IBC Meeting Minutes
August 21, 2025 (Thursday) at 11:00 A.M.
via Zoom Conference Bridge

IBC members present:

Tom Greenough (Chair)	X	Shaoguang Li		Carol Schrader	X	Edward Jaskolski (alt)**	X
Lisa Cavacini	X	Philip Tai	X	Mohan Somasundaran		Timothy Kowalik (alt)	
Colleen Driskill	X	Robert Klugman	X	Regino Mercado-Lubo		Richard Ellison III (alt)	
Kris Giaya	X	Amelia Houghton	X	Sharone Green (alt)		Casey Moran (alt)	
Hardy Kornfeld		Eric Rouse*	X	Jennifer Wang (alt)	X		

Non-members present: Patrice Rando (IACUC/IBC Office), Greg Cohan (Resident- Occupational Medicine)

*ER- joined 11:33am
**EJ- Left at 1:40pm

I. Introductory Remarks

1) The Chair brought to the attention of the Committee the action items completed since the previous meeting and those submissions still under review by IBC.
2) The Chair brought to the attention of the Committee the meeting minutes from the previous IBC meeting. **Meeting Decision: Vote to approve July 17, 2025 Meeting Minutes**

II. Report on incidents/accidents from Employee Health Services (EHS)

Past incidents:

052125: BSL-3 PAPR malfunction. Update provided.

070825: Needlestick in BSL-3. Update provided.

Recent incidents:

072925: Isoflurane over-exposure. Chemical safety follow-up

III. Protocols Reviewed Administratively

1) Investigator: Bogunovic, M
Title: Mucosal Macrophages and Post-Infectious Inflammatory Bowel Disease (IBD)
IBC Registration: 739-24, Amendment
Training Verification: Acceptable
Brief Summary: Mice will be injected intraperitoneally or intravenously with LPS to assess the short-term (within 24 hrs) immune (inflammatory) response of macrophages and other cell types in the tissue (spleen, liver, lymph nodes, bone marrow, blood etc). This protocol will be used as a positive control for other ways to activate macrophages in vivo or in vitro, specifically by infectious agents. Mice will be euthanized 6 to 24 hrs after LPS injection, their tissues, cells and body fluids will be collected or isolated and subjected to further analysis, e.g., for upregulation of inflammatory cytokines (gene and protein expression), cell surface markers, changes in tissue distribution by immunofluorescent microscopy etc.

In addition, primary cell cultures of bone marrow derived, or intestinal FACS-purified macrophages or other sell types (e.g., enteric neurons) will be treated with a range of LPS doses (from 1 ng/ml to 1000 ng/ml of cell culture medium) for 6-12 hours. Cells and cell supernatants will be collected, and their inflammatory response will be measured as above.

BSL/ABSL: **BSL-2; BSL-2 flow sorting; ABSL-1 with Administration to animals using BSL-2 precautions/sharps safety**

NIH Guidelines: NA

2) Investigator: Bradley, E (Prev. Haran, J)
Title: Characterization of the human microbiome by sequence-based profiling of microbial communities associated with COVID-19 patient samples

IBC Registration: **770-25, Renewal**
Training Verification: Acceptable
Brief Summary: To understand the interaction between the microbiome of gut and oral microbiota, human transcriptional profiles, and SARS-Cov-2, adding: samples from patients with Influenza, and RSV infection.

BSL/ABSL: **BSL-2**
NIH Guidelines: NA

3) Investigator: Fan, X
Title: Clinical Trials at UMass Mind Clinical and Research Program (previously Psychotic Disorders Research Program)

IBC Registration: **638-25, Renewal**
Training Verification: Acceptable
Brief Summary: The UMass Mind research program conducts clinical trials with patients with a diagnosis of schizophrenia or schizoaffective disorders. As part of our research studies trials, we: Collect, process, transport blood samples from patients for routine lab safety; the collected blood is separated to serum or plasma and is packaged to be shipped either to a central lab (as designated by the study sponsor) or to the UMass Memorial lab. Blood samples are also stored in an -80F freezer for future studies.

BSL/ABSL: **BSL-2**
NIH Guidelines: NA

4) Investigator: Woda, B
Title: Morphology Core

IBC Registration: **926-25, New**
Training Verification: Acceptable pending completion of PI training
Brief Summary: Our lab perform paraffin, frozen sections and staining the slides.

BSL/ABSL: **BSL-2**
NIH Guidelines: NA

Discussed modifying requisition form to include affirmation of IBC approval for experiment and ≤ BSL-2 requirement

IV. Protocols to Discuss

1. Investigator: Fitzgerald, K

Title: Innate immunity to bacteria, viruses, and fungi

IBC Registration: **284-25, Renewal**

Training Verification: Acceptable

Brief Summary: The long-term goal of our research is to obtain a comprehensive understanding of how the innate immune response is activated and regulated following infection with viral, bacterial and fungal pathogens, as well as to understand the factors that initiate autoimmune diseases. The main objective of these studies is to examine basic principles of the immune response to infection both in vitro and by manipulating the mouse model.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: The long-term goal of this research is to achieve a comprehensive understanding of how the innate immune response is activated and regulated following infection with viral, bacterial, and fungal pathogens, as well as to identify the factors that initiate autoimmune diseases. The studies aim to define the basic principles of immune responses to infection through both in vitro experiments and manipulation of mouse models.

Experimental Approach: A combination of biochemical, molecular and genetic approaches will be used to identify key components(receptors, signaling molecules, cytokines etc.) in the innate immune response. A combination of shRNA/siRNA approaches in human cell lines and mouse cell lines as well as the use of cells from mice with targeted deletions in these key components will be used. Live or UV/heat inactivated bacteria, viruses, fungi, and pathogen-derived products like lipopolysaccharide (LPS) will be used to treat cells in vitro. They will be looking at activation of signal transduction pathways in vitro. Additionally, disease models will be used to monitor the role of innate receptors/signaling molecules in vivo. They will be using some of the following pathogens in our studies: Listeria monocytogenes, Salmonella typhimurium, Candida albicans, Sendai Virus (will not be used in vivo), Vaccinia Virus, mouse cytomegalovirus, Herpes Simples Virus-1, Influenza Virus, and Human Coronavirus SARS-CoV-2.

IBC Discussion and Vote

Discussion: Reviewer questioned the status of the Junin virus being listed and whether it has been obtained yet. Another protocol using the same strain was sent back to UVM to be sequenced to ensure it is not considered a select agent strain. A member brought to the attention of the committee that Citrobacter Rodentium no longer requires ABSL-3; downgraded to ABSL-2 per Animal Medicine. The reviewer also mentioned that the flow sorting might need to be two separate addendums as there are a variety of different infectious agents.

Meeting Decision: Vote to approve upon completion of action items.

BSL/ABSL: **BSL-3; BSL-2 Enhanced Flow Sorting; ABSL-3**

NIH Guidelines: III-D, III-E, III-F

2. Investigator: Gao, F

Title: Adenoviral vector production by Vector Core

IBC Registration: **IBC202300015, Amendment**

Training Verification: Acceptable

Brief Summary: Adding in vivo use of adenoviral vectors.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: Adding in vivo use of adenoviral vectors.

Experimental Approach: Mice will receive intravenous adenoviral vector treatment.

IBC Discussion and Vote

Discussion: Reviewer discussed some minor action items. Describe AdV-CapLib in greater detail. Vector checklist needed.
Meeting Decision: Vote to approve upon completion of action items.
BSL/ABSL: **BSL-2; ABSL-1 with Special Precautions; Administration to Animals Using BSL-2 Precautions/ Sharps Safety**
NIH Guidelines: III-D

3. Investigator: Harris
Title: Determining the Factors that Contribute to Autoimmune Skin Disease
IBC Registration: **465-21, Amendment (previously TABLED)**
Training Verification: Acceptable
Brief Summary: We are proposing to add Adeno-Associated Virus (AAV) vector to the IBC protocol. This addition involves injections of AAVs to animals.
Biosafety Considerations:

- AAV is a replication-deficient viral vector with a low risk of pathogenicity.
- The proposed work will adhere to BSL-2 containment as appropriate for the specific serotype and use.
- Proper PPE, engineering controls, and decontamination protocols will be followed to mitigate any potential risks.

Added New Agents: Adeno-associated virus (AAV) vector

Brief Summary and Review by Primary Reviewer

Overview and Objectives: The goal of the study is to recruit regulatory T cells and CAR Tregs to the skin following AAV transduction for the treatment of vitiligo. In addition, the investigators will utilize AAV vectors to treat skin genetic disorders.

The investigators propose to add adeno-associated virus (AAV) vectors to their IBC protocol for the purpose of injecting AAVs into animals

Experimental Approach: In vivo experiments: AAV vectors will be produced by the UMass Viral Vector core. AAV vectors will be directly injected into animals by staff listed on Harris IACUC protocol 201900330. Animals will be followed for development of vitiligo per their described mouse model, and animals will be sacrificed at various timepoints of disease development to observe treatment effects on tissues and cell populations by flow cytometry.

Ex vivo experiments: mouse T cells will be generated ex vivo using lentiviruses or gamma retroviruses (CAR and CAR T-regulatory cells) in Dr. Keeler's lab/UMass Viral Vector Core.

- The proposed work will adhere to BSL-2 containment.

- The proposed work will adhere to BSL-2 containment. Proper PPE, engineering controls, and decontamination protocols will be followed to mitigate any potential risks.

IBC Discussion and Vote

Discussion: Reviewer discussed that the lab had not submitted a vector checklist but if the Car-T cells are being handled by the Keeler lab then it's not necessary. A question was raised about human PBMC's being mentioned in the flow sorting, but none listed for the amendment so there is some confusion around whether old information should be included or a new flow addendum should be submitted with only the relevant information that pertains to the amendment. The committee decided that a separate flow addendum only including what's being added should be submitted with the amendment as it's the easiest and clearest way to determine what is being added.

Meeting Decision: Vote to approve upon completion of action items.

BSL/ABSL: **BSL-2; BSL-2 Enhanced Flow Sorting; ABSL-2**

NIH Guidelines: III-D, III-E

4. Investigator: Kaufman, P

Title: Nucleosome Assembly during DNA synthesis and repair

IBC Registration: **233-25, Renewal**

Training Verification: Acceptable

Brief Summary: We are studying the roles of various proteins in maintaining the normal structure and function of mammalian cell chromosomes.

- We use siRNA duplexes or lentiviral vectors to conduct RNA interference (RNAi)-mediated depletion of different target proteins, or to express recombinant proteins or protein domains.

In other experiments, we are testing chromosomal responses of innate immune cells such as macrophages and monocytes to external stimulation, focusing on the role of lncRNA Neat1 in the context of mouse macrophage biology.

- Using Cas9, we generate deletions in the Neat1 gene in an immortalized macrophage cell line, and test these mutants have defects in inflammatory gene expression after exposure to lipopolysaccharide.
- We plan to test how these gene expression changes are correlated with changes in the ability to withstand infection by a *Salmonella enterica, serotype Typhimurium* bacteria, strain SL1344.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: The researchers are studying the roles of various proteins in maintaining the normal structure and function of mammalian cell chromosomes. siRNA duplexes or lentiviral vectors will be used to conduct RNA interference (RNAi)-mediated depletion of different target proteins, or to express recombinant proteins or protein domains. Additionally, They will be testing chromosomal responses of innate immune cells such as macrophages and monocytes to external stimulation, focusing on the role of lncRNA Neat1 in the context of mouse macrophage biology. Using Cas9, they will generate deletions in the Neat1 gene in an immortalized macrophage cell line, and test whether these mutants have defects in inflammatory gene expression after exposure to lipopolysaccharide. They plan to test how these gene expression changes are correlated with changes in the ability to withstand infection by a Salmonella enterica, serotype Typhimurium bacteria, strain SL1344.

Experimental Approach: For lentivirus experiments, a self-inactivating (SIN) lentivirus that is replication–incompetent will be used. The production of viruses will performed by co-transfecting three helper plasmids (pLP1, pLP2, and pVSVG) encoding essential proteins for virus particle assembly. This four plasmids system (including the plasmid harboring the genetic element of interest) ensures that no replication-competent viruses are made. In some cases where we need a high multiplicity of infection, viral particles will be collected by ultracentrifugation and use the concentrated lentivirus to infect target cell lines.

To verify that no replication-competent lentiviruses are produced a well-established RT-PCR assay (Li and Rossi, Methods in Enzymology v. 392: 218-226 (2005), which tests for production of viral core proteins, will be used. Specifically, the assays will test for expression of nucleotides 4723-5092 from the pLP1 helper plasmid.

Baculovirus vectors will be used to express recombinant proteins in insect tissue culture cells.

For macrophage infection experiments, an inoculum will be prepared by growing Salmonella enterica, serotype Typhimurium bacteria, strain SL1344 to log phase in LB broth and adjusted to 1.0 at OD600. Bacteria will be washed and suspended in PBS and added to the macrophages at various multiplicities of infection. Following infection. cells will be washed with PBS, and fresh media containing gentamycin (100 µg/mL) will added for an hour to kill extracellular bacteria. At various times post-infection, cells will be washed with PBS and lysed in pure water. Bacterial CFU will be assessed by plating dilutions onto LB agar plates, or via immunofluorescence microscopy of fixed samples using anti-bacterial antibodies. All liquid cultures will be bleach treated and materials in contact with the bacteria will be placed in biohazard waste for autoclaving.

IBC Discussion and Vote

Discussion:	Reviewer noted the PI is missing a flow addendum and vector checklist. Minor action items mentioned.
Meeting Decision:	Vote to approve upon completion of action items.
BSL/ABSL:	**BSL-2; BSL-2 Enhanced Flow Sorting**
NIH Guidelines:	III-D, III-E, III-F

5. Investigator: McKitterick, A
Title: Bacterial Envelope Assembly
IBC Registration: **924-25, New**
Training Verification: Acceptable pending completion of PI training
Brief Summary: The aim of my research is to understand bacterial cell envelope synthesis. This envelope is a complex series of layers that structurally support the cell as well as act as a dominant interface for human immune sensing and phage infection. Not only are the components of the bacterial cell envelope the most important and successful antibiotic targets, but they also act as a permeability barrier that can prevent antibiotic uptake. Thus, further study of this critical layer has the potential to lead to key innovations in multi-drug resistant bacterial infection treatments.

Within this realm, I am interested to uncover:

- What are key factors that contribute to bacterial cell envelope synthesis and regulation?
- How has bacterial envelope evolved between phylogenetic branches?
- What are the mechanisms that bacteria use to grow and divide?
- How have phages have evolved to infect specific bacteria?
- How do phage infections shape the ecology of the bacterial cell envelope?

Brief Summary and Review by Primary Reviewer

Overview and Objectives: To understand bacterial cell envelope synthesis.

This envelope is a complex series of layers that structurally support the cell and act as a dominant interface for human immune sensing and phage infection. Components of synthesis are antibiotic targets. They also act as a permeability barrier that can prevent antibiotic uptake. There is potential for innovations in multi-drug resistant bacterial infection treatments.

Questions being addressed include:

- What are key factors that contribute to bacterial cell envelope synthesis and regulation?
- How has bacterial envelope evolved between phylogenetic branches?
- What are the mechanisms that bacteria use to grow and divide?
- How have phages have evolved to infect specific bacteria?
- How do phage infections shape the ecology of the bacterial cell envelope?

Experimental Approach: To understand the factors that contribute to bacterial cell envelope synthesis, bacteria will be transposon mutagenized. Transposons will be delivered from phage, plasmid, or PCR-derived delivery vehicles. Mutants will be screened either by microscopy or using Tn-Seq procedures. Mutants with transposon insertions will be plated on media with the appropriate antibiotic. The collection of mutants will be pooled and stored at -80ºC.

Screening involves plating the mutated collection on indicator medium designed to detect an elevated level of cell lysis or challenge the collection with a phage. Mutants with phenotypes of interest will be investigated further by standard microbiological methods to determine the location of the transposon and monitor the extent of the assembly defects. Standard genetic manipulation of bacterial strains and phage isolates will be performed, along with standard microbiological processes involving liquid and solid medium culturing. Procedures performed will include phage infection and transduction, transposon mutagenesis, CRISPRi, PCR, plasmid cloning, chromosomal engineering (generation of knockouts), transformation, time-lapse and fluorescence microscopy, and protein purification and pulldown. Phages will be isolated from soil and compost environmental samples, which will be processed by diluting in water and filter-sterilizing to isolate the phages. Cultures of bacteria of interest will be infected by the phages; Corynebacterium glutamicum and Amycolatopsis mediterranei, in both solid and liquid cultures to screen for plaque formation (phage infection). The phages that infect bacterium of interest will be characterized by growing up high-titer stocks, and conducting whole genome sequencing on the phage.

IBC Discussion and Vote

Discussion:	Reviewer discussed that the bacteria to be studied were detailed nicely but 2 are risk group 2 agents (not RG1 as listed). The volumes are small scale
Meeting Decision:	Vote to approve upon completion of action items.
BSL/ABSL:	**BSL-2**
NIH Guidelines:	III-D, III-E, III-F

6. Investigator: Silverman, N
Title: Innate Immune Recognition and Signaling
IBC Registration: **226-25, Renewal**
Training Verification: Acceptable
Brief Summary: The projects in the lab utilize Drosophila and Drosophila cell lines, as well as human, non-human primate and mouse cell lines for the study of innate immune recognition and signaling. Often we express genes of insect, mammalian or bacterial origin in transfected cell lines or transgenic Drosophila. For these expression experiments, we use pUC based expression plasmids. For

transgenic Drosophila work, these plasmids also contain P-transposon or Phage-PhiC31 based elements. We also produce recombinant proteins for in vitro characterization using bacterial, Drosophila cell culture or Baculovirus expression systems. Cell lines used include S2 (Drosophila), Sf9 (lepidopterean), HEK293, THP-1, HCT116 (human), BSC-1, BSC-40, Vero (primate, non-human). We also regularly infect Drosophila, by parenteral injections or natural infection, with E. coli, Micrococcus luteus, Erwinia carotovora carotovora, Providencia rettgeri Beauveria bassiana , Lactococcus lactis (all BSL-1), as well as with viruses (BSL-2), such as Sindbis virus (SV), Vesicular stomatitis virus (VSV), Insect Iridescent virus 6 (IIV-6), Amsacta moorei entomopoxvirus (AMEV). These infection experiments are performed to monitor innate immune responses in flies.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: To utilize Drosophila and Drosophila cell lines, as well as human, non-human primate and mouse cell lines for the study of innate immune recognition and signaling. Genes of insect, mammalian or bacterial origin are expressed in transfected cell lines using pUC-based expression plasmids or in transgenic Drosophila with P-transposon or Phage-PhiC31 based elements. Recombinant proteins for in vitro characterization are generated using bacterial, Drosophila cell culture or Baculovirus expression systems. Cell lines used include insect (S2, Sf9); human (HEK293, THP-1, HCT116), and NHP (BSC-1, BSC-40, Vero). The lab also regularly infects Drosophila, by parenteral injections or natural infection, with E. coli, Micrococcus luteus, Pectobacterium carotovora, Providencia rettgeri, Beauveria bassiana, Lactococcus lactis (all BSL-1), as well as with viruses (BSL-2), such as Sindbis virus, vesicular stomatitis virus, Insect Iridescent virus 6 (IIV-6), and Amsacta moorei entomopoxvirus (AMEV). These infection experiments are performed to monitor innate immune responses in flies.

Experimental Approach: Methods include standard molecular biological / cell culture techniques for transient and stable transfections. Infections of Drosophila are performed with either stainless microsurgery needle, dipped in a concentrated bacterial solution or with a pulled-glass-capillary needle syringe. Briefly, adult or larval Drosophila or Drosophila cultured cell lines are challenged with microbes or stimulated with microbial products. Mutant animals (or RNAi-mediated gene knock-down in cultured cells or transgenic animals) are used to analyze the role of particular host genes in the response to these microbial challenges. Responses measured included lethality following infection as well as the induction of antimicrobial peptide genes.

Other common methods involve generating transgenic Drosophila or otherwise altering the Drosophila genome (i.e. with Crispr/Cas9 technology) to study innate immune responses.

In mammalian cells, we plan to use lentiviral vectors to deliver shRNAs, for gene knockdown, or cDNAs of interest for expression. These reagents will be purchased through the UMMS Core or generated in-house.

IBC Discussion and Vote

Discussion:	Reviewer discussed that a vaccinia virus vaccine is available and that a medical alert card is required for vaccinia virus unattenuated strains. The PI should list the individual vaccinia virus strains in the registration.
Meeting Decision:	Vote to approve upon completion of action items.
BSL/ABSL:	**BSL-2**
NIH Guidelines:	III-D, III-E, III-F

7. Investigator: Tang, Q
Title: Therapeutic Application of Chemically Modified Nucleic Acids
IBC Registration: **925-25, New**

Training Verification: Acceptable

Brief Summary: The overall goals of the lab are to develop RNA-based therapeutics for relevant disease indications. This is done by testing modified or formulated RNA oligonucleotides (or RNA/protein complexes) developed in the lab in in vitro experiments using mouse, human, NHP, cat and dog cell lines and primary cells followed by in vivo rodent and large animal models.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: The overall goals of the lab are to develop RNA-based therapeutics for relevant disease indications. This is done by testing modified or formulated RNA oligonucleotides (or RNA/protein complexes) developed in the lab in in vitro experiments using mouse, human, NHP, cat and dog cell lines and primary cells followed by in vivo rodent and large animal models.

Experimental Approach: Project 1: Screening modified RNAs in vitro.

In vitro work involves testing modified RNA oligonucleotides in modified and primary cells of human and mouse origin. The oligonucleotides will either be naked, complexed with mRNA, protein or formulated (liposome, endosome, polymer, small drugs). Also, we will use peripheral blood (PBMCs, serum, and whole blood), fibroblasts etc from patients with and without the conditions of interest as a model to test the modified RNA oligonucleotides in a more relevant biological context.

Project 2: Screening modified siRNAs in vivo.

The overall goals of our work are to develop RNA-based therapeutics for several diseases as well as to use oligonucleotides to study and validate gene function in vivo. Chemically modified oligonucleotides (naked, complexed or formulated) designed to specifically and selectively target a gene involved in a disease are screened and tested in vitro before animal models are used.

Animal models will be used to test toxicity, tissue distribution, pharmacokinetic/pharmacodynamic (PK/PD) parameters, and efficacy. In addition, the ability of specific oligonucleotide formulation or the co-administration of a drug (i.e guanabenz or dextran sodium sulfate (DSS, etc.) to enhance various in vivo parameters, will also be tested.

In vivo experiments involve injection of mice by several routes (i.e IV, SC, IP, transdermal, intradermal, topical, etc.) with the in vitro tested oligonucleotides. Like the in vitro experiments, oligonucleotides can be naked, in liposomes, and/or injected with a drug.

Project 3: Cell type specific delivery of siRNA in mice and human cells

In this project we will inject mice and isolate specific organs to look at various cell populations to determine the amount of uptake of a fluorescently labeling siRNA. We will also use FACS to determine knockdown in human cell lines and the mouse. In addition, we will use FACS sorting of human cell lines and mouse tissues to quantitate uptake in specific cell types.

Project 4. Evaluation of impact of oligonucleotide formulation on AAV and CRISPR delivery and distribution.

This experiment is performed similarly to what is described above, but oligonucleotides are complexed or co-formulated with proteins or AAV. Detection methods expand to evaluation of reporter gene expression and/ or editing activity (RNAseq or targeted PCR). AAVs will be obtained from laboratories at UMass (Drs. Esteves, Gao, etc.) or outside collaborators.

IBC Discussion and Vote

Discussion: Reviewer discussed that further clarification is required for the human cells being used in mice. Medical alert cards are required for B-Virus. There is too much ambiguity as to what is work is being done and the committee agreed that there not enough information to make an appropriate risk assessment.

Meeting Decision: **TABLED**

BSL/ABSL: N/A

NIH Guidelines: N/A

8. Investigator: Yamada, K

Title: Development of RNAi-based Therapeutics

IBC Registration: **927-25, New**

Training Verification: Acceptable

Brief Summary: The overall goals of the lab are to develop RNA-based therapeutics for several disease indications. This is done by testing modified or formulated RNA oligonucleotides (or RNA/protein complexes) developed in the lab in *in vitro* experiments using mouse, human, NHP cell lines and primary cells followed by in vivo rodent and large animal models. There are a variety of targets we work with including huntingtin, sFLT1, ApoE etc. RNA-based therapeutics development includes overexpression and purification of recombinant Ago2 for use in *in vitro* experiments as well as study of the impact of siRNA gene modulation on ex vivo perfused organs.

Brief Summary and Review by Primary Reviewer

Overview and Objectives: The overall goals of the work are to develop RNA-based therapeutics for several disease indications as well as use oligonucleotides to study and validate gene function in vivo.

Experimental Approach: The approach includes testing modified or formulated RNA oligonucleotides (or RNA/protein complexes) developed in the lab in in vitro experiments using mouse, human, NHP cell lines and primary cells followed by in vivo rodent and large animal models. There are a variety of targets we work with including huntingtin, sFLT1, ApoE etc. RNA-based therapeutics development includes overexpression and purification of recombinant Ago2 for use in in vitro experiments as well as study of the impact of siRNA gene modulation on ex vivo perfused organs.

IBC Discussion and Vote

Discussion: There is too much ambiguity as to what work is being done and the committee agreed that there not enough information to make an appropriate risk assessment.

Meeting Decision: **TABLED**

BSL/ABSL: **N/A**

NIH Guidelines: N/A

V. Report on incidents/accidents/issues involving BSL-3 & ABSL-3 Facilities

1) Card reader incident in the BSL-3: Actively being addressed. A programming error was identified.

Dates to track:

2) SAT Site Visit- September 21, 2025 Select Agent Inspection

VI. Information from the field (Senior Biosafety Officer)

N/A

VII. Other Business

1) AAALAC: visit preparation is well under way
2) MTB medical SOP was updated and will be posted

Acknowledgement Items:

1) Caricchio 880-23- CAB-201 - for SLE ICF v3.0 & IB v2.0
   - **A Phase 1/2, Open-label Study to Evaluate the Safety and Efficacy of Autologous CD19-specific Chimeric Antigen Receptor T cells (CABA-201) in Subjects with Active Systemic Lupus Erythematosus**

   Changes made in screening procedures

2) Hayward 917-25- protocol amendment (EPI-321-02)
   - **A Phase 1/2, Open-label, Dose-escalation Study to Evaluate the Safety, Tolerability, and Biological Activity of EPI-321, an AAVrh74-delivered Epigenetic Editing Therapy in Adult FSHD Patients**
   - EPI-321 is an investigational drug product comprising a recombinant adenoassociated viral vector, serotype rh74 (AAVrh74), for the delivery of genetic material CK8e-dCasONYX-KLb_hU6-D4Z4gRNA encoding an epigenetic editing system for the treatment of FSHD… designed to selectively bind the D4Z4 repeat region via the accompanying guide RNA, methylate CpG groups within the region on chromosome 4q35, and thus repress the expression of the DUX4 gene,
   - ameliorating the downstream pathology that drives FSHD.

4.0 (30APR2025) 1) Updated the advised duration of antibiotic prophylaxis from 2 weeks to 8 weeks after final eculizumab dose (for participants who may be administered eculizumab), per guidance from Alexion, which markets eculizumab (Soliris).
2) Corrected an error in the Schedule of Assessments regarding PGI-C and CGI-C, which are not performed at baseline; updated the PGI-C and CGI-C related endpoints to reflect they are not collected at baseline.
3) Specified in the schedule of assessments that if EPI-321 dosing is unexpectedly delayed, some completed baseline day -7 to -4 assessments will remain valid and need not be repeated should actual dosing occur within 90 days of the assessment.
4) Modified Appendix B, which provides a recommended sequence of clinical outcome assessments for a given study visit, to better balance and distribute the assessments over the visit.
5) Modified AAVrh74 pre-existing immunity exclusion criterion to anti-AAVrh74 total binding antibody titer to □ _1:400 (was previously > 1:400); this updated criterion reflects the intended eligibility based on clinical AAVrh74 experience.

5.0 (10JUN2025) 1) Modified exclusion criterion 14 to be more clinically rationale as it aims to cap the total vector genome dose that may be administered to participants on the study for safety purposes. Specified that the previous criterion (>90 kg) applies to participants dosed at the higher dose level (4 x 10^{13} vg/kg IV); this retains the previous cap or maximum total vector genome dose administered to participants on the study. Added a new criterion for participants dosed at the lower dose level (2 x 10^{13} vg/kg), to exclude participants who are both > 90 kg and meet CDC criteria for obesity (Class 1, or higher).

6.0 (24JUN2025)	1) Added blood draws to measure circulating KHDC1L, a protein activated in the DUX4 pathway and recently reported to be a potential therapeutic response biomarker for FSHD, at the following timepoints: Baseline, Month 3, Month 6, and Month 12. 2) Modified the Potential Risks section related to AAV gene therapy class risks to direct the investigator to the Investigator's Brochure (IB) for detailed information, as the IB is periodically updated for the field's evolving understanding of AAV gene therapy class risks. 3) Modified the Potential Use of Eculizumab and Rationale section, Synopsis, and Potential Risks section to reflect that while TMA has predominantly been reported in AAV9 products, it is not exclusively reported in the AAV9 serotype.

Adjourned at 1:52pm