### IBC MEETING COMMENTS

July 2, 2025 – Zoom Conference Call

Attendance – IBC Committee					
Present	Name	Expertise	Role		
	Paul Gulig	Microbiology	Member		
Х	Luis Martinez	Microbiology	Member		
	Jason Clements		Member		
	Norman Beatty	Infectious Diseases	Member		
	Steeve Boulant	Virology	Member		
х	Sanford L. Boye	Virology	Member		
_	Amber Duren		Community Member		
Х	Mariola Ferraro	Microbiology	Member		
Х	Gary Heil	Plants	Member		
Х	Michael McIntosh		Member – joined		
			1:07pm		
	John D. McVay	Virology	Community Member		
	Mark Moehle	Plants	Member		
X	Kamal A. Mohammed	Microbiology	Member		
X	Christopher Overend	Microbiology	Member		
Х			Member		
		Animals	Member		
	Elias J. Sayour	Clinical Trials	Member		
х	Clay Smith	Virology	Chair		
	Daniel R. Swale	Insects	Member		
_	Amy Vittor	Infectious Diseases	Member		

	Attendance – Staff and Guests				
Present	Name	Affiliation/Position			
X	Asha Rani	EH&S Staff			
х	Pratibha Srivastava	EH&S Staff			
X	Savannah Hardiman	EH&S Staff			
X	Jason Clements	EH&S Staff			
X	Jennifer Jackson	EH&S Staff			
х	Kendra Rooks	EH&S Staff			
X	Craig Moneypenny	EH&S Staff			
х	Michael Mahoney	EH&S Staff			
х	Laura Castillo	EH&S Staff			
х	Kindra Kelly-Quagliana	EH&S Staff			
Х	Artiom Chacon	EH&S Staff - joined1:26 pm			
	Kevin Ross	EH&S Staff – joined 1:36 pm			
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# Agenda:

	Full Committee Projects	PI
1	The Role of CD70 in Antitumor Response	– Renewal BIO5681
2	Development of vaccines against animal and food safety pathogens	- Renewal BIO5627
3	Novel metal homeostasis responses	Keith Choe
4	AAV-mediated retinal gene therapy	Shannon Boye – Renewal BIO5626
5	Molecular pathogenesis of Burkholderia pseudomallei and Burkholderia mallei	
6	Pulmonary endothelial Cell culture	Mohamed Ahmed
	Amendments	Pl
1	The Role of Mitochondria in Malignant Glioma Biology	Serendipity Rinonos
2	Mechanisms of proteostasis-mediated neurodegeneration	
3	In vitro comparison of human and axolotl tau aggregation	
4	Use of AAV constructs in the study of Neuroimmune acis contribution to the pathophysiology of pulmonary hypertenstion	

**Minutes** 

Meeting was called to order at: 1:01pm

**Project Review** 

Principal Investigator:

**Project Title:** The Role of CD70 in Antitumor Response

Renewal BIO5681

<u>Vector/Agent(s) to be used:</u> Herpes Simplex Virus (HSV) type 1, Gamma retrovirus, Lentivirus, Vesicular Stomatitis Virus (VSV),

Name and Function of Transcribed Nucleic Acids: HSVsr39tk - PET imaging reporter. MouseCD70 CAR - help T cell to target CD70 molecule. mouse CXCR2 - increase T cell chemotaxis. Cre: lox-specific DNA recombinase. PDGFb - oncogene. CD70 - target of CAR, overexpress target for CART cells. Firefly Luciferase - *In vivo* detectable marker. Human CAR - helper T cell to target CD70 molecule. LAIR2 - inhibit binding of immunosuppressive molecule LAIR1 with its ligand. LCMV-GP - minimize neurotropism and neurotoxicity of VSV. GFP - fluorescent marker. mlgG2a Fc region - dimerization scaffold, improve solubility and stability. IL2 - activate T cells. DDR1 - specifically bind collagen in tumor extracellular matrix. DS - specifically bind collagen in tumor extracellular matrix.

<u>Host(s) to be used:</u> Rodent model, rodent brain, Cultured rodent cell lines (KR158, KR158-CD70-luc, KR158-CD70, GL261, GL261-CD70-luc, GL261-CD70), Cultured human cell lines (BT5, SSC163), rodent and human T lymphocytes, *E. coli* 

**NIH Guidelines**: III-D-1-a for experiments involving RG2 agents as a host vector system, III-D-1-b use of rDNA in a RG3 vector, III-D-3-a for the generation of lentiviral vectors, III-D-4-b use of rDNA in rodents.

Biosafety Level and Any Additional Requirements: BSL-2 containment and practices are appropriate for *in vitro* work involving Human cell lines. BSL-2+ is recommended for the work with transduced cell lines and *in vitro* experiments with LVV and Retrovirus. The "+" practices require mucous membrane protection and substituting plastic labware for glassware where possible. Required personal protective equipment (PPE) includes laboratory coat, disposable gloves, and mucous membrane protection. A proper protocol for inactivation/decontamination of biohazard waste is emphasized. Animal procedures involving human cells lines are approved at ABSL-2+ (requires the use of safe sharps and wiping of the injection sight), and ABSL-2 subsequent housing for lentivirus vectors (LVV) and Retrovirus experiments. ABSL-1+Hu is designated for housing rodents inoculated with human cells. Ensure the label "Use of Human Cells in Animals" is displayed on both the animal caging and at the entrance to the room.

**<u>Training:</u>** All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

Principal Investigator:

Project Title: Development of vaccines against animal and food safety pathogens

Renewal BIO5627

<u>Vector/Agent(s) to be used:</u> Salmonella spp., Porcine Circovirus 2 (PCV2),

<u>Name and Function of Transcribed Nucleic Acids:</u> O-antigen coding region - Responsible to produce *S. kentucky* O8,20-antigens, Responsible to produce *S. infantis* O6,7-antigens & prokaryatic experession from a plasmid. Tcf fimbriae coding region – Fimbriae. BlaSS - Type 2 Secretion System antigen delivery. Asd and MurA - complementation for the Salmonella backbone ability to replicate. SopE N-80 - enables Type 3 Secretion System antigen delivery.

Host(s) to be used: Rodents, Salmonella spp, E. coli DH5a

<u>NIH Guidelines</u>: III-D-1-a for use of a Risk Group 2 agent as a host-vector system (i.e., recombinant/synthetic nucleic acids in Salmonella enterica-based RASVs) and III-D-4-a for use of recombinant/synthetic nucleic acids containing no more than two-thirds of any eukaryotic viral genome in non-human vertebrates (i.e., RASVs in animals). Note: III-E-1 was previously assigned but use of recombinant/synthetic nucleic acids in tissue culture is not in scope of the project. III-D-2-a was previously assigned as well, but plasmids are cloned in a K-12 strain of *E. coli*, which falls under III-F-8 Appendix C-II, and use of the same plasmids in RASVs is covered under section III-D-1-a.

Biosafety Level and Any Additional Requirements: BSL-2/ABSL-2 for work with pathogenic challenge strains, BSL-1/ABSL-1-i for work with RASVs, with ABSL-1-i housing requiring the inactivation of all animal bedding and waste throughout the vaccination period (non-biomedical biological waste), prior to pathogenic challenge after which ABSL-2 procedures require bedding/waste to be treated as biomedical waste. Note that the RASVs have previously received approval for BSL-1/ABSL-1 containment by the NIH OSP under section III-D-1-a. Please note that any future acquisition of USDA APHIS VS 16-6 permits for transport of challenge agents will require attachment of the permit to this project in an amendment.

**Training:** All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

**Principal Investigator**: Keith Choe

Project Title: Novel metal homeostasis responses

BIO number not yet assigned.

Vector/Agent(s) to be used: Plasmids, CRISPR/Cas-9.

Name and Function of Transcribed Nucleic Acids: GFP - Fluorescent reporter. Scarlet - Fluorescent reporter. CRISPR/Cas-9 – to delete W03G1.5.

Host(s) to be used: Caenorhabditis elegans, E. coli DH5a.

**NIH Guidelines**: III-E for the work involving CRISPR/Cas-9. III-F-8 Appendix C-II for cloning work utilizing K-12 strain *E. coli* (HB101 and DH5alpha).

Biosafety Level and Any Additional Requirements: BSL-1 biosafety containment practices

<u>Training:</u> All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

Principal Investigator:

<u>Project Title:</u> Molecular pathogenesis of *Burkholderia pseudomallei* and *Burkholderia mallei* 

BIO number not yet assigned.

<u>Vector/Agent(s) to be used:</u> Burkholderia mallei, Burkholderia pseudomallei, plasmids

Name and Function of Transcribed Nucleic Acids: purM - Purine metabolism

Host(s) to be used: Burkholderia mallei, Burkholderia pseudomallei

**NIH Guidelines**: III-D-1-b applies to the use of *B. pseudomallei* as a host-vector system (both *in vitro* and *in vivo*). III-D-2-a applies to the cloning of plasmid DNA containing *Burkholderia* nucleic acids into *E. coli*.

Biosafety Level and Any Additional Requirements: All manipulation of *B. mallei* and *B. pseudomallei*, including all derivatives, must be conducted inside of the BSL-3/ABSL-3 suites assigned to the PI, following all facility-specific plans and lab-specific SOPs applicable to those locations. Derivatives of *B. mallei/pseudomallei*, including those from infected animals, must be subjected to a validated inactivation or select agent removal method with a signed Certificate of Inactivation prior to removal of such materials from the BSL-3/ABSL-3 lab to lower containment for further analysis.

Cloning of plasmids containing *B. pseudomallei* DNA into *E. coli* must be conducted using BSL-2 containment and practices. All other procedures in the PI's BSL-2 lab may be conducted using BSL-1 practices, with the exception of use of BSL-2 practices with lipopolysaccharide samples *in vitro*.

Any new ΔpurM B. pseudomallei mutants are subject to all select agent regulations until excluded by the Federal Select Agent Program through the exclusion submission process. B. mallei and B. pseudomallei, including nucleic acids, are subject to export control regulations.

All new scope additional to that covered in the Pl's paper projects must be addressed in new or updated lab-specific SOPs and approved by the RO/ARO prior to beginning work.

**Training:** All training completed

Approval: A conditional approval for clarification on a vector and safety requirements (Yes - 10, No-0, Abstain-0)

Principal Investigator: Mohamed Ahmed

**Project Title:** Pulmonary endothelial cell culture

BIO number not yet assigned.

Vector/Agent(s) to be used: Moloney Murine Leukemia Virus

Name and Function of Transcribed Nucleic Acids: BMI1 - Immortalization of cells.

Host(s) to be used: Human Endothelial Cells (HPAEC)

**NIH Guidelines**: III-E-1 for immortalization of the cell line by insertion of the BMI-1gene into the genome by Moloney murine leukemia virus.

Biosafety Level and Any Additional Requirements: BSL-2 biosafety containment and practices for the use of the immortalized human cell line.

**Training:** All training completed

**Approval**: All approved as recommended (Yes- 10, No-0, Abstain-0)

**Principal Investigator**: Shannon Boye

**Project Title:** AAV-mediated retinal gene therapy

Renewal BIO5626

Approval: Mistakenly worked up and reviewed as an amendment rather than a 5-year renewal. Tabled until further review. No vote taken.

#### **Amendments**

**Principal Investigator**: Serendipity Rinonos

<u>Title:</u> The Role of Mitochondria in Malignant Glioma Biology

#### **BIO6921**

**Summary**: Adding three primary human glioblastoma patient-derived cell lines (L0, L1, L2) from the Florida Central Brain Tumor Registry, a human astrocyte cell line immortalized with SV40T (CSC-C12025Z, Creative Bioarray), in order to perform co-culture experiments with astrocytes and GBM cells to study their interactions and the impacts on mitochondria. The addition of these cell lines this will not alter the previous determined biosafety containment practices, nor the NIH guidelines and they remain as stated:

BSL-1 for plasmid maintenance using K-12 lineage nonpathogenic *E. coli*. BSL-2+ for the work with transduced cell lines and *in vitro* experiments with LVV.

This project falls under the following NIH guidelines: The production of the recombinant lentivirus falls under III-D-3-b.

Use of Lentiviruses in cells under III-E-1. The molecular cloning using K-12 lineage E. coli falls under section III-F-8 Appendix C-II.

**Training:** All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

## Principal Investigator:

<u>Title:</u> Mechanisms of proteostasis-mediated neurodegeneration

#### **BIO5670**

**Summary**: Adding new plasmids. As a result of these new plasmids this will not alter the previously determined biosafety containment practices nor the NIH guidelines and they remain as stated:

BSL-1 for work with AAV vectors; BSL-2 containment for experiments involving HEK-293 cells; ABSL-1 for intracerebroventricular injections of AAV into rodents.

The work is covered under NIH Guidelines sections: III-E-1 (formation of rDNA molecules containing no more than 2/3 of the genome of any eukaryotic virus), III-D-4-a (use of RG1 vector in <u>rodents</u>) for the AAV work.

**Training:** All training completed

Approval: All approved as recommended (Yes- 10, No-0, Abstain-0)

### Principal Investigator:

Title: In vitro comparison of human and axolotl tau aggregation

**BIO7272** 

**Summary**: Adding a new plasmid to amplify their gene of interest and then linearize it and send it to the University of Pennsylvania along with human HEK293T cells for *in vitro* expression experiments.

This work can safely be performed by employing BSL-2 biosafety containment and practices for the *in vitro* work with HEL293T cells and BSL-1 for the work involving BL21 (DE3) cells will be included in the study for protein expression using plasmids.

This amendment will include the following NIH Guidelines: III-E for the use of non-K12 *E. coli* strain [T7 Express Competent to express and BL21 (DE3)], III-F-8, Appendix C-II for *E. coli* K-12 host vector system (DH10B).

As a general reminder, the lab must maintain a current and accurate LATCH hazard assessment. All personnel must stay up to date with the training courses required in the hazard assessment.

**Training:** All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

### Principal Investigator:

<u>Title:</u> Use of AAV constructs in the study of Neuroimmune acis contribution to the pathophysiology of pulmonary hypertension

#### **BIO7137**

<u>Summary</u>: Personnel are being added to the registration (being removed. Dr. is adding over 50 new AAV's, these primarily are targeting reporters for neuronal imaging. These changes will not alter the previously determined biosafety containment practices nor NIH guidelines and they remain as stated.

BSL-1 for work with AAVs, ABSL-1+ for AAV injection into mice and rats with subsequent ABSL-1 housing. NIH Guidelines III-D-4-a for the administration of AAV to rodents

**Training:** All training completed

Approval: All approved as recommended (Yes-10, No-0, Abstain-0)

The meeting adjourned at 2:05 pm