Institutional Biosafety Committee:

Present: Dr. Steven Albelda, Dr. Julian Baptiste, Dr. Jessica Buchanan, Dr. Joseph Fraietta, Dr. Daniel Kessler, Dr. Andrew Maksymowych, Dr. Maureen O'Leary, Ms. Jessa Yoos

Absent: Dr. Paul Bates, Dr. Sara Cherry, Dorothy Kaplan, , Dr. David Pegues, Ms. Denene Wambach

Invited Guests: Ms. Stephanie Adams, Dr. Sarah Capasso, Dr. Tucker Piergallini, Ms. Amanda Wong, Ms. Kimberly Craig, Dr. Clayton Otter

The Institutional Biosafety Committee Meeting was called to order by Dr. Daniel Kessler at **10:02 AM**.

1. IBC Minutes: **08-25-2025**

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

2. Registrations for Review:

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

1. Bagley ..... **#25-256** **C-1**

Dr. Stephen Bagley– NEW HGT Protocol Registration ..... **FULL REVIEW**

PROTOCOL TITLE: Phase 1b, Open-Label Study of CART-EGFR-IL13Rα2 Cells Administered with Lymphodepleting Chemotherapy or Prior to Surgical Resection in Patients with EGFR-Amplified Recurrent Glioblastoma. (Protocol V1 dated September 9, 2025; Main ICF dated September 2, 2025; Apheresis ICF dated September 2, 2025; Retreatment ICF dated September 2, 2025.)

IBC #25-256, IRB # 850297, IND # 28055, Sponsor# 10325, NCT# Pending

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

"**Project Overview:** This is an open-label, phase 1b study to evaluate different approaches for CART-EGFR-IL13Ra2 dosing and further characterize the safety, feasibility, preliminary efficacy, and pharmacokinetics of CART-EGFR-IL13Ra2 cells in patients with EGFR-amplified glioblastoma that has recurred following prior radiotherapy. CART-EGFR-IL13Ra2 cells are autologous T cells co-expressing two CARs targeting the cryptic EGFR epitope 806 and IL13Ra2. CART-EGFR-IL13Ra2 cells will be administered following lymphodepletion (Arms A and B) or prior to surgical resection of the tumor (Arm C).

This study will evaluate three different approaches for administering CART-EGFR-IL13Ra2 cells in the setting of recurrent glioblastoma. Each dosing approach will be evaluated as a separate treatment arm as outlined below:
• Arm A: Single Fixed-Dose Administration Following Lymphodepletion
• Arm B: Repeat Dose Administration Following Lymphodepletion
• Arm C: Single Fixed-Dose Administration in the Pre-Operative Setting

Subjects will be assigned to each treatment arm sequentially according to their planned treatment date, starting with Arm A. Once Arm A is fully enrolled and all required safety evaluations have been completed, the Clinical PI and Sponsor Medical Director will review the cumulative Arm A data and approve the opening of Arm B.

• Arm A (Single Fixed-Dose Administration Following Lymphodepletion):
Up to 4 evaluable subjects will receive a single fixed dose of CART-EGFR-IL13Ra2 cells via intracerebroventricular delivery, following their routine care surgical intervention and lymphodepletion with fludarabine and cyclophosphamide. All subjects will be treated at
Dose Level 1 (2.5x107 CART-EGFR-IL13Ra2 cells) and monitored for treatment-limiting toxicities (TLTs) for up to 28 days post-CART-EGFR-IL13Ra2 cell administration.

• Arm B (Repeat Dose Administration Following Lymphodepletion):
Up to 4 evaluable subjects will receive a single dose of CART-EGFR-IL13Ra2 cells via intracerebroventricular delivery, following their routine care surgical intervention and lymphodepletion with fludarabine and cyclophosphamide. Subjects will then receive a
second dose of CART-EGFR-IL13Ra2 cells on Day 14 (+/-1d), without additional lymphodepletion. Both doses will be administered via intracerebroventricular administration. All subjects will be treated at Dose Level 1 (2.5x107 CART-EGFR-IL13Ra2
cells), unless the CART-EGFR-IL13Ra2 dose used in Arm A was de-escalated.

• Arm C (Single Fixed-Dose Administration in the Pre-Operative Setting):
Up to 4 evaluable subjects will receive a single fixed-dose of CART-EGFR-IL13Ra2 cells via intracerebroventricular delivery, prior to their routine care surgical intervention. All Arm C participants will be treated at Dose Level 1 (2.5x107 CART-EGFR-IL13Ra2 cells) without lymphodepleting chemotherapy.

Retreatment
Subjects who demonstrate clinical benefit after initial treatment with CART-EGFR-IL13Ra2 cells may also be eligible to receive retreatment with CART-EGFR-IL13Ra2 cells at the physician-investigator's discretion.

Agent Description: Autologous CART-EGFR-IL13Ra2

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

Gene transfer agent delivery method: Intracerebroventricular Delivery. ICV injection is a route of administration for drugs via injection into the cerebral ventricles so that it reaches the cerebrospinal fluid (CSF).

Intended target: Tumor. Glioblastoma

Other material to be used in preparation of the agent: N/A

Preclinical studies: N/A

Potential for shedding: N/A

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.

2. Bivalacqua..... **#25-226** **C-1**
Dr. Trinity Bivalacqua – HGT Protocol Registration Amendment V6.....**AMENDMENT**
PROTOCOL TITLE: Phase 3b, Randomized, Controlled Trial of Nadofaragene Firadenovec vs. Observation in Subjects with Intermediate Risk (IR) Non-Muscle Invasive Bladder Cancer (NMIBC). (Protocol V6 dated June 16, 2025; Main ICF dated April 9, 2025.)
IBC #25-226, IRB #857776, IND #12547, Protocol #ABLE-32-000423

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

"**Project Overview:** Nadofaragene firadenovec is a replication-deficient recombinant type 5 adenovirus vector containing the human interferon-alpha2b (IFN- α2b) gene developed for the treatment of adult patients with high-risk Bacillus Calmette-Guérin (BCG)-unresponsive non-muscle invasive bladder cancer with carcinoma in situ with or without papillary tumors.

Nadofaragene firadenovec is instilled quarterly into the bladder by intravesical instillation via an indwelling urinary catheter to deliver the IFN-α2b gene into the urothelial lining of the bladder to expose tumor cells to high concentrations of IFN-α2b protein, to limit systemic exposure, and to minimize dosing frequency. Upon instillation, the bladder epithelium is expected to synthesize and secrete sustained local concentrations of IFN-α2b protein. High local concentrations of IFN-α2b protein and increased duration of exposure are expected to potentiate durable therapeutic responses.

Nadofaragene firadenovec was approved by the US FDA in 2022 as ADSTILADRIN® for the treatment of high-risk BCG-unresponsive non-muscle invasive bladder cancer with carcinoma in situ with or without papillary tumors. However, no therapies are approved by the FDA for the maintenance treatment of intermediate risk non-muscle invasive bladder cancer (IR NMIBC). Instead, the recommended treatment for subjects with intermediate risk non-muscle invasive bladder cancer includes tumor resection, with or without peri-operative chemotherapy followed by

consideration for adjuvant intravesical chemotherapy or immunotherapy and periodic surveillance for recurrence. Approximately 50% of subjects with IR NMIBC will experience recurrence, and there is an unmet clinical need for this at-risk population.

Up to 454 subjects to be randomized 1:1 into treatment and observation groups with 120 North America and international study sites.

Primary Objective: To evaluate the efficacy of nadofaragene firadenovec administered every 3 months vs. observation in subjects with IR NMIBC. Primary outcome is the difference in time to recurrence, progression, or all cause death estimated by hazard ratio.

Secondary Objective: To evaluate the safety of nadofaragene firadenovec vs. observation in subjects with IR NMIBC. There are 3 additional exploratory clinical and quality of life objectives.

Agent Description: The investigational medicinal product (IMP) nadofaragene firadenovec is a replication-deficient recombinant type 5 adenovirus vector containing the IFN-α2b gene and the excipient Syn3NODA that enhances gene transfer across the urothelium.

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

Gene transfer agent delivery method: 75 mL (3×10^11 vp/mL) of sterile suspension instilled quarterly into the bladder via a urinary catheter. Bladder dwell time is 1 hour. The duration of treatment is 24 months in the absence of NMIBC disease recurrence, progression, or death.

Intended target: Bladder urothelial cells.

Other material to be used in preparation of the agent: N/A

Preclinical studies: Adverse reactions (>10%) in patients with NMIBC in the P3 trial (per the FDA label) include: instillation site discharge, fatigue, chills, fever and urinary AEs (bladder spasm, urgency, hematuria and dysuria). There is no information regarding safety in pregnancy and male and female subjects will be advised to use effective contraception during treatment.

Potential for shedding: IFN-α2b is delivered via a replication incompetent adenovirus vector. Study protocol and FDA label note “possible presence of low levels of replication-competent adenovirus in nadofaragene firadenovec.” States: Individuals who are immunosuppressed or immune-deficient should not come into contact with nadofaragene firadenovec.

Protocol notes that potential transient and low-level shedding of nadofaragenefi radenovec may occur in urine, potentially leading to transmission to third party.

FDA label states: ADSTILADRIN biodistribution and shedding were investigated in two clinical studies. Only a single patient receiving a second dose in one study (Phase 2 study) at dose level of 3 x 10^11 vp/mL had measurable vector DNA in blood. In urine, measurable vector DNA was detected in both studies. Generally, a higher frequency of detection of urine samples positive for vector-derived DNA, and persistence of vector-derived DNA correlated with increasing dose level. At the dose level of 3 x 10^11 vp/mL, 16 subjects (out of 19 at visit) had detectable levels of vector DNA at Day 12 in the Phase 2 study.

As a precaution, the ICF instructs subjects “to add about half a cup of bleach to the toilet bowl before you urinate. After urinating, wait 15 minutes before flushing the toilet. Repeat these steps every time you urinate for the first 2 days after treatment.”

There are no new biosafety concerns in protocol amendment v6.

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. Chapin ..... #**23-153 ..... C–1**

Dr. William J Chapin– HGT Protocol Registration Amendment V6 ..... **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 V6 dated June 27, 2025; Main ICF dated August 8, 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.

  "**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 add an additional patient group-biliary tract cancer who would be treated with RP2 and durvalumab (anti-PDL1 antibody) along with some small administrative changes.

  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 or biliary tract cancer carcinoma cells

  Other material to be used in preparation of the agent: 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

  Preclinical studies: 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.

  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.

4. Eads ..... #**24-153**……**C–1**

Dr. Jennifer R Eads– HGT Protocol Registration Amendment V2 ..... **AMENDMENT**

PROTOCOL TITLE: UPCC 06224: A Phase 1/2 Study to Evaluate CHM-2101, an Autologous Cadherin 17(CDH17) Chimeric Antigen Receptor (CAR) T Cell Therapy for the Treatment of Relapsed or Refractory Gastrointestinal Cancers. (Protocol V1.3 dated January 10, 2025; Protocol V1.4 dated February 18, 2023; Main ICF V1 dated March 31, 2025.)

IBC #24-153, IRB #856105, IND #029359, STUDY #CHM-2101-001

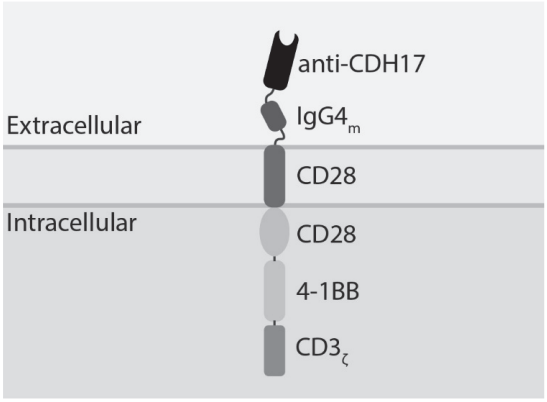
- Dr. Daniel Kessler introduced the submission.
- Dr. Jessica Buchanan provided a summary and analysis.

"**Project Overview:** Chimeric Therapeutics is developing CHM-2101 to address the continuing unmet medical need for effective therapy against relapsed or refractory gastrointestinal (GI) cancers. CDH17 is a glycoprotein composed of 7 extracellular repeat domains and a short cytoplasmic domain that contributes to embryogenesis, cellular adhesion, and tissue morphogenesis and maintenance. It is increased in many GI cancers correlated with poor clinical outcomes in numerous cancer subtypes.

This is a phase 1/2 open-label student to evaluate CHM-2101 for the treatment of advanced gastrointestinal cancers that are relapsed or refractory to at least 1 standard treatment regimen in the metastatic or locally advanced setting. Phase 1 is a dose escalation and expansion, and Phase 2 will confirm the safety of the recommended Phase 2 dose and evaluate anti-tumor activity of CHM-2101 using a Simon 2 stage design.

Changes since last full review: 15-year, long-term follow-up period, other minor changes.

Agent Description: CHM-2101 is a novel peptide-based CAR that binds to CDH17 expressed on the surface of tumor cells. It is composed of autologous CD4+ and CD8+ T cells engineered to express a CAR construct that specifically targets the human CDH17 protein. The CDH17-DAR was engineered using the sequence of a CDH17-binding singly VHH (anti-CD17), a modified IgG4 hinge region ($IgG4_m$), a CD28 transmembrane domain, and intracellular CD28, 4—1BB and CD3zeta signaling domains.



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

Gene transfer agent delivery method: Autologous CHM-2101 will be infused through an IV catheter at the indicated dose over ~1-3 minutes, without the use of a filter or pump. Participants will be monitored for at least 4 hours prior to being discharged.

Intended target: CDH17-positive cancer cells in patients with gastrointestinal cancers.

Other material to be used in preparation of the agent: Autologous T cells will be isolated from each participant's peripheral blood mononuclear cells and will undergo immunomagnetic selection to enrich for CD4+ and CD8+ T cells. The resulting cell preparation will then be activated with anti-CD3/CD28 polymeric nanomatrix and undergo lentiviral transduction to express the human CDH17 CAR. The resulting CHM-2101 will be expanded in vitro to achieve cell numbers sufficient for the participant's planned clinical cell dose and all related product release testing. The cells will then be harvested, washed, and formulated for cryopreservation until time of infusion.

Preclinical studies: No change since previous review.

Potential for shedding: Biosafety concerns related to CAR-T therapy are well known. No additional concerns identified since previous review.

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.

5. Gill ..... **#24-358** **C-1**

Dr. Saar Gill– HGT Protocol Registration Amendment V4 ..... **AMENDMENT**

PROTOCOL TITLE: Phase I Trial of CART123 cells Given in Combination with Ruxolitinib in Patients with Relapsed or Refractory Acute Myeloid Leukemia (AML). (Protocol V4 dated August 28, 2025; Main ICF V3 dated August 28, 2025; Retreatment ICF V3 dated August 28, 2025.)

IBC #24-358, IRB #857450, IND #18515, Sponsor #50424

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

"**Project Overview:** This is a Phase I, open-label study to assess the safety, feasibility, pharmacokinetics, and preliminary efficacy of CART123 cells (targeting CD123) given in combination with ruxolitinib (JAK inhibitor) in patients with relapsed or refractory acute myeloid leukemia (AML). All subjects will receive a single infusion of CART123 cells following ruxolitinib administration and lymphodepletion. A single dose of CART123 cells ($1.3x10^8$ CAR T cells) will be used in all subjects, however the optimal dose of ruxolitinib used in combination with CART123 cells will be further explored.

As of Amendment V4, the study will be divided into two treatment arms. This change was based on a preliminary review of safety, efficacy, and correlative data in the first two subjects treated on study. The original ruxolitinib dosing strategy will now be retrospectively identified as Treatment Arm A, and a modified ruxolitinib dosing strategy will be introduced as Treatment Arm B. Given concerns that higher doses of ruxolitinib may have an immunosuppressive effect on CAR T cell expansion, Treatment Arm A will be closed as of Amendment V4 and all subjects will be assigned to Treatment Arm B moving forward. Ruxolitinib dosage in Arm B is 5 mg, half of the dose used in Arm A. Ruxolitinib treatment will occur on day -1 through day 7 relative to CART123 infusion.

Agent Description: CART123. Active Ingredient: autologous chimeric antigen receptor (CAR) T-cell product targeting the CD123 antigen

Pharmacological Class: Modified autologous T-cell

Viral Construct: Lentiviral transduction of T-cells to introduce chimeric antigen receptor

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

Gene transfer agent delivery method: Intravenous infusion of CAR-T product.

Intended target: Acute Myeloid Leukemia cells expressing the CD123 antigen in vivo.

Other material to be used in preparation of the agent: Manufacturing and Formulation: CART123 is manufactured using standard CAR-T methods. CD4 and CD8 T-lymphocytes are collected from subject leukapheresis material. Following isolation and activation cell culture is initiated and cells are transduced with LVV to express the CD123 CAR and expanded for 4 days. The cells are then washed and concentrated to make CART123, which is a cryopreserved liquid cell suspension intended for intravenous infusion.

Preclinical studies: N/A

Potential for shedding: No 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.

6. Hass ..... #**25-179 ..... C-1**

Dr Andrew R Haas – HGT Protocol Registration Amendment V2 ..... **AMENDMENT**

PROTOCOL TITLE: Phase I Clinical Trial of Autologous Folate Receptor-Alpha Redirected T Cells in Patients with FRa+ Cancers. (Protocol V2 dated August 15, 2025; Main ICF dated August 15, 2025; Pre- Screen ICF V2 dated August 15, 2025.)

IBC #25-179, Sponsor# 06525, IRB# 858800, IND # 14802

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

"**Project Overview:** Evaluate safety/tolerability, feasibility, and preliminary anti-tumor activity of MOv19-BBz CAR T cells delivered intrapleurally for FRα-positive malignancies associated with pleural effusions. Secondary/exploratory objectives include feasibility metrics and time-to-event/response readouts as outlined in the analysis plan.

Agent Description: Source/Vector System (*ex vivo*): Autologous T cells lentivirally transduced to express the MOv19-BBz chimeric antigen receptor. Manufacturing at UPenn CVPF; product is cryopreserved in infusion bags.

Construct/Modifications: scFv specific for Folate Receptor-α (FRα) (murine MOv19 origin) fused to 4-1BB co-stimulatory domain and CD3ζ signaling domain; human intracellular domains; expression typically under EF-1α in the lentiviral platform used at Penn.

Transgene/Cargo: CAR sequence encoding MOv19 scFv–4-1BB–CD3ζ; redirects autologous T-cells to FRα-expressing tumor cells.

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

Gene transfer agent delivery method: Single intrapleural (IP) infusion of MOv19-BBz on Day 0, approximately 3 days (±1) after completion of lymphodepleting cyclophosphamide (300 mg/m²/day ×3) and fludarabine (30 mg/m²/day ×3); recommended infusion rate 10–20 mL/min; outpatient administration expected, with inpatient at investigator's discretion. Dose Level 1: 5×10^7 CAR^+ cells; potential de-escalation DL-1: 2.5×10^7 if ≥2 TLTs at DL1; minimum acceptable dose 2.5×10^7. Dosing is staggered per protocol.

Intended target: *Ex vivo* transduction of autologous T cells with lentiviral vector; *in vivo* target is FRα on tumor cells within the pleural space. Preclinical work demonstrates specific lysis of FRα-positive tumor lines and activity in models. Routine transduction efficiencies >60% reported in preclinical development; clinical-grade efficiency varies by lot and is not specified as a fixed threshold here.

Other material to be used in preparation of the agent: At the clinical site, only cryopreserved cell product is handled (no helper virus on site). Cryomedia excipients include human serum albumin, DMSO, and Dextran-40 (hypersensitivity to these is an exclusion). Lymphodepleting agents cyclophosphamide and fludarabine are administered per institutional standards before infusion. Refer to the Investigational Product Handling Manual for detailed handling/disposal procedures.

Preclinical studies: Penn's CAR platform experience and MOv19-BBz preclinical data show FRα-specific cytotoxicity and cytokine production; lentiviral transduction and EF-1α-driven expression achieve high CAR surface expression in T cells. Prior clinical experience with MOv19-BBz by intraperitoneal route in $FR\alpha^+$ ovarian cancer showed tolerability at tested doses; the current study evaluates intrapleural safety/feasibility.

Potential for shedding: Not viral shedding (no replicating vector is administered at the site); rather, persistence of genetically modified T cells is monitored by qPCR in blood and pleural fluid. The protocol's schedule specifies qPCR persistence serially and notes RCL (VSV-G) testing is not performed routinely. Blood is banked for future RCL testing if indicated. Recommendation: ensure nursing/pharmacy SOPs specify closed-system pleural drainage handling and BSL-2–equivalent precautions for pleural effluent during the immediate post-infusion period.

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, with recommendations detailed below."

The proposed amendments to the protocol are as follows: Clarifies pause/stop rules (adds explicit triggers incl. RCL detection; testing if suspected; samples banked) and TLT framework.

Codifies DSMB cadence (q6 months during primary follow-up) and reporting path to local committees.

Specifies dose/regimen table (IP route; 5×10^7 CAR^+ cells at DL1; lymphodepleting Cy/Flu timing and dosing; staggering).

Strengthens pregnancy/contraception language (≥1 year after infusion; pregnancy = SAE, follow to outcome; include paternal exposures).

Biosafety summary and recommendation: No replicating virus is administered at the clinical site; the product is *ex vivo* engineered autologous cells. The protocol monitors CAR T-cell persistence by qPCR in blood and pleural fluid; RCL (VSV-G) testing is not routine. Blood is banked for testing if indicated. Safety oversight is robust (CRS/ICANS/IEC-HS grading and management, explicit pause/stop criteria including RCL detection, new possibly related malignancy, and treatment-related death). Standard Precautions/BSL-2–equivalent practices are appropriate for product handling and pleural drainage/waste management; staff risk is limited to routine exposure to body fluids and cryomedia. Recommendation: Approve amendment as submitted, with recommendations to (1) place the IP Handling/Administration Manual and manufacturing QC (incl. RCL strategy) on file; (2) confirm

closed-system pleural drainage and BSL-2 handling of effluent in the immediate post-infusion period; and (3) ensure post-infusion restrictions (donation and ≥1-year contraception) are consistently documented in patient instructions.

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

HGT Administrative Actions: #13

Research Administrative Actions: #43

SECTION III–D. Experiments that Require IBC Approval Before Initiation:

7. Berger .....25-243 .....**D-1,4 O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is the generation and/ or use of 2nd generation lentiviral vectors in various human melanoma cell lines for delivery of CRISPR elements. The lab will create methylation-competent and methylation deficient cells using CRISPR mediated technology that will help address whether methylation of p53 (a post-translational modification found in cancer) drives tumor growth. The lab will utilize xenograft mouse models (NSG immune-deficient mice) injected with the modified human cells to study tumor growth following p53 perturbation. 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.

8. Chatterjee.....25-235 .....**D-4**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/ or use of mRNA-LNPs in human cancer cell lines and direct injection *in-vivo.* The lab will be encapsulating protein editing enzymes as mRNA in lipid nanoparticles for intertumoral injection of material into CDX mice. The protein editors will modulate the expression of protein, either by causing degradation in overexpressed oncogenic fusion proteins such as PAX3::FOXO1 and EWS::FLI1 which drive the tumor's development; or prevention of degradation of proteins such as the tumor suppressor P53. The catalytic and deubiquitination domains of those expressed proteins are connected to binding peptides which are AI for the lab to design new enzymes to stop tumor growth. Containment has been set to BSL-2, ABSL-2 for administration of human cells into animal, and ABSL-1 for injection of mRNA-LNPs into animal directly.
- The registration was discussed by committee members.
- Training was complete.

- All members were in favor of approval.
- The IBC registration was approved.

9. Dowling .....25-240 .....**D-4, O-1**

- Ms., Amanda Wong presented the registration. This registration is for the generation and/or use of mRNA transcripts with CRISPR/Cas 9. The lab will administer lipid nanoparticle delivery system to deliver mRNA transcripts, guide RNAs with Crispr/Cas9 machinery and antisense oligonucleotides to mice IV or IM to create an in vivo model of neuromuscular disease. Following treatment, mice will be analyzed using behavioral/strength parameters that are standard for looking at progression of muscular diseases. At termination of the experiment, tissues will be analyzed for histopathology and gene/protein expression to assess treatment efficacy. In Zebrafish, the lab will inject guideRNAs, mRNAs encoding Crispr/Cas9 editing machinery into early-stage embryos as a system to correct genetic mutations of neuromuscular disease and use morpholinos for gene silencing into early-stage embryos to suppress expression of toxic gene products that result in disease. Zebrafish development can be monitored using live imaging to look at muscle development and using molecular techniques following euthanasia to track phenotypic improvements with the treatments. 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.

10. Good .....25-209 .....**D-1,3**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of 2nd generation lentiviral vectors in cell culture. The lab is exploring how lactase oxidizing enzymes and NAD+/NADH ratio affect immune cell proliferation and immune function in Jurkat T cells by transducing the cells with lentivirus to express LbNOX (water-forming NADH oxidase from Lactobacillus brevis. The lab will also investigate how constant TYK2 activation causes cancer cell and immune cell exhaustion. The lab will insert "engineered" RGG condensates and tagged TYK2 genes to enable precise control over the signaling by transducing mouse B16 and Res499 cells, as well as human CD4/8 T cells with TYK2 and the TsCC-RGRR scaffold via Lentivirus. 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. Harty .....25-270 .....**D-1,2**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the storage of replication-defective vesicular stomatitis virus (VSV) that have their glycoprotein open reading frame (ORF) on the genome replaced with the gene for red fluorescent protein (RFP) and are pseudotyped on their surface with the glycoprotein (GP) of Ebola virus Zaire strain or the Spike (S) protein of SARS-CoV-2. This VSV was used and generated to further the Harty's lab understanding of Ebola virus and SARS-CoV-2 pathology and mechanism of viral entry. 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.

12. Mitchell.....25-258 .....**D-1**

- Ms. Maire-Luise Faber prepared the registration and Dr. Sarah Capasso presented the registration. This registration is for the generation and/or use of 3rd generation lentiviral vectors. The lab aims to establish nanoparticle systems for the intracellular delivery of the CRISPR-Cas9 gene editing system. These nanoparticle systems are lipid-based, and include excipients such as ionizable lipids, neutral lipids, lipid-anchored polymers, and carbohydrates. These excipients are varied in ratio and type to optimize delivery efficiency. The CRISPR-Cas9 system is delivered as either RNA only (nuclease-encoding RNA and guide RNA) or as an RNP (Cas9 protein and guide RNA complex).
  This optimization effort is currently performed on cultured human cells, utilizing a CRISPR-Cas9 system that is targeted for a deletion of a reporter system, such as GFP, which facilitates monitoring by visual imaging. For continued expansion of this work, the lab will test the compatibility of these lipid nanoparticle systems across a variety of different cell types (immortalized cancer lines as well as disease model lines) in vitro using lentivirus: to create new fluorescent model human cell lines, such as GFP, that will be used to evaluate (and potentially re-optimize) our LNP system and for the production of CD19 CAR+ cell lines, to use in experiments as controls and to evaluate our LNPs system. Containment has been set to BSL-2.
- The registration was discussed by committee members. Dr. Sarah Capasso noted that several sections of the registration were incomplete or not entered correctly it was recommended to table registration for further review. Dr. Daniel Kessler requested a more detailed title.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was tabled. Administrative approval pending Biosafety officer review.

13. Murray .....25-080 .....**D-4, O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the creation of transgenic C. elegans. The lab aims to understand the zygotic regulation of lineage specification during the period from ~gastrulation through terminal differentiation. Current plans involve dissecting the regulation of the C. elegans tab-1 gene (a homolog of mammalian BSX, a Brain-Specific Homeobox gene, which encodes a transcription factor that is essential for proper brain development and function). The lab will generate transgenic C. elegans expressing fusion

proteins, such as HIS-24::mCherry under the control of predicted tab-1 regulatory sequences and will delete regulatory sequences using CRISPR/Cas9 to determine which are necessary for tab-1 expression and function. They create transgenic C. elegans strains using microinjection of plasmid DNA or PCR products. 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.

14. Sehgal-Field...25-262.....**D-4**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of adeno associated viral vectors. AAV serotypes 1 and 2 are acquired from the Penn Vector core and are either directly injected *in-vivo* by iv or administered to cell culture. The lab hypothesize that the blood-brain barrier is regulated through intracellular magnesium levels, which are controlled by the circadian clock. To determine Mg2+ levels in vivo, the lab will use AAV, which induces the genomic incorporation of a fluorescence sensor for Mg2+. Using this sensor, the lab will be able to monitor changes in [Mg2+] levels in the mouse blood-brain barrier endothelium throughout the circadian day in WT and mutants with defective circadian rhythms. 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.

15. Wherry.....25-236.....**D-1,4**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of modified Listeria monocytogenes to study T cell memory and vaccination strategies for infectious disease *in-vivo*. Mice will be used to examine the dynamics of pathogenesis and the interactions between pathogenic agents and the host. In this study, recombinant Listeria monocytogenes, modified to express known T cell epitopes, will be used to facilitate tracking of specific immune responses. Listeria will be administered by the intra-peritoneal, intravenous, and intranasal routes. The recombinant Listeria was donated by colleagues at Emory University. 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.

SECTION III–O. Experiments that Require IBC Approval Before Initiation:

16. Booker.....25-182 .....**E-1, O-1**

- Ms. Stephanie Adams-Tzivelekidis presented the registration. This registration is for the generation and/or use of plasmids. The lab is transforming E. coli BL21 or transfecting HEK293T to overexpress non-hazardous, recombinant iron-sulfur cluster proteins. Culture volume is as follows: 4 L for E coli, 1 L for HEK cells (suspension). Additionally, CRISPR elements dCas9 and sgRNA are expressed in bacteria. This is a deactivated form of Cas9 and has no editing function. Containment has been set to BSL-2 for work with human tissue culture and BSL-1 for work with E. coli.
- The registration was discussed by committee members.
- Training was complete.
- All members were in favor of approval.
- The IBC registration was approved

3. New Business:

(a) No New Business Scheduled.

4. Old Business:

(a) No Old Business Scheduled.

5. End Meeting:

- The Institutional Biosafety Committee was adjourned by Dr. Daniel Kessler at **10:49 AM**.

Our next meeting scheduled for Monday, October 27th, 2025, will be held on site at the EHRS Office with a Teleconference option, at 10:00 am. A light Brunch will be provided.