type or recombinant) will be used for *in vivo* experiments in mice to determine how the inserted or deleted genes affect viral spread throughout the mouse. Recombinant DNA procedures include the cloning of viral chemokines, host genes, and chemokine receptors into *E. coli* for sequencing, baculoviruses for

overexpression/purification, and eukaryotic cell lines for *in vitro* expression studies. For the latter, third-generation, replication-incompetent lentiviral vector delivery systems will be used. Containment was set at BSL-2/ABSL-2. The committee voted to approve the registration pending the removal of IACUC Protocol# 1668 and references to tumor modeling in animals.

#IBC-06-280-2 (Barry Rouse) Recombinant DNA & Infectious Agents, III-D-2-a; 4-b, 3-year rewrite

Dr. Rouse's registration uses the mouse model to study herpes simplex virus (HSV) induced immunopathology via ocular HSV infection. The goals of this research are 1) to design an approach that can inhibit virus induced ocular immunopathology, and 2) design an efficacious vaccine against HSV. Briefly, recombinant constructs encoding for viral proteins (e.g. glycoprotein B) or murine cytokines (e.g. IL-15) will be injected into mice intranasally or intramuscularly to enhance the immune response to virus infection and reduce the viral loads. Animals will then be challenged with HSV to determine DNA vaccine efficacy. Containment was set at BSL-2/ABSL-2. The committee voted to approve the registration pending removal of IACUC Protocol# 2264; 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 autoclave validation dates.

#IBC-09-342-2 (Jonathan Wall) Recombinant DNA & Human Derived Materials, III-D-4-a & III-E, 3-year rewrite

Dr. Wall's registration includes the use of a commercial phage library system to generate random peptides or antibody fragments (scFv), specifically those that can bind ligands of interest, such as amyloid or heparan sulfate, a sugar molecule that is found in amyloid deposits such as those that form in the brains of patients with Alzheimer's disease. Secondly, the study will involve isolation of RNA from patient tissues or cells, generation of cDNA by reverse transcription (primed for genes encoding amyloidogenic proteins, including immunoglobulin light chains, ODAM, apolipoprotein, and galactin-7), and cloning into a bacterial expression system (pET27B) for *E. coli*-based protein expression and isolation. Ultimately, these proteins are used to study cancer and amyloidosis and to design new effective methods of diagnosis and treatment. Thirdly, the study will involve expressing proteins of interest isolated from patient samples, or synthetic genes that are commercially produced, in human cells (e.g. HEK, MCF7, HeLa, and A375). Recombinant cell lines will then be xenografted into mice to generate tumors and study the effects of expressed proteins. These studies will lead to a better understanding of the disease (cancer and amyloidosis) and provide test beds for developing new therapies. Lastly, the study involves the extraction of human amyloid or light-chain proteins from donated organs obtained at autopsy, or urine, respectively. This material will be used in laboratory studies and injected into mice to generate mouse models of the disease. The committee approved the registration pending updates to autoclave validations and spill response. Work with scFv and recombinant *E. coli* was approved at BSL-1; procedures involving human derived materials, human cells lines, and xenografted mice were approved at BSL-2.

#IBC-12-382-1 (Neal Stewart) Recombinant DNA, III-E-2-a, 3-year rewrite

Dr. Stewart's research involves the development of new biotechnology for targeting genome-modification in plants. Briefly, the research involves using genetically modified transcription activation-like effectors (TALEs) to target the promoter region of select plant genes, leading to activation of gene expression. Additionally, the TALE-specific activation domain will be replaced with an appropriate nuclease (TALENs) that could then be used for genome editing. Containment was set at BSL-1/BL-1-P. The committee approved the registration as written pending a minor edit to the sharps container section. There was one abstention.

#IBC-12-384-1 (Rebecca Trout Fryxell) Infectious Agents, 3-year rewrite

Dr. Rebecca Trout Fryxell's research involves the survey of arthropods for various bacterial (Rickettsial), viral (La Crosse), or protozoan (Plasmodium/Haemeaproteus) pathogens by collecting vectors (mosquitoes, ticks, black flies) from the field, identifying them and storing them in 80% ethanol. Molecular techniques (DNA extraction, PCR) will be used to identify the different pathogens. Additional procedures such as dissections of insect tissue (e.g. dissecting mosquito salivary glands from the head & thorax) may be conducted for species identification. Also, Dr. Trout Fryxell will be establishing breeding colonies of endemic flies and mosquitoes. Live arthropods will be maintained/handled in accordance with ACL-1 guidelines. The committee voted to approve the registration as written.

#IBC-12-392-1 (Josh Bembenek) Recombinant DNA, III-E, 3-year rewrite

Dr. Bembenek's research involves the study of cell cycle genes (e.g. *sep-1*) and regulatory pathways that control genetic material and physical separation during cytokinesis. Dr. Bembenek will be using a well-established model, *Caenorhabditis elegans*, for his studies. Recombinant procedures will include RNAi introduction via *E. coli* "feeding" and biolistic transformation with fluorescent bioreporters (GFP fusions) to measure early gene expression. The committee approved the registration as written pending clarification of assays mentioned in the Technical Summary and an update to autoclave validation dates. Containment was set at BSL-1.

#IBC-15-430-2 (Oudessa Kerro DeGo) Recombinant DNA & Infectious Agents, III-D-1-a, new registration

Dr. Kerro DeGo's registration covers the testing and identification of known virulence factors of *Escherichia coli*, *Staphylococcus aureus*, 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 the proposed assays. The committee voted to table the registration pending the clarification of gene deletion procedures in the nontechnical and technical summaries; clarification of enterotoxin-encoding strains of *S. aureus*; addition of ATCC reference numbers; corrections to the spill response; update of the autoclave validation dates; clarification as whether this is future work or a current IACUC protocol is being used; and a statement in the health surveillance section about risks of exposure from *L. monocytogenes* to pregnant women.

Old Business:

Administrative Report

i. Contingencies

Following up on May 16, 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 updated to the occupational health section, including the Hepatitis B offer for those working with human blood and a routine TB testing for those working with *M. bovis* (BCG). Dr. Maria Prado's registration (#12-381-2) was edited to include a clarification about REV in the technical summary and that PPE would be disinfected and washed in the departmental washing machine. Dr. Richard Gerhold's registration (#13-397-2) was edited to include a clarifying statement about risks to pregnant mothers and the correction of typographical errors. Dr. Graham Hickling's registration (#18-520-2) was edited to include the use of respirators for field collection of wild rodents.

ii. Administrative Approvals

Dr. Shawn Campagna's amendment (#08-323-2) was administratively approved by the Biosafety Officer to include human-derived materials (blood, plasma/serum, tissue) from the Forensic Anthropology Center. Dr. Tarek Hewezi's amendment (#13-398-1) was administratively approved by the IBC Vice Chair to include vectors (Addgene; MTA approved 5/21/18) for CRISPR/Cas9 editing. Vectors will be used to splice in various low-risk gene cassettes for studying gene function. Dr. Stacy Stephenson's amendment (#14-423-2) was administratively approved by the IBC Chair to include the incorporation of fluorescent protein coding regions (i.e. GFP, RFP) into lentiviral vectors and transduction of mesenchymal stem cells for *in vitro* & *in vivo* imaging. Dr. Jeremiah Johnson (#16-441-2) and Dr. Heidi Goodrich-Blair (#16-442-2) provided location updates (labs moved to Mossman Bldg.), which were administratively approved by the Biosafety Office.

iii. Administrative Terminations

None.

iv. Administrative Exemptions:

Dr. Bode Adebowale Olukolu's registration (#06-286-1) was administratively exempted by the Biosafety Office for the use of *E. coli* DH10B for sequencing DNA/genes from sweet potato (III-F-8/Appendix C-II).

v. Accidents, Injuries/Exposures:

Reports of *Cryptosporidium parvum* cases in CVM students. The Biosafety Office is/will be working with LACS faculty to review training materials, safety procedures and PPE provided to students assisting with field services.

vi. *Laboratory Report (Hamilton)*
None.

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

Charter Revision Update

Brian notified the committee that the SOP for Registration Reporting, Review and Recordkeeping has been drafted and requires some revision. Brian will share the draft of this and other new/revised SOPs prior to the August meeting.

WLS and JHB Lab Moves to the New Mossman Bldg. Update

Brian updated the committee on the Mossman laboratory moves, and informed the committee about issues with the air handling system and other laboratory design limitations in the Mossman Building.

IBC Form Edit - Redesign of Host/Vector Table

Brian notified the committee that the Biosafety Office will be demoing the BIORAFT IBC Module and encouraged the committee to review the module as well.

Peer Review with Clemson University

Brian shared the results from the peer review with Clemson University performed in May of this year. No major concerns were noted, but there were some recommendations to review programmatic organization and resourcing.

New Business:

Proposed New Approach to Refresher Training

Linda notified the committee that the Biosafety Office would like to do in-person refresher training based on lab-specific needs. The training will accompany laboratory audits if possible. Linda will prototype this new platform with select PIs/laboratories and get feedback before full implementation (~October).

CVM Immunology/Infectious Disease Candidates

Brian notified the committee that CVM is looking to hire two candidates for immunology and/or infectious disease positions. Both top candidates are virologists and the University of Tennessee who may require enhanced facilities and biosafety oversight. The IBC will be updated as necessary.

External Audit - Dr. Bob Emery

Brian notified the committee that Dr. Bob Emery, Professor of Occupational Health & Vice President for Safety, Health, Environment & Risk Management with the University of Texas Health Science Center at Houston, will be at the University of Tennessee at Knoxville the week of August 13th to perform an external audit and gap analysis of current lab safety and compliance programs.

Review Assignment & Due Dates

Jessica reminded the committee about due dates for reviews and comments. The committee suggested sending calendar reminders along with the PDF copy of the registration and review questions to reviewers.

August Meeting

The next meeting has been tentatively scheduled for August 15, 2018, at 3 pm in the Plant Biotechnology Building, Room 410. Jessica will send out a calendar reminder to committee members.

The meeting was adjourned at 1 PM.