Institutional Biosafety Committee:

Present: Dr. Steven Albelda, Dr. Julian Baptiste, Dr. Jessica Buchanan, Dr. Joseph Fraietta, Dorothy Kaplan, Dr. Daniel Kessler, Dr. Andrew Maksymowych, Dr. Maureen O'Leary, Dr. David Pegues

Absent: Dr. Paul Bates, Dr. Sara Cherry,

Invited Guests: Ms. Stephanie Adams, Dr. Tucker Piergallini, Mr. Edwin Siu, Ms. Amanda Wong, Ms. Kimberly Craig, , Ms. Jessa Yoos, Ms. Denene Wambach

The Institutional Biosafety Committee Meeting was called to order by Dr. Daniel Kessler at **10:00 AM**.

1. IBC Minutes: **06-23-2025**

- The IBC reviewed the IBC Minutes.
- All members are in favor of approval as submitted.
- Minutes approved as submitted.

2. Registrations to Review:

SECTION III–C. Experiments Involving Human Gene Transfer that Require IBC & IRB Approval <u>Prior to</u> Initiation:

1. Bagley ....**#25-105**................. **C–1**

Dr. Stephen Bagley – HGT Protocol Registration Amendment V2 .... **AMENDMENT**
PROTOCOL TITLE: Phase 1, Open-Label Study Evaluating the Safety of CART-EGFR-IL13Ra2 Cells in Patients with Newly Diagnosed, EGFR-Amplified, MGMT-Unmethylated Glioblastoma Following Completion of Initial Radiotherapy. (Protocol V2 July 10, 2025; Main ICF V2 July 10, 2025; Retreatment ICF V2 July 10, 2025.)
IBC #25-105, IRB #858463, NCT #06973096, Sponsor #03325, IND #28055

- Dr. Daniel Kessler introduced the submission.
- Dr. David Pegues provided a summary and analysis.

"Phase 1, Open-Label Study Evaluating the Safety of CART-EGFR-IL13Ra2 Cells in Patients with Newly Diagnosed, EGFR-Amplified, MGMT-Unmethylated Glioblastoma Following Completion of Initial Radiotherapy

Project Overview: Targeted Disease and/or Clinical Aim of Project

The treatment standard for newly diagnosed glioblastoma multiforme (GBM) has been maximal safe surgical resection followed by adjuvant radiation therapy and temozolomide, a DNA methylating drug. However, 50%-60% of GBM tumors have an unmethylated O6-methylguanine-DNA methyltransferase (MGMT) promoter, and these patients do not benefit from temozolomide treatment. Currently, there are no approved immunotherapies for treatment of GBM.

This is an open-label phase 1 study to assess the safety, feasibility, pharmacokinetics and preliminary efficacy of autologous T cells co-expressing two CARs targeting the cryptic EGFR epitope 806 and IL13Ra2 (referred to as "CART-EGFR-IL13Ra2 cells") in patients with newly diagnosed, EGFR-amplified, MGMT-unmethylated glioblastoma, without evidence of disease recurrence/progression following completion of initial radiotherapy.

Agent Description (Detailed in the Investigator's Brochure)

The active ingredient of CART-EGFR-IL13Rα2 cells is the autologous, patient-derived T cells genetically modified by lentivirus transduction to express a bicistronic CAR transgene from the same transcript, using P2A, the 2A self-cleaving mechanism of the Porcine teschovirus. Each independent CAR transgene consists of:

- a single chain variable fragment (scFv) (the human-murine chimeric EGFR scFv specifically binds the conformationally-dependent EGFR epitope 806 and the humanized IL13Rα2 scFv specifically binds IL13Rα2)

- a hinge derived from CD8α, and

- a tandem signaling domain comprised of the TCRζ signaling module linked to the 4-1BB costimulatory domain

EGFR epitope 806 is cryptic, conformation binding epitope that is overexpressed in numerous cancers, including GBM; monoclonal antibody 806 does not bind to normal EGFR. IL13Ra2 expression is limited to malignancies and adult testes and is expressed in ~82% of GBM cases.

Is a novel vector system, approach or technology used for this clinical trial? NO

Gene transfer agent delivery method: Third-generation lentivirus

Intended target, cells: Patient-derived autologous T cells

Biosafety Concerns applicable to clinical staff, subjects, subject families, community

(Please note concerns in this section)

Potential for shedding: Replication competent lentivirus, insertional oncogenesis, uncontrolled T cell proliferation, complications of intracerebral delivery, febrile reactions, CRS and acute neurologic toxicity.

No new or unanticipated safety concerns.

Are "Standard Precautions," Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted."

- The amendment was discussed by the committee members.
- All members were in favor of approval.
- The HGT registration amendment is approved as submitted.

2. Chapin............**#23-153** **C-1**

Dr. William J Chapin – HGT Protocol Registration Amendment 5... **AMENDMENT**

PROTOCOL TITLE: UPCC 12223: A Phase 2, Open-label, Multicenter Study Investigating RP2 Oncolytic Immunotherapy in Combination with Second-line Therapy in Patients with Locally Advanced Unresectable, Recurrent and/or Metastatic Hepatocellular Carcinoma. (Protocol V5 dated April 17, 2025; Main ICF dated February 3, 2025)
IBC #23-153, IRB #854065, IND #028278, UPCC #12223, Protocol #RP3-003

- Dr. Daniel Kessler introduced the submission.
- Dr. Steven Albelda provided a summary and analysis.

"This is A Phase 2, Open-label, Multicenter Study Investigating RP2 Oncolytic Immunotherapy in Combination with Second-line Therapy in Patients with Locally Advanced Unresectable, Recurrent and/or Metastatic Hepatocellular Carcinoma

Project Overview: This study will evaluate whether treatment with RP2 can provide efficacy as a second line treatment combined with atezolizumab plus bevacizumab in patients with locally advanced unresectable, recurrent, and/or metastatic HCC.

The primary reason for this protocol amendment was to revise the guidance around esophagogastroduodenoscopy (EGD) to ensure all varices (irrespective of size) are assessed and treated regardless of grade before enrollment, rather than allowing patients to be treated per local standards. Bevacizumab has been associated with variceal bleeding.

Agent Description:

RP2 is a selectively replication competent, acyclovir-sensitive HSV-1 virus. RP2 is the second in a series of viruses (RP1, RP2, and RP3) being developed by Replimune, with each virus modified as compared to its predecessor by the insertion (or deletion) of additional transgenes.

RP2 was constructed using a new strain of HSV-1 (strain RH018). The neurovirulence factor (infected cell protein [ICP] 34.5) encoding genes and the ICP47-encoding gene are deleted from the virus making replication tumor-selective. The virus also contains a codon-optimized sequence for human granulocyte-macrophage colony-stimulating factor (hGM-CSF) in addition to a codon-optimized sequence for the gibbon ape leukemia virus surface protein (GALV-GP) with the R sequence deleted (R-). Cell-to-cell fusion is caused by GALV-GP-R-, resulting in accelerated cell death. Further, RP2 expresses a human cytotoxic T lymphocyte antigen 4 blocking antibody-like molecule (ahCTLA-4), which interferes with the interaction of cytotoxic T lymphocyte antigen 4 (CTLA-4) with B7 molecules on professional antigen presenting cells.

A first generation HSV1 virus (lacking the neurovirulence genes ICP 34.5 and ICP47, and expressing GM-CSF) was FDA approved in 2015 for IT injection in melanoma and has proven safe.

RP2 consists of (a) a lipid bilayer envelope derived from host cell membranes, including polyamines, lipids, and glycoproteins; (b) a tegument of amorphous material; (c) a capsid made of capsomers

arranged in icosapentahedral symmetry; (d) an internal core containing double-stranded DNA of ~160 Kb pairs

Is a novel vector system, approach or technology used for this clinical trial? YES

Gene transfer agent delivery method: Intratumoral injection

Intended target: Hepatocellular carcinoma cells

Any other material to be used in preparation of the agent (vector and transgene) to be administered to research participants (e.g., helper virus, packaging cell line, carrier particles): Preparation details were not provided other than RP2 is a preparation of a genetically modified live herpes simplex 1 virus that is cultured in Vero cells

Summary of preclinical studies conducted in support of the proposed clinical trial: Antitumor efficacy of a mouse version of RP2 has been demonstrated in immune-competent mice with HSV-1-permissive mouse tumors (A20) and with RP2 in hCTLA-4 knock-in immune-competent mice with MC38 mouse tumors. Consistently, tumor regression or clearance was seen following intratumoral administration of RP2 at a range of virus doses in both injected and noninjected tumors. No serious toxicity was seen.

A number of patients have been treated with RP2 in a clinical trial (RP2-001-18) that included patients with advanced or metastatic solid tumors (including melanomas, sarcomas, pancreas tumors, colorectal, and head and neck cancers). As of the cutoff date of 20 December 2024, 85 patients had been treated in Study RP2-001-18. 25 patients with RP2 monotherapy and 60 patients with RP2 and nivolumab. Of the 60 patients enrolled in a cohort that received RP2 in combination with nivolumab, all received at least 1 dose of RP2, and 50 patients have received at least 1 dose of nivolumab. No serious adverse events were noted.

Biosafety Concerns applicable to clinical staff, subjects, subject families, community

Potential for shedding

RP2 levels (copies of DNA or viral titer) will be determined in blood, urine, and potentially other bodily fluids if needed at time points. Levels of RP2 will be determined in saliva/oral mucosa, injection sites, injection-site dressings, and lesions that appear to be herpetic.

Are "Standard Precautions," Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted."

- The amendment was discussed by the committee members.
- All members were in favor of approval.
- The HGT registration amendment is approved as submitted.

3. Clark ....**#25-171** **C–1**

Dr. Amy S Clark – NEW HGT Protocol Registration .... **FULL REVIEW**

PROTOCOL TITLE: UPCC 02110: I-SPY 2 TRIAL - Investigation of Serial Studies to Predict your Therapeutic Response with Imaging and Molecular Analysis 2. (Protocol V39 date March 12, 2025; Main ICF dated March 18, 2025; **IBC #25-171, IRB #844215, IND #105139, UPCC #02110**

- Dr. Daniel Kessler introduced the submission.
- Dr. Paul Bates provided a summary and analysis.
- Dr. Paul Bates provided a summary and analysis.

"I-SPY 2 Trial (Investigation of Serial Studies to Predict Your Therapeutic Response with Imaging and Molecular Analysis 2).

I-SPY 2 TRIAL is sponsored by Quantum Leap Healthcare Collaborative and designed to evaluate and develop biomarkers of early response to therapeutic regimens, and to develop a strategy to improve outcomes of adults who do not have an optimal response to current standard therapy. I-SPY 2 TRIAL will specifically examine the efficacy of investigational agents/agent combinations in adults with locally advanced Stage II or III breast cancer with high risk for early recurrence. This is a platform trial that will continue until agents or combinations of agents are discovered that achieve the endpoint of over 90% pathologic complete response (pCR) rates in all high risk breast cancer subtypes. The trial uses a study design, I-SPY 2.2, which consists of 3 sequential regimens or blocks. Participants are screened and adaptively randomized based on breast cancer subtype to receive one of several novel targeted agents (without concomitant chemotherapy) in Block A. When these agents and regimens are not successful after 3-6 weeks of minimal response, the novel therapy is halted and participants can be re-randomized to receive a second targeted rescue agent representing the best-in-class therapy for their particular sub-type (Block B). Following the completion of the 12-week Block B treatment, participants with complete response will proceed to surgery while those who still have significant residual disease will receive standard of care anthracycline chemotherapy (Block C) prior to surgery. One of the investigational agents used in this trial is VSV-IFNβ-NIS (also known as VV1) which is based on a recombinant, conditionally replication-competent vesicular stomatitis virus (VSV) engineered to enhance the immune response and to allow imaging of tumors supporting vector replication. VV1 is used in combination with cemiplimab, a human monoclonal antibody against PD-1 in participants with breast cancer that are HER2 negative. The study agent VV1 is given as an intratumoral (IT) administration at a dose of 3 x109 transforming units (TCID50) in a volume of 3 mL. In patients with multiple tumors, the viral dose (3 mL) can be divided between the injectable lesions proportionately to the largest of the

respective lesion on MRI at the physician's discretion. The intratumoral VV1 injection is administered 60 minutes to 24 hours following a fixed dose of intravenous (IV) cemiplimab every 21 days for a maximum of 4 cycles. MRI will be performed according to schedule of study assessments as a non-invasive serial measurement of response during the course of treatment; and tissue samples will be collected including blood and tumor biopsies for molecular analyses including genomic DNA mutation, SNP profiling, and gene expression profiling. There is a 10-year follow-up period for all participants in the trial.

Project Overview: This protocol is part of a larger study which aims to determine optimal treatment strategies for adults with locally advanced Stage II or III breast cancer with high risk for early recurrence.

Agent Description: The agent to be administered is VSV-IFNβ-NIS; also referred to as Voyager-V1™ or VV1. Vesicular Stomatitis Virus (VSV) is a member of the Rhabdoviridae family. VV1 is a recombinant VSV that has been engineered to express both interferon (IFN) beta (β) and the human thyroidal sodium iodide symporter (NIS) for optimal oncolytic efficacy, safety and feasibility of noninvasive in-vivo tracking of virotherapy.

IFNβ released by infected cells induces an antiviral state in surrounding normal cells, thereby curtailing virus spread. Cancer cells are often hyporesponsive to IFNβ, such that the VV1 infection spreads more efficiently in cancerous tissue where it is selectively destructive (oncolytic). In addition, IFNβ has anti-tumor effects including enhancement of innate immune responses, antiproliferative activities and the priming of T-cell responses.

Expression of the NIS protein by the VSV infected tumor cell enables cellular uptake of radiotracers such as 123I and 99mTc pertechnetate, thereby allowing noninvasive detection of sites of VSV infection using SPECT/CT imaging. Imaging NIS with 123I or 99mTc pertechnetate has been a widely used clinical procedure to diagnose thyroid diseases or cancer for more than 60 years and does not impose additional safety concerns for the patient.

Is a novel vector system, approach or technology used for this clinical trial? NO

Gene transfer agent delivery method: Intratumoral (IT) administration at a dose of 3 x10E9 transforming units (TCID50).

Intended target: In vivo infection of cells near injection site. VSV is cytopathic and is cleared very rapidly by host immune system so replication in vivo is limited. VSV is an RNA virus so there is no integration of foreign genetic information into host nor any residual viral genetic material in the cells. Tumor cells are inherently deficient in innate immune responses compared to non-tumor tissues, therefore VV1 replicates preferentially in tumor cells.

Any other material to be used in preparation of the agent (vector and transgene) to be administered to research participants (e.g., helper virus, packaging cell line, carrier particles)

VV1 will be used +/- cemiplimab; a PD-1 targeting mAb that acts as a checkpoint inhibitor.

Summary of preclinical studies conducted in support of the proposed clinical trial: Preclinical toxicology studies in mice have shown no limiting toxicity or adverse events (AEs) in any treatment groups at any time points when a dose up to 5x1010 TCID50/kg was used. Importantly, no infectious virus was recovered from brain, spinal cord, spleen or liver.

A preclinical rapid dose-escalation study was carried out in dogs to (i) determine maximum tolerated dose (MTD), (ii) characterize the adverse event profile and (iii) describe route(s) and duration of virus shedding following IV VV1 administration in purpose-bred healthy, immune-competent dogs. This study indicated that an IV dose of 1010 TCID50 is well-tolerated in dogs. At a dose of 1011 TCID50, severe dose limiting toxicities (DLTs) were observed. The primary DLT was hepatotoxicity (aspartate transaminase (AST) and alanine aminotransferase (ALT) >10 x upper limit of normal (ULN), possibly caused by particle toxicity and/or release of inflammatory cytokines resulting in abnormal clotting with necropsy indicating disseminated intravascular coagulation. Virus shedding analysis indicated low levels of VSV-N genomes detectable in buccal swabs and urine with no detectable infectious virus.

A veterinary study of client-owned dogs demonstrated s similarly strong saftery profile for VV1 or a related virus carrying the canine IFNβ gene. Buccal, blood, stool and urine samples were collected to monitor virus PK, shedding, and immune response and additionally therapeutic efficacy was monitored by weekly measurement of disease burden. VSV treatment was well tolerated with all dogs developing mild, transient fevers within about 3 hours following treatment that resolved by 24 hours. One dog developed self-limiting hepatotoxicity. Overall, there was a modest effect on tumor growth: two dogs with T-cell lymphoma had transient disease remission while all others (6) has stable or progressive disease. Safety studies indicated no infectious virus was detectable in urine and buccal swab samples, but low levels of virus genomes were detectable in buccal swabs, urine and feces.

To date, a total of 183 patients have received VV1 in clinical trial participation. The most commonly reported VV1-related AEs were: CRS (48%), lymphocyte count decreased (34%), fatigue (30%), and nausea (27%). All other AEs were reported in <20% of patients. There has been one VV1 related death in 183 patients treated, a patient who developed CRS, attributed to IFNβ toxicity and subsequent tumor lysis syndrome (TLS). In this regard, IFNβ toxicity mitigation measures are included in this protocol.

In summary, clinical experience data from the five clinical trials with available data have shown acceptable VV1-related adverse events at doses up to 1.7x1011 TCID50 IV and up to 3x109 TCID50 IT. Approximately 14% of patients experienced a serious adverse reaction (SAR) and there was one fatal event (<1%). Given the potential for therapeutic benefit in patients with advanced solid tumors and TCL based on promising early efficacy data, tumor model data and a compelling mechanism of action, the safety data available at present support the continuation of the development program as monotherapy and in combination.

Potential for shedding: Safety studies in mice, dogs and pigs demonstrate that there is no infectious virus shed.

Are "Standard Precautions," Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted."

- The new HGT registration was discussed by the committee members.
- All members were in favor of approval.
- The new HGT registration is approved as submitted.

4. Haas ....**#25-179** **C–1**
Dr. Andrew R Haas – NEW HGT Protocol Registration .... **FULL REVIEW**
PROTOCOL TITLE: Phase I Clinical **Trial** of Autologous Folate Receptor-Alpha Redirected T Cells in Patients with FRa+ Cancers. (Protocol V1 dated June 12, 2025; Main ICF V1 dated June 12, 2025; Tissue Consent Script V1 dated June 18, 2025; Verbal Consent Plan V1 dated June 18, 2025; Pre- Screen ICF V1 dated June 12, 2025.)
IBC #25-179, IRB #858800, IND #14802, Sponsor #06525

- Dr. Daniel Kessler introduced the submission.
- Dr. Maureen O'Leary provided a summary and analysis.

"Phase I Clinical Trial of Autologous Folate Receptor-Alpha Redirected T Cells in Patients with FRa+ Cancers

This is a clinical trial to assess the safety, feasibility, and preliminary efficacy of intrapleural administration of MOv19-BBz CAR T cells in patients with FRa+ cancers (I will refer to it as MOV-19 CART in my review). This study will be initiated in patients with metastatic or recurrent non-small cell lung cancer only.

Subjects will receive a single dose of 5x107 MOv19-CAR T cells on Day 0
via intrapleural infusion following lymphodepleting chemotherapy. Subjects will be monitored for treatment-limiting toxicities for up to 28 days post-MOv19- CAR T cell administration.

Primary Objective:
• Evaluate the safety of intrapleural administration of MOv19-CAR T cells
Secondarily:
• Evaluate study feasibility
• Describe the preliminary efficacy of MOv19-CAR T cells

Investigational Products:
• MOv19- CAR T cells are autologous T cells lentivirally transduced with chimeric anti-alpha folate receptor immunoreceptor MOv19 scFv fused to the 4-1BB and CD3 signaling
domains. MOv19-CAR T cells will be manufactured at the University of Pennsylvania
Clinical Cell and Vaccine Production Facility.

Non-small cell lung cancer accounts for approximately 85% of all lung cancers and encompasses different histologies. Most Non small lung cancer patients present with loco-regionally advanced or metastatic disease where response rates and median overall survival remain dismal. Platinum-based chemotherapy has been the mainstay of treatment; however, over the last few years, improvements have been made in overall survival of patients with metastatic disease due to the availability of immune therapies and targeted therapies based on patients' individual molecular profile.

Minimal toxicity is expected in this study based on the restricted polarized expression of FRa . This is further supported by previous clinical safety data specifically with FRa specific immunochemotherapy.

I therefore recommend approval of this protocol.

Potential for shedding: Safety studies in mice, dogs and pigs demonstrate that there is no infectious virus shed.

Are "Standard Precautions," Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted."

- The new HGT registration was discussed by the committee members.
- All members were in favor of approval.
- The new HGT registration is approved as submitted.

5. Lynch ....**#25-196** **C–1**
Dr. David R Lynch – NEW HGT Protocol Registration .... **FULL REVIEW**
PROTOCOL TITLE: A Phase 1b First-in-Human, Open-Label, Dose-Finding Trial to Evaluate the Safety and Tolerability of SGT-212 Delivered via Dual Intradentate Nucleus (IDN) and Intravenous (IV) Administration to Participants With Friedreich's Ataxia (FA) (Protocol V3 dated March 24, 2025; Main ICF dated June 5, 2025; Screening Adult ICF dated June 5, 2025; Pregnant Partner ICF dated June 5, 2025.)
IBC #25-196, IRB #858805, IND #30949, PROTOCOL #SGT-212-101

- Dr. Daniel Kessler introduced the submission.
- Due to Dr. Sara Cherry missing the IBC meeting this submission was tabled pending a review by the Institutional Biosafety Officer, Dr. Andrew Maksymowych.
- Dr. Andrew Maksymowych provided the following summary and analysis for IBC review.

"Project Overview: Solid Biosciences Inc. (Solid) is developing SGT-212 for the treatment of Friedreich's ataxia (FA). SGT-212, also referred to as GTP-212 or rAAVhu68.CB7.CI.hFXNco.rBG, is a recombinant non-replicating adeno-associated virus serotype AAVhu68 containing a codon-optimized complementary DNA (cDNA) that encodes the human frataxin protein (FXN) under control of the hybrid chicken β-actin promoter with human cytomegalovirus immediate-early enhancer (CB7) promoter and enhancer elements, flanked by adeno-associated virus (AAV) serotype 2 (AAV2) inverted terminal repeats (ITRs). AAV serotupe-hu68 (AAVhu68) is 99% identical to AAV serotype-9 (AAV9) at the protein level. Intravenous (IV) administration of AAV9 has been shown to result in transduction of the heart, as well as to the brain, spinal cord, and dorsal root ganglia (DRG) (Foust et al., 2009; Gray et al., 2011; Samaranch et al., 2012). Furthermore, AAV9 has been shown to transduce central nervous system (CNS) tissues when delivered directly into the CNS (Hinderer et al., 2014). The AAVhu68 AAV9-based vector is, thus, capable of transduction in both the periphery and in the CNS and ideally suited for the proposed dual route of administration intended for SGT-212. The increased expression of the transgene and subsequent expression of FXN is expected to repair the underlying mitochondrial dysfunction, which is a hallmark of FA, a genetic disease of FXN deficiency.

Agent Description: SGT-212 is a recombinant non-replicating adeno-associated virus serotype hu68 (AAVhu68) containing a codon-optimized complementary deoxyribonucleic acid (cDNA) that encodes the human frataxin protein (FXN). The vector capsid exhibits broad tissue tropism for effective transduction of a wide range of host cell types, including cardiomyocytes and neurons. With broad targeting and strong expression of the transgene, administration of the drug product is expected to lead to expression of FXN in disease affected tissues. Increasing FXN expression to therapeutic levels is expected to repair the underlying mitochondrial dysfunction, which is a hallmark of FA.

Product Name: SGT-212 (rAAVhu68.CB7.CI.hFXNco.rBG, GTP-212)

Gene Inserts: Codon-optimized complementary DNA encoding human frataxin protein (FXN)

Control Element: CB7 promoter composed of a hybrid between a human cytomegalovirus immediate-early enhancer (CMV IE) enhancer and a chicken beta actin (CBA) promoter

Other Elements: Chimeric intron consisting of a chicken beta-actin splice donor and rabbit beta-globin splice acceptor elements; Polyadenylation (PolyA) signal derived from the rabbit beta-globin (rBG) gene; Two AAV2 inverted terminal repeat sequences (ITRs)

AAV Serotype: AAVhu68

Abbreviations: AAV = adeno-associated virus; CMV IE = cytomegalovirus immediate-early; hFXNco, = codon-optimized human frataxin complementary DNA, codon-optimized; ITR = inverted terminal repeats; PolyA =, polyadenylation

Is a novel vector system, approach or technology used for this clinical trial? Novel approach – YES. Novel vector system – NO.

Gene transfer agent delivery method

SGT-212 drug product is intended as a dual administration: IDN infusion using an MRI-guided device followed by an IV infusion. Suspension for infusion into Intradentate nucleus (IDN) and intravenous (IV).

Intended target: Treatment of Friedreich's ataxia (FA). FA is a serious, life-threatening, progressive multisystem disorder that is classically known to affect both cardiac and neurologic systems but also has involvement with endocrine, musculoskeletal, and other organs. The disease is due to autosomal recessive variants in the frataxin (FXN) gene.

Summary of preclinical studies: The nonclinical program was designed to support the initiation of human clinical trials in adult patients with FA and included studies evaluating the pharmacodynamics, pharmacokinetics (PK), and toxicity of SGT-212 (Table 2, IB). To establish proof-of-concept (POC) for SGT-212 in the treatment of FA, 2 non-Good Laboratory Practice (GLP) in vivo pharmacology studies were performed in in well-characterized cardiac and neuronal Fxn conditional knock out mouse models of FA (Fxn cardiac conditional knockout [cKO] and neuronal conditional knockout [nKO]). Both the cardiac conditional knock out mouse model (Jackson Laboratory Stock #029720) and neurologic conditional knock out mouse model (Jackson Laboratory Stock #029721)(Piguet et al., 2018) used are well-characterized models of FA. The POC studies included evaluations of transgene expression and efficacy. The non GLP studies were performed in laboratories with expertise in the FA mouse models and/or in performing functional efficacy measurements and were conducted according to a prospectively written protocol in an unbiased manner and included measures to ensure data quality and integrity.

SGT-212 has been engineered to enable expression of a functional FXN protein in relevant tissues primarily affected in patients with FA including the heart and dentate nuclei. A single, dual IV/IDN administration of SGT-212 in non-human primates (NHPs) at doses up to 3.0 x 1012 GC/kg IV and 4.0 x 1010 genome copies/DN (GC/DN) IDN up to 365 days post-administration was well-tolerated with no adverse toxicities. In 2 mouse models of FA, treatment with SGT-212 led to the production of detectable levels of human FXN messenger ribonucleic acid (mRNA) that corresponded with reduced disease pathology and improvements in cardiac and neuromuscular function. NHP studies were performed in adult animals at an age range correlating with the proposed clinical study population and utilizing the same IDN delivery device and components planned for the first-in-human (FIH) clinical study.

Results from the nonclinical program establish that SGT-212 leads to the production of a functional FXN protein and prevention of functional consequences of disease, and the NHP toxicity study establishes no-observed-adverse-effect level (NOAEL) doses for both routes of administration. The nonclinical program establishes that SGT-212 shows the probability of benefit with an acceptable safety profile. The nonclinical program supports the clinical evaluation of SGT-212 in adult patients with FA at the proposed first in human clinical dose, given the high unmet need for effective therapeutic interventions that can target both the cardiac and neurological manifestations of disease.

Potential for shedding: SGT-212 has not yet been administered to humans; therefore, the safety profile has not been characterized.

The vector was detectable in urine and feces of all SGT-212-treated animals with peak levels on Day 5. Vector shedding was dose-dependent and undetectable by Day 150. Test article-related neutralizing antibody (Nab) response was seen in all dosed animals with peak NAb titers at Day 28 for the short-term cohort and Day 90 for the long-term cohort. Broad distribution of vector genomes was observed by qPCR with highest levels in the liver, followed by spleen and heart. Vector genomes and/or frataxin expression was detected in the spinal cord (SC), DRG, and DN (NHPs) demonstrating sufficient delivery of SGT-212 to neuronal tissue which is important for the treatment of FA.

Are “Standard Precautions,” Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES.

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted.”

- The new HGT registration was discussed by the committee members.
- All members were in favor of approval.
- The new HGT registration is approved as submitted.

6. Owens ....#**25-199** **C-1**
Dr. Anjali T Owens – NEW HGT Protocol Registration .... **FULL REVIEW**
PROTOCOL TITLE: A Phase 1/2, Open-Label, Multicenter, Dose Finding and Dose Expansion Study to Investigate the Safety, Tolerability, and Efficacy of ALXN2350 Gene Therapy in Adult Participants with BAG3 Mutation Associated Dilated Cardiomyopathy. (Protocol V1 dated March 3, 2025; Main ICF dated June 2, 2025.)
IBC #25-199, IRB #pending, IND #031051, Protocol #ALXN2350-DCM-201

- Dr. Daniel Kessler introduced the submission.
- Dr. Joseph Fraietta provided a summary and analysis.

“A Phase 1/2, Open-Label, Multicenter, Dose Finding and Dose Expansion Study to Investigatethe Safety, Tolerability, and Efficacy of ALXN2350 Gene Therapy in Adult Participants withBAG3 Mutation Associated Dilated Cardiomyopathy

Detailed Project Description: Mutations in BAG3 have been associated with a highly penetrant DCM in humans (80% in genotype-positive patients > 40 years of age) (Dominguez, 2018). ALXN2350 is being developed with the intent to be a one-time disease-modifying gene therapy for the treatment of patients with DCM who have confirmed pathogenic or likely pathogenic BAG3 mutations. This study aims to explore safety, tolerability, and efficacy across the dose ranges starting from the anticipated minimally effective dose to the top dose. ALXN2350 is being developed for use in addition to SoC.

Project Overview: This Phase 1/2, open-label, multicenter in vivo gene-therapy study evaluates ALXN2350, an AAV9-based BAG3 transgene, delivered by single IV infusion in adults with genetically confirmed BAG3-associated dilated cardiomyopathy (DCM). Part A (dose escalation; 2–6 × 10^{13} vg/kg cohorts) assesses safety and tolerability, and Part B (dose expansion at the selected dose) evaluates preliminary efficacy versus external control with endpoints including change from baseline in VO_2 peak, LVEF, and patient-reported outcomes through Week 78.

Agent Description: Recombinant, replication-deficient AAV9 with a cardiac-troponin-T (cTNT) promoter driving human BAG3 expression in cardiomyocytes. Standard AAV9 design (no helper virus genes; packaged in a cell line via triple transfection). Full-length human BAG3 coding sequence under cTNT promoter; no truncations or engineered mutations.

Is a novel vector system, approach or technology used for this clinical trial? NO

Gene transfer agent delivery method: Single IV infusion (2–6 × 10^{13} vg/kg) over 1–2 hours on Day 1.

Intended target: In vivo cardiomyocytes; ≥ 20% cardiomyocyte transduction and ≥ 14% normal BAG3 protein expression required for functional benefit.

Any other material to be used in preparation of the agent (vector and transgene) to be administered to research participants (e.g., helper virus, packaging cell line, carrier particles): No helper virus or carrier particles. Vector production via HEK293 packaging cell line; purified by affinity chromatography.

Summary of preclinical studies conducted in support of the proposed clinical trial: Bag3 knockout mice demonstrated dose-responsive stabilization of cardiac structure/function when ≥ 20% of cardiomyocytes were transduced and ≥ 14% normal BAG3 protein was expressed.

Non-human primate (GLP toxicology): Defined a NOAEL with immunosuppression (methylprednisolone + sirolimus ± rituximab), supporting safety margins at clinical dose levels.

Potential for shedding: Yes; monitoring of vector DNA in whole blood, saliva, urine, feces, and semen until 3 consecutive negatives.

Are "Standard Precautions," Biosafety Level 2 (BSL-2) equivalent, practices appropriate for ensuring clinical trials personnel safety? YES

This Registration Meets IBC Criteria for Approval: YES. I recommend approval, as submitted."

- The new HGT registration was discussed by the committee members.
- All members were in favor of approval.
- The new HGT registration is approved as submitted.

<u>HGT Administrative Actions:</u> #32

<u>Research Administrative Actions:</u> #70

SECTION III–D. Experiments that Require IBC Approval <u>Before</u> Initiation:

7. Asangani ....25-147 .... **D-1,3,4**
8. Asangani ....25-159 .... **D-1,4 O-1**

- The registrations were prepared by Dr. Sarah Capasso and presented by Ms. Amanda Wong. The registrations are for the generation and/ or use retroviral vectors with an amphotropic envelope (25-147) and 3rd generation lentiviral vectors expressing CRISPR/Cas9 (25-159). Retroviral vectors with an amphotropic envelope are used to overexpress epigenetic regulators in a human prostate cancer cell line. Modified cells will be administered to mice to generate in vivo xenograft models of prostate cancer. These models will allow the lab to determine the role of epigenetic regulators in prostate cancer and to develop novel treatment strategies. 3rd generation lentiviral vectors expressing CRISPR/Cas9 components are used to target expression of epigenetic regulators or shRNA targeting genes of interest in human and murine cancer cells. Luciferase or GFP will be used as reporter genes. Modified cells will be administered to mice to generate in vivo xenograft models of prostate cancer. This will allow the lab to study epigenetic dysregulation and tumor recurrence as well as to develop novel treatment strategies. Containment for both registrations has been set to BSL-2 and ABSL-2.
- The registrations were discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registrations, 25-147 and 25-159 were approved.

9. Brodsky..........25-183.................. **D-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/ or use of mutant Shigella flexneri strains. The lab will infect murine macrophages or human cell lines with these modified microorganisms to examine the response of the murine or human host cells' ability to mount inflammasome responses, release cytokines, and engage in cell death. Modifications are made to regulatory enzymes that orchestrate lipid A modification. The modification of Lipid A can change the host's ability to sense the pathogen and activate inflammasomes and cell death pathways. The research does not enhance the transmissibility, virulence, toxicity, stability, host range, diagnostic evasion, treatment resistance, immune evasion, or host susceptibility of the pathogen and/or toxin. Containment has been set to BSL-2.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved.

10. Brown ....24-369.................. **D-1,3**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of 3rd generation lentiviral vectors with CRISPR/ Cas9 in murine and human cell lines. The lab is investigating DNA damage response pathways which play critical function in maintaining genomic stability and cell survival. They identified high-priority genes such as WRN, ARID1A, MTAP, SLX4 using iPOND (Isolation of Proteins on Nascent DNA; this is a biochemical method used to study protein dynamics at DNA replication forks and in response to DNA damage. These identified genes will be knock out using a lentiviral vector expressing CRISPR elements and GFP. Containment has been set to BSL-2.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

11. Bugaj.............24-255.................. **D-1,3,4 O-1**
12. Bugaj.............25-198.................. **D-1,3,4 O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registrations. The registrations are for the generation and/or use of 2nd generation lentiviral vectors with CRISPR/ Cas9 (25-198) and retroviral vectors with amphotropic envelopes (24-255). The lentiviral vectors are from Dr. Wendell Lim at UCSF, and the lab has plans to transfer their work into the 3rd generation Lentivirus system. The retroviral vectors are subcloned from a CLPIT vector obtained from Dr. David Schaffer at UC Berkeley. The lab develops molecular probes for external control of mammalian cells, with the goal of understanding the logic of mammalian cells and signaling breakdown in cancer. Those probes respond to either light or temperature, which are of plant or fungal origin. In other work, they generate fusions of temperature-sensitive proteins to effectors of cell death pathways (caspase1/4/5/8/9), conferring thermal control of cell death, or use CRISPR to add a fluorescence tag to G3BP1, a key protein involved in the formation and dynamics of stress granules, in cell culture. In some experiments modified cells will be introduced into mice for xenograft studies. The lab transduces a variety of cells including human and mouse cells and cancer cells. These are sourced from either ATCC or collaborating labs. Containment for both registrations has been set to BSL-2 and ABSL-2.

- The registrations were discussed by committee members. Dr. Daniel Kessler requested “LNP” be removed from both project descriptions since it is listed in a separate registration.
- Training was complete.
- All members were in favor of approval.
- The IBC registrations, 24-255 and 25-198, were approved pending update to project description

13. Bugaj.............25-211.................. **D-1,4 O-2**

- This registration was prepared by Ms. Marie- Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and/or use of LNP-mRNA. The lab develops proteins for external or autonomous control of mammalian cells. The lab is looking to manipulate cancer cells for basic studies and for therapy. Engineered proteins will be delivered via an mRNA construct in lipid nanoparticles to cancer cells, both in vitro and in vivo. These proteins will activate specific pathways of cell death in the cancer when triggered by external or autonomous thermal stimuli.
- Additionally, 'smart' proteins that sense oncogenic states will be delivered inside cancer cells and execute cell death programs in response. For in vivo studies these proteins are given either intravenously or intratumorally. All LNPs will be made by the UPenn LNP core facility. Containment has been set to BSL-2 and ABSL-1.
- The registration was discussed by committee members. Dr. Daniel Kessler requested a more descriptive title.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved pending update to title.

14. Busino...........25-185.................. **D-1,4 O-1**

- This registration was prepared by Ms. Marie - Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and/or use of 2nd and 3rd generation lentiviral vectors with CRISPR/ Cas9. The lab is studying the mechanism behind the alteration of protein degradation through the ubiquitin proteasome system (UPS) caused by hematological disease, such as lymphoma and leukemia. The lab will study Cullin-RING-Ubiquin Ligases in the context of hematologic malignancies with the aim of characterizing the role of these proteins. Using CRISPR lentiviral vectors to knock out ligases in human cell lines. The modified cells lines will then be subcutaneously injected into NSG and C57Bl/6 mice. Containment has been set to BSL-2 and ABSL-2.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

15. Chatterjee.......25-167.................. **D-1,3**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of 3rd generation lentiviral vectors acquired from Addgene. The lab is developing precise protein binders for biologics and targeted protein degradation (TPD) therapies mainly for Ewing sarcoma, Alexander disease and for the oncoprotein Beta-catenin. They established a human cell-based screening platform using genetically encoded, doxycycline-

inducible ubiquibodies (uAbs). These uAbs fuse computationally designed peptide binders to an E3 ubiquitin ligase domain for targeted protein degradation in mammalian cells, enabling rapid and physiologically relevant screening of AI-designed peptides. Lentiviral vectors will be used to deliver and express peptide binders. Peptide libraries (2000 peptides total per target) are computationally designed using PepMLM and moPPIt algorithms developed in-house and synthesized by Twist Bioscience. All peptide binders are BLASTed to confirm they are not found in nature. The lentiviral vector is also used to transduce HEK293T to generate stable cell lines expressing target-mCherry fusion proteins for screening. Containment has been set to BSL2.

- The registration was discussed by committee members. Ms. Dorothy Kaplan asked what an AI designed peptide is. Ms. Stephanie Adams-Tzivelekidis clarified that the program lets AI synthesize the peptide using crystallized structures.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

16. Dowling ....25-116 .... **D-4**

- This registration was prepared by Ms. Marie- Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and/or use of adeno- associated viral vectors. The Lab is studying multiple neuromuscular and neurogenic disorders and looking to use AAV to deliver gene replacement therapy in a mouse disease model. They will introduce expressed genes involved in various diseases, including PURA-syndrome (AAV9-PURA), X-linked myotubular myopathy (PikC32b and associated miRNA miR-81), Nemaline Myopathy (Nebulin), and Duchenne muscular dystrophy (DMD). The AAV vectors will either be made in the lab or transferred from other collaborating labs. AAV particles will be introduced into mice by either IV or IM injections. Containment has been set to BSL-1 and ABSL-1.
- The registration was discussed by committee members. Dr. Daniel Kessler requested a more descriptive title.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved pending update to title.

17. Edwards ....25-201 .... **D-4**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of adeno- associated viral vectors. AAV2/9 will be expressing CRISPR elements to knock-down or overexpress specific structural proteins in mice. AAVs serotype 2/9 was purchased from SignaGen and Vector Biolabs. The vector is directly injected into mice retro-orbitally or IP (interperitoneally). The lab aims to unravel the molecular underpinnings of Right Ventricular Heart Failure (RVF) to identify novel mechanism-driven therapeutic targets. Areas of focus include noncanonical WNT signaling in proteostasis, regulators of cardiomyocyte contractile/relaxation function, remodeling of cardiomyocytes, and metabolic regulation. Containment has been set to BSL-1 and ABSL-1.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.

- The IBC registration was approved

18. Gao................24-296.................. **D-1 O-1**

- This registration was prepared by Ms. Marie-Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and/or use of 3rd generation lentiviral vectors with CRISPR/ Cas9. The lab will use the lentiviral vectors with CRISPR technology to delete various genes/regions to study therapeutic genome editing of sickle cell disease and beta thalassemia. Additionally, they will use the lentivector to create disease model cell lines by integrating sections of DNA harboring a pathogenic mutation for subsequent therapeutic editing. This will be done by performing PCR on genomic DNA near the gene of interest, generating a DNA fragment before the desired mutation and a second fragment after the desired mutation. Containment has been set to BSL-2.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

19. Jensen............25-206.................. **D-4**

- This registration was prepared by Ms. Marie- Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and/or use of adeno- associated viral vectors. The lab aims to find therapeutic treatments for CDKL5 Deficiency Disorder (CDD). Cdkl5(R59X) neonatal mice will be treated with the AAV-9 vector developed by the GTP program. The cassette consists of a human synapsin or Ubiquitin C promoter, the human CDKL5 coding sequence, a WPRE element and an SV40 polyA sequence. About 3 months after injection, mice will be subjected to a neurobehavior tests to assess treatment outcome, followed by molecular and histological tissue analysis. A second aim of the study is to determine if reducing neuronal activity by expressing inhibitory designer receptors exclusively activated by designer drugs (DREADDs) affects disease progression in mouse models of epilepsy and Alzheimer’s disease. They will use FosTRAP and 5XFAD mice to inject intracerebrally with an AAV vector coding for an inhibitory receptor, which will be followed by both seizure producing and then inhibiting treatment to assess the outcome post gene therapy treatment. Containment has been set to BSL-1 and ABSL-1.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

20. Karni ....25-157.................. ~~D-3,~~ O-1, corrected to **D-4 O-1**

- This registration was prepared by Ms. Marie- Luise Faber and presented by Ms. Amanda Wong. This registration is for the generation and or use of modified cancer cells with CRISPR/ Cas9. The lab will focus on alternative splicing and how it is misregulated in cancer cells and whether it can be revered through therapies. The lab will look at the manipulation of RNA express effects in cancer cell growth. They will use human and murine cancer cells engineered to express altered levels of RNA splicing regulators (such as hnRNPH1) or mispliced products (such as Tma7) via overexpression vectors. The modified cells were made at Dr. Karni's lab in Hebrew University of Jerusalem. The cells will be analyzed in vitro and implanted into flanks of NOD-SCID, BALB/c,

or C57BL/6 mice either subcutaneously or orthotopically to induce tumor growth and test for potential therapeutics. Containment has been set to BSL-2 and ABSL-2.

- The registration was discussed by committee members. Dr. Tucker Piergallini asked for clarification on NIH guidelines since D-3 was selected and connected IACUC was assigned and ABSL-1 containment. Dr. Daniel Kessler stated BSL-2 and ABSL-2 are correct based on provided information. Ms. Stephanie Adams-Tzivelekidis stated that since this is an in vivo study it should be listed as D-4 and O-1.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved pending update to NIH guidelines selection.

21. Lengner..........25-189.................. **D-4**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of retrovirally modified cells. The lab is studying the cellular and molecular basis of resistance of colorectal cancers to immune checkpoint inhibitors, with a focus on the immunosuppressive role of stromal cells and the extracellular matrix (ECM) they generate. The cancer associated fibroblasts express the cell surface molecule FAP, which is not expressed in normal tissue. The lab will use FAP-CAR T cells pioneered by the Puré lab to treat mouse models of colorectal cancer using ecotropic retroviral vectors to transduce mouse t cells in vitro. These cells will be modified to express chimeric antigen receptors (CARs) targeting antigens of interest (i.e. mesothelin control or FAP), along with fluorescent reporters. Modified cells are administered to mice to assess their ability to recognize and kill cells expressing the antigen of interest testing whether these CAR Ts have efficacy against colorectal cancer, and whether combination with anti-PD1 antibodies (checkpoint inhibitor) increases efficacy. Containment has been set to BSL-1 and ABSL-1.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

22. Moncla..........25-169.................. **D-1,3,7**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of modified influenza virus. The lab will generate a recombinant, replication-deficient Influenza A virus by reverse genetics in a co-culture of 293T and MDCK cells that stably express PB1 (an influenza polymerase subunit). These viruses will also contain GFP in place of their PB1 gene segment, rendering them unable to replicate outside of these cell lines. The lab will generate viruses that express the HA and NA glycoproteins of human seasonal viruses (namely the H1N1 A/California/07/2009) with internal genes (except PB1) from the A/Puerto Rico/8/1934 strain to assess inhibitory properties of polymers containing sialic acid. The lab hypothesizes that such polymers would interfere with viral infection by competing with cellular receptors (entryway) and inhibit the growth of influenza virus in vitro, so these polymers may act as antivirals. To investigate this, the viruses will be propagated in the presence of near-inhibitory concentrations of polymer to determine if influenza viruses can mutate to escape inhibition. Viral stocks will be sequenced after each propagation step to determine the presence of such mutations. This will allow us to determine if it is possible for influenza viruses to escape these polymers and, if so, what evolutionary mechanism(s) they use for escape. The Lab

anticipates that these mutants would be less infectious, as they will likely need to compromise some aspect of receptor binding for escape. In the case of HA mutants, the lab expects mutants that bind less strongly to sialic acid and thus are less inhibited by the polymer. Containment has been set to BSL-2.

- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

23. Naji ....25-172 .... **D-4**

- Dr. Tucker Piergallini presented the registration. This registration is for the generation and/or use of mRNA-LNPs. Lipid nanoparticle (LNP)–delivered mRNAs encoding immune-regulatory proteins (such as CD47, IL-10, TGF-β, and PD-L1) to modulate immune responses and promote immune tolerance. mRNA-LNPs will be administered to mice. This approach is designed to induce antigen-specific and tissue-localized immune tolerance, with the goal of both promoting long-term transplant acceptance and preventing or treating autoimmune diseases such as type 1 diabetes by protecting pancreatic β cells from immune-mediated destruction. The mRNA-LNPs will be obtained from the laboratories of Dr. Drew Weissman and Dr. Hamideh Parhiz. Containment has been set to BSL-2 and ABSL-2 for human cells; BSL-1 and ABSL-1 for LNP injection and mouse cells.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

24. Scott ....25-186 .... **D-1**

- This registration was prepared by Ms. Marie- Luise Faber and presented by Ms. Amanda Wong. This registration is for the storage of Modified Leishmania. The Leishmania was altered to express mCherry to track infectious and 2WS1 to track the t cell response. Work with Leishmaniasis in animals has ended in the Scott Lab. Reference to the parasite has been removed from the IACUC protocol. The vector will remain in storage for the foreseeable future. Containment has been set to BSL-2.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

25. Sundstrom......25-204 .... **D-4 O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of adeno- associated viral vectors with the modification of cells using CRISPR. The lab aims to understand the mechanisms of vision loss in various retinal dystrophies, including age-related macular degeneration (AMD), and identify new therapies for patients with AMD. This project focuses on examining the factors that drive AMD progression. The rsNA material will be acquired from Addgene and Dr. Quinn (U. Penn). AAV, expressing the RPE65

gene, will be directly injected in vivo subretinal or intravitreally. Additionally, ARPE and iPSC-derived (retinal pigment epithelial cells) RPE cells will be transduced with AAV. Modified ARPE cells or Extracellular Vesicles (EV) extracted from iPSC cells will also be injected in vivo. Containment has been set to to BSL-2, ABSL-1 for direct injection of AAV, and ABSL-2 for injection of modified human ARPE cells.

- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

26. Wherry ....24-085 .... **D-1,4**
27. Wherry ....24-225 .... **D-1,4**

- The registrations were prepared by Dr. Sarah Capasso and presented by Ms. Amanda Wong. These registration are for the generation and/or use of Vaccinia viruses expressing epitopes GP33, GP61 and OVA to track and study immune responses in mice after administration (24-085) and of recombinant Vesicular Stomatitis Virus (rVSV), modified to express known T cell epitopes, which will facilitate tracking and studying of specific immune responses when administered to mice or studied in cell culture (24-255). The use of vaccina viruses will allow the lab to study the factors that influence memory T cell quality and ultimately improve vaccination and immunotherapeutic strategies aimed at improving immunity to infectious agents. Recombinant Vaccinia viruses were obtained from the lab of Dr. J. LINDSAY WHITTON (Department of Neuropharmacology, CVN-9, The Scripps Research Institute, La Jolla, California). Use of rVSV will allow the lab to study the role of co-stimulatory and co-inhibitory pathways with respect to the development of antiviral immunity. Recombinant Vesicular Stomatitis Viruses were donated by Dr. David Masopust (Center for Immunology, Department of Microbiology, University of Minnesota). Containment for both registrations has been set to BSL-2 and ABSL-2.
- The registrations were discussed by committee members. Mr. Edwin Siu asked if the USDA permits were listed on the registrations for the Vaccina virus. Dr. Andrew Maksymowych stated they should be listed on the MTA agreement, but it is not required when registering with the IBC. Dr. Daniel Kessler asked if this is something we should start reviewing. Dr. Andrew Maksymowych stated IBC reviews the safety of the work and not the transportation of material.
- Training was complete.
- All members were in favor of approval.
- The IBC registrations, 24-085 and 24-225, were approved.

SECTION III–O. Experiments that Require IBC Approval Before Initiation:
28. Seykora ....25-151 .... **E-3 O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation of transgenic animals using CRISPR. The lab studies the etiology of UV-induced skin cancer and mechanisms of wound healing in skin and the cornea. They are generating a floxed allele of the P2RX7 and Srcasm gene using CRISPR technology through the gene targeting core at Penn. They will cross this mouse harboring the floxed allele to mice expressing Cre recombinase to generate deletion of the P2RX7 or Srcasm genes in tissues, then study the impact of loss of this gene on wound healing. Containment has been set to BSL1 and ABSL-1.

- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved.

29. Susztak...........25-200.................. **E-3 O-1**

- Mr. Edwin Siu presented the registration. This registration is for the generation and/or use of guide RNA and CRISP/Cas9. The lab investigates how non-coding genetic variants influence predisposition for later-onset kidney diseases such as diabetic and hypertensive kidney disease. The lab has 27 putative kidney disease risk genes identified from GWAS or eQTL and will generate knock-outs of these genes in mice using CRISPR/Cas9. The lab plans to examine how these genetic variants affect the mouse kidney phenotype, both under normal conditions and after kidney injury. Guide RNA for targeting the genes will be synthesized by a commercial vendor and submitted to the Penn Transgenic mouse core for microinjection to generate the CRISPR KO mice. Containment has been set to BSL-1 and ABSL-1.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved.

SECTION III–E. Experiments that Require IBC Notice <u>Simultaneous with</u> Initiation:

30. Cherry ....25-194.................. **E-1**

- Dr. Tucker Piergallini presented the registration. This registration is for the generation and/or use of plasmids. The lab will test if drugs inhibit the Nipah and Cedar virus polymerase, allowing them to potentially identify drugs active against these diseases. Live Nipah virus requires BSL-4 containment, so the lab will use the tetracistronic minigenomes (plasmid) that does not use the live virus. The lab will also study Cedar virus. In brief, this system has 4 vectors. The minigenome that is expressed from T7 polymerase that has the signals used for the viral polymerase to amplify to detect a reporter. The proteins required to amplify this RNA are supplied in trans. This includes the N, P and L protein to replicate the minigenome. These are produced from a CMV promoter. The system is obtained from Dr. Lee (found in publication attached in PIERS). The plasmids will be transfected into Hamster BHK cells to study the activity of antivirals. Nipah virus will not be reconstituted, and infectious Nipah virus will not be produced or used. They will not show active drugs against live virus, only the polymerase. Containment has been set to BSL-2.
- The registration was discussed by committee members. Dr. Daniel Kessler requested that project description be cleaned up. Ms. Stephanie Adams-Tzivelekidis asked why this is E-1 and not D-1. Dr. Tucker Piergallini clarified that this is not a vector system but a plasmid expressing a protein.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved pending update to project description.

3. New Business:

(a) No New Business Scheduled.

4. Old Business:

(a) Using the 'new' HGT Summary Form. (Andrew)

5. End Meeting:

- The Institutional Biosafety Committee Meeting was adjourned by Dr. Daniel Kessler at **11:21 AM**.

Our next meeting scheduled for Monday, August 25th, 2025, will be held on site at the EHRS Office with a Teleconference option, at 10:00 am. A light Brunch will be provided.