CLINICAL PROTOCOL

Study Title:	Phase 1b & 2 Study with GL-ONC1 in Patients with Recurrent or Refractory Ovarian Cancer (VIRO-15)
Protocol Number:	GL-ONC1-015
Development Phase:	Phase 1b & 2
Indication:	Platinum-resistant, platinum-refractory and intermediate platinum-sensitive ovarian cancer and peritoneal carcinomatosis
Sponsor:	Genelux Corporation
Sponsor Signatory:	Tony Yu, PhD
Sponsor's Responsible Medical Officer:	Paul Scigalla, MD, PhD
Study duration:	24 months
Clinical Protocol Date:	December 18, 2018
Version:	Version 1.10
IND Number:	014680
NCT Number:	NCT02759588

GOOD CLINICAL PRACTICE

This study will be conducted according to the principles of Good Clinical Practice as described in International Conference on Harmonisation guidelines, including the archiving of essential documents, and in accordance with the Food and Drug Administration's Code of Federal Regulations.

PROTOCOL SIGNATURE PAGE

I have read and understand the contents of the indicated clinical protocol and will adhere to the trial requirements as presented, including all statements regarding confidentiality. I and my Sub-investigator(s) agree to conduct the study as outlined herein, in accordance with Good Clinical Practice (GCP), the Declaration of Helsinki, and the Belmont Report, in compliance with the obligations and requirements of Investigators and all other requirements listed in Title 21 Code of Federal Regulations (CFR) *Part 50—Protection of Human Subjects; Part—Financial Disclosure by Clinical Investigators; Part 56—Institutional Review Boards;* and *Part 312—Investigational New Drug Application.* In addition, the *NIH Guidelines for Research Involving Recombinant DNA Molecules,* which is under the regulatory oversight of the National Institutes of Health Office of Biotechnology Activities shall be followed regarding the use, storage and handling of the investigational product, GL-ONC1.

Investigator:

Name: Robert W. Holloway, MD

Signature: _____

Date: _____

Genelux's Sponsor Signatory:

Name: Tony Yu, PhD

Signature: _____

Date: 18-December-2018

Title: Vice President of Clinical Trial Operations

Genelux's Responsible Medical Officer:

Name: Paul Scigalla, PhD, MD

Signature: _____

Date: _____

Title: Chief Medical Officer

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STUDY SYNOPSIS

Title of Study: Protocol GL-ONC1-015: Phase 1b & 2 Study with GL-ONC1 in Patients with Recurrent or Refractory Ovarian Cancer (VIRO-15)

IND Number: 014680

Investigational Product: GL-ONC1, an oncolytic vaccinia virus.

Study Phase: Phase 1b & 2.

Methodology: This is an open-label, non-randomized Phase 1b & 2 study evaluating the effect of the oncolytic virus, GL-ONC1, administered via an intraperitoneal (IP) catheter or intravenously as a bolus infusion in patients diagnosed with recurrent or refractory ovarian cancer and peritoneal carcinomatosis as a monotherapy or as a combination therapy.

Research Hypothesis: That GL-ONC1 is well tolerated with antitumor activity in patients diagnosed with recurrent or refractory ovarian cancer and peritoneal carcinomatosis.

Primary Objective:

Phase 1b: Investigate the safety and tolerability.
Phase 2 (Cohorts A, B & D): Investigate Progression-free Survival (PFS) with therapeutic intent.
Phase 2 (Cohort C & D): Overall Response Rate (ORR) by RECIST1.1 and by GCIG CA-125 criteria with therapeutic intent.

Secondary Objectives:

Phase 1b & Phase 2 Cohorts A & B: Response to treatment with therapeutic intent will be determined based on Response Evaluation Criteria in Solid Tumors (RECIST) 1.1, by the Immune-related Response Criteria (irRC) as an exploratory endpoint, and by CA-125 according to the Gynecologic Cancer Intergroup (GCIG) CA-125 response criteria in patients with measurable disease. Patients who enter with non-measurable disease will be evaluated for response to treatment by PET/PET CT scan, physical examination and CA-125 levels. Antitumor activity (if evaluable by CT, PET/PET-CT scans) will be evaluated by progression-free survival (Phase 1b only), Clinical Benefit Rate (CBR: CR + PR + SD), best overall response (BOR), duration of response (DoR), objective overall response rate (ORR), disease control rate (DCR = CR + PR + SD≥15 weeks). Additionally, overall survival (OS) and time to treatment failure (TTF) will be assessed. In the Phase 2 study, safety assessment will continue.

Phase 2 Cohort C & D:

- Progression-free Survival by RECIST1.1 (Ch C)
- Duration of Response
- Clinical Benefit Rate
- Disease Control Rate
- Overall Survival

Exploratory Objectives (optional):

All cohorts:

(1) To evaluate the immune response to treatment, immune assays will be performed to study immune activation and antitumor immune response in blood, ascites and tumor biopsies;

(2) To evaluate antitumor activity at the tissue/cellular levels (proliferation index, apoptosis, etc.);
(3) To confirm the presence of GL-ONC1 within the tumor by viral plaque assay (VPA), immunohistochemistry (IHC), and/or quantitative polymerase chain reaction (qPCR).
(4) To determine the possible prognostic value (e.g., as a predictive value of survival outcome) of circulating tumor cells (CTCs) in patients diagnosed with ovarian cancer. In addition, to demonstrate the correlation of CTC number with radiological outcome as early pharmacodynamic and response rate indicators in the context of GL-ONC1 treatment.

Treatment Plans:

Phase 1b		
Cohort	**GL-ONC1 (dosing on 2 consecutive days)**	**N**
1	3×10^9 pfu per day	3
2	1×10^{10} pfu per day	3
3	2.5×10^{10} pfu per day	3
4	4×10^{10} pfu per day	3
	Subtotal	**12-18** (based on optional cohort expansion)
• There is a 2-4 week safety observation window, with a minimum 2 weeks between each cohort. • As an optional expansion, up to 6 more patients may be treated in any of the above cohorts.		

Phase 2		
Cohort[1]	**GL-ONC1[2] (dosing on 2 consecutive days)**	**N**
A	3×10^9 pfu per day	20
B	1×10^{10} pfu per day	20
	Subtotal	**40**

[1] Enrollment into Cohorts A and B is by Sponsor assignment.

[2] An optional second cycle of GL-ONC1 could be given through a temporary intraperitoneal percutaneous catheter for intraperitoneal delivery at the discretion of the Investigator and the Sponsor. The catheter will be inserted and removed by Interventional Radiology. Patients who have demonstrated disease control (CR + PR + SD ≥ 15 weeks) from first cycle of GL-ONC1 but start to show disease progression with or without further chemotherapy and/or bevacizumab, will be considered for a second cycle of GL-ONC1. Optional CT or ultrasound-guided biopsies will be obtained prior to (within 2 weeks or on day of catheter placement) and ~4 weeks after second cycle of GL-ONC1.

Disease progression could be determined by RECIST 1.1 or based on confirmed upward trend of CA-125 values. For example, when CA-125 increases to more than twice the nadir value and above 70 u/mL (CA-125 nadir < 100); or when there is an increase in CA-125 levels by >100 when the nadir was >100 u/mL. Repeat CA-125 2-4 weeks after an elevation to confirm a trend upwards. When disease progression is suspected clinically based on persistent rise in CA-125, physical exam findings, and /or symptoms, a confirmation CT scan will be obtained. In pts whose CA-125 has been shown non-diagnostic (low levels despite RECIST lesions on CT scans), the decision to proceed with second cycle of GL-ONC1 will be made on progression by CT scan coupled with physical exam findings and symptoms.

Phase 2				
Cohort	**GL-ONC1**	**Chemotherapy (chemo)**	**Bevacizumab (bev)**	**N**
Safety run-in cohort	1 cycle @ 3 ×10^9 pfu per day × 2 consecutive days	Carboplatin doublet, up to 6 cycles	With chemotherapy, plus maintenance as single agent	3-6
C				Up to **35**

- Safety run-in cohort (SRIC) (with staggered enrollment and treatment): There is at least 28-day interval for safety observation between each patient. Enrollment into Cohort C occurs only after completing the safety run-in cohort.
- Combination with bevacizumab is allowed but not required. If developing toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment, e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel, to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if tolerated well and determined to be clinically beneficial after 6 cycles.
- Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

Phase 2				
Cohort	**GL-ONC1**	**Chemotherapy (chemo)**	**Bevacizumab (bev)**	**N**
Safety run-in cohort	1 cycle @ 2,3,5 ×10^9 pfu by IV route for 3 consecutive days	Carboplatin doublet, 6 cycles	With chemotherapy, plus maintenance as single agent	3-6
D				15

- Safety run-in cohort (with staggered enrollment and treatment): There is at least 28-day interval for safety observation between each subject. Enrollment into Ch D occurs only after completing the safety run-in cohort.
- GL-ONC1 is administered intravenously as an undiluted bolus infusion.
- Progression-free Survival (PFS) is to be assessed after IV doses of GL-ONC1 until disease progression, if clinically applicable.
- Chemo +/- bev will start either at time of disease progression from GL-ONC1, or will start at the discretion of the PI as clinically indicated based on clinical assessment (shall consult with Sponsor if falls into this category).
- Details of chemo +/- bev regimen are the same as to Ch C.
- Combination with bev is allowed but not required. If developing toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment, e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel, to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if tolerated well and determined to be clinically beneficial after 6 cycles.
- Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

In the Phase 1b portion, a temporary intraperitoneal catheter will be inserted 5 to 7 days prior to Day 1. For Phase 2 (except Cohort D), a temporary intraperitoneal catheter is inserted 3 to 7 days (5 days preferred) prior to administration of the first GL-ONC1 dose. Patients who have an existing indwelling peritoneal catheter or port at screening will have it surgically removed and replaced with a temporary peritoneal catheter by laparoscopic procedure. During the baseline laparoscopy, photos will be taken to document the extent of disease and assessed by the Sugarbaker *Peritoneal Cancer Index (Appendix 3)*. The catheter may be removed on or before Day 17 for patients treated in Phase 1b, or 14 days after date of the first GL-ONC1 dose for patients treated in the Phase 2 portion. If medically required to manage ongoing ascites, timing of catheter removal is at the Investigator's discretion.

Patients treated in Phase 2 Cohort D will receive GL-ONC1 intravenously as an undiluted bolus infusion.

Tumor Biopsies: A baseline tumor biopsy is required (e.g., during surgical implantation of the intraperitoneal catheter). Depending on the location of the tumor(s) selected by the Investigator for sampling, biopsies may be performed through the laparoscope (preferred method, if feasible) or by needle biopsy (e.g., CT-guided or ultrasound-guided biopsy), if tumor is safely accessible. Tumor tissue

that is safely accessible by needle biopsy will be obtained from consenting patients. During the course of the study, unscheduled biological samples (e.g., tumor tissue, ascites, pleural fluid, blood) provided from consenting patients will be kept for translational research purposes. If additional tumor biops(ies) are obtained from the same patient at later time point(s), it is preferred that tumor samples are from the same baseline body location if feasible. The number of patients to obtain optional tumor biopsies will be at the discretion of the Sponsor.

- PHASE 1b Post-treatment Tumor Biopsies: Post-treatment CT-guided biopsies may occur either on Day 10 or 17, and at Week 36.
- PHASE 2 COHORTS A & B -- Post-treatment Tumor Biopsies (CT-guided or ultrasound-guided biopsies): Post-treatment needle biopsies may occur either 7 or 14 days after date of the first GL-ONC1 dose, and either at Week 6 or Week 15 (± 7-day variance at each time point). For patients who receive a second cycle of GL-ONC1, optional needle biopsies will be obtained prior to (within 2 weeks or on day of catheter placement) and at approximately 4 weeks after second cycle of GL-ONC1.
- PHASE 2 COHORT C -- Post-treatment Tumor Biopsies (CT-guided or ultrasound-guided): A preferred but optional post-treatment needle biopsy may occur between Weeks 2 to 5 after date of the first GL-ONC1 dose, and prior to initiation of further chemotherapy. Another optional post-treatment needle biopsy may be obtained after second cycle (either during Week 11 for q3w, or Week 13 for q4w with ± 1 week for both time points) or after the third cycle (Week 14 for q3w, or Week 17 for q4w ± 1 week for both time points) of chemotherapy. Biopsy will be obtained after CT scan if they are conducted on same day.
- PHASE 2 COHORT D -- Post-treatment Tumor Biopsies (CT-guided or ultrasound-guided): A preferred but optional post-treatment needle biopsy may occur at the time points listed above for Phase 2 Cohorts A & B.

Phase 1b Study Requirements:

3+3 Dose Escalation Design:

Decisions to escalate to the next dose level will be based on whether dose-limiting toxicities (DLTs) were encountered, and available clinical (i.e., safety data) and non-clinical data (e.g., vital signs, routine lab tests). Safety and non-clinical data will be reviewed by the Cohort Management Committee (CMC) following the two to four-week safety observation window, with a minimum of 2 weeks between cohorts to allow sufficient time to monitor acute and sub-acute adverse events. Patients can be consented and screened for eligibility during the safety observation window to prepare for enrollment in the next dose cohort following a positive CMC vote.

- Dose escalation (i.e., opening a new cohort for patient treatment) will be based on the toxicities encountered within each cohort according to the following dose escalation plan: If one patient out of three in a dose group experiences a DLT, three more patients will be added to that dose group.
- If the next patient at this dose level experiences a DLT (this may be patient 4, 5, or 6 in the second group), this dose level will be closed (i.e., no further treatments administered).
- If two or more patients in a dose group of three to six patients experience a DLT, a one-half log lower dose, or the previous dose level will be employed.
- If tolerated, the lower dose level will be defined as the Maximum Tolerated Dose (MTD).
- If MTD is reached, a minimum of total 6 patients will be treated at the determined viral product dose level to investigate and confirm the observations seen in the first 3 patients of the intended cohort.

In each Phase 1b cohort, a week for safety evaluation begins following the first patient receiving the last GL-ONC1 treatment before the second patient is treated. Subsequent patients in a cohort may be treated in parallel with the second patient. Intrapatient dose escalations are not permitted.

Dose Limiting Toxicities (DLT):

Toxicities will be graded according to the National Cancer Institute (NCI) Common Toxicity Criteria for Adverse Events (CTCAE) version 4.03. If a grade is not provided in the CTCAE for a particular adverse event, the Investigator will use his/her best medical judgment in assessing the toxicity grade.

GL-ONC1 administration should be discontinued if a patient experiences at least one of the following dose limiting toxicities (DLT). A DLT is defined as any of the following adverse events having a degree of attribution to GL-ONC1 (i.e., probably, possibly or definitely) occurring within 30 days after the last GL-ONC1 treatment. These include:

- Any Grade 3 non-hematologic adverse events lasting more than 72 hours (excluding nausea/vomiting and diarrhea; exceptions may be made for transient (e.g. lasting < 7 Days) Grade 3 elevations of ALT/AST in the presence of known liver metastases and without evidence of other hepatic injury, if agreed by the Investigator);
- Any Grade 4 or higher (≥ 4) non-hematologic adverse events;
- Any Grade 4 or higher (≥ 4) hematologic adverse events lasting more than 7 days;
- Grade 2 or greater bronchospasm requires discontinuation of the viral product.
- All Grade 2 hypersensitivity reactions to GL-ONC1 treatment.
- Any adverse event that leads to a discontinuation of GL-ONC1 infusion.

Study Stopping Rules:

The following rules will apply, if warranted:

- Individual Treatment: Any patient who develops a DRUG-RELATED dose limiting toxicity (DLT), as defined in this protocol, will not receive further administration of the investigational product and the patient are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible.
- Study Stopping Rules: If 2 or more out of the six patients (≥ 33%) experience a DLT at any dose level, the Investigator, Medical Monitor and the Sponsor may elect to employ a one-half log lower dose or the prior dose level. The site may proceed with this lower dose level only following written notification from the Sponsor.

Study Pausing Rule:

If the following events occur during the Treatment Period (i.e., Week 1 Days 1 to 5) up to 30 days post last GL-ONC1 infusion, the study will be placed on hold until an appropriate evaluation of the cause of the toxicity is determined, and a plan of correction, if necessary, is established:

- Death (other than death related to progressive disease);
- Grade 3 to 4 adverse event rate > 70% in the first 30 days.

Revision to Enrollment Criteria:

If the study is stopped or paused to evaluate the occurrence of an event(s) defined above, revisions to the enrollment criteria may be required through a protocol amendment to exclude individuals who might be at a higher risk of developing particular adverse reactions. This decision will be made in conjunction between the CMC and the Sponsor.

Maximum Feasible Dose (MFD):

The MFD is defined as the highest dose level when either dose limiting toxicities (DLTs) or the maximum tolerated dose (MTD) are not reached.

Maximum Tolerated Dose (optional):

The dose one level lower will be declared as the MTD, if a DLT is observed in ≤ 1 patient of the total 6 patients in the dose level.

Cohort Management Committee (CMC):

Phase 1b: Decisions to escalate to the next level, or, when appropriate, to an intermediate level are made by the Investigator following consultation with the Cohort Management Committee (CMC) members. CMC membership will consist of the Investigator, additional clinical site qualified medical personnel, as well as two qualified and experienced sponsor representatives (e.g., Medical Monitor and a senior staff member with knowledge of GL-ONC1's development) as voting members. CMC members will follow procedures described in the 'Cohort Management Committee Charter' for review of safety data. Judgments on whether the trial should be stopped or paused based on review of safety data will be made jointly between the CMC and the Sponsor.

Phase 2: The CMC will review safety data on an as needed basis determined by the frequency, severity and type of suspected serious adverse reactions. The safety data report reviewed by the CMC and the committee's recommendations will be documented in the Sponsor's trial master file (TMF) and the Investigator's intuitional site files (ISF).

Analysis of CTCs in Ovarian Cancer:

Optional analysis of circulating tumor cells (CTC) may be included in any cohorts. As this is an optional Exploratory Objective, the number of patients in any cohorts to be tested for CTC will be at the discretion of the Sponsor. Fresh whole blood samples will be immediately shipped to testing facility for analysis using the CellSearch system

Several studies have shown that circulating tumor cells (CTCs), over a specified cut-off value, predicts for a worse prognosis in terms of progression free survival (PFS) and overall survival (OS) in breast, prostate and colorectal cancer population (Cohen *et al., J Clin Oncol* 2008;26:3213-21; de Bono *et al., Clin Cancer Res* 2008;14:6302-09; Cristofanilli *et al., N Engl J Med* 2004;351:781-91).

Common to all of these studies is that CTCs, prior to commencing a new line of treatment, have prognostic value in breast, prostate and colorectal cancer. Furthermore, changes in CTCs during treatment predict survival outcome, highlighting their potential as a surrogate endpoint biomarker (Krebs *et al., Ther Adv Med Oncol* 2010;2(6):351-65).

Studies correlated CTCs with response to imaging according to Response Evaluation Criteria in Solid Tumors (RECIST) have also been performed (De Giorgi *et al., J Clin Oncol* 2009;27:3303-11; Budd *et al., Clin Cancer Res* 2006;12; Cohen *et al., Ann Oncol* 2009;20:1223-29). Although these studies were heterogeneous, the results favored CTCs over conventional imaging to be the most significant factor in determining survival prognosis. Results should be interpreted with caution in patients with stable disease (SD) and with no CTC changes.

CTCs may be used to predict the activity of new drugs in the drug development setting, potentially facilitating short time go/no-go decision-making between baseline CTCs and assessment of CTCs at subsequent time points (Krebs *et al., Ther Adv Med Oncol* 2010;2(6):351-65). Whilst data are emerging

on several cancer types, there remains little information regarding CTCs and other malignancies (melanoma, non-small cell lung cancer, etc.), so much more research is required (Krebs *et al., J Clin Oncol* 2011;29(12):1556-63; Mandrekar *et al., J Thorac Oncol* 2010;5:3-9).

Cui *et al.,* (*J Ovarian Res* 2015:8:38 DOI 10.1186/s13048-015-0168-9) performed a meta-analysis of clinical studies which included CTCs and disseminated tumor cells (DTC) that showed a strong relationship of CTCs and DTCs with advanced staging, response to treatment and poor prognosis in patients with ovarian cancer. Poveda *et al.* (*Gyn Onco* 2011;122(3):567-72) found that CTCs were prognostic during ovarian cancer therapy and that patients with ≥ 2 CTCs at baseline had a shorter overall survival and shorter time to progression.

Therefore, an optional exploratory endpoint is to investigate the correlation of the number of CTC in survival outcome and response to treatment in women diagnosed with ovarian cancer.

Study Population:

Sponsor allows the rescreening of patients, if necessary. The Sponsor will determine the final cohort allocation on an *ad hoc* basis based on the clinical information provided during eligibility review.

Key inclusion criteria for the Phase 1b & 2 include (refer to *Section 4.7 Inclusion Criteria* for full listing of inclusion criteria):

- Signed, written informed consent.
- Women ≥ 21 years.
- History of histologically confirmed (from prior treatment) non-resectable ovarian, fallopian tube or primary peritoneal cancer.
- Patient population: High-grade serous (including Malignant Mixed Mullerian Tumor (MMMT) with metastasis that contains high grade epithelial carcinoma), endometrioid, or clear-cell ovarian cancer that is recurrent or refractory, which include:

  - *Platinum-resistant* (recurrence or progression< 6 months) or *platinum-refractory* (progression while on platinum-based therapy): Patients must have failed either at least 2 consecutive therapies or are not eligible for additional cytotoxic therapies (exception is for patients registered into Cohorts C & D).
  - *Intermediate platinum-sensitive patients* (recurrence of disease 6 to 12 months from last platinum compound treatment): Recurrent ovarian carcinoma with at least four prior individual treatment regimens including at least two separate platinum-based therapies with recurrence from the last platinum-based regimen less than 12 months, who are unwilling or unable to undergo additional platinum-based cytotoxic therapy (this sub-population is not applicable for Cohorts C & D).

- Performance status ECOG is at 0 or 1.
- Patient has grossly visible tumors located in the peritoneal cavity which are accessible by the peritoneal catheter **(eligible to 'Ch D' if not meeting this criterion).**
- Patient has measurable disease in the peritoneal cavity (e.g., stomach, spleen, liver, ovaries, fallopian tubes, tail of the pancreas, uterus, bulb of the duodenum, jejunum, ileum, transverse colon, and sigmoid colon) as defined by RECIST 1.1 with at least one lesion that can be accurately measured in one dimension (longest dimension recorded) by contrast spiral CT scan. **Patients who do not meet this criterion are eligible for enrollment in 'Ch D' if (1) there is RECIST 1.1 measurable extra-peritoneal disease such as mediastinal nodes, liver metastasis, lung, etc,; (2) RECIST 1.1 measurable intra-abdominal disease not easily accessible by laparoscopy in the opinion of the PI (dome of liver/diaphragm post liver diaphragm resection, rigid abdomen with multiple laparotomies, etc)**.

Phase 1b only: non-measurable disease in the peritoneal cavity that is identifiable by PET/PET-CT scan with contrast and can be confirmed by laparoscopy and/or elevated CA-125. Patients who have non-measurable disease that is not identifiable by PET/PET-CT scan, but who have elevated CA-125, and/or ascites, with visible disease confirmed by laparoscopy are also eligible.

- Able to undergo IP or IV bolus infusion, and all administration procedures.
- Adequate renal, hepatic, bone marrow, and immune functions:

  a. Renal function:
     - Creatinine ≤ 1.8 mg/dL or calculated creatinine clearance using the Cockcroft-Gault formula ≥ 45 mL/min, or measured creatinine clearance ≥ 45 mL/min

       $$\text{Female CrCl} = \frac{(140 - \text{age in years}) \times \text{weight in kg} \times 0.85}{72 \times \text{serum creatinine in mg/dL}}$$

     - Absence of clinically significant hematuria on urinalysis: dipstick <2+ (exception to this criterion is for presence of renal stent(s) and/or otherwise good renal function as assessed by Investigator);
     - Absence of clinically significant proteinuria on urinalysis: dipstick < 2+.

  b. Adequate hepatic function
     - Serum bilirubin <1.5 × ULN;
     - AST and ALT ≤ 3 × ULN.

  c. Adequate bone marrow function:
     - ANC ≥ 1.5 × 10^9/L;
     - Platelets ≥ 100 × 10^9/L;
     - Hemoglobin ≥ 90 g/L.

  d. Adequate coagulation tests:
     - INR ≤ 1.5 × ULN.

  e. Adequate immune function by lymphocyte count:
     - Absolute lymphocyte count (ALC) ≥ 0.5 × 10^3/mm^3;
     - Relative lymphocyte count (RLC) ≥ 10%.

- Life expectancy of at least 6 months.
- A baseline tumor biopsy is required.
- Adequate nutritional status as determined by Investigator (e.g., assessed based on Body Mass Index (BMI), or Prognostic Nutritional Index (PNI)).
- **Phase 2 only:** Documented progressive disease status at baseline (based on RECIST 1.1 or clinical progression).

Key exclusion criteria include (refer to *Section 4.8 Exclusion Criteria* for full listing of exclusion criteria):

- Tumors of mucinous subtypes, or non-epithelial ovarian cancers (e.g., Brenner tumors, Sex-cord tumors).
- Unresolved bowel obstruction.
- Undiagnosed gastrointestinal bleeding.
- Known central nervous system (CNS) metastasis.
- Inflammatory diseases of the bowel.
- Concurrent therapy with any other investigational anti-cancer agent or treatments.
- Known seropositive for active viral infection with human immunodeficiency virus (HIV); or active hepatitis B virus (HBV) or hepatitis C virus (HCV) infection within 4 weeks prior to study initiation.
- History of thromboembolic event within the last 3 months.
- Pregnant or breast-feeding women.
- Small pox vaccination within 1 year of study therapy.

- At the time of eligibility assessment, have clinically significant cardiac disease (New York Heart Association Class III or IV; refer to *Appendix 5 New York Heart Association (NYHA) Functional Classification* for classification of symptoms).
- Oxygen saturation <90% measured by pulse oximetry at rest.
- Have received prior gene therapy or therapy with cytolytic virus of any type.
- Be receiving concurrent antiviral agent active against vaccinia virus (e.g., cidofovir, vaccinia immunoglobulin, imatinib, ST-246) during the course of study.
- Have known allergy to ovalbumin or other egg products.
- Have clinically significant dermatological disorders (e.g., eczema, psoriasis, or any unhealed skin wounds or ulcers) as assessed by the Investigator during screening and during the study.
- Symptomatic malignant ascites defined as rapidly progressive ascites with abdominal distention and gastrointestinal dysfunction, breathing difficulties, and/or requiring frequent paracentesis more than once every 14 days.
- Non-manageable pleural effusion and/or oxygen dependence on a routine basis. Stable manageable pleural effusions with or without a catheter can be allowed as long as other performance status requirements are met.
- **Cohorts C & D -- Regarding bevacizumab if applicable:** Known hypersensitivity to bevacizumab, uncontrolled hypertension, history of stroke, or clinical findings suggestive of excessive risk for GI perforation (uncontrolled peptic ulcer disease, partial small bowel obstruction, etc.) that would make risks of bevacizumab unacceptable in the opinion of the Investigator.

Study Periods:

This study consists of 4 periods: screening, treatment, post-treatment follow-up and long-term follow-up. For specific procedures as well as timing of events during each period, refer to *Section 15.0 Study Assessments and Procedures*.

General Statements

Phase 1b & Phase 2 Cohorts A & B -- Weeks 2 to 48, and Years 2 & 3 Long-term Follow-up: If a patient moves on to receive other anti-cancer treatment after GL-ONC1, clinic visits are no longer required as follow-up is conducted by a telephone call to assess survival, disease and anti-cancer treatment status. Telephone calls occur at the clinic visit time points specified for each study period. In order to determine if other anti-cancer treatment may have a synergistic effect following GL-ONC1 treatment, the sponsor asks that study sites continue to provide de-identified radiologic imaging reports and clinical labs, including CA 125 results, on an *ad hoc* basis when available.

Assessment of SAE(s) that occurred since the last telephone call (or visit if first follow-up call). If the Investigator determines a SAE has a degree of attribution to GL-ONC1 treatment, submit SAE Report to the Pharmacovigilance Team via electronic data capture (EDC) system or by paper CRF if EDC not available. Non-serious AEs are not collected. In order to determine if other anti-cancer treatment may have a synergistic effect following GL-ONC1 treatment, the sponsor asks that sites provide in a timely manner de-identified radiologic imaging reports and clinical labs, including CA 125 results, on an *ad hoc* basis when available.

ALL PHASE 2 COHORTS: Ongoing treatment may be administered following carboplatin doublet therapy with either the non-platinum single agent chemotherapy, single agent bev, or combination of single agent chemotherapy with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

All Cohorts off-site treatment with chemo +/- bev: It is highly preferred that all subsequent platinum-based doublet therapy, non-platinum therapy and bev be administered at the Investigator's institution.

However, for patients traveling significant distances to the clinical site, only the first two treatment cycles will be required to be administered at the Investigator's institution. Further treatments can continue locally by a patient's primary practitioner if the Investigator and the local primary practitioner have an agreement on a specific treatment regimen to follow. Regular follow-up visits at the Investigator's institution for monitoring purposes are required, and all follow-up CT scans are to be performed at the Investigator's institution.

All Cohorts – Quality of Life (QoL) Questionnaire: The *Functional Assessment of Cancer Therapy-Ovarian* (FACT-O) Quality of Life Questionnaire (refer to *Appendix 7*) is obtained at baseline and at each clinic visit when radiologic imaging is scheduled.

Screening Period – All Phases/All Cohorts (unless otherwise specified):

- Begins by signed informed consent form (ICF) and establishing the patient's initial eligibility.
- Patients are registered by the Investigator or designee by submitting the registration form and applicable eligibility documents to the Sponsor for review of eligibility.
- Patients with an existing intraperitoneal catheter/port will have it surgically removed at the time of the laparoscopic procedure.
- PHASE 1b: A temporary intraperitoneal catheter will be inserted during the laparoscopic procedure within 5 to 7 days prior to of Treatment Day 1 and photos will be obtained to document the extent of the disease for assessment by the Peritoneal Cancer Index (Sugarbaker *et al., Peritoneal carcinomatosis: principals of management,* Sugarbaker PH (Ed); Kluwer Academic, Boston, MA USA; pp359-74).
- PHASE 2 Cohorts A, B & C: Patients will have a temporary peritoneal catheter surgically implanted by laparoscopic procedure within 3 to 7 days (5 days preferred) prior to administration of the first GL-ONC1 dose with photos taken to document the extent of disease for assessment by the *Peritoneal Cancer Index* (40) *(*refer to *Appendix 3)*.
- PHASE 1b: A biopsy of tumor tissue is obtained (e.g., during laparoscopy) for analysis of the cancer gene profile and immune status of the tumor.
- PHASE 2: Depending on the location of the tumor(s) selected by the Investigator for sampling, biopsies may be performed through the laparoscope (preferred method for all cohorts except Cohort D, if feasible) or by needle biopsy (e.g., CT-guided or ultrasound-guided biopsy) if tumor is safely accessible.
- It is preferred that baseline radiographic assessment occur as close to treatment as allowable (ideally within 2 weeks).
- Patient is assessed for complete study eligibility within the required timeframe (see *Section 15.0 Study Assessments and Procedures*).
- Screening Assessments: For patients screened and enrolled into study within a short period of time prior to treatment, it is not considered a protocol deviation if all screening procedures are collected at one-time point instead of two-time points (i.e., toxicity, physical examination, weight, performance status, vital signs, serum pregnancy and clinical labs, concomitant medications).

Treatment Period (Week 1) -- Phase 1b and Phase 2 Cohorts A & B:

- *PHASE 1b:* This is a dose escalating study with GL-ONC1 administered IP on two consecutive days to determine the MFD or MTD (MTD is optional).
- *PHASE 2:* Sponsor assignment into either Cohort A or B.
- Clinical labs, pharmacodynamic (PD) and pharmacokinetic (PK) samples are collected according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.
- For patients who have peritoneal ascites, as much ascites as possible will be drained prior to the instillation of the first GL-ONC1 dose. On the day after the second GL-ONC1 dose, 10 mL of ascites will be collected, if available.

- All patients will have 1 L to 1.5 L of Ringer's Lactate instilled and drained through the IP catheter prior to the first GL-ONC1 dose as a peritoneal wash.
- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. Non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team.
- Deaths that occur during Treatment Week 1 are reported as an expedited safety report to the Pharmacovigilance Team regardless of causality (refer to *Section 21.11 Deaths* for reporting requirements).

Post-treatment Period Week 2 to Week 48 -- Phase 1b and Phase 2 Cohorts A & B:

- For patients with measurable disease, imaging by spiral CT scan is performed after initiation of treatment at Weeks 6, 15, 24, 36 and 48. For patients allergic to IV CT contrast, it is recommended that oral CT contrast (preferred) or MRI may be used. Per RECIST 1.1, the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up.
- For patients with non-measurable disease (Phase 1b only), imaging will be performed by either PET scan or PET/CT scan at any of the time points listed for the spiral CT scan per the Investigator's discretion.
- PHASE 2: Either 7 or 14 days after date of first GL-ONC1 dose, an optional needle biopsy (e.g., CT-guided or ultrasound-guided biopsy) to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- PHASE 1b: Either on Day 10 or 17, an optional CT-guided biopsy to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- PHASE 2: An optional post-treatment needle biopsy (by CT-guided or ultrasound-guided biopsy) may be obtained either during Week 6 or Week 15 (± 7 days).
- PHASE 2 COHORTS A & B: For patients who receive a second cycle of GL-ONC1, optional needle biopsies will be obtained prior to (within 2 weeks or on day of catheter placement) and at approximately 4 weeks after second cycle of GL-ONC1.
- PHASE 1b: On Day 248 (Week 36), an optional CT-guided biopsy to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- Clinical labs, PD and PK samples are collected according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.
- PHASE 2: For patients who have peritoneal ascites, 10 mL of ascites will be collected 7 days after date of first GL-ONC1 dose, if available. If the IP catheter is in place 14 days after date of the first GL-ONC1 dose, 10 mL can be drawn if ascites is available.
- PHASE 1b: For patients who have peritoneal ascites, 10 mL of ascites will be collected on Day 10, if available. If the IP catheter is in place on Day 17, 10 mL can be drawn if ascites is available.
- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. During this period, non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team. From Weeks 6 to 48, for patients who receive other anti-cancer treatment, only SAEs that the Investigator determines have a degree of attribution to GL-ONC1 treatment are reported; non-serious adverse events are not reported regardless of causality.
- Regardless of causality, deaths that occur from Week 2 to Week 5 (30 days post last GL-ONC1 infusion) are reported as an expedited safety report to the Pharmacovigilance Team.
- Deaths that occur from Week 6 to Week 48 are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the Death Report electronic Case Report Form (eCRF).

ANY COHORT: After completion of 'safety run-in cohort' for Ch D, and after treatment with GL-ONC1 and further chemo +/- bev, a second cycle of GL-ONC1 at 2,3,5 $\times 10^9$ pfu as an undulated bolus administered by IV route for 3 consecutive days is allowed (time point beyond Week 48 is also allowed) for 3 patients and expandable to another 3 patients upon approval by the Sponsor. PFS, OS, and Objective Response by RECIST1.1 and CA-125 should be documented after the second cycle of GL-ONC1 until disease progression, if clinically applicable. Since the second cycle of GL-ONC1 is given via IV route of administration, follow Cohort D study calendar for procedures and follow-up specifics.

If a patient receives other anti-cancer treatment, see the **General Statement** section above for procedures to follow.

Long-term Follow-up Period -- Phase 1b and Phase 2 Cohorts A & B:

- *During Year 2*, patients should come in on a quarterly basis for clinic visits as indicated in *Section 15.0 Study Assessments and Procedures*. The timing of the first follow-up visit is based on the timing of the Post-treatment Week 48 visit, or the last visit performed during the Post-treatment Period. If a patient receives other anti-cancer treatment, see the **General Statement** section above for procedures to follow.
- *During Year 3*, patients are telephoned quarterly to collect information on survival, disease and treatment status. The first quarterly follow-up call is based on the last Year 2 Long-term Follow-up visit.
- Deaths that occur during the Long-term Follow-up Period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the *Death Report* eCRF.

GL-ONC1 Treatment Period (Week 1) through Post-GL-ONC1 Treatment (Week 5) -- Phase 2 Cohort C & D (unless otherwise specified):

General Statement: The following procedures occur prior to and following treatment. Refer *Section 15.0 Study Assessments and Procedures* and relevant study calendars for more detailed information. Refer instructions provided in the **General Statement** section above for chemotherapy ± bev administered by local primary practitioner.

Enrollment will be by Sponsor assignment into either Cohort C or D.

Week 1 GL-ONC1 Treatment:

- Virus dose: 3 × 10^9 pfu/day for 2 consecutive days by IP route (except Cohort D). For Cohort D: 2,3,5 × 10^9 pfu in 3 consecutive days by IV route.
- **COHORT C:** For patients who have peritoneal ascites, as much ascites as possible will be drained prior to the instillation of the first GL-ONC1 dose. On the day after the second GL-ONC1 dose, 10 mL of ascites will be collected, if available. 10 mL of ascites will be collected 7 days (Week 2) after date of first GL-ONC1 dose, if available. If the IP catheter is in place 14 days (Week 3) after date of the first GL-ONC1 dose, 10 mL can be drawn if ascites is available.
- **COHORT C:** All patients will have 1 L to 1.5 L of Ringer's Lactate instilled and drained through the IP catheter prior to the first GL-ONC1 dose as a peritoneal wash.
- A preferred but optional post-treatment needle biopsy (e.g., CT-guided or ultrasound-guided) may occur between Week 2 to Week 5 after date of the first GL-ONC1 dose, and prior to initiation of further

chemotherapy to obtain a post-GL-ONC1 treatment tumor tissue sample from consenting patients if tumor is located in a reasonably safe area.

- Clinical labs, pharmacodynamic (PD) and pharmacokinetic (PK) samples are collected prior to treatment according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.
- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. Non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team.
- Deaths that occur during Treatment Week 1 through Week 5 (30 days post last GL-ONC1 infusion) are reported as an expedited safety report to the Pharmacovigilance Team regardless of causality (refer to *Section 21.11 Deaths* for reporting requirements).

Post-GL-ONC1 Treatment Period (Week 6 through Week 59 (q3W) or Week 65 (q4W) -- Phase 2 Cohort C:

Carboplatin doublet chemotherapy +/- bevacizumab treatment plan: refer to Study Calendars for *Treatment q3 Weeks and Treatment q4 Weeks* schedules. An example of regimen and dosing schedule is the OCEANS (Aghajanian C *et al. J. Clin Oncol* (2012) Jun 10;30(17):2039-2045) phase 3 trial evaluating the efficacy and safety of bevacizumab combined with gemcitabine + carboplatin (GC) for patients with platinum-sensitive recurrent ovarian cancer (ROC). Previous treatments with any of the regimens below are allowed. Cycle 1 of chemotherapy +/- bevacizumab treatment begins at Wk 6, irrespective of disease status from Wk 6 CT scan. The number of cycles of chemotherapy +/- bevacizumab may be expanded at the discretion of the Investigator if tolerated well and clinically determined to be beneficial after 6 cycles. Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

- Carboplatin doublet chemotherapy: Up to 6 cycles of carboplatin + a taxane (e.g., docetaxel (i.e., Taxotere)), paclitaxel (i.e., Taxol), *nab*-paclitaxel (i.e., Abraxane), or + gemcitabine (i.e., Gemzar), or + PLD (i.e., Doxil). Chemotherapy component could be substituted if due to toxicity or by Investigator's choice. If due to platinum-allergy, a single-agent non-platinum chemotherapy is allowed.
- Suggested chemotherapy intravenous (IV) dose: carboplatin (AUC 4 to 5, q3w) with gemcitabine (800 mg/m^2, D1&D8, q3w); carboplatin AUC 5 to 6 with docetaxel (60 to 75 mg/m^2 q3w), or with paclitaxel (175 mg/m^2, q3w), or with nab-paclitaxel (260 mg/m^2, q3w), or with PLD (40 mg/m^2, q4w). Dose of chemotherapy may be adjusted to allow better tolerability.
- Suggested bevacizumab (bev) dose: 10 mg/kg, q3w with chemotherapy (7.5 mg/kg, q2w with PLD), and as single agent maintenance q3w for another 9 months. Dose adjustment is allowed per Investigator's discretion.

Chemotherapy doublet +/- bevacizumab (bev), both q3 weeks:

Baseline **CT scan**, ideally within 2 weeks (3-4 weeks allowed) prior to 1st dose of GL-ONC1
Days 1 & 2 (W1), GL-ONC1
↓
W6, **CT scan** immediate prior to 1st cycle of chemo + bev
↓
W9, 2nd cycle of chemo + bev
↓

W12, 3rd cycle of chemo + bev
↓ W14, **CT scan**
W15, 4th cycle of chemo + bev
↓
W18, 5th cycle of chemo + bev
↓
W21, 6th cycle of chemo + bev
↓ W23, **CT scan**
W24, start of maintenance bev for additional 9 mos
↓ W35, 47, 59, then PRN for **CT scans**

For chemotherapy doublet at q4 weeks with bev at q2 weeks:

Baseline **CT scan**, ideally within 2 weeks (3-4 weeks allowed) prior to 1st dose of GL-ONC1
Days 1 & 2 (W1), GL-ONC1
↓
W6, **CT scan** immediate prior to 1st cycle of chemo + bev
↓ W8, bev
W10, 2nd cycle of chemo + bev
↓ W12, bev
W14, 3rd cycle of chemo + bev
↓ W16, bev
↓ W17, **CT scan**
W18, 4th cycle of chemo + bev
↓ W20, bev
W22, 5th cycle of chemo + bev
↓ W24, bev
W26, 6th cycle of chemo + bev
↓ W28, bev
↓ W29, **CT scan**
W30, start of maintenance bev for additional 9 mos
↓ W41, 53, 65, then PRN for **CT scans**

Post-GL-ONC1 Treatment Period -- Phase 2 Cohort D:

Treatment with chemotherapy ± bevacizumab will start at the time of disease progression (preferred by RECIST 1.1) from GL-ONC1, or will start at the discretion of the Investigator as clinically indicated based on clinical assessment (in consultation with Sponsor if disease progression is clinically determined). Combination with bev is allowed but not required. If patient develops toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment (e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel) to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if well tolerated and determined to be clinically beneficial after 6 cycles. Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev until progression or

unacceptable toxicity, at the discretion of the Investigator. **Refer to information provided above for Cohort C for examples of chemotherapy regimens and time lines.**

Post-GL-ONC1 Treatment Period (Week 6 through Week 59 (q3W) or Week 65 (q4W) -- Phase 2 Cohorts C & D (timeline adjusted accordingly based start of chemo):

- Deaths that occur during this period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements) taking into consideration that chemotherapy agents and bevacizumab are administered during this study period. If no attribution to GL-ONC1, report death on the Death Report electronic Case Report Form (eCRF).
- Imaging by spiral CT scans & RECIST assessment are performed according to time points of treatment depending on whether time frame for chemotherapy is q3 weeks or q4 weeks. During this period, time points include: following baseline, immediately prior to cycle 1, 1-wk prior to cycle 4, 2-3 weeks post cycle 6 (i.e., 1-wk prior to start of bev maintenance), and then every 3 mos for 3 more scans. Further scans will be PRN and at the discretion of the Investigator and the Sponsor. Refer to specific study calendar for imaging time points. For patients allergic to IV CT contrast, it is recommended that oral CT contrast (preferred) or MRI may be used. Per RECIST 1.1, the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up.
- Another optional post-treatment needle biopsy may be obtained after second cycle (either during Week 11 for q3w, or Week 13 for q4w with ± 1 week for both time points) or after third cycle (Week 14 for q3w, or Week 17 for q4w with ± 1 week for both time points) of chemotherapy. Biopsy will be obtained after CT scan if they are conducted on same day.

Long-term Follow-up Period -- Phase 2 Cohorts C & D:

- *During Year 2 and Year 3*, patients should come PRN at Investigator discretion (e.g., quarterly basis) for clinic visits as indicated in *Section 15.0 Study Assessments and Procedures*. The timing of the first follow-up visit is based on the timing of the 30-day following the last cycle.
- If patients are unable to come to clinic, quarterly telephone calls occur to collect information on survival, disease and treatment status. The first quarterly follow-up call is based on the last Long-term Follow-up visit.
- Deaths that occur during the Long-term Follow-up Period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the *Death Report* eCRF.

Allowable Window of Time around Study Visits: During Week 1 of the Treatment Period, a window of ± 1 day for each procedure is allowed. During the Post-treatment Period, there is a ± 2-day window, and a ± 1 week for imaging studies. There is a ± 7-day window of variance for the optional post-treatment biopsies. For Phase 2 Cohorts C & D, in general there is a ± 7-day window allowable for treatments, procedures, and imaging scans, unless there is a further delay due to toxicity from chemo +/- bev requiring additional recovery time. The allowable window of variance during the Long-term Follow-up Period is 2 weeks. Documented delays that occur outside of the allowable window of variance due to holidays, weekends, weather, or other unforeseen circumstances do not constitute a protocol deviation; refer to *Section 4.5 Treatment Compliance Criteria* for exceptions.

Treatment Compliance Criteria: Treatment breaks must be clearly indicated in the treatment record along with the reason(s) for the treatment break(s). Missed treatments due to holidays or logistic reasons can be compensated for by delivering the additional treatments as soon as the

Investigator determines is feasible, or by treating on a Saturday or Sunday. Treatment breaks should be allowed for resolution of severe acute toxicity and/or intercurrent illness, and ideally should not exceed 5 treatment days at a time (see exception above for Phase 2 Cohort C allowable treatment break).

Patient Replacement: Replacement patients may be enrolled in a cohort if a patient does not receive at least 1 dose of GL-ONC1 and have been withdrawn from investigational treatment for a reason that is unrelated to study agent toxicity.

Optional In-patient Stay: A patient is recommended to have an optional in-patient stay during and/or after treatment for observation with the duration determined by the study-affiliated treating physician (suggested duration is for 48 hours post-second dose of GL-ONC1). Any in-patient or out-patient care (e.g., hydration, etc.) will be in accordance with local standard medical practices.

1.0 INTRODUCTION AND RATIONALE

1.1 Study Rationale

Treatment agents: GL-ONC1-015 is an open-labeled, non-randomized Phase 1b & 2 study of GL-ONC1, an oncolytic vaccinia virus, administrated intraperitoneally in patients with recurrent or refractory ovarian cancer and peritoneal carcinomatosis.

GL-ONC1 as monotherapy: It was shown preclinically that GL-ONC1 (laboratory name: GLV-1h68) could efficiently infect (1) and kills ovarian cancer cells *in vitro* and *in vivo* (unpublished results). Enhanced tumor tissue necrosis and delayed tumor growth were observed in OVCAR-3 xenograft model from virus treatment. The rationale is to further test GL-ONC1 at the proposed dosing schedule and route of administration. Based on early phase information from 4 separate Phase 1 trials, we do not anticipate any new toxicity signals.

GL-ONC1 with combination therapy: The rationale for combining oncolytic vaccinia virotherapy, chemotherapy, and targeted therapy (bevacizumab) is to use drugs that work by different mechanisms to reduce and/or reverse resistant mechanisms, and at the same time mutually potentiate the therapeutic effects of each agent. Combining GL-ONC1 with well-timed chemotherapy of choice, such as those could remove inhibitory Tregs and MDSCs, one can envision an optimal treatment regimen with significantly enhanced clinical results. In a completed Phase 1 trial, we have combined GL-ONC1 with cisplatin and radiotherapy as standard of care (SOC) in newly diagnosed pts with locally advanced head and neck cancer (Mell *et al*., Clin Cancer Res. 2017;23(19):5696-702). We have shown that the combination is well tolerated, with a favorable trend of survival benefits as compared to well-documented historical numbers from SOC. Combination therapy of bevacizumab with chemotherapy in platinum-resistant recurrent ovarian cancer (≤ 2 prior lines of treatment) was studied in the AURELIA Phase 3 trial (NCT00976911). Comparing chemo alone vs. with bevacizumab, median PFS was 3.4 mos vs. 6.7 mos, ORR was 11.8% vs. 27.3%, and GCIG CA-125 response was 11.6% vs. 31.8%, indicating the clinical benefits of combination therapy.

Data (see *Section 2.6 Overview of Clinical Trial Experience with Oncolytic Viruses*, updated summary for **GL-ONC1-015** trial) from Phase 1b part of this trial have demonstrated clinically significant results in this heavily pretreated patient population (median ≥ 5 prior lines of treatment), including (1) evidence of anti-tumor activities, e.g., stabilized and/or reduced CA-125 tumor biomarker, tumor shrinkage by RECIST 1.1 (including objective response), reduction in circulating tumor cells (CTC), and encouraging Disease Control Rate (DCR = OR + SD≥15 weeks) of 55%. The Phase 2 expansion of this trial is to further investigate anti-tumor response of GL-ONC1 monotherapy or combination therapy to observe trend of clinical benefits in a larger number of patients.

Method of delivery for GL-ONC1: GL-ONC1 has shown tumor-selective replication if administered intravenously (IV) or intraperitoneally (IP) in animal models. GL-ONC1 has also been administered IP as a single dose of 1×10^7 pfu to 1×10^9 pfu in a Phase 1 study (NCT01443260) in 9 patients with various malignancies (total number of IP infusions was 24). GL-ONC1 was well tolerated with the majority of adverse events being mild-to-moderate which included transient flu-like symptoms and increased abdominal pain (resulting from treatment-induced peritonitis). No deaths were attributed to GL-ONC1, and no dose limiting toxicities (DLTs) were reported. Furthermore, no viral shedding was observed. In 8 out of 9 patients, effective intraperitoneal infection and in-patient replication of GL-ONC1 was demonstrated by viral plaque assay (VPA), as well as subsequent oncolysis demonstrated by the release of GL-ONC1 encoded ß-glucuronidase. However, all 9 patients developed neutralizing antibodies (NAb) against GL-ONC1 with the earliest detection beginning on Day 8 following initiation of treatment.

For the Phase 1b clinical trial, it is intended to intensify the administration of GL-ONC1. GL-ONC1 was also shown to be well tolerated with individual IV doses up to 5×10^9 pfu (NCT00794131), or as individual intrapleural (IPL) doses up to 6×10^9 pfu (NCT01766739) in two other Phase 1/1b trials. The highest cumulative dose we have tested in previous trials is at 4×10^{10} pfu (intravenous route; NCT00794131), which was shown to be well tolerated. In addition, 5 patients have received and tolerated cumulative doses of 9×10^9 pfu and above given within the first week (3 patients received intrapleural dose total of 9×10^9 pfu; 1 received IV dose total of 1×10^{10} pfu; 1 received intrapleural dose total of 1.8×10^{10} pfu). Therefore, we expect the starting IP dose of 3×10^9 pfu through a regional delivery route, as proposed in this trial, will be well tolerated.

Phase 2 will use the same dose levels as the Phase 1b Cohorts 1 and 2: Cohorts A and C (3×10^9 pfu × 2 consecutive doses) and Cohort B (1×10^{10} pfu × 2 consecutive doses), respectively. Safety evaluation from the Phase 1b study documented these dose levels were well tolerated with no DLT or MTD.

In the Phase 2 Cohort D, oncolytic virus by intravenous (IV) delivery could reach distal metastatic sites beyond the peritoneal cavity. This would allow enrollment of patients in this trial who do not have measurable disease in the peritoneal cavity, but do have measurable disease outside the peritoneal cavity. IV delivery of GL-ONC1 has been investigated in early phase clinical trials (see *Section 2.6 Overview of Clinical Trial Experience with Oncolytic Viruses).* The aims of Cohort D is (1) to assess safety and clinical efficacy of IV delivered GL-N/C1 followed by chemotherapy ± bevacizumab, and (2) to investigate the mechanism of interaction between IV delivered GL-ONC1 and chemotherapy ± bevacizumab.

1.2 Research Hypothesis

It is hypothesized that GL-ONC1 is well tolerated with enhanced antitumor activity as monotherapy or combination therapy in patients diagnosed with recurrent or refractory ovarian cancer and peritoneal carcinomatosis.

1.3 Study Objectives

Primary Objective:

- Phase 1b: To investigate the safety and tolerability.
- Phase 2 (Cohorts A, B & D): Investigate Progression-free Survival (PFS) with therapeutic intent.
- Phase 2 (Cohort C & D): Overall Response Rate (ORR) by RECIST1.1 and by GCIG CA-125 criteria with therapeutic intent.

Secondary Objectives:

Phase 1b & Phase 2 Cohorts A & B: Response to treatment with therapeutic intent will be determined based on Response Evaluation Criteria in Solid Tumors (RECIST) 1.1 (2), by the Immune-related Response Criteria (irRC) (3) as an exploratory endpoint, and by CA-125 according to the Gynecologic Cancer Intergroup (GCIG) CA-125 response criteria (4) in patients with measurable disease. Patients who enter with non-measurable disease will be as evaluated for response to treatment by PET/PET CT scan, physical examination and CA-125 levels. Antitumor (if evaluable by CT, PET/PET-CT scans) activity will be evaluated by PFS, Clinical Benefit Rate (CBR = CR + PR + SD), best overall response (BOR), duration of response (DoR), objective overall response rate (ORR), disease control rate (DCR = CR + PR + SD ≥15 weeks). Additionally, overall survival (OS) and time to treatment failure (TTF) will be assessed. In Phase 2, safety assessments will continue.

Phase 2 Cohort C & D:

- Progression-free Survival by RECIST1.1 (Ch. C)
- Duration of Response
- Clinical Benefit Rate
- Disease Control Rate
- Overall Survival

1.4 Exploratory Objectives (optional):

All cohorts:

(1) To evaluate the immune response to treatment, immune assays will be performed to study immune activation and antitumor immune response in blood, ascites and tumor biopsies;
(2) To evaluate antitumor activity at the tissue/cellular levels (proliferation index, apoptosis, etc.);
(3) To confirm the presence of GL-ONC1 within the tumor by VPA, IHC, and/or qPCR;
(4) To determine the possible prognostic value (e.g., as a predictive value of survival outcome) of circulating tumor cells (CTCs) in patients diagnosed with ovarian cancer. In addition, to demonstrate the correlation of CTC number with radiological outcome as early pharmacodynamic and response rate indicators in the context of GL-ONC1 treatment.

1.5 Rationale for Immune-related Response Criteria (irRC) and Evaluation as an Exploratory Endpoint

Accumulating clinical evidence indicates that some patients treated with agents that activate antitumor immune responses may develop progression of disease (by conventional response criteria) before demonstrating clinical objective responses and/or durable stable disease (3).

Two hypotheses have been put forth to explain this phenomenon. First, enhanced inflammation within tumors could lead to an increase in tumor size which would appear as enlarged index lesions and as newly visible small non-index lesions. Over time, both the malignant and inflammatory portions of the mass may then decrease leading to overt signs of clinical improvement. Alternatively, in some individuals the kinetics of tumor growth may initially outpace antitumor immune activity. With sufficient time, the antitumor activity will dominate and become clinically apparent.

As an example, following treatment with ipilimumab, serial biopsies provided histopathological evidence that radiographically-defined enlarging tumor lesions can be the result of an influx of tumor infiltrating lymphocytes (3). Therefore, early increases in lesion size detected radiographically, or upon gross examination, could be misinterpreted as progressive tumor growth and precede objective tumor

shrinkage. In addition, the appearance of new lesions may have categorized a patient to have progressive disease using conventional tumor assessment criteria despite the concurrent observation of objective tumor responses in preexisting lesions and a net reduction in global tumor burden that includes the new lesions. Hence, the appearance of new lesions, in and of themselves, may not necessarily constitute progressive disease.

Based on the distinct patterns of clinical responses observed in patients treated with immunotherapy agents such as ipilimumab, and which differ from those seen in patients treated with other classes of anti-cancer agents, new exploratory immune-related Response Criteria (irRC) are described *(Appendix 2: Immune-related Response Criteria)*. The irRC is a refinement of conventional response criteria created to systematically capture tumor response in patients on immunotherapy (3). The major modifications to the conventional criteria are the following: (a) a requirement to 'confirm' progression at least 4 weeks after scan indicating initial progression and, (b) not scoring new, small non-target lesions as evidence of progression. Rather, the net tumor burden (which may include new small non-target lesions) is used to gauge progression. In the case of ipilimumab, the irRC identified 9.7% of patients (22/227 treated patients) from studies CA184022 and CA184008 who demonstrated disease control in the form of stable or reduced measurable tumor burden, including new lesions, at or after disease progression.

In this study, irRC will be evaluated on an exploratory basis by the Sponsor. The primary mode of tumor response assessment will be based on RECIST 1.1 criteria (2).

1.6 Rationale of Circulating Tumor Cells as an Exploratory Endpoint

Optional analysis of circulating tumor cells (CTC) may be included in any cohorts. As this is an optional Exploratory Objective, the number of patients in any cohorts to be tested for CTC will be at the discretion of the Sponsor. Fresh whole blood samples will be immediately shipped to testing facility for analysis using the CellSearch system.

Several studies have shown that circulating tumor cells (CTCs), over a specified cut-off value, predicts for a worse prognosis in terms of progression free survival (PFS) and overall survival (OS) in breast, prostate and colorectal cancer population (5-7).

Common to all of these studies is that CTCs, prior to commencing a new line of treatment, have prognostic value in breast, prostate and colorectal cancer. Furthermore, changes in CTCs during treatment predict survival outcome, highlighting their potential as a surrogate endpoint biomarker (8).

Studies correlated CTCs with response to imaging according to Response Evaluation Criteria in Solid Tumors (RECIST) have also been performed (9-11). Although these studies were heterogeneous, the results favored CTCs over conventional imaging to be the most significant factor in determining survival prognosis. Results should be interpreted with caution in patients with stable disease (SD) and with no CTC changes.

In conclusion, CTCs may be used to predict the activity of new drugs in the drug development setting, potentially facilitating short time go/no-go decision-making between baseline CTCs and assessment of CTCs at subsequent time points (8). Whilst data are emerging on several cancer types, there remains little information regarding CTCs and other malignancies (melanoma, non-small cell lung cancer, etc.), so much more research is required (12,13).

Cui *et al.* (14) performed a meta-analysis of clinical studies which included CTCs and disseminated tumor cells (DTC) that showed a strong relationship of CTCs and DTCs with advanced staging, response to treatment and poor prognosis in patients with ovarian cancer. Poveda *et al.* (15) found that CTCs were

prognostic during ovarian cancer therapy and that patients with ≥ 2 CTCs at baseline had a shorter overall survival and shorter time to progression.

Therefore, an optional exploratory endpoint is to investigate the correlation of the number of CTC in survival outcome and response to treatment in women diagnosed with ovarian cancer.

Phase 1b: Whole blood samples for CTC analysis are collected based on the number of CTCs reported at baseline prior to the first GL-ONC1 infusion:

1. 5+ CTCs: Collect samples on W3/D17, W15/D101 and W24/D164.
2. 1-5 CTCs: Collect samples on W3D17. If this result is 5+ CTCs, continue sample collection on W15/D101 and W24/D164.
3. 0 CTCs: Do not collect any additional samples.

Phase 2 (Cohorts A & B): At the Sponsor's discretion, and regardless of baseline CTC count, samples are collected at baseline and at W3 (14 days after date of first GL-ONC1 dose), W6, W15 and W24.

Phase 2 (Cohort C): At the Sponsor's discretion, and regardless of baseline CTC count, samples are collected at baseline and at relevant time points in relationship to different treatments if exactly following schedule: W3 (14 days after date of first GL-ONC1 dose) and W5 to W6. For q3W schedule collect at W12 and W21. For q4W schedule collect at W17 and W29.

Phase 2 (Cohort D): Follow time lines above for Phase 1b until chemo ± bev is administered and then follow Phase 2 Cohort C time lines.

2.0 BACKGROUND

2.1 Ovarian Cancer

Ovarian cancer (OC) remains the most lethal gynecologic malignancy owing to late detection, intrinsic and acquired chemo-resistance and remarkable heterogeneity. Despite optimization of surgical and chemotherapy protocols, and initiation of clinical trials incorporating targeted therapy, only modest gains have been achieved in prolonging of survival of OC. Therefore, there is an unmet medical need to develop new therapy modalities.

In platinum-resistant ovarian cancer, the combination of non-platinum single agent therapies with bevacizumab have shown a significant increase of PFS from 3.4 months to 6.7 months, and an increase of survival from 13.3 months to 16.6 months in patients had ≤ 2 prior lines of treatment (16). On the basis of these results, BEV, a vascular endothelial growth factor receptor antibody, was approved in November 2014 by the US Food and Drug Administration (FDA) for recurrent platinum-resistant ovarian cancer in combination with single-agent chemotherapy.

2.2 Oncolytic Viruses

Oncolytic viral therapy is a novel approach to the treatment of cancer. By targeting target genetic abnormalities in cancer cells, oncolytic viruses selectively infect and replicate in cancer cells and spare adjacent, normal cells. Viral replication, subsequent cell lysis and further viral spread then occurs selectively within cancer cells. Oncolytic viruses may kill cancer cells by a number of mechanisms including virus replication-associated necrosis (i.e., 'oncolysis'), induction of tumor-specific T lymphocytes, induction of bystander cell killing, and by viral induction of changes in tumor-associated vasculature (17,18). Currently, a number of oncolytic viruses are at various stages of clinical development. At least three oncolytic viruses are currently being investigated in Phase 3 clinical trials in

a range of clinical settings (Onyx-015 adenovirus, GM-CSF-encoding herpes virus, and reovirus in combination with chemotherapy). Concerning this worldwide effort, the ideal oncolytic virus would be completely tumor-specific in its action with negligible systemic or local toxicities and would be able to infect or exert cytopathic effects on all tumor cells within an organism. It would be easily manufactured and stored for widespread use and would be able to circumvent potential immune down-regulation. None of the viruses currently under investigation have the ability to fulfil this idealistic phenotype. However, recent work investigating vaccinia virus as an oncolytic agent suggests that it may satisfy many of the above criteria.

2.3 Vaccinia Virus

Vaccinia virus (VACV) is the prototype member of the genus Orthopoxvirus (subfamily Chordopoxviridae; family Poxviridae). VACV is a large and complex particle containing a single linear double-stranded DNA genome of approximately 190 kb with inverted terminal repeats and terminal hairpin loops. The DNA genome of a number of strains of VACV has been sequenced and found to encode approximately 150-200 proteins. VACV replicates entirely in the cytoplasm of infected cells and the virus particle contains all enzymes required for the synthesis of capped (methylated) poly-adenylated mRNAs. This particular characteristic of VACV mitigates its potential for mutagenesis by incorporation into the host genome.

VACV has many characteristics desirable in an oncolytic virus for clinical applications:

- short, well-characterised life cycle, spreading very rapidly from cell to cell;
- highly cytolytic for a broad range of tumor cell types;
- large insertion capacity (> 25 kb) for the expression of exogenous genes;
- high genetic stability;
- amenable to large scale production of high levels of active virus;
- lack of a known natural host;
- not pathogenic in humans;
- remains in the cytoplasm and does not enter the host cell nucleus during the entire life cycle, and thus does not integrate into the host genome;
- used previously as smallpox vaccine in millions of people with well-documented and low incidence side effects;
- susceptible to rescue drugs (e.g. vaccinia immunoglobulin, cidofovir, ST-246) if necessary for any potential vaccinia-related infections.

2.4 Description of GL-ONC1

Genelux Corporation has genetically engineered an oncolytic vaccinia virus, designated as GLV-1h68 (laboratory name for GL-ONC1). GLV-1h68 was derived from the LIVP strain (Lister strain obtained from the Institute of Viral Preparations, Moscow) by inserting *Ruc-GFP* (a fusion gene of *Renilla* luciferase and green fluorescent protein), *lacZ* (β-galactosidase gene from *E. coli*), and gusA (β-glucuronidase gene from *E. coli*) expression cassettes into *F14.5L* (located between *F14L* and *F15L*), thymidine kinase (*TK*), and hemagglutinin (*HA*) loci; respectively. Disruption of these nonessential genes and expression of the foreign gene expression cassettes not only attenuated the virus but also enhanced its tumor-specific targeting ability. GLV-1h68 has been utilized in many *in vitro* cytotoxicity studies and preclinical efficacy studies in animals (19-22).

The Good Manufacturing Practices (GMP)-derived material of this same virus is called GL-ONC1, which is also its proprietary name. GL-ONC1 has been used primarily for safety, pharmacological, and toxicological experiments performed at the Bioservice Scientific Laboratories GmbH (BSL), and Comparative Biosciences Inc. (CBI), both of which are contracted Good Laboratory Practices (GLP)

laboratory). It has also been investigated in *in vitro* potency comparisons (in cell cultures) and *in vivo* potency comparisons (in tumor-bearing animals).

GL-ONC1 is considered a Biosafety Level 2 (BSL-2) substance.

2.5 Non-clinical Efficacy Studies

In vitro, a large panel of tumor cells could be killed efficiently by GLV-1h68 (1).

Various *in vivo*, preclinical experiments in which GLV1h68 was delivered via intravenous, intratumoral or intrapleural delivery of this virus resulted in the complete eradication or significant growth inhibition of several types of large subcutaneous xenograft tumors (including breast, lung, colon, prostate, ovarian, pancreatic, thyroid, and skin), and an orthotopic mesothelioma model in nude mice with no significant adverse effects. Refer to the *GL-ONC1 Investigator's Brochure* for more information.

Effect of GL-ONC1 on Ovarian Xenograft Growth with or without Cisplatin

The therapeutic effect of GL-ONC1 on the progression of human ovarian OVCAR-3 tumors was evaluated in a direct *in vivo* study with female nude mice by measuring the volume of the tumor at various time points. Administration of GLV-1h68 intravenously at 1×10^7 pfu per animal was able to slow tumor growth by 65% at 23 days after virus injection. In addition to single dose virus treatment, combination therapy of ovarian tumors with virus (2×10^6 pfu of GLV-1h68 per animal via tail vein) and cisplatin (5 mg/kg, IP once per day on days 10, 11, 13, and 14 after the virus injection) in comparison to virus or cisplatin treatment alone was also conducted. Tumor eradication was achieved in the combination therapy group approximately two months after the initiation of virus treatment.

Effect of GL-ONC1 combined with bevacizumab

Bevacizumab (Avastin) is a target-therapy against VEGF. Vaccinia has both vascular disrupting and anti-angiogenic properties. In preclinical studies, we have demonstrated robust anti-tumor effect when GL-ONC1 (i.e., GLV-1h68) was combined with Avastin (Frentzen *et al*., PNAS 2009;106(31):12915-20). Vaccinia-induced vascular collapse within the tumor is both rapid and extensive (Hou *et al*., Int J Cancer. 2014;135(5):1238–46). It has been hypothesized that direct viral effects on vasculature and VEGF levels are capable of suppressing angiogenesis during the period of viral infection, and that further combination with additional anti-angiogenic therapies can be highly effective, especially when these are added at times "after" viral clearance (Hou *et al*., Int J Cancer. 2014;135(5):1238–46). In addition to bevacizumab, such anti-angiogenic therapies may also include tyrosine kinase inhibitors targeting VEGF and PDGF pathways.

The sequence and timing of anti-VEGF therapy in relationship to vaccinia virotherapy need to be carefully considered. It has been shown that VEGF derived from tumor cells acts to increase vaccinia internalization, resulting in increased replication and cytotoxicity in an AKT-dependent manner (Hiley *et al*., J Virol. 2013;87(5):2781-90). Concurrent treatment with bevacizumab could therefore hinder initial virotherapy. The VEGF molecule is considered immunosuppressive because it directly and indirectly inhibits T-cell function (Bamias *et al*., Gynecol Oncol. 2008;108(2):421-7). Also, VEGF stimulates immunosuppressive regulator T cells, inhibits dendritic cell function, reduces the adhesion of lymphocytes to vessel walls, and induces abnormal tumor vasculature. Therefore, inhibition of VEGF with bevacizumab subsequent to virotherapy could positively impact the immune system and aid virotherapy.

Non-clinical Safety Studies

Biodistribution, persistence, safety, pharmacology, toxicity, and horizontal transmissibility studies were conducted in both immunocompetent and immunocompromised animals by Genelux laboratories, BSL,

and CBI. Details of these non-clinical safety studies are found in the *GL-ONC1 Investigator's Brochure*.

Horizontal Transmissibility

To study the potential transmissibility of the virus, the single-dose toxicology studies each included an additional group of three male and three female animals that were caged together with the animals of the high dose group.

In both mouse and rat studies, the animals were observed for 21 days after dosing. A careful clinical examination was made once daily. At the end of the observation period, the animals were sacrificed and necropsy was carried out to record gross pathological changes.

In both studies, there were no signs of horizontal transmissibility. All animals survived. Normal weight gain was observed and no detectable amounts of the virus were found in organs, blood or feces.

2.6 Overview of Clinical Trial Experience with Oncolytic Viruses

Early evidence of oncolytic activity of vaccinia strains

Several clinical investigations (see below) have used recombinant vaccinia virus for the treatment of cancer. A 1987 study in the *Japanese Journal of Experimental Medicine* reported successful treatment of a 67-year-old male patient with multiple myeloma using an intravenous injection of the AS strain of VACV (1×10^8 pfu) in varying volumes daily for one week and every other day for the next 13 weeks (23). By the 106th day of treatment, the patient's NK cell activity had risen to 33% from the initial 20% on the tenth day. No adverse effects were observed. The patient remained in remission three years after treatment. Later that year, the same research group reported that they had achieved favorable antitumor effects using the AS strain in two cases of advanced adenocarcinoma without observing any adverse reactions except high fever and general malaise after the first administration in one of the patients (24). A 76-year-old male patient received a total of 22 intravenous injections of the virus (1×10^8 pfu) every other day, followed by 20 daily injections of 2×10^8 pfu. The second patient, a 55-year-old woman, received an intravenous injection of 2×10^8 pfu three times per week for about three weeks, then daily administration of 4×10^8 pfu for 12 months. These studies drew upon earlier evidence of vaccinia viruses as potential therapy for human cancers. In 1978, a case study reported that a 78-year-old man with untreated chronic lymphocytic leukemia went into complete remission after revaccination for smallpox. He endured a severe local reaction and generalized rash following the vaccination, but responded well to treatment with human vaccinia immune globulin (25). Other medical practitioners have confirmed these findings with responses of patients vaccinated for smallpox (26).

Safety profile of vaccinia virus as a smallpox vaccine

The role of vaccinia virus as a vaccine for smallpox has provided the medical and research community with a long history of clinical use from which to draw safety data. In 1968, when the vaccine was still a common immunization tool, epidemiologists collected data on adverse events related to vaccination both in a national and in a multi-state survey (27,28). The results of their work are represented in the table below.

Smallpox Vaccine: Adverse Event Rates, 1968 (number per million vaccinees)

	NATIONAL SURVEY		TEN STATE SURVEY	
	All primary (i.e., first-time) vaccinees	**Vaccinees ≥ 1 yr old**	**All primary (i.e., first-time) vaccinees**	**Vaccinees ≥ 1 yr old**
	Serious, but not life-threatening reactions			
Inadvertent Inoculation	25.4	27.1	529.2	532.0
Generalized Vaccinia	23.4	17.7	241.5	222.8
Erythema Multiforme	N/A	N/A	164.6	131.3
Total number of serious, but not life-threatening reactions	**48.8**		**935.3***	
	Life-threatening reactions			
Postvaccinal Encephalitis	2.9	2.4	12.3	8.6
Progressive Vaccinia (Vaccinia Necrosum)	0.9	1.0	1.5	1.7
Eczema Vaccinatum	10.4	10.6	38.5	41.5
Total number of life-threatening reactions	**14.2***		**52.3***	
Deaths	**1.1***	0.6	1.5	none reported

*Adverse event statistics cited in table are marked with an asterisk.

These two epidemiological studies used different methodologies in data collections. In the ten-state survey, clinicians were actively contacted and urged to report all adverse reactions, including those considered less severe, therefore, it may have overestimated the risks of severe adverse reactions. The range of frequencies for these two studies, however, provides an estimate of the frequencies of adverse reactions that might be expected with the New York City Board of Health (NYCBOH) strain vaccination for first-time vaccinees.

Vaccinia virus for cancer therapy

Lattime and Mastrangelo performed the initial studies utilizing VACV for cancer gene therapy in human patients (29-32). In some of these studies, injection of vaccinia recombinants expressing immune-stimulating cytokines, such as GM-CSF, were shown to be well tolerated. More recently, additional safety data have been obtained in clinical studies using the same vaccinia-GM-CSF (33,34).

Human Clinical Experience with GL-ONC1

GL-ONC1 has been investigated in early stage clinical trials in the United States and Europe via systemic delivery as monotherapy and in combination with other therapies, and via regional delivery as monotherapy. The table below summarizes the GL-ONC1 clinical trial designs. The total number of patients treated with GL-ONC1 at the time of this submission was 126 patients. The starting dose used in the first-in-human clinical trial was based on preclinical results.

GL-ONC1 Human Clinical Trial Summary

Protocol Number	Indication	Modality & Route	Dose & Regimen	Number of Patients Treated / Number of Doses
GL-ONC1-002/MA (United Kingdom; first-in-human)	Advanced solid tumors	Monotherapy as intravenous infusion or bolus injection	**Single Dose/Cycle** - 28 day cycle - Dose: 1 × 10^5 pfu up to 3 × 10^9 pfu **Multiple Dose/Cycle** - 28 day cycle - Dose: 1.667 × 10^7 pfu to 1.667 × 10^8 pfu × 3 consecutive days/cycle **Multiple/Single Dose/Cycle** - Either 28, 14 or 7 day cycles - Cycle 1 Dose: 1.667 × 10^9 pfu × 3 consecutive days - Cycles 2-6 Dosing: Single dose at either 3 × 10^9 pfu or 5 × 10^9 pfu/cycle	43 # of Doses: 233
GL-ONC1-003/MSK (United States) (Investigator-initiated)	Malignant pleural effusion including primary, metastases, & mesothelioma	Monotherapy as intrapleural catheter delivery	- Single dose, 3 consecutive doses - Dose: 1 × 10^7 pfu to 6 × 10^9 pfu (multiple dose cohort)	18 # of Doses: 32
GL-ONC1-004/TUE (Germany)	Peritoneal carcinomatosis	Monotherapy as intraperitoneal catheter delivery	- Treatment once per cycle (every 28 days) for up to 4 cycles - Dose: 1 × 10^7 pfu up to 1 × 10^9 pfu/cycle	9 # of Doses: 24
GL-ONC1-005 (United States)	Head and neck cancer	Combination therapy with intravenous or bolus injection with cisplatin and radiotherapy	- 1, 2 or up to 4 treatments - Dose: 3 × 10^8 pfu up to 3 × 10^9 pfu	19 # of Doses: 34
GL-ONC1-011 (United States) (Investigator-initiated)	Solid Organ Cancer	Monotherapy as intravenous bolus infusion	- 3 or 5 daily treatment during Week 1 - Dose: 2 × 10^9 pfu × 5 daily doses or 2,3,5 × 10^9 pfu	5 # of Doses: 21
GL-ONC1-015 (United States)	Recurrent/ Refractory Ovarian Cancer	Monotherapy as intraperitoneal catheter delivery	- 2 consecutive doses during Week 1 - Dose: 3 × 10^9 pfu up to 2.5 × 10^{10} pfu	26 # of Doses: 52
GL-ONC1-021 (EAP Study) (United States)	Advanced cancers (solid & blood cancer)	Monotherapy as intravenous bolus infusion	- 3 or 5 daily treatment during Week 1 - Dose: 2 × 10^9 pfu × 5 daily doses or 2,3,5 × 10^9 pfu	6 # of Doses: 28

Safety: The data presented in the table below represents adverse reactions attributed by the Investigator to GL-ONC1 reported to date for all adverse reactions with ≥ 5 occurrences (N=represents total number of reported adverse reactions; N% = fractional percent of reported adverse reactions per total adverse reactions).

Reported GL-ONC1 Adverse Reactions

Reaction	GL-ONC1-002/MA N = 262 (%)	GL-ONC1-003/MSK N = 91 (%)	GL-ONC1-004/TUE N = 109 (%)	GL-ONC-005 N = 45 (%)
General disorders and administration site conditions (n=153 occurrences)				
Pyrexia	48 (19.3%)	16 (17.6%)	18 (16.5%)	7 (15.6%)
Chills	23 (9.2%)	10 (11.0%)	-	8 (17.7%)
Rigors	18 (7.4%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Flu-like symptoms	0 (0.0%)	5 (5.5%)	0 (0.0%)	0 (0.0%)
Investigations (n= 103 occurrences)				
aPTT increased	11 (4.4%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
GGT increased	11 (4.4%)	0 (0.0%)	-	0 (0.0%)
ALP increased	10 (4.0%)	8 (8.8%)	-	0 (0.0%)
AST increased	9 (3.6%)	6 (6.6%)	-	0 (0.0%)
ALT increased	8 (3.2%)	7 (7.7%)	-	0 (0.0%)
C-reactive protein increased	8 (3.2%)	0 (0.0%)	20 (18.3%)	0 (0.0%)
Creatinine increased	5 (2.0%)	0 (0.0%)	-	0 (0.0%)
Blood and lymphatic system disorders (n= 70 occurrences)				
Lymphocyte count decreased	0 (0.0%)	-[1]	26 (23.8%)	0 (0.0%)
Lymphopenia	20 (8.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Fatigue	14 (5.6%)	-	-	5 (11.1%)
Thrombocytopenia	5 (2.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Gastrointestinal disorders (n=38 occurrences)				
Vomiting	13 (5.2%)	0 (0.0%)	-	-
Nausea	12 (4.8%)	-	5 (4.6%)	-
Abdominal pain	0 (0.0%)	0 (0.0%)	8 (7.3%)	0 (0.0%)
Vascular disorders (n= 16 occurrences)				
Hypotension	16 (6.4%)	0 (0.0%)	0 (0.0%)	-
Cardiac disorders (n=11 occurrences)				
Tachycardia	11 (4.4%)	-	0 (0.0%)	0 (0.0%)
Respiratory, thoracic and mediastinal disorders (n=7 occurrences)				
Low oxygen saturation	7 (2.8%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Musculoskeletal & connective disorders (n=7 occurrences)				
Myalgia	7 (2.8%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Skin and subcutaneous tissue disorders (n=6 occurrences)				
Rash	6 (2.4%)	-	-	-
Nervous system disorders (n=5 occurrences)				
Headache	0 (0.0%)	5 (5.5%)	0 (0.0%)	0 (0.0%)

- refers to incidence of occurrence < 5 reported adverse reactions.

GL-ONC1-011: Below is a summary of adverse reactions reported by the Investigator for patients receiving 5 consecutive GL-ONC1 doses as a bolus intravenous infusion at 2×10^9 pfu/dose. Of the 63 total reported adverse reactions, 54 (87%) adverse reactions are within the current known toxicity profile for GL-ONC1 and were determined by the Investigator to be mild (n=40; 63%) to moderate (n=23; 37%).

AE Reported Term	G1	G2	G3	G4	TOTAL	%
General disorders and administration site conditions						
Fever	12	6	0	0	18	29%
Body pain	1	0	0	0	1	2%
Fatigue	1	0	0	0	1	2%
Gastrointestinal disorders						
Nausea	8	5	0	0	13	21%
Vomiting	2	0	0	0	2	3%
Musculoskeletal and connective tissue disorders						
Rigors	4	7	0	0	11	17%
Chills	2	1	0	0	3	5%
Nervous system disorders						
Headache	5	1	0	0	6	10%
Migraine headache (all AEs in same pt)	1	2	0	0	3	5%
Dizzy	0	1	0	0	1	2%
Infections and infestations						
Fever blisters (lips)	2	0	0	0	2	3%
Injury, poisoning and procedural complications						
Blisters (foot, hand, shoulder)	1	0	0	0	1	2%
Respiratory, thoracic and mediastinal disorders						
Pressure on back of throat	1	0	0	0	1	2%
Toxicity Grade Totals	**40**	**23**	**0**	**0**	**63**	**100%**
% Total Adverse Reactions by Toxicity Grade	**63%**	**37%**	**0%**	**0%**	**100%**	

Serious Adverse Events (SAEs), Results of Viral Shedding, PK and PD Laboratory Testing and Response to Treatment

GL-ONC1-002/MA (United Kingdom: Phase I Advanced solid tumors; NCT00794131): GL-ONC1 was administered as single or multiple intravenous doses

Phase I: Sixteen SAEs were reported for 9 out of 27 treated patients.

Safety:
Intravenous administration of GL-ONC1 is well tolerated. MTD was not reached in this trial in either the Phase I or the Phase Ib cohorts.

A total of five SAEs were reported for three patients, determined by the Investigator to have a degree of relatedness to GL-ONC1:

- *Cohort 2 Patient (single dose per cycle of 1 × 10^6 pfu):* Patient received the third cycle of GL-ONC1 and 6.3 hours later developed mild **pyrexia** of 39.1°C (Grade 1), sinus tachycardia and developed difficulty passing urine resulting in placement of a catheter. Patient was hospitalized for continued monitoring. On the next day, the patient's temperature lowered to 36.4 °C and was administered oral, and subsequently IV antibiotics and treated as community acquired pneumonia. Results of sepsis screen were negative and there was not symptomology consistent with sepsis. Two days following onset of pyrexia, the patient was apyrexial, the catheter was removed, and the IV antibiotics were changed to oral route of administration. Patient was released from the hospital on the third day following GL-ONC1 treatment with resolution of symptoms without sequelae. Investigator reported only pyrexia as an SAE. As this adverse event prolonged post-GL-ONC1 hospitalization, this adverse event qualified as an SAE.
- *Cohort 5 (single dose per cycle of 1 × 10^9 pfu):* Following Cycle 6, this patient experienced severe (Grade 3) **stiffness and pain in left leg**. The next day, the patient began experiencing numbness and coldness of the left leg from the knee to all toes during regular post-GL-ONC1 hospital admission. Later that same day, the left foot became blue and cold with no palpable pulse and was completely numb. An ultrasound and vascular thrombotic screening examination revealed an **arterial embolism** (severe Grade 3) of the common femoral artery in the left leg which was surgically removed. These serious adverse events resulted in prolonging inpatient hospitalization following GL-ONC1 treatment. Although the patient would have been eligible to receive additional GL-ONC1 treatment past Cycle 6 due to objective evidence of stable disease at Weeks 12 and 24, the Investigator withdrew the patient from receiving any further GL-ONC1 treatment due to these SAEs. The outcome was unknown at the time of these events. As this adverse event prolonged post-GL-ONC1 hospitalization, this adverse event qualified as an SAE. As arterial embolism is not currently listed as a known toxicity for GL-ONC1, this event was reported to U.S. and European regulatory authorities and Investigators as a Suspected Unexpected Serious Adverse Reaction (SUSAR).
- *Cohort 5a (single dose per cycle of 1 × 10^9 pfu):* Three days following administration of Cycle 1, this patient was diagnosed with moderate (Grade 2) **thrombocytopenia.** Concomitant medication was not required to treat this adverse event which spontaneously improved the following day. As this adverse event prolonged post-GL-ONC1 hospitalization for Cycle 1, this adverse event qualified as an SAE. Due to the thrombocytopenia not currently listed as a known toxicity for GL-ONC1, this event was reported to U.S. and European regulatory authorities and Investigators as a Suspected Unexpected Serious Adverse Reaction (SUSAR). It was also reported that following Cycle 1, this patient experienced a Grade 3 **aspartate aminotransferase (AST)** elevation which was transient (resolved within 48 hours) and self-limiting. Patient was otherwise clinically fine. This event qualified as a Dose Limiting Toxicity (DLT) and the patient was withdrawn from treatment after receipt of Cycle 1. This patient's disease history is remarkable for extensive liver metastases which required a hepatectomy. The Sponsor's Medical Monitor who reviewed this information stated that the rise in AST is reflective of metabolic activity and response in the patient's liver which is compromised by invasive tumor masses. Furthermore, the Sponsor's Medical Monitor stated that there is considerable variability from patient-to-patient in terms of the liver response to similar or identical provoking entities, and that it is important to note that liver enzyme levels were not particularly severe, and that the levels plateaued on day 2 to 3 with decline then being indicated. The potential results of this finding is that this may be an indicator of aggressive and rapid damage to tumor tissue in the liver, and does not demonstrate any correlation of clinical deterioration associated with any change in liver enzyme levels, or any other abnormal blood work. In summary, this response may possibly be an indicator of a destructive impact on hepatic metastases. Outcome was provided as recovered without sequelae.

Phase Ib: Nine SAEs were reported for 6 out of 16 treated patients.

One SAE was determined by the Investigator to have a degree of relatedness to GL-ONC1:

- *Cohort 8c (Cycle 1 at three consecutive doses per cycle of 1.667 × 10^9 pfu); Cycles 2-6 single doses at 5 × 10^9 pfu):* Sixteen days following administration of the Cycle 6, this patient, diagnosed with esophageal carcinoma, was hospitalized after developing an **tracheal airway obstruction** following the symptomatic growth of disease during the later cycles of GL-ONC1 treatment, and a tracheal stent was put in place. The Investigator wrote in the *SAE Report* "Possibly related because immunotherapy/virotherapy may cause transient tumor swelling before regression". The opinion of the Sponsor's Medical Monitor was "In contrast to the PI, I assess the event of airway obstruction trachea was due to the typical progression of the disease and therefore to be assessed as not related." As tracheal airway obstruction is not currently listed as a known toxicity for GL-ONC1, this event was reported to U.S. and European regulatory authorities and Investigators as a Suspected Unexpected Serious Adverse Reaction (SUSAR).

Viral Shedding:
Viral shedding was analyzed by Viral Plaque Assay (VPA) of blood, urine, stool and sputum. No virus shedding was detected in patients treated with GL-ONC1 at doses less than 1 × 10^9 pfu. Virus shedding was detected at none or minimal levels at the higher doses (i.e., 1 × 10^9 pfu and up to 5 × 10^9 pfu) and only in Cycle 1 of treatment.

Vaccinia Virus Positive Skin Rash:
A maculopapular skin rash comprising vaccinia pustules appeared in two patients in Cohort 5 (1 × 10^9 pfu × 1 dose/cycle) after Cycle 1 infusion of GL-ONC1. In both cases, the rash resolved without treatment by the end of Cycle 1. Some of the skin lesions of these patients were positive for GL-ONC1 by VPA and GFP imaging.

Pharmacokinetics and Pharmacodynamics:
Viral DNA was detected in blood of patients over the first 10 hours post-treatment in Cycles 1 and 2. The presence of viral DNA in blood was transient before being cleared from blood circulation. There was an exception for one patient who demonstrated delayed clearance. In Cohorts 3 to 5b, 6, 7, 8a and 8b, it was evident that substantial levels of viral titer were transiently maintained after the second treatment cycle as the durations of transient viral presence in blood was similar to that seen after Cycle 1 treatment. VACV neutralizing antibodies increased in all but one patient. However, except for Cohort 1, the antibody titers plateaued and did not result in further increase after the second cycle of treatment. Comparison of baseline neutralizing antibody levels between patients with objective evidence of stable disease versus patients with progressive disease showed that patients with stable disease had a trend toward lower levels of NAb than patients with progressive disease. There was a trend toward patients with higher levels of serum beta-glucuronidase having stable disease post-GL-ONC1 treatment versus lower levels of beta-glucuronidase seen in patients with progressive disease (p=0.14). Circulating tumor cells (CTC) were analyzed in the Phase 1b portion of this trial which showed evidence of virus infected CTCs with GFP+ CTC cells in 5 treated patients.

Tumor Response to Treatment:
The majority of patients presented with Stage IV cancers, and a small fraction with Stage III cancers. These patients had failed previous treatment(s) with disease progression when entering the GL-ONC1 trial. Thirteen patients from early to later dose cohorts had radiographic evidence of stable disease by CT scans from 8, 12, 13, 24 weeks as well as up to 48 weeks as compared to baseline tumor imaging. Clear changes in tumor growth rate post GL-ONC1 treatment were documented by CT scans. In such cases, patients failed previous therapy(ies) with progressive diseases, but experienced significant reduction in tumor growth after receiving GL-ONC1 treatment. Overall survival was compared in patients with progressive diseases (PD) or with stable diseases (SD). A statistically significant difference (p =

0.024) was documented between the two groups, indicating a potential clinical benefit of GL-ONC1 therapy in this groups of patients who have entered the trial with progressive diseases.

Immune Response Analysis:
Peripheral blood mononuclear cells (PBMCs) were examined in two groups of patients, with lower level of baseline anti-vaccinia antibody (NAb) titers (i.e. <1:40), or with higher baseline NAb titers (i.e. ≥1:40). 50% of the patients with baseline NAbs <1:40 were recorded to have disease control (as 'stable disease' (SD)) at week 12 after initial virus treatment, versus only 20% in patients with higher baseline Nabs.

Patients with baseline NAb <1:40 exhibited an up-regulation in the CD4+CD69+ and CD8+CD69+ (either CD3+ or CD3-) subsets on day 8 after virus injection, which is associated with disease control (stable diseases) at week 12 after the first virus injection, as compared to patients with baseline NAb ≥1:40 who tend to have progressive diseases (PD) after treatment. The fact that a subset of CD4+ and CD8+ cells carrying newly expressed CD69 on day 8 after virus injection suggests an activation of immune cells in response to either an ongoing virus replication, and/or tumor cell lysis.

In addition, we have found that serum levels of IL-10 are higher in PD patients than in SD patients, either before or after virotherapy, suggesting extra up-regulation of Tregs and suppression of CD4+ T cells in PD patients than in SD patients.

We have also noted higher level of IL-5 and IL-13 levels in patients with disease control (SD) than patients with PD under low baseline NAb level. IL-5 has been associated with enhanced tumor surveillance and suppression of tumor metastases (35). High IL-13-immunoreactivity has been noted to be associated with a better overall survival, and low IL-13 expression correlated with worse overall survival in colorectal patients (36). Therefore, both IL-5 and IL-13 may serve as serum markers to indicate favorable antitumor response from virotherapy.

GL-ONC1-003/MSK (United States – Investigator Initiated: Phase I malignant pleural effusion: primary, metastases and mesothelioma; NCT01766739): GL-ONC1 was administered as single or multiple infusions via intrapleural catheter

Safety:
Intrapleural treatment of GL-ONC1 was well tolerated. MTD was not reached in this trial.

SAEs were reported for 10 of the 18 patients. None of the SAEs were determined by the Investigator to be related to GL-ONC1 treatment. No DLTs have been reported to date in this clinical trial.

Viral Shedding:
Urine, sputum, and blood samples tested negative for viral shedding for all patients.

Vaccinia Virus Positive Skin Rash:
No skin rashes or lesions positive for vaccinia were reported.

Virus Infected Tumor Tissue:
Tissue from 13 out of 17 patients biopsied in this study were shown to be positive for GL-ONC1 infection. Of these patients, 7 patients were positive by GFP imaging, 8 patients by anti-VACV immunohistochemical staining (3/8 patients were also positive by GFP imaging), and 4 patients by tissue viral plaque assay (3/4 patients also positive for IHC staining; 1/4 patients showed positivity by GFP imaging).

Tumor Response to Treatment:
This trial is ongoing with 18 patients enrolled and treated to date. Ten out of 16 patients evaluated for response to treatment had a best overall response of stable disease at the Day 60 follow-up visit.

Historical survival data in patients with epithelioid subtype of mesothelioma is well documented in recent publication by Meyerhoff *et al.* (37), after reviewing data from 1183 patients (811 with epithelioid subtype). It shows that there are essentially no survival differences among disease stages of epithelioid mesothelioma. Without surgery, median survival is around 8-11 months. With surgery, median survival is around 18-21 months. In this trial, thirteen patients have epithelioid subtype of mesothelioma of different stages. When compared to historical data, there is a clear trend of survival advantage for patients who received GL-ONC1 treatment with or without further follow-up treatments (all epithelioid mesothelioma patients survived at least 12 months in this trial; only patients having > 12 months of survival is summarized and compared to historical data). Five of the 9 pts with epithelioid MPM had time to progression > 9 months (18 months in one patient).

GL-ONC1-004/TUE (Germany: Phase I/II Peritoneal carcinomatosis; NCT01443260): GL-ONC1 was administered as a single intraperitoneal dose per cycle

Safety:
Intraperitoneal treatment of GL-ONC1 is well tolerated. No virus-related organ toxicity was observed by clinical or serologic parameters. No DLT was reported, and no MTD was reached in this trial.

Twelve SAEs were reported in 6 of 9 treated patients. Patients were required by the German Competent Authority, Paul Ehrlich Institut (PEI), to have an extended hospital stay following treatment with GL-ONC1 for viral shedding testing.

Two SAEs was determined by the Investigator to have a degree of relatedness (i.e., possibly, probably, definitely related) to GL-ONC1. Both SAEs are within the currently known GL-ONC1 toxicity profile. Each adverse reaction reported below qualified as SAEs due to prolongation of required post-GL-ONC1 treatment hospitalization period.

- *Cohort 1 (1×10^7 pfu):* Patient experienced Grade 3 **fatigue** which was not relieved by rest and limited activities of daily living (ADL) that began 9 days after Cycle 1 was administered and lasted 4 days before resolving. This event was determined by the Investigator to be probably related to GL-ONC1 treatment.
- *Cohort 2 (1×10^8 pfu):* Patients was admitted to the hospital 8 days following administration of GL-ONC1 for Cycle 1 for a **mild fever** which resolved without sequelae within 1 day. The Investigator determined that this adverse event was possibly related to GL-ONC1.

Viral Shedding / Replication:
No viral shedding was reported in urine, sputum and stool/anal swabs from any patients.

Vaccinia Virus Positive Skin Rash:
No virus positive skin rashes were reported.

Pharmacokinetics and Pharmacodynamics:
Nine out of nine patients showed induction of Nab with the earliest being at Cycle 1 / Day 8. Beta-glucuronidase assays showed proof of in-patient expression of virus-encoded transgene in 8 out of 9 study patients.

Tumor Response to Treatment:
Killing of tumor cells in peritoneal fluid by the virus was confirmed based on cytology analysis. The single dose of intraperitoneal administration of GL-ONC1, even at the lowest dose of 1×10^7 pfu, significantly reduced the number of tumor cells in the ascites of treated patients.

Immune Response Analysis:
Cytological analysis of ascitic cells revealed also massive infiltration of immune cells into peritoneal cavity, indicating a profound GL-ONC1-induced peritonitis. Such inflammatory response is believed to be a key factor from virus-mediated immunotherapy for immune activation and responses against cancer.

GL-ONC1-005 (United States: Phase I newly diagnosed head and neck cancer; NCT01584284): GL-ONC1 was administered as single or multiple intravenous doses in combination with cisplatin and radiotherapy

Safety:
Intravenous treatment of GL-ONC1 along with chemoradiation therapy was shown to be well tolerated. No MTD was reached in this trial.

Nine SAEs were reported from 5 of 19 treated patients.

Two patients were reported to have experienced SAEs (n=5) with a degree of causality to GL-ONC1 treatment in this combination treatment clinical trial. A patient treated in Cohort 4 experienced Grade 3 (severe) myocardial infarction with acute renal failure and emesis following administration of the second dose (i.e., the last dose for this cohort) of GL-ONC1 which the Investigator determined to be unlikely related to GL-ONC1. The patient recovered and continued with the chemoradiotherapy standard of care portion of this treatment program. Under the protocol at that time, these events were considered a DLT. The DLT definition was revised in a subsequent protocol amendment which required a degree of attribution to GL-ONC1 to be classified as a DLT. This modification was accepted and approved by the FDA and local regulatory review committees and was placed into practice.

- *Cohort 1 (3×10^8 pfu × 1 infusion):* Patient was hospitalized for treatment of mild fever which resolved without sequelae.
- *Cohort 5 (3×10^9 pfu × 4 infusions):* Following the second infusion of GL-ONC1, a patient experienced severe orthostatic hypotension, dizziness, moderate pyrexia and moderate tachycardia from which he recovered although the Investigator withdrew the patient from receiving that last two GL-ONC1 treatments.

Viral Shedding:
No viral shedding has been reported in urine or oral swab samples from any patients.

Vaccinia Virus Positive Skin Rash:
Positive virus pox-like skin rash confirmed by VPA and GFP imaging was reported for a patient treated in Cohort 3, and one patient in Cohort 5, which resolved in a short period of time.

Tumor Response to Treatment:
An 84% response rate (16 out of 19) was observed in this group of patients; with 81% (13 out of 16) have complete response (CR). Since response to standard of care for this group of patients is well-documented in the literature (38), we compared our GL-ONC1 trial data with the historical data, on both response rate and survival. Clear advantages were observed with GL-ONC1 treatment on response rate (RR), progression free survival (PFS) and overall survival (OS) as compared to historical data, particularly in the difficult-to-treat subpopulation of HPV-negative patients.

Tumor Biopsy:
Tumor biopsy samples from 4 patients were shown to be positive for viral DNA by qPCR analysis. A lesion swab from the tumor lesion on the tongue of one patient in Cohort 5 was shown to be positive for GL-ONC1 by VPA.

GL-ONC1-015 (VIRO-15) (United States: Phase Ib & 2 recurrent ovarian cancer; NCT02759588): GL-ONC1 administered as two consecutive doses via intraperitoneal catheter as a monotherapy)

As of July 20, 2018, a total of 25 patients have been treated in the trial with 6 treated in Cohort 1 (3×10^9 pfu/day), 5 in Cohort 2 (1×10^{10} pfu/day), and 1 in Cohort 3 (2.5×10^{10} pfu/day). In the Phase 2 portion, 11 patients were treated in Cohort A (3×10^9 pfu/day).

Safety:
IP infusions of GL-ONC1 up to 2.5×10^{10} pfu/day for 2 consecutive days were well tolerated. There were no DLTs and MTD reports to date, and no deaths were attributed to GL-ONC1 treatment. Multiple IP infusions of GL-ONC1 (up to 1×10^9 pfu/cycle × 4 cycles) was also shown to be well tolerated in a prior Phase I trial performed in Germany (GL-ONC1-004/TUE; NCT01443260). With these two clinical trials, we have treated and evaluated the safety of GL-ONC1 delivery through IP route in 34 patients with well documented safety data.

The table below provides reported adverse reactions for GL-ONC1 treatment in 22/25 patients (88%). Out of the total reported AEs (n=772), 164 (21%) were determined to have a degree of attribution to GL-ONC1 treatment. The majority of adverse events were mild (53%).

Verbatim AE Term (n=164)	Body System	# of Pts	% of Pts	G1	G2	G3	G4	TOTAL	%
Abdominal pain	Gastrointestinal disorders	13	59%	7	12	5	0	24	15
Nausea	Gastrointestinal disorders	16	73%	9	10	1	0	20	12
Fever	General disorders & administration site conditions	14	64%	15	5	0	0	20	12
Vomiting	Gastrointestinal disorders	3	14%	4	9	4	0	17	11
Abdominal distension	Gastrointestinal disorders	8	36%	7	4	0	0	11	7
Chills	Musculoskeletal & connective tissue disorders	9	41%	11	0	0	0	11	7
Fatigue	General disorders & administration site conditions	5	23%	3	3	0	0	6	4
Ascites	Gastrointestinal disorders	3	14%	2	3	0	0	5	3
Body Aches	Musculoskeletal & connective tissue disorders	3	14%	1	2	0	0	3	2
Dehydration	Metabolism & nutrition disorders	2	10%	0	2	1	0	3	2
Intermittent abdominal cramping	Gastrointestinal disorders	1	5%	2	1	0	0	3	2
Generalized muscle weakness	Nervous system disorders	2	10%	2	0	0	0	2	1
Abdominal bloating	Gastrointestinal disorders	2	10%	2	0	0	0	2	1
Anorexia	Metabolism & nutrition disorders	2	10%	1	1	0	0	2	1
Diarrhea	Gastrointestinal disorders	2	10%	2	0	0	0	2	1
Dyspepsia	Gastrointestinal disorders	2	10%	0	2	0	0	2	1
Flank pain	Renal and urinary disorders	1	5%	1	1	0	0	2	1
Headache	Nervous system disorders	2	10%	1	1	0	0	2	1
Hypomagnesemia	Metabolism & nutrition disorders	1	5%	2	0	0	0	2	1
Rhinitis	Respiratory, thoracic and mediastinal disorders	2	10%	2	0	0	0	2	1
Somnolence	Psychiatric disorders	2	10%	0	2	0	0	2	1
Upper airway cough syndrome	Respiratory, thoracic and mediastinal disorders	2	10%	1	1	0	0	2	1

Verbatim AE Term (n=164)	Body System	# of Pts	% of Pts	G1	G2	G3	G4	TOTAL	%
Weakness	General disorders & administration site conditions	1	5%	1	1	0	0	2	1
Abdominal tenderness	Gastrointestinal disorders	1	5%	1	0	0	0	1	0.62
Constipation	Gastrointestinal disorders	1	5%	0	1	0	0	1	0.62
Cough	Respiratory, thoracic and mediastinal disorders	1	5%	1	0	0	0	1	0.62
Dry mouth	Gastrointestinal disorders	1	5%	1	0	0	0	1	0.62
Dyspnea	Cardiac disorders	1	5%	1	0	0	0	1	0.62
Flu-like symptoms	General disorders & administration site conditions	1	5%	1	0	0	0	1	0.62
Generalized pain	General disorders & administration site conditions	1	5%	1	0	0	0	1	0.62
GERD	Gastrointestinal disorders	1	5%	0	1	0	0	1	0.62
Head pain (NOS)	Nervous system disorders	1	5%	1	0	0	0	1	0.62
Hypoalbuminemia	Hepatobiliary disorders	1	5%	0	1	0	0	1	0.62
Hypokalemia	Hepatobiliary disorders	1	5%	1	0	0	0	1	0.62
Intermittent confusion	Psychiatric disorders	1	5%	1	0	0	0	1	0.62
Insomnia	Psychiatric disorders	1	5%	0	1	0	0	1	0.62
Lower leg lymphedema	Blood & lymphatic system disorders	1	5%	0	1	0	0	1	0.62
Mid-back pain	Musculoskeletal and connective tissue disorders	1	5%	0	1	0	0	1	0.62
Myalgia	Musculoskeletal and connective tissue disorders	1	5%	1	0	0	0	1	0.62
Tingling sensation abdominal area	Nervous system disorders	1	5%	1	0	0	0	1	0.62
			Totals	**87**	**66**	**11**	**0**	**164**	**100**
			Total Percentages	**53**	**40**	**7**	**0**	**100**	

Suspected Serious Adverse Reactions (SSARs):
Three SSARs were determined by the Investigator and the Sponsor's Medical Monitor to have a degree of attribution to GL-ONC1.

COHORT 1:

- *Patient 15A-02 (77 years old):* Patient was admitted for severe vomiting (expected adverse reaction for GL-ONC1) on the day of the second GL-ONC1 dose which lasted for 1 day and resolved without sequelae.
- *Patient 15A-03 (76 years old):* Patient was admitted for treatment of severe dehydration, moderate asthenia (expected adverse reaction for GL-ONC1), and mild abdominal pain 1 day following the last GL-ONC1 dose. Dehydration resolved within 72 hours, with asthenia remaining an ongoing event and abdominal pain resolving 25 days after onset.

COHORT 2:

- *Patient 15A-09 (75 years old):* Patient was admitted for moderate pyrexia (expected adverse reaction for GL-ONC1) and observation 3 days following the last dose of GL-ONC1 which resolved without sequelae the same day. The Investigator's determination of moderate severity was based a patient self-report (i.e., 103.4°F) prior to admission for observation. Upon hospital admission, patient's documented pyrexia was 99.2°F, which was below Grade 1 (mild) in toxicity.

Viral Shedding:
Oral and anal samples tested negative for all patients, except for one anal swab sample from a Cohort 2 patient showing minimal positivity (18 pfu/swab).

Skin Rash:
No skin rashes were reported.

Efficacy Data:
Below are efficacy results from Phase 1b portion of this trial, which was presented at American Society of Clinical Oncologists (ASCO) 2018.

Intraperitoneal infusion of GL-ONC1 monotherapy was given at higher repeated doses compared to a previous Ph1 trial in patients (pts) with platinum refractory/resistant disease. Primary endpoint: adverse events; Secondary endpoints: anti-tumor response by RECIST1.1 & survival. Eleven heavily pretreated pts with end-stage ROC were enrolled: 3-4 prior lines (n = 3), ≥ 5 lines (n = 8), ECOG 0 (n = 7) or 1 (n = 4), ascites/pleural effusion (n = 9) & progressive disease (PD) at baseline (n = 10). There were two dose cohorts: 3×10^9 (n = 6) or 1×10^{10} (n = 5) plaque forming units/day on 2 consecutive days.

Results: (1) Adverse reactions included Grade 1-2 chills (n = 7), nausea (7), fever (6), abdominal pain/distention (4), & vomiting (3). There were no differences in toxicity for the two dose levels. (2) GL-ONC1 colonized and replicated in the tumor, as indicated by a virus-encoded glucuronidase assay. (3) Clearance of tumor cells in ascites with induction of lymphocyte infiltration was shown in 5 pts with ascites. (4) Reduction of circulating tumor cells (CTC) was identified in 6/8 (75%) pts who had baseline CTC, ranging 1-42 per 7.5 mL blood. (5) Enhanced infiltration of CD8+ T cells into tumor tissue was demonstrated by repeat biopsy. (6) Disease Control Rate (DCR = partial response (PR) + stable disease (SD) ≥ 15 weeks) was 6/11 (55%). (7) Extended progression-free survival (PFS) of 23, 35, 59 (with confirmed PR) & 71 weeks were observed in 4 pts, respectively. (8) A tumor-specific T cell response was absent at baseline but confirmed at Week-30 in the PR patient by IFN-γ ELISPOT assay. (9) More than doubling of PFS compared to the last chemotherapy regimen was recognized in 4/11 (36%) pts.

Conclusions: Promising safety data, anti-tumor activity, and immune activation mechanisms were documented in this Ph1b trial, and a Ph2 trial (VIRO-15) is currently enrolling. Future studies combining GL-ONC1 and other immune therapies and/or chemotherapy are under consideration.

SUMMARY OF ALL CURRENT GL-ONC1 CLINICAL TRIALS

Overall, treatment with GL-ONC1 oncolytic vaccinia virus was well tolerated across different malignancies, routes of administration, as well as monotherapy and combination therapy regimens. There were no deaths reported to have any causality to treatment with GL-ONC1. MTD was not reached in any trial. The ability of GL-ONC1 to infect tumor tissue and kill tumor cells was demonstrated. In addition, virus-induced immune activation and elevation of serum markers linking to favorable antitumor immune response have been observed. Evidence of antitumor efficacy as monotherapy (including objective response) and clinical benefits have also been documented. Overall, these clinical trials have successfully met both primary and secondary objectives as originally designed. More importantly, data from these trials also provided further insights on treatment schedule/dosing, routes of delivery, etc. The Phase 2 portion of this trial is ongoing to further investigate anti-tumor response of GL-ONC1 as a monotherapy or combination therapy to observe trends of clinical benefits in a larger number of patients.

3.0 INVESTIGATIONAL AGENT INFORMATION

3.1 GL-ONC1

Please refer to the *GL-ONC1 Investigator's Brochure* for more comprehensive information.

Investigational Product

USAN/INN name: Not yet assigned
Internal product name: GL-ONC1
Substance: Live recombinant vaccinia virus
Route of administration: Intraperitoneal or intravenous infusion
Manufacturer: IDT Biologika GmbH, Germany
Sponsor: Genelux Corporation, San Diego, California, USA
Biosafety Level (BSL): BSL-2

3.2 GL-ONC1 Laboratory Name

GLV-1h68 (laboratory name).

3.3 GL-ONC1 Composition

An example for informational purposes from the GMP produced batch 007 08 12, by IDT is shown below.

Ingredients	1.0 mL contains	Function
Active substances		
Live recombinant LIVP strain GL-ONC1	1.1×10^8 pfu	Active ingredient
Excipients		
Tris (hydroxymethyl)-amino methane, pH 7.7	1.21 mg	Component of dilution buffer
Sodium chloride	8.18 mg	Component of dilution buffer

3.4 Biosafety Level

As GL-ONC1 is a genetically modified, attenuated vaccinia virus, it is considered a BSL-2 substance. Refer to *Precautions and Administration of the Investigational Medication* and the *GL-ONC1 Drug Handling Manual* for required BSL-2 precautions. The National Institutes of Health's Office of Biotechnology Activities lists general requirements for the handling and containment of BSL-2 specified material in *Appendix G-II-B. Biosafety Level 2 (BL-2)* of the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines)*.

3.5 Method of Preparation

The product is prepared by infection of chicken embryo fibroblast (CEF) cells with the primary inoculum (for master seed virus: MSV) or with MSV (for working seed virus: WSV). After incubation, the product was harvested and refined through cell suspension, centrifugation and other processes. Tests performed before final release include:

- Sterility
- Infection titer
- Mycoplasma (culture method)
- Mycobacteria
- Extraneous agents using chicks
- Total protein content
- Abnormal toxicity
- pH value
- Identity and purity
- Inserted genes (protein)

- Extraneous agents *in vitro*
- Extraneous agents *in vivo*
- Endotoxin
- Appearance
- Gentamycin content
- Extractable volume
- General Safety (21 CFR 610.11)
- Osmolality
- Benzonase content
- Total DNA content
- QF-PERT
- Specific activity calculation

3.6 Manufacturing Agent

GL-ONC1 is produced for Genelux in accordance with Good Manufacturing Practices (GMP) by IDT Biologika GmbH, Germany.

Address:

Am Pharmapark
D-06861Dessau-Roßlau
Germany
Phone: +49 (34901) 885-0
Fax: +49 (34901) 8855-323

3.7 GL-ONC1 Provision

GL-ONC1 will be provided to the Investigator in glass vials for storage at -70°C ±10°C. All GL-ONC1 handling procedures will be undertaken with appropriate approved biohazard precautions including masks, gowns and gloves in accordance with instructions listed in the *GL-ONC1 Drug Handling Manual.* Information on the receipt, storage, use and destruction of GL-ONC1 vials is documented on forms that can be found in the *GL-ONC1 Drug Handling Manual* and which will be inspected by the Study Monitor assigned to the Study Site at each monitoring visit. BSL-2 precautions are required for GL-ONC1 when handling and reconstituting GL-ONC1 for administration to study participants.

3.8 GL-ONC1 Batch Description

Information is provided as an example as other GL-ONC1 batches may be used throughout the life of this clinical trial.

Batch No:	**007 08 12**
Package quantity	Six vials per carton
Nominal batch size	4,900 vials with 1.1×10^8 pfu/mL
Appearance	Pale, milky-colored to light brown homogenous suspension
Storage	-70ºC ± 10ºC
Dosage form	Aqueous suspension
Container	2 mL injection vials, 2R, made of clear borosilicate glass, FIOLAX®-klar, WKB 1, KIN ISO 8362/1. The vials are supplied by Thüringer, Pharmaglass, Germany. The vials are labelled with a printed self-adhesive paper label.
Closure system	13 mm rubber injection stoppers, FM 257/2, V 9218, SAF 1, DIN ISO 8362-5-13, supplied by Helveot Pharma, Belgium. 13 mm aluminium caps, gold, DIN ISO 8362/3, supplied by BICO Pharma Verpackungen GmbH.
Expiration	At least 102 months, with regular checks for viability. The expiration date may be extended by conducting stability analyses.

3.9 GL-ONC1 Vial and Box Label Information

As other batches may be used during the duration of the treatment period, the following information is an example of what will be listed on the investigational product label in accordance with a particular batch that may be used in this clinical trial:

VIAL LABEL:
GL-ONC1 (#.# × 10E# pfu/mL)
Volume: 1.0 mL
Batch Number: ### ## ##
Retest Date: mm/yyyy
Medication Number: ####
For Clinical Trial Use Only
Manufacturer: IDT Biologika GmbH

BOX LABEL:
GL-ONC1 (#.# × 10E# pfu/mL)
Volume: 1.0 mL
Batch Number: ### ## ##
Retest Date: mm/yyyy
Medication Number: ####
Caution: New Drug—Limited by Federal (or United States) law to investigational use
To be stored at -70°C ± 10°C
Use as instructed in the Drug Handling Manual
Manufactured for Genelux Corporation by: IDT Biologika GmbH, Am Pharmapark D-06861 Dessau-Roßlau Germany; Phone: +49 (34901) 885-0
Supplier: Genelux Corporation, 3030 Bunker Hill Street, Suite 310, San Diego, CA 92109 USA; Phone: 858-210-6800

3.10 Clinical Supply

Genelux will supply the Investigator with GL-ONC1 packaged as a 1.2 mL aqueous suspension in vials. Full details of the requirements for dispensing are listed in the *GL-ONC1 Drug Handling Manual* (DHM).

3.11 GL-ONC1 Route of Administration

GL-ONC1 is administered as a diluted IP bolus infusion or an undiluted IV bolus infusion.

3.12 GL-ONC1 Precaution Requirements

- Refer to the *GL-ONC1 Drug Handling Manual* for IP preparation procedures.
- For investigational use only.
- All personnel are to be advised that vaccinia virus (GL-ONC1) is classified as a BSL-2 biosafety hazard and all universal biohazard precautions (e.g., wearing of personal protection equipment (PPE) such as mask, gown and gloves) and procedures for a BSL-2 laboratory must be observed when handling vaccinia virus (GL-ONC1), or contaminated equipment and objects that have been in contact with GL-ONC1.
- From the time the virus is thawed, the virus should be administered to patients within 3.5 hours, but optimally as soon as possible.
- Unused material remaining in the vial must be disposed of according to Institutional universal precautions for biohazardous material under BSL-2 containment standards.

All patients who have received virus treatment should be advised of the appropriate safety precautions to follow outside of the clinic (refer to *Appendix 4: Safety Issues*). These instructions are also incorporated into the patient consent form.

3.13 GL-ONC1 Accountability, Return and Retention

Genelux Corporation will provide the investigational site with sufficient amounts of study drug. The Investigator or qualified designee will administer the study drug only to patients enrolled in this study (i.e., are eligible and provided signed informed consent) and according to the procedures set forth in this study protocol and in the *GL-ONC1 Drug Handling Manual*. The Investigator or designee will confirm receipt of all batches of study drug by completing the applicable forms provided with the *GL-ONC1 Drug Handling Manual*, and is responsible for ensuring that accurate and adequate records including dates, lot numbers and quantity received, storage conditions, and individual usage, etc., are maintained throughout the life of the clinical trial. This documentation will be inspected by the assigned Study Monitor during routine scheduled monitoring visits. The site pharmacy may use institutional forms to track study drug accountability following review and approval by the Sponsor.

The Investigator and designee may be instructed by the Sponsor to either return or destroy unused study drug. If Genelux requests that all remaining GL-ONC1 vials are returned, instructions and the address for study drug return will be provided at the applicable time. However, if the Sponsor instructs that the remaining study drug is destroyed on-site, destruction will occur in accordance to institutional policy for a BSL-2 biohazardous product. The Investigator is responsible for verifying that all unused study drug supplies have been either returned or destroyed according to the Sponsor's instruction, and that no remaining supplies are in his/her possession. This verification will be reviewed by the Study Monitor at the trial close-out visit.

3.14 Stability of Genetic Identity of GL-ONC1

Quality control of GMP-manufactured GL-ONC1, using PCR and sequence testing by MICROMUN GmbH (Walther-Rathenaustraße 49a, Biotechnikum Greifswald, 17489 Greifswald, Germany), has shown that there is some evidence of viral DNA with non-recombinant *F14.5L*-insertion region present, but that the amount is regarded as minor. This finding has no adverse effects, as shown by the results of the toxicology and safety pharmacology testing in mice and rats using the clinical trial material conducted by the contracted GLP animal facility. Furthermore, there is no impact on the tumor targeting or the oncolytic capacity (potency).

3.15 Risk of Recombinations and Deletions

Genomic differences among VACV strains are mainly due to variability within the terminal regions of the DNA molecule. The central part of the genome is highly conserved. Cell culture studies have demonstrated that functions coded within the DNA from the left-hand and right-hand terminal regions of the orthopoxvirus genome are not essential for replication of the virus, but rather for determination of host range and virulence (39).

The risk that GL-ONC1 will revert into more virulent mutants is very low, as the parental LIVP strain, a descendant of the Lister strain, was attenuated by frequent passages on calf skin (more than 500 times) and was widely used as a vaccine, with excellent documented safety, during the World Health Organization (WHO) smallpox eradication program. In addition, because orthopoxviruses are not endemic in the human population, it is unlikely that GL-ONC1, as a clonal strain, will recombine with a wild-type orthopoxvirus to produce a more virulent strain. Despite worldwide use of the live virus vaccine, no adverse events due to mutation to a more aggressive phenotype have ever been reported. Various

strains of VACV (such as New York City Board of Health (NYCBOH)) have been detected in humans shortly after receiving vaccinia-based vaccines.

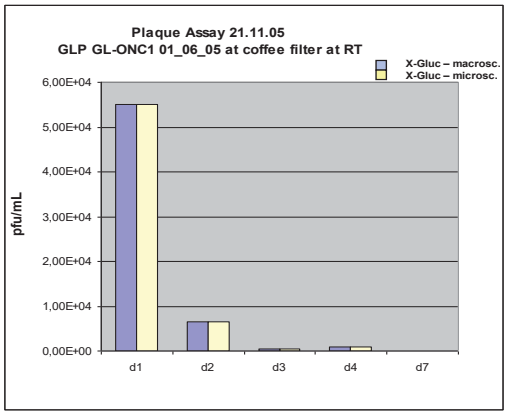
Recombination of orthopoxviruses with other DNA/RNA-viruses has been reported very rarely and appears to be very unusual. In addition, since poxviruses use unique promoters, gene activation by inserted foreign promoters is also very unlikely.

3.16 Physical Stability of GL-ONC1

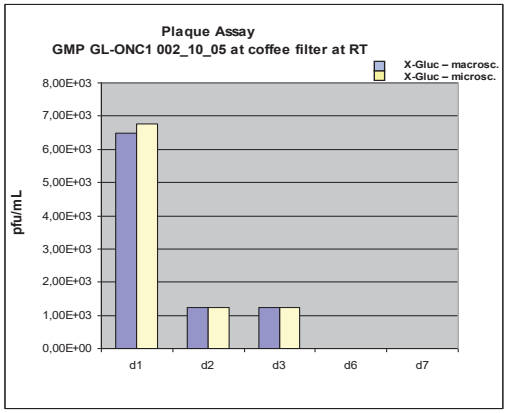
Samples of GLP and GMP manufactured GL-ONC1 were spotted onto filter paper to examine the stability after release into environment. At the respective time points, the virus samples were retrieved from the filter paper and the amount of active virus was analysed by plaque assay.

The experimental results showed that the titers of both GLP and GMP manufactured GL-ONC1 were decreased by 99.99% within 24 hrs when released into the environment at room temperature (see Figure below). By days 6-7, all viruses were disintegrated. Therefore, it is unlikely that virus shed from patients, if shedding occurs, would be of significant environmental concern or health concern to others.

Stability of GL-ONC1 after release into environment



A) GLP GL-ONC1 - Starting titer: 3,92 E+08 pfu/ml



B) GMP GL-ONC1 - Starting titer: 6,17E+07 pfu/ml

Figure. GL-ONC1 quickly disintegrates when released into the environment at room temperature.

4.0 INVESTIGATIONAL PLAN

4.1 Patient Population Characteristics

This is an open-label, non-randomized Phase 1b & 2 study evaluating the effect of the oncolytic virus GL-ONC1 as a monotherapy or as a combination therapy with carboplatin doublet plus bevacizumab. GL-ONC1 is administered via an intraperitoneal catheter or as an intravenous bolus infusion in patients diagnosed with recurrent or refractory ovarian cancer and peritoneal carcinomatosis. Eligible patients must have histologically confirmed (from prior treatment(s)) non-resectable ovarian, fallopian tube or primary peritoneal cancer who are platinum-resistant, platinum-refractory or intermediate platinum-sensitive with good performance status (ECOG of 0 or 1). Patients with platinum-resistant disease (i.e., recurrence or progression < 6 months) or platinum-refractory disease (progression while on platinum-based therapy) must have either (1) failed at least two consecutive therapies, or (2) are not eligible for additional cytotoxic therapies. Intermediate platinum-sensitive patients (recurrence of disease 6 to 12 months from last platinum compound) have recurrent ovarian carcinoma with at least four prior individual

treatment regimens including at least two separate platinum-based therapies with recurrence from the last platinum-based regimen less than 12 months, who are unwilling or unable to undergo additional platinum-based cytotoxic therapy. Patients treated with chemotherapy, radiotherapy or any anti-cancer biologic therapies will require a 4-week washout period prior to administration of first GL-ONC1 dose.

For IP route of virus delivery, patients must have sufficient peritoneal space to allow instillation of GL-ONC1 via an intraperitoneal catheter. However, to ensure sufficient peritoneal space, the surgeon can tent the peritoneal wall up by lifting the Halstead mosquito artery forceps that are holding the edges of the peritoneal opening. The potential space can then be directly visualized. In addition, a blunt instrument, for example, Watson Cheyne dissector, can be used to confirm if there is a potential space.

4.2 Optional In-patient Stay

A patient is recommended to have an optional in-patient stay during and/or after treatment for observation with the duration determined by the study-affiliated treating physician (suggested duration is for 48 hours post-last dose of GL-ONC1). Any in-patient or out-patient care (e.g., hydration, etc.) will be in accordance with local standard medical practices.

4.3 Study Periods

This study consists of 4 periods: screening, treatment, post-treatment and long-term follow-up. For specific procedures as well as timing of events during each period, refer to *Section 15.0 Study Assessments and Procedures*.

4.3.1 General Statements:

Phase 1b & Phase 2 Cohorts A & B -- Weeks 2 to 48, and Years 2 & 3 Long-term Follow-up: If a patient moves on to receive other anti-cancer treatment after GL-ONC1, clinic visits are no longer required as follow-up is conducted by a telephone call to assess survival, disease and anti-cancer treatment status. Telephone calls occur at the clinic visit time points specified for each study period. In order to determine if other anti-cancer treatment may have a synergistic effect following GL-ONC1 treatment, the sponsor asks that study sites continue to provide de-identified radiologic imaging reports and clinical labs, including CA 125 results, on an *ad hoc* basis when available.

Assessment of SAE(s) that occurred since the last telephone call (or visit if first follow-up call). If the Investigator determines a SAE has a degree of attribution to GL-ONC1 treatment, submit SAE Report to the Pharmacovigilance Team via electronic data capture (EDC) system or by paper CRF if EDC not available. Non-serious AEs are not collected. In order to determine if other anti-cancer treatment may have a synergistic effect following GL-ONC1 treatment, the sponsor asks that sites provide in a timely manner de-identified radiologic imaging reports and clinical labs, including CA 125 results, on an *ad hoc* basis when available.

ALL PHASE 2 COHORTS: Ongoing treatment may be administered following carboplatin doublet therapy with either the non-platinum single agent chemotherapy, single agent bev, or combination of single agent chemotherapy with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

All Cohorts off-site treatment with chemo +/- bev: It is highly preferred that all subsequent platinum-based doublet therapy, non-platinum therapy and bev be administered at the Investigator's institution. However, for patients traveling significant distances to the clinical site, only the first two treatment cycles will be required to be administered at the Investigator's institution. Further treatments can continue locally by a patient's primary practitioner if the Investigator and the local primary practitioner have an agreement on a specific treatment regimen to follow. Regular follow-up visits at the Investigator's institution for

monitoring purposes are required, and all follow-up CT scans are to be performed at the Investigator's institution.

All Cohorts – Quality of Life (QoL) Questionnaire: The *Functional Assessment of Cancer Therapy-Ovarian* (FACT-O) Quality of Life Questionnaire (refer to *Appendix 7*) is obtained at baseline and at each clinic visit when radiologic imaging is scheduled.

4.3.2 Screening Period – All Phases/All Cohorts (unless otherwise specified):

- Begins by signed informed consent form (ICF) and establishing the patient's initial eligibility.
- Patients are registered by the Investigator or designee submitting the registration form and applicable eligibility documents to the Sponsor for review of eligibility.
- Patients with an existing intraperitoneal catheter/port will have it surgically removed at the time of the laparoscopic procedure.
- PHASE 1b: Patients will have a temporary peritoneal catheter surgically implanted by laparoscopic procedure within 5 to 7 days prior to of Treatment Day 1 with photos taken to document the extent of disease for assessment by the *Peritoneal Cancer Index* (40) *(*refer to *Appendix 3).*
- PHASE 2 Cohorts A, B & C: Patients will have a temporary peritoneal catheter surgically implanted by laparoscopic procedure within 3 to 7 days (5 days preferred) prior to administration of the first GL-ONC1 dose with photos taken to document the extent of disease for assessment by the *Peritoneal Cancer Index* (40) *(*refer to *Appendix 3).*
- PHASE 1b: A biopsy of tumor tissue is obtained (e.g., during laparoscopy) for analysis of the cancer gene mutation profile and immune status of the tumor.
- PHASE 2: Depending on the location of the tumor(s) selected by the Investigator for sampling, biopsies may be performed through the laparoscope (preferred method for all cohorts except Cohort D, if feasible) or by needle biopsy (e.g., CT-guided or ultrasound-guided biopsy) if tumor is safely accessible.
- It is preferred that baseline radiographic assessment occur as close to treatment as allowable (ideally within 2 weeks)
- Patient is assessed for complete study eligibility within the required timeframe (see *Section 15.0 Study Assessments and Procedures*).
- Screening Assessments: For patients screened and enrolled into study within a short period of time prior to treatment, it is not considered a protocol deviation if all screening procedures are collected at one-time point instead of two-time points (i.e., toxicity, physical examination, weight, performance status, vital signs, serum pregnancy and clinical labs, concomitant medications).

4.3.3 Treatment Period (Week 1) -- Phase 1b and Phase 2 Cohorts A & B:

- *PHASE Ib:* This is a dose escalating study with GL-ONC1 administered IP on two consecutive days during Week 1 to determine the MFD or MTD (MTD is optional).
- *PHASE 2:* Sponsor assignment into either Cohort A or Cohort B.
- Clinical labs, pharmacodynamic (PD) and pharmacokinetic (PK) samples are collected according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.
- For patients who have peritoneal ascites, as much ascites as possible will be drained prior to the instillation of the first GL-ONC1 dose. On the day after the second GL-ONC1 dose, 10 mL of ascites will be collected, if available.
- All patients will have 1 L to 1.5 L of Ringer's Lactate instilled and drained through the IP catheter prior to the first GL-ONC1 dose as a peritoneal wash.
- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. Non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team. Clinical labs, PD and PK

samples are collected according to the schedule listed in *Section 15.0 Study Assessments and Procedures*.
- Deaths that occur during Treatment Week 1 are reported as an expedited safety report to the Pharmacovigilance Team regardless of causality (refer to *Section 21.11 Deaths* for reporting requirements).

4.3.4 Post-treatment Period (Week 2 to Week 48) -- *Phase 1b and Phase 2 Cohorts A & B*:

- For patients with measurable disease, imaging by spiral CT scan is performed after initiation of treatment at Weeks 6, 15, 24, 36 and 48. For patients allergic to IV CT contrast, it is recommended that oral CT contrast (preferred) or MRI may be used. Per RECIST 1.1, the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up.
- For patients with non-measurable disease (Phase 1b only), imaging will be performed by either PET scan or PET/CT scan at any of the time points listed for the spiral CT scan per the Investigator's discretion.
- PHASE 2: Either 7 or 14 days after date of first GL-ONC1 dose an optional needle biopsy (e.g., CT-guided or ultrasound-guided) to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- PHASE 1b: Either on Day 10 or 17 an optional CT-guided biopsy to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- PHASE 2: An optional post-treatment needle biopsy (CT-guided or ultrasound-guided) may be obtained either during Week 6 or Week 15 (± 7 days).
- PHASE 2 COHORTS A & B: For patients who receive a second cycle of GL-ONC1, optional needle biopsies will be obtained prior to (within 2 weeks or on day of catheter placement) and at approximately 4 weeks after second cycle of GL-ONC1.
- PHASE 1b: On Day 248 (Week 36), an optional CT-guided biopsy to obtain a post-treatment tumor tissue sample from consenting patients can be collected if tumor is located in a reasonably safe area.
- Clinical labs, PD and PK samples are collected according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.
- PHASE 2: For patients who have peritoneal ascites, 10 mL of ascites will be collected 7 days after date of first GL-ONC1 dose, if available. If the IP catheter is in place 14 days after date of the first GL-ONC1 dose, 10 mL can be drawn if ascites is available.
- PHASE 1b: For patients who have peritoneal ascites, 10 mL of ascites will be collected on Day 10, if available. If the IP catheter is in place on Day 17, 10 mL can be drawn if ascites is available.
- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. During the Post-treatment Period, non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team. From Weeks 6 to 48, for patients who receive other anti-cancer treatment, only SAEs that the Investigator determines have a degree of attribution to GL-ONC1 treatment are reported; non-serious adverse events are not reported regardless of causality.
- Regardless of causality, deaths that occur from Week 2 to Week 5 (30 days post last GL-ONC1 infusion) are reported as an expedited safety report to the Pharmacovigilance Team.
- Deaths that occur from Week 6 to Week 48 are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the *Death Report* eCRF.

ANY COHORTS: After completion of 'safety run-in cohort' for Cohort D, and after treatment with GL-ONC1 and further chemo +/- bev, a second cycle of GL-ONC1 at 2,3,5 $\times 10^9$ pfu as an undiluted bolus administered by IV route for 3 consecutive days is allowed (time point beyond Week 48 is also

allowed) for 3 patients and expandable to another 3 patients upon approval by the Sponsor. PFS, OS, and Objective Response by RECIST1.1 and CA-125 should be documented after the second cycle of GL-ONC1 until disease progression, if clinically applicable. Since the second cycle of GL-ONC1 is given via IV route of administration, follow Cohort D study calendar for procedures and follow-up specifics.

If a patient receives other anti-cancer treatment, see **General Statement** section above for procedures to follow.

4.3.5 Long-term Follow-up Period -- Phase 1b and Phase 2 Cohorts A & B:

- During Year 2, patients should come in on a quarterly basis for clinic visits as indicated in *Section 15.0 Study Assessments and Procedures*. The timing of the first follow-up visit is based on the timing of Post-treatment Week 48 visit, or the last visit performed during the Post-treatment Period. If a patient receives other anti-cancer treatment, see **General Statement** section above for procedures to follow
- In Year 3, patients are telephoned quarterly to collect information on survival, disease and treatment status. The first quarterly follow-up call is based on the last Year 2 Long-term Follow-up visit.
- Deaths that occur during the Long-term Follow-up Period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the *Death Report* eCRF.

4.3.6 GL-ONC1 Treatment Period (Week 1) through Post-GL-ONC1 Treatment (Week 5) -- Phase 2 Cohort C & D (unless otherwise specified):

General Statement: The following procedures occur prior to and following treatment. Refer *Section 15.0 Study Assessments and Procedures* and relevant study calendars for more detailed information. Refer instructions provided in the **General Statement** section above for chemotherapy ± bev administered by local primary practitioner.

Enrollment will be by Sponsor assignment into either Cohort C or D.

Week 1 GL-ONC1 Treatment:

- Virus dose: 3×10^9 pfu/day for 2 consecutive days (except Cohort D). For Cohort D: $2,3,5 \times 10^9$ pfu in 3 consecutive days.
- COHORT C: For patients who have peritoneal ascites, as much ascites as possible will be drained prior to the instillation of the first GL-ONC1 dose. On the day after the second GL-ONC1 dose, 10 mL of ascites will be collected, if available. 10 mL of ascites will be collected 7 days (Week 2) after date of first GL-ONC1 dose, if available. If the IP catheter is in place 14 days (Week 3) after date of the first GL-ONC1 dose, 10 mL can be drawn if ascites is available.
- COHORT C: All patients will have 1 L to 1.5 L of Ringer's Lactate instilled and drained through the IP catheter prior to the first GL-ONC1 dose as a peritoneal wash.
- A preferred but optional post-treatment needle biopsy (e.g., CT-guided or ultrasound-guided) may occur between Week 2 to Week 5 after date of the first GL-ONC1 dose, and prior to initiation of further chemotherapy to obtain a post-GL-ONC1 treatment tumor tissue sample from consenting patients if tumor is located in a reasonably safe area.
- Clinical labs, pharmacodynamic (PD) and pharmacokinetic (PK) samples are collected prior to treatment according to the scheduled listed in *Section 15.0 Study Assessments and Procedures*.

- Toxicity assessments are conducted at each patient contact point. Patients are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible. Non-serious adverse event and serious adverse event information are recorded. All SAEs regardless of causality are reported to the Pharmacovigilance Team.
- GL-ONC1 (study drug): Deaths that occur during Treatment Week 1 through Week 5 (30 days post last GL-ONC1 infusion) are reported as an expedited safety report to the Pharmacovigilance Team regardless of causality (refer to *Section 21.11 Deaths* for reporting requirements).

4.3.7 Post-GL-ONC1 Treatment Period (Week 6 through Week 59 (q3W) or Week 65 (q4W) -- Phase 2 Cohort C:

Carboplatin doublet chemotherapy +/- bevacizumab treatment plan: refer to Study Calendars for *Treatment q3 Weeks and Treatment q4 Weeks* schedules. An example of regimen and dosing schedule is the OCEANS (Aghajanian *et al. J. Clin Oncol*. 2012;30(17):2039-2045) phase 3 trial evaluating the efficacy and safety of bevacizumab combined with gemcitabine + carboplatin (GC) for patients with platinum-sensitive recurrent ovarian cancer (ROC). Previous treatments with any of the regimens below are allowed. Cycle 1 of chemotherapy +/- bevacizumab treatment begins at Wk 6, irrespective of disease status from Wk 6 CT scan. The number of cycles of chemotherapy +/- bevacizumab may be expanded at the discretion of the Investigator if tolerated well and clinically determined to be beneficial after 6 cycles. Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev until progression or unacceptable toxicity, at the discretion of the Investigator.

- Carboplatin doublet chemotherapy: Up to 6 cycles of carboplatin + a taxane (e.g., docetaxel (i.e., Taxotere)), paclitaxel (i.e., Taxol), nab-paclitaxel (i.e., Abraxane), or + gemcitabine (i.e., Gemzar), or + PLD (i.e., Doxil). Chemotherapy component could be substituted if due to toxicity or by Investigator's choice. If due to platinum-allergy, a single-agent non-platinum chemotherapy is allowed.
- Suggested chemotherapy intravenous (IV) dose: carboplatin (AUC 4 to 5, q3w) with gemcitabine (800 mg/m^2, D1&D8, q3w); carboplatin AUC 5 to 6 with docetaxel (60 to 75 mg/m^2, q3w), or with paclitaxel (175 mg/m^2, q3w), or with nab-paclitaxel (260 mg/m^2, q3w), or with PLD (40 mg/m^2, q4w). Dose of chemotherapy may be adjusted to allow better tolerability.
- Suggested bevacizumab (bev) dose: 10 mg/kg, q3w with chemotherapy (7.5 mg/kg, q2w with PLD), and as single agent maintenance q3w for another 9 months. Dose adjustment is allowed per Investigator's discretion.

Chemotherapy doublet +/- bevacizumab (bev), both q3 weeks:

Baseline **CT scan**, ideally within 2 weeks (3-4 weeks allowed) prior to 1st dose of GL-ONC1
Days 1 & 2 (W1), GL-ONC1
↓
W6, **CT scan** immediate prior to 1st cycle of chemo + bev
↓
W9, 2nd cycle of chemo + bev
↓
W12, 3rd cycle of chemo + bev
↓ W14, **CT scan**
W15, 4th cycle of chemo + bev
↓
W18, 5th cycle of chemo + bev
↓

W21, 6th cycle of chemo + bev
↓ W23, **CT scan**
W24, start of maintenance bev for additional 9 mos
↓ W35, 47, 59, then PRN for **CT scans**

For chemotherapy doublet at q4 weeks with bev at q2 weeks:

Baseline **CT scan**, ideally within 2 weeks (3-4 weeks allowed) prior to 1st dose of GL-ONC1
Days 1 & 2 (W1), GL-ONC1
↓
W6, **CT scan** immediate prior to 1st cycle of chemo + bev
↓ W8, bev
W10, 2nd cycle of chemo + bev
↓ W12, bev
W14, 3rd cycle of chemo + bev
↓ W16, bev
↓ W17, **CT scan**
W18, 4th cycle of chemo + bev
↓ W20, bev
W22, 5th cycle of chemo + bev
↓ W24, bev
W26, 6th cycle of chemo + bev
↓ W28, bev
↓ W29, **CT scan**
W30, start of maintenance bev for additional 9 mos
↓ W41, 53, 65, then PRN for **CT scans**

4.3.8 Post-GL-ONC1 Treatment Period -- Phase 2 Cohort D:

Treatment with chemotherapy ± bevacizumab will start at the time of disease progression (preferred by RECIST 1.1) from GL-ONC1, or will start at the discretion of the Investigator as clinically indicated based on clinical assessment (in consultation with Sponsor if disease progression is clinically determined). Combination with bev is allowed but not required. If patient develops toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment (e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel) to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if well tolerated and determined to be clinically beneficial after 6 cycles. Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev until progression or unacceptable toxicity, at the discretion of the Investigator. **Refer to information provided above for Cohort C for examples of chemotherapy regimens and time lines.**

4.3.9 Post-GL-ONC1 Treatment Period (Week 6 through Week 59 (q3W) or Week 65 (q4W) -- Phase 2 Cohorts C & D (timeline adjusted accordingly based start of chemo):

- GL-ONC1 (study drug): Deaths that occur during this period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements) taking into consideration that chemotherapy agents and bevacizumab are administered during this study period. If no attribution to GL-ONC1, report death on the Death Report electronic Case Report Form (eCRF).
- Imaging by spiral CT scans & RECIST assessment are performed according to time points of treatment depending on whether time frame for chemotherapy is q3 weeks or q4 weeks. During this period, time points include: following baseline and immediately prior to cycle 1, 1-wk prior to cycle 4, 2-3 weeks post cycle 6 (i.e., 1-wk prior to start of bev maintenance), and then every 3 mos for 3 more scans. Further scans will be PRN and at the discretion of the Investigator and the Sponsor. Refer to specific study calendar for imaging time points. For patients allergic to IV CT contrast, it is recommended that oral CT contrast (preferred) or MRI may be used. Per RECIST 1.1, the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up.
- Another optional post-treatment needle biopsy may be obtained after second cycle (either during Week 11 for q3w, or Week 13 for q4w with ± 1 week for both time points) or after third cycle (Week 14 for q3w, or Week 17 for q4w with ± 1 week for both time points) of chemotherapy. Biopsy will be obtained after CT scan if they are conducted on same day.

4.3.10 Long-term Follow-up Period -- Phase 2 Cohorts C & D:

- *During Year 2 and Year 3*, patients should come PRN at Investigator discretion (e.g., quarterly basis) for clinic visits as indicated in *Section 15.0 Study Assessments and Procedures*. The timing of the first follow-up visit is based on the timing of the 30-day following the last cycle.
- If patients are unable to come to clinic, quarterly telephone calls to collect information on survival, disease and treatment status. The first quarterly follow-up call is based on the last Year 2 Long-term Follow-up visit.
- Deaths that occur during the Long-term Follow-up Period are reported as an expedited safety report if the Investigator feels the death has a degree of attribution to GL-ONC1 treatment (refer to *Section 21.11 Deaths* for reporting requirements). If no attribution to GL-ONC1, report death on the *Death Report* eCRF

4.4 Allowable Window of Time around Study Visits

During Week 1 of the GL-ONC1 Treatment Period, a window of ± 1 day for each visit is allowed. During the Post-treatment Period, there is a ± 2-day allowable window, and a ± 1 week for imaging studies. Unless otherwise specified, there is a ± 7-day window around the optional post-treatment needle biopsies. For Phase 2 Cohorts C & D, in general there is a ± 7-day window allowable for treatments, procedures, and imaging scans, unless there is a further delay due to toxicity from chemo + bev requiring additional recovery time. The allowable window of variance during the Long-term Follow-up Period is 2 weeks. Documented delays that occur outside of the allowable window of variance due to holidays, weekends, weather, or other unforeseen circumstances do not constitute a protocol deviation; refer to *Section 4.5 Treatment Compliance Criteria* for exceptions.

4.5 Treatment Compliance Criteria

Treatment breaks must be clearly indicated in the treatment record along with the reason(s) for the treatment break(s). Missed treatments due to holidays or logistic reasons can be compensated for by delivering the additional treatments as soon as the Investigator determines is feasible, or by treating on

a Saturday or Sunday. Treatment breaks should be allowed for resolution of severe acute toxicity and/or intercurrent illness, and ideally should not exceed 5 treatment days at a time (see exception above for Phase 2 Cohorts C & D allowable treatment break).

4.6 Population

Patients diagnosed with recurrent or refractory ovarian cancer and peritoneal carcinomatosis. Sponsor allows the rescreening of patients, if necessary. The Sponsor will determine the final cohort allocation on an *ad hoc* basis based on the clinical information provided during eligibility review.

4.7 Inclusion Criteria

Patients must meet all inclusion criteria to participate:

1. Signed, written informed consent.
2. Women ≥ 21 years.
3. History of histologically confirmed (from prior treatment) non-resectable ovarian, fallopian tube or primary peritoneal cancer.
4. Patient population: High-grade serous (including MMMT with metastasis that contains high grade epithelial carcinoma), endometrioid, or clear-cell ovarian cancer that is recurrent or refractory, which includes:
   - Platinum-resistant (recurrence or progression < 6 months), or platinum-refractory (progression while on platinum-based therapy) patients who failed at least 2 consecutive therapies or are not eligible for additional cytotoxic therapies (exception is for patients registered into Cohorts C & D).
   - Intermediate platinum-sensitive patients (recurrence of disease 6 to 12 months from last platinum compound) with recurrent ovarian carcinoma with at least four prior individual treatment regimens including at least two separate platinum-based therapies with recurrence from the last platinum-based regimen less than 12 months, who are unwilling or unable to undergo additional platinum-based cytotoxic therapy (this sub-population is not applicable for Cohort C).
5. Performance status ECOG is at 0 or 1.
6. Patient has grossly visible tumors located in the peritoneal cavity which are accessible by the peritoneal catheter **(eligible to 'Ch D' if not meeting this criterion).**
7. Patient has either measurable disease in the peritoneal cavity (e.g., stomach, spleen, liver, ovaries, fallopian tubes, tail of the pancreas, uterus, bulb of the duodenum, jejunum, ileum, transverse colon, and sigmoid colon) as defined by RECIST 1.1 with at least one lesion that can be accurately measured in one dimension (longest dimension recorded) by contrast spiral CT scan. **Patients who do not meet this criterion are eligible for enrollment in 'Ch D' if (1) there is RECIST 1.1 measurable extra-peritoneal disease such as mediastinal nodes, liver metastasis, lung, etc,; (2) RECIST 1.1 measurable intra-abdominal disease not easily accessible by laparoscopy in the opinion of the PI (dome of liver/diaphragm post liver diaphragm resection, rigid abdomen with multiple laparotomies, etc)**.

   Phase 1b only: non-measurable disease in the peritoneal cavity that is identifiable by PET/PET-CT scan with contrast and can be confirmed by laparoscopy and/or elevated CA-125. Patients who have non-measurable disease that is not identifiable by PET/PET-CT scan, but who have elevated CA-125, and/or ascites, with visible disease confirmed by laparoscopy are also eligible.

8. Able to undergo IP or IV bolus infusion, and all administration procedures.

9. Adequate renal, hepatic, bone marrow, and immune functions:
   a. Renal function:
      - Creatinine ≤ 1.8 mg/dL or calculated creatinine clearance (CrCl) using the Cockcroft-Gault formula ≥ 45 mL/min, or measured creatinine clearance ≥ 45 mL/min:

$$\text{Female CrCl} = \frac{(140 - \text{age in years}) \times \text{weight in kg} \times 0.85}{72 \times \text{serum creatinine in mg/dL}}$$

      - Absence of clinically significant hematuria on urinalysis: dipstick <2+ (exception to this criterion is for presence of renal stent(s) and/or with otherwise good renal function as assessed by Investigator).
      - Absence of clinically significant proteinuria on urinalysis: dipstick < 2+.
   b. Adequate hepatic function:
      - Serum bilirubin <1.5 × ULN;
      - AST and ALT ≤ 3 × ULN.
   c. Adequate bone marrow function:
      - ANC ≥ 1.5×10^9/L;
      - Platelets ≥ 100×10^9/L;
      - Hemoglobin ≥ 90 g/L.
   d. Adequate coagulation tests:
      - INR ≤ 1.5 × ULN.
   e. Adequate immune function by lymphocyte count:
      - Absolute lymphocyte count (ALC) ≥ 0.5×10^3/mm^3;
      - Relative lymphocyte count (RLC) ≥ 10%.

10. Life expectancy of at least 6 months.
11. Have NO continuing acute toxic effects of any prior therapy, including but not limited to biological therapy, radiotherapy, chemotherapy, or surgical procedures, i.e., all such effects must have resolved to *Common Terminology Criteria for Adverse Events* (CTCAE, Version 4.03) Grade ≤ 1 (with the exception of peripheral neuropathy). Any other surgery (except biopsies) must have occurred at least 28 days prior to study enrollment.
12. Women of child-bearing potential (WOCBP) has negative pregnancy test prior to initiating study drug dosing.
13. Women of child-bearing potential must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry, for the duration of study participation, and for 90 days following completion of therapy. Should a woman become pregnant or suspect she is pregnant while participating in this study, she should inform her treating physician immediately.

    a. A woman of child-bearing potential is any female (regardless of sexual orientation, having undergone a tubal ligation, or remaining celibate by choice) who meets the following criteria:
       - Has not undergone a hysterectomy or bilateral oophorectomy; or
       - Has not been naturally postmenopausal for at least 12 consecutive months (i.e., has had menses at any time in the preceding 12 consecutive months).

14. Have negative test result for active HIV and Hepatitis B or C infection.
15. Be willing and able to comply with scheduled visits, the treatment plan, imaging and laboratory tests.
16. A baseline tumor biopsy is required.
17. Adequate nutritional status as determined by Investigator (e.g., assessed based on BMI* or PNI**).
18. **Phase 2 only:** Documented progressive disease status at baseline (based on RECIST 1.1 or clinical progression).

* BMI: Body Mass Index. Suggested minimal BMI is ≥ 20, with an ideal value of ≥ 25.

** PNI: Prognostic Nutritional Index (PNI = 10 × serum albumin (g/dL) + 0.005 × peripheral lymphocyte count (per mm^3)). Suggested minimal PNI is ≥ 40, with an ideal value of ≥ 44.5.

4.8 Exclusion Criteria

Patients meeting any of the exclusion criteria at baseline will be excluded from study participation.

1. Tumors of mucinous subtypes, or non-epithelial ovarian cancers (e.g., Brenner tumors, Sex-cord tumors).
2. Unresolved bowel obstruction.
3. Undiagnosed gastrointestinal bleeding.
4. Known CNS metastasis.
5. Inflammatory diseases of the bowel.
6. Concurrent therapy with any other investigational anti-cancer agent or treatments.
7. Known seropositive for active viral infection with human immunodeficiency virus (HIV); or active hepatitis B virus (HBV) or hepatitis C virus (HCV) infection within 4 weeks prior to study initiation.
8. History of thromboembolic event within the last 3 months.
9. Pregnant or breast-feeding women.
10. Small pox vaccination within 1 year of study therapy.
11. Any non-oncology vaccine therapy used for prevention of infectious diseases including seasonal (influenza) vaccinations within 2 weeks of the first dose of study drug.
12. At the time of eligibility assessment, have clinically significant cardiac disease (New York Heart Association Class III or IV; refer to *Appendix 5: New York Heart Association (NYHA) Functional Classification* for classification of symptoms).
13. Oxygen saturation <90% measured by pulse oximetry at rest.
14. Have received prior gene therapy or therapy with cytolytic virus of any type.
15. Be receiving concurrent antiviral agent active against vaccinia virus (e.g., cidofovir, vaccinia immunoglobulin, imatinib, ST-246) during the course of study.
16. Have known allergy to ovalbumin or other egg products.
17. Have clinically significant dermatological disorders (e.g., eczema, psoriasis, or any unhealed skin wounds or ulcers) as assessed by the Investigator during screening and during the study.
18. Prior malignancy (i.e., metastatic disease) active within the previous 3 years except for locally curable cancers that have been apparently cured, such as basal or squamous cell skin cancer, superficial bladder cancer, or carcinoma *in situ* of the cervix or breast, or any other stage I/II local malignancies.
19. Patients who have received chemotherapy, radiotherapy or any anti-cancer biologic therapies within 4 weeks prior to entering the study or has not recovered from adverse events due to agents administered more than 4 weeks earlier.
20. Patients who are less than 4 weeks from surgery (excluding the surgical placement of the indwelling catheter required for the application of the study drug and laparoscopy) or have insufficient recovery from surgical-related trauma or wound healing.
21. Patients who are receiving additional immunosuppressive therapy or any steroids (exception is acute concurrent corticosteroid usage if no more than 20 mg per day for medical management of patient, prednisolone equivalent is applied).
22. Have dementia or altered mental status that would prohibit informed consent.
23. Severe or uncontrolled medical disorder that would, in the Investigator's opinion, impair ability to receive study treatment (i.e., uncontrolled diabetes, chronic renal disease, chronic pulmonary disease or active, fever, systemic and/or uncontrolled infections, psychiatric illness/social situations that would limit compliance with study requirements).
24. Known drug or alcohol abuse.
25. Symptomatic malignant ascites defined as rapidly progressive ascites with abdominal distention and gastrointestinal dysfunction, breathing difficulties, and/or requiring frequent paracentesis more than once every 14 days.
26. Non-manageable pleural effusion and/or oxygen dependence on a routine basis. Stable manageable pleural effusions with or without a catheter can be allowed as long as other performance status requirements are met.

27. **Cohorts C & D -- Regarding bevacizumab if applicable**: Known hypersensitivity to bevacizumab, uncontrolled hypertension, history of stroke, or clinical findings suggestive of excessive risk for GI perforation (uncontrolled peptic ulcer disease, partial small bowel obstruction, etc.) that would make risks of bevacizumab unacceptable in the opinion of the Investigator.

4.9 Women of Childbearing Potential

Women of childbearing potential include any female who has experienced menarche and who has not undergone successful surgical sterilization (hysterectomy, bilateral tubal ligation, or bilateral oophorectomy) or is not postmenopausal. Postmenopausal is defined as:

- Amenorrhea ≥ 12 consecutive months without another cause and a documented serum follicle stimulating hormone (FSH) level > 35 mIU/mL or;
- Women with irregular menstrual periods and a documented serum follicle stimulating hormone (FSH) level > 35 mIU/mL NOTE: FSH level testing is not required for women ≥ 62 years old with amenorrhea of ≥ 1 year or
- Women on hormone replacement therapy (HRT).

Women who are using oral contraceptives, other hormonal contraceptives (vaginal products, skin patches, or implanted or injectable products), or mechanical products such as an intrauterine device or barrier methods (diaphragm, condoms, spermicides) to prevent pregnancy, or are practicing abstinence or where their partner is sterile (e.g., vasectomy) should be considered to be of childbearing potential.

5.0 PHASE 1B TREATMENT PLAN

Cohort	GL-ONC1 (dosing on 2 consecutive days)	N
1	3×10^9 pfu per day	3
2	1×10^{10} pfu per day	3
3	2.5×10^{10} pfu per day	3
4	4×10^{10} pfu per day	3
	Subtotal	**12-18** (based on optional cohort expansion)
• There is a 2-4 week safety observation window, with a minimum of 2 weeks between each cohort. • As an optional expansion, up to 6 more patients may be treated in any of the above cohorts.		

A temporary intraperitoneal catheter will be inserted 5 to 7 days prior to Day 1. Patients who have an existing indwelling peritoneal catheter or port at screening will have it surgically removed and replaced with a temporary peritoneal catheter by laparoscopic procedure. During the baseline laparoscopy, photos will be taken to document the extent of disease and assessed by the Sugarbaker *Peritoneal Cancer Index (Appendix 3)*. The catheter may be removed on or before Day 17. If medically required to manage ongoing ascites, timing of catheter removal is at the Investigator's discretion.

Post-treatment tumor biopsies (i.e., either on Day 10 or 17, and at Week 36) by CT-guided biopsy are optional. Tumor tissue that is safely accessible will be obtained from consenting patients.

5.1 3+3 Dose Escalation Design

Decisions to escalate to the next dose level will be based on whether dose-limiting toxicities (DLTs) were encountered, and available clinical (i.e., safety data) and non-clinical data (e.g., vital signs, routine lab tests). Safety and non-clinical data will be reviewed by the Cohort Management Committee (CMC) following the two- to four-week safety observation window, with a minimum of 2 weeks between cohorts

to allow sufficient time to monitor acute and sub-acute adverse events. Patients can be consented and screened for eligibility during the safety observation window to prepare for enrollment in the next dose cohort following a positive CMC vote.

- Dose escalation (i.e., opening a new cohort for patient treatment) will be based on the toxicities encountered within each cohort according to the following dose escalation plan: If one patient out of three in a dose group experiences a DLT, three more patients will be added to that dose group.
- If the next patient at this dose level experiences a DLT (this may be patient 4, 5, or 6 in the second group), this dose level will be closed (i.e., no further treatments administered).
- If two or more patients in a dose group of three to six patients experience a DLT, a one-half log lower dose, or the previous dose level will be employed.
- If tolerated, the lower dose level will be defined as the Maximum Tolerated Dose (MTD).
- If MTD is reached, a minimum of 6 patients will be treated at the determined viral product dose level to investigate and confirm the observations seen in the first 3 patients of the intended cohort.

In each cohort, a week for safety evaluation occurs between the first and second patients before treatment is initiated. Subsequent patients in a cohort may be treated in parallel with the second patient. Intrapatient dose escalations are not permitted.

5.2 Dose Limiting Toxicities (DLTs)

Toxicities will be graded according to the National Cancer Institute (NCI) Common Toxicity Criteria for Adverse Events (CTCAE) version 4.03. If a grade is not provided in the CTCAE for a particular adverse event, the Investigator will use his/her best medical judgment in assessing the toxicity grade.
GL-ONC1 administration should be discontinued if a patient experiences at least one of the following dose limiting toxicities (DLT). A DLT is defined as any of the following adverse events having a degree of attribution to GL-ONC1 (i.e., probably, possibly or definitely) occurring within 30 days after the last GL-ONC1 treatment. These include:

- Any Grade 3 non-hematologic adverse event lasting more than 72 hours toxicity (excluding nausea/vomiting and diarrhea; exceptions may be made for transient (e.g. lasting < 7 Days) Grade 3 elevations of ALT/AST in the presence of known liver metastases and without evidence of other hepatic injury, if agreed by the Investigator); Any Grade 4 (≥ 4) non-hematologic adverse event;
- Any Grade 4 (≥ 4) hematologic adverse event lasting more than 7 days;
- Grade 2 or greater bronchospasm requires discontinuation of the viral product;
- All Grade 2 hypersensitivity reactions to GL-ONC1 treatment;
- Any adverse event that leads to a discontinuation of GL-ONC1 infusion.

5.3 Study Stopping Rules

The following rules will apply, if warranted:

- Individual Treatment: Any patient who develops a DRUG-RELATED dose limiting toxicity (DLT), as defined in this protocol, will not receive further administration of the investigational product and the patient are followed for drug related toxicities until resolution, return to baseline values, or if the drug related toxicity is deemed irreversible.
- Study Stopping Rules: If 2 or more out of the six patients (≥ 33%) experience a DLT at any dose level, the Investigator, Medical Monitor and the Sponsor may elect to employ a one-half log lower dose or the prior dose level. The site may proceed with this lower dose level only following written notification from the Sponsor.

5.4 Study Pausing Rule

If the following events occur during the Treatment Period (i.e., Week 1 Days 1 to 5) up to 30 days post last GL-ONC1 infusion, the study will be placed on hold until an appropriate evaluation of the cause of the toxicity is determined, and a plan of correction, if necessary, is established:

- Death (other than death related to progressive disease);
- Grade 3 to 4 adverse event rate > 70% in the first 30 days.

5.5 Revision to Enrollment Criteria

If the study is stopped or paused to evaluate the occurrence of an event(s) defined above, revisions to the enrollment criteria may be required through a protocol amendment to exclude individuals who might be at a higher risk of developing particular adverse reactions. This decision will be made in collaboration between the CMC and the Sponsor.

5.6 Maximum Feasible Dose (MFD)

The MFD is defined as the highest dose level when either dose limiting toxicities (DLTs) or the maximum tolerated dose (MTD) are not reached.
Maximum Tolerated Dose (optional)

The dose one level lower will be declared as the MTD, if a DLT is observed in ≤ 1 patient of the total 6 patients in the dose level.

6.0 PHASE 2 TREATMENT PLAN

Cohort[1]	GL-ONC1[2] (dosing on 2 consecutive days)	N
A	3×10^9 pfu per day	20
B	1×10^{10} pfu per day	20
	Subtotal	**40**

[1] Enrollment into Cohorts A and B is by Sponsor assignment.

[2] An optional second cycle of GL-ONC1 could be given through a temporary intraperitoneal percutaneous catheter for intraperitoneal delivery at the discretion of the Investigator and the Sponsor. The catheter will be inserted and removed by Interventional Radiology. Patients who have demonstrated disease control (CR + PR + SD ≥ 15 weeks) from first cycle of GL-ONC1 but start to show disease progression with or without further chemotherapy and/or bevacizumab, will be considered for a second cycle of GL-ONC1. Optional CT or ultrasound-guided biopsies will be obtained prior (within 2 weeks or on day of catheter placement) to and ~4 weeks after second cycle of GL-ONC1.

Disease progression could be determined by RECIST 1.1 or based on confirmed upward trend of CA-125 values. For example, when CA-125 increases to more than twice the nadir value and above 70 u/mL (CA-125 nadir < 100); or when there is an increase in CA-125 levels by >100 when the nadir was >100 u/mL. Repeat CA-125 2-4 weeks after an elevation to confirm a trend upwards. When disease progression is suspected clinically based on persistent rise in CA-125, physical exam findings, and /or symptoms, a confirmation CT scan will be obtained. In pts whose CA-125 has been shown non-diagnostic (low levels despite RECIST lesions on CT scans), the decision to proceed with second cycle of GL-ONC1 will be made on progression by CT scan coupled with physical exam findings and symptoms.

Cohort	GL-ONC1	Chemotherapy (chemo)	Bevacizumab (bev)	N
Safety run-in cohort	1 cycle @ 3 ×10^9 pfu per day × 2 consecutive days	Carboplatin doublet, up to 6 cycles	With chemotherapy, plus maintenance as single agent	3-6
C				Up to **35**

- Safety run-in cohort (with staggered enrollment and treatment): There is at least 28-day interval for safety observation between each subject. Enrollment into Ch C occurs only after completing the safety run-in cohort.
- Combination with bevacizumab is allowed but not required. If developing toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment, e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel, to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if tolerated well and determined to be clinically beneficial after 6 cycles.
- Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo, single agent bev, or combination single agent chemo with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

Phase 2				
Cohort	**GL-ONC1**	**Chemotherapy (chemo)**	**Bevacizumab (bev)**	**N**
Safety run-in cohort	1 cycle @ 2,3,5 ×10^9 pfu by IV route for 3 consecutive days	Carboplatin doublet, 6 cycles	With chemotherapy, plus maintenance as single agent	3-6
D				15

- Safety run-in cohort (with staggered enrollment and treatment): There is at least 28-day interval for safety observation between each subject. Enrollment into Ch D occurs only after completing the safety run-in cohort.
- GL-ONC1 is administered intravenously as an undiluted bolus infusion.
- Progression-free Survival (PFS) is to be assessed after IV doses of GL-ONC1 until disease progression, if clinically applicable.
- Chemo +/- bev will start either at time of disease progression from GL-ONC1, or will start at the discretion of the PI as clinically indicated based on clinical assessment (shall consult with Sponsor if falls into this category).
- Details of chemo +/- bev regimen are the same as to Ch C.
- Combination with bev is allowed but not required. If developing toxicity to any component(s) of the regimen, treatment could continue with the remaining component(s). In addition, replacement of a doublet component with carboplatin is allowed after start of treatment, e.g., replacing pegylated liposomal doxorubicin (PLD) with docetaxel, to complete all 6 treatment cycles. Schedules will then be adjusted accordingly. The number of cycles of chemo +/- bev may be expanded for up to 3 more cycles at the discretion of the Investigator if tolerated well and determined to be clinically beneficial after 6 cycles.
- Ongoing therapy may be administered following carboplatin doublet +/- bev with either the non-platinum single agent chemo agent, single agent bev, or combination single agent chemo with bev as maintenance until progression or unacceptable toxicity, at the discretion of the Investigator.

For Cohorts using IP route of delivery of virus, a temporary IP catheter will be inserted around 3 to 7 days (5 days preferred) prior to administration of the first GL-ONC1 dose. Patients who have an existing indwelling peritoneal catheter or port at screening will have it surgically removed and replaced with a temporary peritoneal catheter by laparoscopic procedure. During the baseline laparoscopy, photos will be taken to document the extent of disease and assessed by the Sugarbaker *Peritoneal Cancer Index* (*Appendix 3*). The catheter may be removed on or before 14 days after date of the first GL-ONC1 dose. If medically required to manage ongoing ascites, timing of catheter removal is at the Investigator's discretion.

7.0 COHORT MANAGEMENT COMMITTEE

Phase 1b: For Cohorts 1 to 4, decisions to escalate to the next level, or, when appropriate, to an intermediate level are made by the Investigator following consultation with the Cohort Management Committee (CMC) members. CMC membership will consist of the Investigator, additional clinical site qualified medical personnel, as well as two qualified and experienced sponsor representatives (e.g., Medical Monitor and a senior staff member with knowledge of GL-ONC1's development) as voting

members. CMC members will follow procedures described in the 'Cohort Management Committee Charter' for review of safety data. Judgements on whether the trial should be stopped or paused based on review of safety data will be made jointly between the CMC and the Sponsor.

Phase 2: The CMC will review safety data on an as needed basis determined by the frequency, severity and type of suspected serious adverse reactions.

The safety data report reviewed by the CMC and the committee's recommendations will be documented in the Sponsor's trial master file (TMF) and the Investigator's intuitional site files (ISF).

8.0 REPLACEMENT OF PATIENTS

All patients must receive GL-ONC1 to be considered evaluable for safety and efficacy. Replacement patients may also be enrolled in a cohort if a patient is withdrawn from investigational treatment for a reason that is unrelated to study agent toxicity.

9.0 CONCOMITANT THERAPY

9.1 Permitted Concomitant Therapy

As the presence of fevers, rigors, nausea, etc., following treatment may show a body's response to viral therapy that may be favorable toward an antitumor response, it is requested that the Investigator not prescribe medications such as analgesics, antipyretics, opioids, benzodiazepines, phenothiazines, and general anesthetics (e.g., Propofol) for 48 hours prior to the first dose of GL-ONC1 and for 14 days following the last dose of GL-ONC1 to suppress these symptoms, unless symptoms are severe requiring urgent medical intervention.

Medications required to treat adverse events and manage cancer symptoms, concurrent stable disease (e.g., controlled hypertension), and supportive care agents such as erythropoietin or blood transfusion, and pain medications are allowed. The patient needs to notify the investigational site about any new medications she takes after the start of the study medication. All medications (other than study drug) and significant non-drug therapies (including physical therapy and blood transfusions) administered after the patient starts treatment with study drug must be recorded in the patient's medical record.
Below is a list of permitted supportive therapy/procedures:

- Antiemetics;
- Antidiarrheals;
- Antibiotics;
- Nutritional and fluid supplementation;
- Myeloid growth factors (except routine use of G-CSF and GM-CSF);
- Use of granulocyte colony–stimulating factors (G-CSF) is allowed at Investigator's discretion, but not mandated. G-CSF could lower rates of febrile neutropenia; reduce the incidence of infection, antibiotic use, and hospital admission, and allow for maintenance of dose intensity;
- Bisphosphonates;
- Blood transfusions and the use of erythropoietin are permitted at the discretion of the treating physician;
- Prophylactic anticoagulant therapy (low dose) and full anticoagulation are allowed.
- Hydration as needed (e.g., administration of 1 Liter of 0.9% normal saline).

9.2 Prohibited Concomitant Therapy

Other investigational therapies must not be used while the patient is on the study. Anti-cancer therapy (e.g., chemotherapy, biologic or radiation therapy, surgery) other than the study treatments must not be given to patients while the patient is on the study medication. If such agents are required for a patient, then the patient must be discontinued from the treatment portion of the study.

The list of non-permitted concurrent supportive therapy is listed below:

- Surgery;
- Other investigational treatment;
- Other anti-cancer treatments;
- Systemic immunosuppressants (except concurrent corticosteroid usage, if no more than 20 mg per day, prednisolone equivalent is applied);
- Immunotherapy (other than specified in the protocol);
- Routine prophylactic use of growth factor (G-CSF or GM-CSF).

9.3 Antidote - Rescue Medications

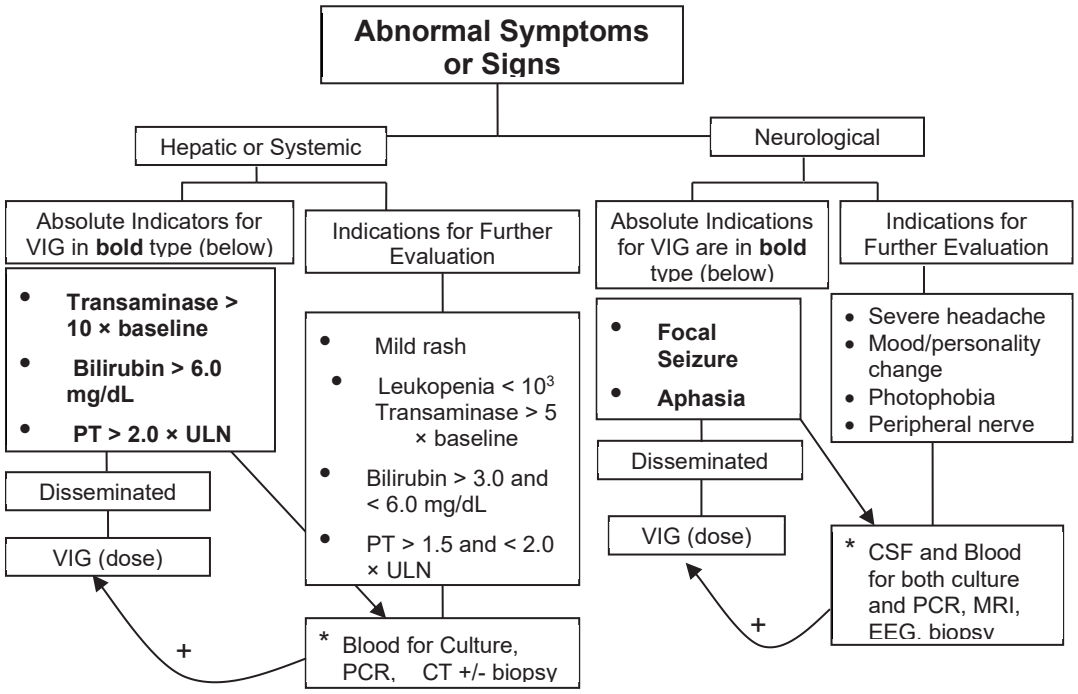
Patients who develop serious harmful side effects due to viral administration will be treated by the Investigator.

9.4 Treatment Guidelines for Use of Vaccinia Immunoglobulin (VIG)

Since GL-ONC1 is a replication-competent virus, it may cause generalized vaccinia infection.

A standard treatment of generalized vaccinia infection is the use of vaccinia immunoglobulin (VIG) (https://www.cdc.gov/mmwr/preview/mmwrhtml/rr5204a1.htm). In addition, cidofovir has also been listed as a potential medication. A copy of the package insert for vaccinia virus immunoglobulin will be held in the Investigator Site File (ISF) which lists information on the use, storage requirements, efficacy and known adverse reactions to treatment with VIG.

Anti-vaccinia therapy will only be used if there is evidence of a systemic vaccinia infection. The figure below provides a management decision tree in cases where systemic vaccinia infection occurs in the treated patient population.



* These tests will be performed based on the clinical judgment of the Site Investigator.

10.0 PERITONEAL CATHETER AND ASCITES MANAGEMENT

10.1 Placement of Peritoneal Catheter

For patient enrolled in cohorts with IP delivery, the peritoneal catheter will be surgically placed requiring light general or extensive anesthesia. Thereby, visualization of the peritoneal cavity, precise placement of the catheter tip, lysis of adhesions and omentectomy, if required, are ensured. Institutional surgical practices and after care for the placement of the peritoneal catheter will be followed.

10.2 Management of Ascites if Clinically Indicated

If urgent management of ascites is clinically indicated, drainage of ascites may be ideally performed (e.g., paracentesis, existing catheter, etc.) no sooner than 20 hours after each dose of virus treatment.

10.3 Drainage of Ascites Prior to Administration of the First Dose of GL-ONC1

For patients who have ascites prior to the intraperitoneal infusion of 500 mL of GL-ONC1, the following procedures will be conducted:

Patients must have sufficient peritoneal space to allow instillation of GL-ONC1 via an intraperitoneal catheter. However, to ensure sufficient peritoneal space, the surgeon can tent the peritoneal wall up by lifting the Halstead mosquito artery forceps that are holding the edges of the peritoneal opening. The potential space can then be directly visualized. In addition, a blunt instrument, for example, Watson Cheyne dissector, can be used to confirm if there is a potential space.

11.0 GL-ONC1 ADMINISTRATION

11.1 Justification of Route of Administration

Phase 1b Cohorts and Phase 2 Cohorts A, B & C: The intraperitoneal route of administration was chosen to deliver GL-ONC1 directly to its intended site of action. In addition, data obtained in a Phase 1 trial of GL-ONC1 conducted at the University of Tübingen, Germany in 9 patients with peritoneal carcinomatosis (including 4 patients with ovarian cancer), and clinical data from using other biological agents such as measles vaccine virus agents, showed that intraperitoneal infusions are in general well tolerated and safe (41).

Phase 2 Cohort D: Extensive pre-clinical studies testing IV dose(s) of GL-ONC1 have demonstrated robust anti-tumor efficacy in a wide range of human cancer types in animal models. Clinically, IV dose(s) of GL-ONC1 has been administered to 67 patients in three phase 1/1b clinical trials with advanced cancers, demonstrating good tolerability, tumor cell and tissue infection, evidence of tumor shrinkage, and a favorable trend of clinical benefit. In the proposed Cohort D, GL-ONC1 will be administered as an undiluted IV bolus infusion over 3 days at 2,3,5 × 10^9 pfu/dose/day, with a cumulative dose of 1 × 10^{10} pfu total. Patients will be followed for safety and efficacy until disease progression from GL-ONC1 before given further chemotherapy +/- bevacizumab. The previous GL-ONC1-002 trial (Royal Marsden Hospital, UK), the GL-ONC1-005 trial (UCSD; Mell *et al*., Clin Cancer Res. 2017;23(19):5696-702), the GL-ONC1-011 trial (UCSD), and the ongoing GL-ONC1-021 EAP study (Florida Hospital Cancer Institute) support this proposed IV treatment schedule and doses. For example, IV doses of GL-ONC1 up to 3 × 10^9 pfu or 5 × 10^9 pfu administered to 29 patients with advanced solid tumors (GL-ONC1-002 and GL-ONC1-005 studies; safety summary provided in the Annual Development Safety Update Report) have been shown to be well tolerated. In addition, condensed dosing schedule has been tested previously with demonstrated safety and promising pharmacokinetics and pharmacodynamics profiles. Cumulative IV dose of 1 × 10^{10} pfu given within 3-5 days has been tested in 8 patients with advanced solid tumors (GL-ONC1-011 and GL-ONC1-021 studies), and was shown to be tolerated. No deaths were determined to be related to GL-ONC1 treatment. No Maximum Tolerated Dose was reached in any of these studies. In summary, we anticipate the proposed GL-ONC1 treatment regimen followed by further chemotherapy +/- bevacizumab will be tolerated and yield potential benefits to this advanced stage patient population.

11.2 Mode of Administration

Phase 1b cohorts and Cohorts A, B & C: GL-ONC1 (final reconstituted volume of 500 mL) will be administered as a bolus via a peritoneal catheter. In order to support distribution of the study drug solution within the peritoneal space, patients should, at the Investigator's discretion, regularly change position from prone position to lateral and supine position for about 30 minutes after the completion of the infusion of the study drug.

Phase 2 Cohorts A & B: An optional second cycle of GL-ONC1 could be given through temporary percutaneous catheter for intraperitoneal delivery at the discretion of the Investigator and the Sponsor. Insertion and removal of catheter will be by Interventional Radiology.

Phase 2 Cohort D: GL-ONC1 is administered as an intravenous undiluted bolus infusion.

Any cohort: After completion of ‘safety run-in cohort’ for Cohort D, and after treatment with 1st cycle of GL-ONC1 and further chemo +/- bev, a second cycle of GL-ONC1 by IV route is allowed for 3 patients and expandable to another 3 patients upon approval by the Sponsor.

12.0 TREATMENT COMPLIANCE

Treatment compliance will be monitored by drug accountability as well as by a review of the patient's medical records and case report forms (electronic).

13.0 REMOVAL OF PATIENTS FROM STUDY TREATMENT AND STUDY

Patients must discontinue investigational product for any of the following reasons:

- Withdrawal of informed consent (patient's decision to withdraw for any reason).
- Patient voluntarily withdraws from treatment but allows follow-up.
- Patient is unable to comply with protocol requirements.
- Any clinical adverse event (AE), laboratory abnormality or intercurrent illness which, in the opinion of the Investigator, indicates that continued participation in the study is not in the best interest of the patient.
- Pregnancy.
- Termination of the study by Genelux.
- Loss of ability to freely provide consent through imprisonment or involuntarily incarceration for treatment of either a psychiatric or physical (e.g., infectious disease) illness.
- Protocol defined reasons for discontinuation.
- *Lost to follow-up:* If a patient cannot be located, the patient may be considered 'lost to follow-up'. All attempts to contact the patient during this time must be documented.

All patients who discontinue should comply with protocol specified follow-up procedures as outlined in *Section 15.0 Study Assessments and Procedures.* The only exception to this requirement is when a patient withdraws consent for all study procedures including follow-up, or loses the ability to consent freely (i.e., is imprisoned or involuntarily incarcerated for the treatment of either a psychiatric or physical illness).

If a patient was withdrawn before completing the study, the reason for withdrawal must be entered on the appropriate case report form (CRF) page.

14.0 PREMATURE TERMINATION OF THE STUDY

If the Sponsor or the Investigator becomes aware of conditions or events that suggest a possible hazard to patients, they must notify the applicable parties immediately. The study may be terminated early at the Sponsor or the Investigator's discretion in the absence of such findings. Conditions that may warrant termination include, but are not limited to:

- Meet the criteria as defined in *Section 5.3, Study Stopping Rules.*
- Unsafe or unethical practices.
- A decision on the part of the Sponsor or the Investigator to suspend or discontinue the development of the program.
- Inadequate drug supply to continue trial.

15.0 STUDY ASSESSMENTS AND PROCEDURES

15.1 Time and Events Schedule

The study calendars below show the timing of the study procedures and tests performed during Year 1. Refer to *Section 4.4 Allowable Window of Time around Study Visits* and *Section 4.5 Treatment Compliance Criteria* for allowable variance windows.

Screening Assessments: For patients screened and enrolled into study within a short period of time prior to treatment, it is not considered a protocol deviation if all screening procedures are collected at one-time point instead of two-time points (i.e., toxicity, physical examination, weight, performance status, vital signs, serum pregnancy and clinical labs, concomitant medications).

Assessment of Response to Treatment: Eligible patients who enter with measurable disease at baseline will have contrast spiral CT scans performed at the times listed in the table below. Patients with non-measurable disease (Phase 1b only) will be assessed for response to treatment by PET scan, physical examination, and CA-125 level monitoring. The performance of either a PET, PET/CT or CT scan with contrast is at the discretion of the Investigator, as clinically indicated. If a patient is allergic to intravenous CT contrast, a preferred recommendation is the use of oral CT contrast or secondarily by MRI. RECIST 1.1 indicates that the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up.

Tumor Biopsies:
For patients treated in the Phase 2 portion of this study, a baseline tumor biopsy is required. A baseline tumor biopsy is optional in the Phase 1b portion. Depending on the location of the tumor(s) selected by the Investigator for sampling, biopsies may be performed through the laparoscope (preferred method if treated in Phase 1b cohorts or Phase 2 Cohorts A, B & C, if feasible) or by CT-guided or ultrasound-guided needle biopsy (e.g., Phase 2 Cohort D), if tumor is safely accessible. Tumor tissue that is safely accessible by needle biopsy will be obtained from consenting patients. If additional tumor biops(ies) are obtained from the same patient at later time point(s), it is preferred that tumor samples are from the same baseline body location if feasible. The number of patients to obtain optional tumor biopsies will be at the discretion of the Sponsor.

- PHASE 1b Post-treatment Tumor Biopsies: Post-treatment CT-guided biopsies may occur either on Day 10 or 17, and at Week 36.
- PHASE 2 COHORTS A & B -- Post-treatment Tumor Biopsies (CT-guided or ultrasound-guided biopsies): Post-treatment needle biopsies may occur either 7 or 14 days after date of the first GL-ONC1 dose, and/or at Week 6 or Week 15 (± 7-day variance at each time point). For patients who receive a second cycle of GL-ONC1, optional needle biopsies will be obtained prior to (within 2 weeks or the same day as catheter placement) and at approximately 4 weeks after second cycle of GL-ONC1.
- PHASE 2 COHORTS C & D *(timeline adjusted accordingly based start of chemo)* -- Post-treatment Tumor Biopsies (CT-guided or ultrasound-guided): A preferred but optional post-treatment needle biopsy may occur between Weeks 2 to 5 after date of the first GL-ONC1 dose, and prior to initiation of further chemotherapy. Another optional post-treatment needle biopsy may be obtained after second cycle (either during Week 11 for q3w, or Week 13 for q4w) or after third cycle (Week 14 for q3w, or Week 17 for q4w) of chemotherapy. Biopsy will be obtained after CT scan if they are conducted on same day.

ALL COHORTS -- Unscheduled Biological Sample Collection: During the course of the study, unscheduled biological samples (e.g., tumor tissue, ascites, pleural fluid, blood, etc.) provided from consenting patients will be kept for translational research purposes. Preferably, unscheduled research

blood is collected before initiation of chemotherapy, then after completion of every 2 cycles of chemo, including the last cycle. Beyond that, blood can be drawn on a quarterly basis for up to another 12 mos. Consult with Sponsor before collecting and shipping of unscheduled ascites or pleural fluid samples.

Phase 1b & Phase 2 Cohorts A & B -- Post-treatment Period Week 2 to Week 48 & Year 2 Long-term Follow-up: If a patient has documented disease or clinical progression, and receives other anti-cancer treatment, follow-up is conducted by telephone to assess survival, disease and anti-cancer treatment status, and assessment if a SAE(s) occurred since the last telephone call (or visit if first follow-up call). If the Investigator determines a SAE has a degree of attribution to GL-ONC1 treatment, submit SAE Report to the Pharmacovigilance Team via the electronic data capture (EDC) system or by paper SAE Report CRF if EDC not available. Non-serious AEs are not collected. In order to determine if other anti-cancer treatment may have a synergistic effect following GL-ONC1 treatment, the sponsor asks that sites timely provide de-identified radiologic imaging reports and clinical labs, including CA 125 results, on an *ad hoc* basis when available.

Phase 1b & Phase 2 Cohorts A & B -- Year 2 Long-term Follow-up Visits (± 2 weeks): Procedures are performed quarterly with the first clinic visit occurring 3 months after Week 48, or the last visit performed during the Post-treatment Period.

- Toxicity assessment (SAEs deemed to have a degree of attribution to GL-ONC1 Treatment)
- Disease assessment by contrast spiral CT scan or PET/PET-CT scan with contrast (at Investigator's discretion);
- Physical exam;
- ECOG performance status evaluation;
- Vital signs;
- Concomitant medications;
- Clinical labs.

Phase 1b & Phase 2 Cohorts A, B -- Year 3 Long-term Follow-up Telephone Calls (± 2 weeks): During this time, patients are called quarterly to assess survival, disease and treatment status. Timing of the first call is based on the timing of the last Year 2 Post-treatment follow-up visit. Toxicity assessment is obtained for SAEs deemed to have a degree of attribution to GL-ONC1 Treatment. If applicable, submit SAE Report to the Pharmacovigilance Team via the electronic data capture (EDC) system or by paper CRF if EDC not available. Non-serious AEs are not collected.

Phase 2 Cohorts C & D Year 2 and Year 3 Long-term Follow-up: During this period, imaging studies will be performed at the Investigator discretion (e.g., on quarterly basis) along with other clinical procedures to follow patient's health status and response to treatment. If patient is unable to come to clinic visits, a telephone call should be performed to collect information as listed below for Phase 2 Cohorts A & B for survival, disease and treatment status. Non-serious AEs are not collected.

ALL PHASES/ALL COHORTS -- Clinical lab parameters collected during the study:

- Complete Blood Count (CBC) with differential: Hemoglobin, hematocrit, red blood cell count, white blood cell count, platelet count, neutrophils (%), lymphocytes (%), monocytes (%), eosinophils (%), basophils (%), reticulocytes (%), International Normalized Ratio (INR)
- Comprehensive Metabolic Panel: Calcium, blood urea nitrogen (BUN), creatinine, glucose, chloride, sodium, potassium, carbon dioxide 2 (CO_2), lactate dehydrogenase (LDH), aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin, gamma-glutamyl transferase (GGT), total protein, albumin, alkaline phosphatase (ALP), uric acid, magnesium, phosphorous, creatine kinase.
- Other serum clinical labs: C-reactive protein (inflammatory), Cancer Antigen 125 (CA 125)

- Urinalysis: Specific gravity, pH, glucose, blood, ketones, bilirubin, protein, WBC, RBC, crystals, bacteria, epithelial cells

COHORTS C & D: Clinical labs can be ordered according to the Cohorts C & D study calendars below or in accordance with institutional standards during chemotherapy ± bevacizumab. All the clinical lab parameters listed in the protocol remain the same (e.g., including testing of CA-125, LDH, etc.)

Vital Signs collected (with exception when other anti-cancer treatment occurs):

- Blood pressure
- Pulse rate
- Respiratory rate
- Temperature
- Oxygen saturation (baseline)
- Weight
- Height (baseline)

Phase 1b & Phase 2 Cohorts A & B -- Year 1: Screening, Treatment Period and Post-treatment Period Procedures

	SCREENING		TREATMENT			POST-TREATMENT PHASE								
Procedures	**Within 4 Wks of 1st Dose**	**Within 3-7 Days of 1st Dose**	**W1 (3 consecutive days)**			**W2 D10/ 7 days after 1st dose**	**W3 D17/ 14 days after 1st dose**	**W6 D38**	**W15 D101**	**W24 D164**	**W30 D206**	**W36 D248**	**W42 D290**	**W48 D332**
Informed Consent	√													
Medical & cancer disease history	√													
Demographics	√													
HIV, HBsAg, anti-HCV testing	√													
Toxicity Assessment 1 Related to screening procedures	√[1]	√[1]	√	√	√	√	√	√	√	√	√	√	√	√
Spiral CT scan	√							√	√	√		√		√
PET or PET-CT scan 1 Frequency is performed at the discretion of the Investigator at any of the time points listed	√[1]							√[1]	√[1]	√[1]		√[1]		√[1]
Physical exam, weight, performance status, vital signs	√	√	√			√		√	√	√	√	√	√	√
Height	√													
Vital signs: pre-GL-ONC1 dose & at 30 min (±5 min) intervals up to 120 minutes			√	√										
Serum pregnancy test for WOCBP	√	√												
Clinical labs: CBC, comprehensive metabolic panel, LDH, C-RP, CA-125, urinanalysis 1 Samples collected prior to treatment	√	√	√[1]	√[1]	√	√	√	√	√	√	√	√	√	√
Concomitant medications	√	√	√	√	√	√	√	√	√	√	√	√	√	√
FACT-O Quality of Life Questionnaire	√							√	√	√		√		√
Implantion of peritoneal catheter & Laparoscopy - **1 Phase 1b:** 5 to 7 days prior to start of Treatment Week 1 **2 Phase 2:** 3-7 days prior to 1st GL-ONC1 (5 days preferred)		√[1,2]												
REQUIRED: Tumor biopsy during laparoscopy		√												
Cancer gene mutation profile of tumor biopsy		√												
Photos to assess extent of disease via laparoscopy		√												
Accessment of disease by Peritoneal Cancer Index		√												
Research bloods: Lymphocyte, cytokine analysis, neutralizing antibody titers, beta-glucuronidase 1 Samples collected prior to treatment		√	√[1]	√[1]	√	√	√	√	√	√	√	√		√
Viral shedding (oral & anal swab by VPA, qPCR) collected post-GL-ONC1 treatment					√									

	SCREENING		TREATMENT			POST-TREATMENT PHASE								
Procedures	**Within 4 Wks of 1st Dose**	**Within 3-7 Days of 1st Dose**	**W1 (3 consecutive days)**			**W2 D10/ 7 days after 1st dose**	**W3 D17/ 14 days after 1st dose**	**W6 D38**	**W15 D101**	**W24 D164**	**W30 D206**	**W36 D248**	**W42 D290**	**W48 D332**
OPTIONAL: CT-guided tumor biospy (Ph 2: CT or ultrasound guided biopsy) **1 Ph 1b & 2**: Needle biospy can occur on either day **2 Ph 2:** Needle biopsy can occur on either day **3 Ph1:** For this phase only						√[1]	√[1]	√[2]	√[2]			√[3]		
OPTIONAL: Circulating Tumor Cells (CTC): 2 × 10 mL whole blood samples **1 Ph 1b & Ph2:** Collected up to 7 days prior to the first GL-ONC1 dose **2 Ph 1b:** If baseline CTC sample 0, don't collect post-treatment samples **3 Ph 1b:** Collect samples if baseline CTC was +5 CTCs **4 Ph 1b:** Collect samples if baseline CTC was 1-5 CTCs. If W3D15 5+ collect CTC on W15D101 & W24/D164 **5 Ph 2:** Collect CTC samples regardless of baseline CTC count		√[1,2]					√[3,4,5]	√[5]	√[3,4,5]	√[3,4,5]				
Peritoneal fluid sampling (for patients with ascites) cytology analysis (including tumor cells & immune cells); Viral kinetics (peritoneal fluid by VPA & qPCR) 1 Complete drainage of ascites prior to the first GL-ONC1 dose 2 10 mL ascites sample collected 3 10 mL ascites sample collected if catheter is in place			√[1]		√[2]	√[2]	√[3]							
Peritoneal wash to obtain cell components & supernatant 1 Infusion of 1 to 1.5 of Ringer Lactate & drainage prior to the first GL-ONC1 dose			√[1]											
GL-ONC1 treatment: For Phase 2 Cohorts A & B, 2nd cycle can be given by percutaneous catheter at discrection of PI & Sponsor for patients demonstrated disease control.			√	√										

COHORT C Treatment q3 Weeks	SCREENING		GL-ONC1 TREATMENT			POST-GL-ONC1 TREATMENT PERIOD																										POST-TREATMENT FOLLOW-UP VISIT	FOLLOW-UP
Procedures	Within 4 Wks of 1st Dose, Ideally within 2 weeks	Within 3-7 Days of 1st Dose	W1 D1	W1 D2	W1 D3	W2 to W5	CYCLE 1 W6	CYCLE 2 W9	W11	CYCLE 3 W12	W14	CYCLE 4 W15	CYCLE 5 W18	CYCLE 6 W21	W23	MAIN. CYCLE 1 W24	MAIN. CYCLE 2 W27	MAIN. CYCLE 3 W30	MAIN. CYCLE 4 W33	W35	MAIN. CYCLE 5 W36	MAIN. CYCLE 6 W39	MAIN. CYCLE 7 W42	MAIN. CYCLE 8 W45	W47	MAIN. CYCLE 9 W48	MAIN. CYCLE 10 W51	MAIN. CYCLE 11 W54	MAIN. CYCLE 12 W57	W59	30 Days Following Last Dose/Cycle	PRN Per PI (e.g., Quarterly for Year 2 to Year 3)
Informed Consent	√																															
Medical & cancer disease history	√																															
Demographics	√																															
HIV, HBsAg, anti-HCV testing	√																															
Implantion of peritoneal catheter & Laparoscopy **1** 3-7 days prior to 1st GL-ONC1 (5 days preferred)		√[1]																														
REQUIRED: Tumor biopsy during laparoscopy		√																														
Cancer gene mutation profile of tumor biopsy		√																														
Photos to assess extent of disease via laparoscopy		√																														
Accessment of disease by Peritoneal Cancer Index		√																														
Toxicity Assessment **1** Related to screening procedures **2** Toxicity assessment done at each study visit during this time frame **3** Report SAEs if causality to GL-ONC1	√[1]	√[1]	√	√	√	√[2]	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√[3]
Spiral CT scan **1** Performed immediately prior to 1st cycle of chemotherapy ± bevacizumab	√						√[1]				√				√					√					√					√		√
Physical exam, weight, performance status, vital signs **1** Assessments done at Weeks 2 and 3	√	√	√			√[1]	√	√		√	√		√	√	√		√	√	√	√		√	√	√	√		√	√	√	√	√	√
Height	√																															
Vital signs: pre-GL-ONC1 dose & at 30 min (±5 min) intervals up to 120 minutes			√	√																												
Serum pregnancy test for WOCBP	√	√																														
Clinical labs: CBC, comprehensive metabolic panel, LDH, C-RP, CA-125, urinanalysis **1** Samples collected prior to treatment **2** Samples collected at Weeks 2 and 3	√	√	√[1]	√[1]	√	√[2]	√[1]	√[1]		√[1]	√	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√	√
Concomitant medications **1** Obtained at each study visit during this time frame	√	√	√	√	√	√[1]	√	√		√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	
FACT-O Quality of Life Questionnaire	√						√				√				√					√					√					√		√
Research bloods: Lymphocyte, cytokine analysis, neutralizing antibody titers, beta-glucuronidase **1** Samples collected prior to treatment **2** Samples collected at Weeks 2 and 3		√	√[1]	√[1]	√	√[2]	√[1]	√			√				√					√					√					√	√	

COHORT C Treatment q3 Weeks	SCREENING		GL-ONC1 TREATMENT			POST-GL-ONC1 TREATMENT PERIOD																									POST-TREATMENT FOLLOW-UP VISIT	FOLLOW-UP
Procedures	Within 4 Wks of 1st Dose, Ideally within 2 weeks	Within 3-7 Days of 1st Dose	W1 D1	W1 D2	W1 D3	W2 to W5	CYCLE 1 W6	CYCLE 2 W9	W11	CYCLE 3 W12	W14	CYCLE 4 W15	CYCLE 5 W18	CYCLE 6 W21	W23	MAIN. CYCLE 1 W24	MAIN. CYCLE 2 W27	MAIN. CYCLE 3 W30	MAIN. CYCLE 4 W33	W35	MAIN. CYCLE 5 W36	MAIN. CYCLE 6 W39	MAIN. CYCLE 7 W42	MAIN. CYCLE 8 W45	W47	MAIN. CYCLE 9 W48	MAIN. CYCLE 10 W51	MAIN. CYCLE 11 W54	MAIN. CYCLE 12 W57	W59	30 Days Following Last Dose/Cycle	PRN Per PI (e.g., Quarterly for Year 2 to Year 3)
OPTIONAL: Needle tumor biospy (CT or ultrasound guided biopsy) **1** Needle biospy can occur at 1 time point during this time frame **2** Needle biopsy can occur on either week; after CT scan, if done on the same day (±7 days)						√[1]			√[2]		√[2]																					
OPTIONAL AT SPONSOR'S DISCRETION: Circulating Tumor Cells (CTC): 2 × 10 mL whole blood samples **1** Collected up to 7 days prior to the first GL-ONC1 dose **2** Collected at Week 3 **3** Sample collected prior to treatment		√[1]				√[2]	√[3]			√				√																		
Peritoneal fluid sampling (for patients with ascites) cytology analysis (including tumor cells & immune cells); Viral kinetics (peritoneal fluid by VPA & qPCR) **1** Complete drainage of ascites prior to the first GL-ONC1 dose **2** 10 mL ascites sample collected **3** 10 mL ascites sample collected at Week 2, and at Week 3 if catheter is in place			√[1]		√[2]	√[3]																										
Peritoneal wash to obtain cell components & supernatant **1** Infusion of 1 to 1.5 of Ringer Lactate & drainage prior to the first GL-ONC1 dose			√[1]																													
GL-ONC1 treatment			√	√																												
Viral shedding (oral & anal swab by VPA, qPCR) collected post-GL-ONC1 treatment					√																											
Chemotherapy Investigator's Choice: Carboplatin doublet for up to 6 cycles. Chemotherapy component could be substituted if due to toxicity or by Investigator's choice. If due to platinum-allergy, single-agent non-platinum chemotherapy is allowed.							√	√		√		√	√	√																		
Bevacizumab 10 mg/kg, q3 weeks with chemotherapy agents (7.5 mg/kg q2 weeks with PLD) for up to 6 cycles, and as a single agent maintenace q3 weeks for 9 months. Dose reduction is allowed at Investigator's discretion.							√	√		√		√	√	√		√	√	√	√		√	√	√	√		√	√	√	√			

COHORT C Treatment q4 Weeks	SCREENING		GL-ONC1 TREATMENT			POST-GL-ONC1 TREATMENT PERIOD																																POST-TREATMENT FOLLOW-UP VISIT	FOLLOW-UP
Procedures	Within 4 Wks of 1st Dose, Ideally within 2 weeks	Within 3-7 Days of 1st Dose	W1 D1	W1 D2	W1 D3	W2 to W5	CYCLE 1 W6	CYCLE 1 W8	CYCLE 2 W10	CYCLE 2 W12	W13	CYCLE 3 W14	CYCLE 3 W16	W17	CYCLE 4 W18	CYCLE 4 W20	CYCLE 5 W22	CYCLE 5 W24	CYCLE 6 W26	CYCLE 6 W28	W29	MAIN. CYCLE 1 W30	MAIN. CYCLE 2 W33	MAIN. CYCLE 3 W36	MAIN. CYCLE 4 W39	W41	MAIN. CYCLE 5 W42	MAIN. CYCLE 6 W45	MAIN. CYCLE 7 W48	MAIN. CYCLE 8 W51	W53	MAIN. CYCLE 9 W54	MAIN. CYCLE 10 W57	MAIN. CYCLE 11 W60	MAIN. CYCLE 12 W63	W65	30 Days Following Last Dose/Cycle	PRN Per PI (e.g., Quarterly for Year 2 to Year 3)
Informed Consent	√																																					
Medical & cancer disease history	√																																					
Demographics	√																																					
HIV, HBsAg, anti-HCV testing	√																																					
Implantion of peritoneal catheter & Laparoscopy **1** 3-7 days prior to 1st GL-ONC1 (5 days preferred)		√[1]																																				
REQUIRED: Tumor biopsy during laparoscopy		√																																			
Cancer gene mutation profile of tumor biopsy		√																																			
Photos to assess extent of disease via laparoscopy		√																																			
Accessment of disease by Peritoneal Cancer Index		√																																			
Toxicity Assessment **1** Related to screening procedures **2** Toxicity assessment done at each study visit during this time frame **3** Report SAEs if causality to GL-ONC1	√[1]	√[1]	√	√	√	√[2]	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√[3]
Spiral CT scan **1** Performed immediately prior to 1st cycle of chemotherapy ± bevacizumab	√						√[1]							√							√					√					√					√		√
Physical exam, weight, performance status, vital signs **1** Assessments done at Weeks 2 and 3	√	√	√			√[1]	√	√	√	√		√	√	√		√	√	√	√	√	√		√	√	√	√		√	√	√	√	√		√	√	√	√	√	√
Height	√																																					
Vital signs: pre-GL-ONC1 dose & at 30 min (±5 min) intervals up to 120 minutes			√	√																																	
Serum pregnancy test for WOCBP	√	√																																				
Clinical labs: CBC, comprehensive metabolic panel, LDH, C-RP, CA-125, urinanalysis **1** Samples collected prior to treatment **2** Samples collected at Weeks 2 and 3	√	√	√[1]	√[1]	√	√[2]	√[1]	√[1]	√[1]	√[1]		√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√[1]	√[1]	√[1]	√[1]	√	√	√
Concomitant medications **1** Obtained at each study visit during this time frame	√	√	√	√	√	√[1]	√	√	√	√		√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√
FACT-O Quality of Life Questionnaire	√						√[1]							√							√					√					√					√		√
Research bloods: Lymphocyte, cytokine analysis, neutralizing antibody titers, beta-glucuronidase **1** Samples collected prior to treatment **2** Samples collected at Weeks 2 and 3		√	√[1]	√[1]	√	√[2]	√[1]		√[1]					√							√					√					√					√	√	

COHORT C Treatment q4 Weeks	SCREENING		GL-ONC1 TREATMENT			POST-GL-ONC1 TREATMENT PERIOD																																POST-TREATMENT FOLLOW-UP VISIT	FOLLOW-UP
Procedures	Within 4 Wks of 1st Dose, Ideally within 2 weeks	Within 3-7 Days of 1st Dose	W1 D1	W1 D2	W1 D3	W2 to W5	CYCLE 1 W6	CYCLE 1 W8	CYCLE 2 W10	CYCLE 2 W12	W13	CYCLE 3 W14	CYCLE 3 W16	W17	CYCLE 4 W18	CYCLE 4 W20	CYCLE 5 W22	CYCLE 5 W24	CYCLE 6 W26	CYCLE 6 W28	W29	MAIN. CYCLE 1 W30	MAIN. CYCLE 2 W33	MAIN. CYCLE 3 W36	MAIN. CYCLE 4 W39	W41	MAIN. CYCLE 5 W42	MAIN. CYCLE 6 W45	MAIN. CYCLE 7 W48	MAIN. CYCLE 8 W51	W53	MAIN. CYCLE 9 W54	MAIN. CYCLE 10 W57	MAIN. CYCLE 11 W60	MAIN. CYCLE 12 W63	W65	30 Days Following Last Dose/Cycle	PRN Per PI (e.g., Quarterly for Year 2 to Year 3)
OPTIONAL: Needle tumor biospy (CT or ultrasound guided biopsy) **1** Needle biospy can occur at 1 time point during this time frame **2** Needle biopsy can occur on either day; after CT scan, if done on the same day (± 7 days)						√[1]					√[2]			√[2]																								
OPTIONAL AT SPONSOR'S DISCRETION: Circulating Tumor Cells (CTC): 2 × 10 mL whole blood samples **1** Collected up to 7 days prior to the first GL-ONC1 dose **2** Collected at Week 3 **3** Sample collected prior to treatment		√[1]				√[2]	√[3]							√						√																		
Peritoneal fluid sampling (for patients with ascites) cytology analysis (including tumor cells & immune cells); Viral kinetics (peritoneal fluid by VPA & qPCR) **1** Complete drainage of ascites prior to the first GL-ONC1 dose **2** 10 mL ascites sample collected **3** 10 mL ascites sample collected at Week 2, and at Week 3 if catheter is in place			√[1]		√[2]	√[3]																																
Peritoneal wash to obtain cell components & supernatant **1** Infusion of 1 to 1.5 of Ringer Lactate & drainage prior to the first GL-ONC1 dose			√[1]																																			
GL-ONC1 treatment			√	√																																		
Viral shedding (oral & anal swab by VPA, qPCR) collected post-GL-ONC1 treatment					√																																	
Chemotherapy Investigator's Choice: Carboplatin doublet for up to 6 cycles. Chemotherapy component could be substituted if due to toxicity or by Investigator's choice. If due to platinum-allergy, single-agent non-platinum chemotherapy is allowed.							√		√			√			√		√		√																		
Bevacizumab 10 mg/kg, q2 weeks with chemotherapy agents (7.5 mg/kg q2 weeks with PLD) for up to 6 cycles, and as a single agent maintenace q3 weeks for 9 months. Dose reduction is allowed at Investigator's discretion.							√	√	√	√		√	√		√	√	√	√	√	√		√	√	√	√		√	√	√	√		√	√	√	√			

	SCREENING		TREATMENT				GL-ONC1 POST-TREATMENT PHASE								
COHORT D PROCEDURES & TIME LINES **FOLLOW SCHEDULE UNTIL ADMINISTRATION OF CHEMOTHERAPY ± BEVACIZUMAB AFTER WHICH FOLLOW COHORT C COORESPONDING PROCEDURES/TIME LINES**	**Within 4 Wks of 1st Dose**	**Within 3-7 Days of 1st Dose**	**W1 (3 consecutive treatment days & day after last dose)**				**W2 D10/ 7 days after 1st dose**	**W3 D17/ 14 days after 1st dose**	**W6 D38**	**W15 D101**	**W24 D164**	**W30 D206**	**W36 D248**	**W42 D290**	**W48 D332**
Informed Consent	√														
Medical & cancer disease history	√														
Demographics	√														
HIV, HBsAg, anti-HCV testing	√														
Toxicity Assessment 1 Related to screening procedures	√[1]	√[1]	√	√	√	√	√	√	√	√	√	√	√	√	√
Spiral CT scan	√								√	√	√		√		√
PET or PET-CT scan 1 Frequency is performed at the discretion of the Investigator at any of the time points listed	√[1]								√[1]	√[1]	√[1]		√[1]		√[1]
Physical exam, weight, performance status, vital signs	√	√	√				√		√	√	√	√	√	√	√
Height	√														
Vital signs: pre-GL-ONC1 dose & at 30 min (±5 min) intervals up to 120 minutes			√	√	√										
Serum pregnancy test for WOCBP	√	√													
Clinical labs: CBC, comprehensive metabolic panel, LDH, C-RP, CA-125, urinanalysis 1 Samples collected prior to treatment	√	√	√[1]	√[1]	√[1]	√	√	√	√	√	√	√	√	√	√
Concomitant medications	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√
FACT-O Quality of Life Questionnaire	√								√	√	√		√		√
REQUIRED: Tumor biopsy by needle-guided biopsy		√													
Cancer gene mutation profile of tumor biopsy		√													
Research bloods: Lymphocyte, cytokine analysis, neutralizing antibody titers, beta-glucuronidase 1 Samples collected prior to treatment		√	√[1]	√[1]	√[1]	√	√	√	√	√	√	√	√		√
Viral shedding (oral & anal swab by VPA, qPCR) collected post-GL-ONC1 treatment						√									
OPTIONAL: CT-guided tumor biospy (CT or ultrasound guided biopsy) **1** Needle biospy can occur on either day **2** Needle biopsy can occur on either day							√[1]	√[1]	√[2]	√[2]			√		
OPTIONAL AT SPONSOR DISCRETION: Circulating Tumor Cells (CTC): 2 × 10 mL whole blood samples **1** Collected up to 7 days prior to the first GL-ONC1 dose 2 Follow time lines for Cohort C after initiation of chemotherpy ± bevacizumab		√[1]						√	√[2]	√[2]	√[2]				
GL-ONC1 treatment: For patients receiving 2nd cycle of GL-ONC1 by IV route, this Study Calendar will be used as reference.			√	√	√										
Chemo ± bev treatment									Chemo ± bev begins following disease progression (refer to Cohort C for treatment plan & schedule)						

16.0 MANAGEMENT OF POX-LIKE SKIN LESIONS OR RASHES

At any time after initiation of GL-ONC1 treatment, a patient develops a pox-like lesion/rash that may be suspicious for vaccinia virus positivity, swabs of the lesion/rash are obtained. Genelux is to be immediately notified that these biological sample(s) require virus shedding analysis by Genelux or another qualified lab. Results of this analysis will be sent to the Investigator and designated study staff upon completion of the analysis. In the case of positive virus shedding, the Investigator or designee should immediately notify the patient, and follow-up care should be discussed with the patient. Refer to *Appendix 4. Safety Issues* for a description of vaccinia-specific and non-specific rashes, and home care instructions for the patient (also provided in the informed consent form). Refer to the *Biological Sample Handling & Processing Manual* for information on the method and supplies required to collect this type of biological sample as well as shipping requirements.

17.0 STUDY MATERIALS

Examples of materials that will be provided to the Investigator and designated staff are the following:

- GL-ONC1 Safety Data Sheet;
- NCI CTCAE Version 4.03;
- GL-ONC1 Investigator's Brochure;
- GL-ONC1 Drug Handling Manual;
- Laboratory manual for the collection and handling of blood, tissue specimens and other biological samples;
- Patient Registration Forms;
- Serious Adverse Event (SAE) Report Forms;
- Pregnancy Surveillance Forms.

18.0 SAFETY ASSESSMENTS

Baseline assessments include: medical history to capture relevant underlying conditions, physical examination which captures weight, height, performance status, blood pressure (BP), heart rate (HR) and temperature, and concomitant medications. With the exception of height, these procedures are performed throughout the treatment and during Year 2 post-treatment follow-up Period.

During screening, SAEs related to study procedures are reported.

Patients will be evaluated for safety if they have received the study drug, GL-ONC1.

PHASE 1b AND PHASE 2 COHORTS A & B: Toxicity assessments (i.e., all non-serious and serious adverse events) will be continuous during the Treatment Period and Post-treatment Period. If a patient receives a second cycle of GL-ONC1, AEs/SAEs are reported for 30 days after the last dose if determined to be attributed to GL-ONC1.

PHASE 2 COHORT C: Only non-serious adverse events and serious adverse events that are determined to have causality to GL-ONC1 will be reported during the Post-GL-ONC1 Treatment Period (i.e., q3w group from Weeks 6 to 59 and q4w group from Weeks 6 to 65) and at the 30-day post-treatment visit.

PHASE 2 COHORT D: Adverse event reporting will follow the time lines and requirements listed above for the Phase 1b and Phase 2 Cohorts A & B until chemotherapy ± bevacizumab is administered after which specifics listed above for the Phase 2 Cohort C are followed.

ALL PHASES & COHORTS: During the Long-term Follow-up Period (i.e., Years 2 and 3) SAEs that are determined by the Investigator or designated qualified person (i.e., Sub-investigator) to have a degree of attribution (i.e., possibly, probably, definitely related) to GL-ONC1 treatment are reported on the *SAE Report Form*. Non-serious AEs and unrelated SAEs are not reported during the Long-term Follow-up Period.

Adverse events and laboratory values will be graded according to the NCI-CTCAE Version 4.03. If a toxicity grade is not listed in the NCI-CTCAE, the Investigator/Sub-investigator will use his/her best medical judgement in determining the toxicity grade. Physical examinations, performance status and body weight are assessed at the time points found in *Section 15.0 Study Assessments and Procedures*.

During GL-ONC1 treatment, vital signs are obtained prior to, and at 30-minute intervals post-GL-ONC1 treatment for 120 min (±5 min each time point). The start and stop time of the GL-ONC1 infusion is recorded on the appropriate electronic case report form (eCRF). For the Phase 1b and Phase 2 Cohorts A and B groups, during the Post-treatment Period and Year 2 of the Long-term Follow-up Period, vital signs are collected with the physical examination during clinic visits for patients who have not received anti-cancer treatment during these study periods. In the Phase 2 Cohorts C & D group, vital signs and physical examinations will continue at the Investigator's discretion during the Post-GL-ONC1 Treatment Period and during the Long-term Follow-up Period for patients who are able to come in for clinic visits.

Additional measures including non-study required laboratory tests should be performed as clinically indicated.

Baseline serum chemistries (e.g., comprehensive blood chemistry panel) and hematology (CBC plus differential), urinalysis, LDH, C-RP, and CA-125 will be processed by local or commercial labs. Baseline creatinine clearance (CrCl) based on the Cockcroft-Gault formula may be calculated from the local lab. Serum pregnancy testing (done locally) must be performed within 4 weeks and 7 days prior to administration of first GL-ONC1 dose for WOCBP.

For the Phase 1b and Phase 2 Cohorts A & B, laboratory toxicities (e.g., suspected drug induced liver enzyme elevations) will be monitored during the Year 2 Long-term Follow-up Period via on-site/local labs until all study drug related toxicities resolve, return to baseline levels or are deemed irreversible. Exception is for patients who receive other anti-cancer treatment during this period. In the Phase 2 Cohorts C & D patients, monitoring of laboratory toxicities will continue at the Investigator's discretion during Years 2 and 3 Long-term Follow-up Periods for patients who are able to come in for clinic visits.

19.0 EFFICACY ASSESSMENTS

Study evaluations will take place in accordance with the table in *Section 15.0 Study Assessments and Procedures*. To be considered for efficacy evaluation, at least one post-GL-ONC1 treatment imaging scan must be obtained for RECIST 1.1 assessment (except Phase 1b). Follow-up by imaging and clinical labs (e.g., CA-125) could be up to the end of Year 3 but could stop earlier at the discretion of the Sponsor.

Phase 1b, Phase 2 Cohorts A & B: Patients are evaluated for baseline disease status (within 4 weeks of administration of first GL-ONC1 dose), and for tumor response to treatment by contrast spiral CT scans for patients with measurable disease, at Weeks 6, 15, 24, 36 and 48 (± 1 week) or at the discretion of the Investigator, PET or PET-CT scans with contrast, until tumor progression is documented or the primary endpoint of PFS is analyzed by the Sponsor. If a patient is allergic to IV CT contrast, a preferred recommendation is the use of oral CT contrast or secondarily by MRI. Per RECIST 1.1, the same method of assessment and techniques used to identify and characterize each lesion at baseline is used throughout follow-up. For patients who received or are receiving further anti-cancer therapies, imaging studies preferably by contrast spiral CT scans and evaluation by RECIST1.1. will be at the discretion of

the Investigator or treating physician, ideally once every 3 months or according to routine clinical care practices.

Phase 2 Cohort C: Imaging by spiral CT scans & RECIST assessment are performed according to time points of treatment depending on whether time frame for chemotherapy is q3 weeks or q4 weeks. Time points include: baseline, then immediately prior to Cycle 1, one week prior to Cycle 4, 2 to 3 weeks post Cycle 6 (i.e., 1 week prior to start of bevacizumab maintenance), and then every 3 months for 3 more scans. Further scans will be PRN and at the discretion of the Investigator and the Sponsor. Refer to specific study calendars for imaging time points.

Phase 2 Cohort D: Until patients begin to receive chemotherapy ± bevacizumab, follow time lines listed above for Phase 1b and Phase 2 Cohorts A & B. At initiation of chemo ± bev, follow time lines listed for Phase 2 Cohort C.

Changes in tumor measurements and tumor responses will be assessed by the Investigator or qualified designee (e.g., radiologist) using the Response Evaluation Criteria in Solid Tumors (RECIST) 1.1 criteria (2) as the primary assessment, and by the Immune-response Response Criteria (irRC) as an exploratory assessment (3). Refer to Eisenhauer *et al.* (2) for RECIST 1.1 guidelines. irRC guidelines are provided in *Appendix 2: Immune-related Response Criteria*. Phase 1b patients who enter with non-measurable disease will be evaluated for response to treatment by PET or PET CT scan with contrast, physical examination and CA-125 levels at any of the time points specified for patients with measurable disease.

The following efficacy assessments will be used in this non-randomized trial:

- Progression-free Survival (PFS): is defined as the time (in months) from the date of enrollment to the date of the first observation of 'confirmed' progression by RECIST 1.1 or date of death, regardless of the cause. For the Phase 2 Cohort C, the first assessment of PFS is following initiation of chemo +/- bev, irrespective of disease status from Week 6 scan prior to start of first cycle of chemo +/- bev. If PD is documented at both time points, the earlier time point of PD will be used to calculate PFS. For Phase 1b and Phase 2 Cohorts A & D, PFS1 from GL-ONC1, PFS2 from subsequent chemo+/-bev (if applicable), and combined PFS will be assessed.
- Overall Response Rate (ORR): ORR is defined as the proportion of patients (sum of PRs plus CRs) with best overall response of PR or CR by RECIST 1.1 and/or GCIG CA-125 criteria. ORR is a clinically very relevant endpoint in ovarian cancer due to the burdensome nature of symptoms for patients. Reduction in symptoms improves patient's Quality of Life (QoL) (42). For Phase 2 Cohort C (and Cohorts A & B if received further chemo +/- bev after GL-ONC1), patients are evaluable only if they have at least one CT scan and/or at least two CA-125 reads 28 days apart following initiation of chemo +/- bev, by RECIST or GCIG CA-125 criteria, respectively.
- Duration of Response (DoR): DoR is the time from documentation of response (RECIST 1.1 and/or GCIG CA-125) to disease progression.
- Best Overall Response (BOR): is defined as the best among all overall responses from enrollment until disease progression, treatment toxicity and death.
- Disease Control Rate (DCR): DCR is defined as the proportion of patients with objective evidence of a CR, or PR, or SD ≥ 15 weeks.
- Clinical Benefit Rate (CBR): CBR is defined as the proportion of patients with objective evidence of CR + PR + SD.
- Time-to-Treatment Failure (TTF): TTF is defined as a composite endpoint measuring time from enrollment until disease progression, treatment toxicity and death.
- GCIG CA-125 Response Criteria: A response according to CA-125 has occurred if there is at least a 50% reduction in CA-125 levels from pre-treatment sample. The response must be confirmed and maintained for at least 28 days.

- Overall Survival (OS): OS is defined as the time (in months) from the date of enrollment to the date of death, regardless of the cause. In the absence of death confirmation, survival time will be censored at the date of the last study visit.

20.0 MEASUREMENT OF EFFECT

20.1 Safety/tolerability

Analyses will be performed for all patients having received at least one dose of the study drug, GL-ONC1.

20.2 Research Specimen Studies for Biological Effects

Sample Collection Guidelines

Detailed information regarding obtaining the clinical research samples for processing by the designated labs is provided in the *Biological Sample Handling and Processing Manual* and will be given to the Investigator and designated staff for reference.

All patient biological samples processed by Genelux Corporation are coded by Genelux qualified staff on the *Biological Specimen Transmittal & Receipt Form* with a sample ID number that (1) references the patient ID number, (2) the sample sequence number, (3) H (i.e., to denote a human biological sample), and (4) the date the sample was taken. For example, the first blood sample obtained on August 25, 2015 for the first treated patient would be coded as 15A-01-001-H-082515. When the biological sample arrives at Genelux, a designated and trained laboratory staff member stores the biological sample as required until processing can begin, and enters the information listed below into the secure computerized biological sample tracking database. Only designated laboratory and clinical trial operation staff have access to this folder on the server. Sample tracking may include:

- Sample ID number.
- Study visit;
- Date sample obtained;
- Sample location (i.e., freezer; liquid nitrogen storage tank);
- Rack location;
- Box number and position in box;
- Date and initials of person processing a sample processed;
- Results;
- Information if sample aliquoted, and if so, the volume.
- Automatic link to sample box which is based on type of retained biologic sample. When biological samples are removed for processing, the designated lab person writes the position in the sample was removed from, the date and purpose for removal, and space is provided for the signature of the person processing the biological sample and a witness signature.

20.3 Pharmacokinetics and Cytology Analyses

To determine the effect of GL-ONC1 in patients administered GL-ONC1 via IP catheter, viral pharmacokinetics (PK) are evaluated by VPA/qPCR from peritoneal fluid collected at the time points found in *Section 15.0 Study Assessments and Procedures*. For patients with ascites, as much peritoneal fluid as possible is drained immediately before the first dose of GL-ONC1. At other peritoneal fluid sampling time points, a 10 mL sample is obtained.

PHASE 1b: The catheter may be removed on Day 10, but if it is in place on Day 17, a 10 mL ascites sample is withdrawn, if available.

PHASE 2 COHORTS A, B & C: The catheter may be removed 7 days after date of first GL-ONC1 dose, or if the catheter is still in place, a 10 mL ascites sample may be withdrawn 14 days following first GL-ONC1 dose, if available.

If for medical reasons complete ascites drainage is required following a GL-ONC1 treatment, a wait time of at least 20 hrs after the first virus treatment is needed before complete drainage is performed. For the analysis of cell components and supernatants, all patients will undergo a wash of the peritoneal cavity with 1 L to 1.5 L of Ringer Lactate solution (RLS) prior to the first GL-ONC1 dose with as much RLS drained prior to the administration of the first dose of GL-ONC1. For patients with ascites, the peritoneal cavity wash will occur following the drainage of accumulated ascites.

PHASE 2 COHORT D: Ascites may be collected from patients requiring drainage as part of medical management per institutional practices.

ALL COHORTS: On the day after the last GL-ONC1 dose, viral shedding samples will be collected for analysis from oral and anal swabs.

Collection of pleural fluid: For patients requiring drainage of pleural fluid, samples may be collected and sent to Sponsor to analyze for presence of virus, especially for the Phase 2 Cohort D with systemic delivery of GL-ONC1. Time points of collection will be decided ad hoc with the Sponsor.

20.4 Pharmacodynamic Analyses

To evaluate pharmacodynamic (PD) objectives, assess anti-vaccinia virus and antitumor responses to GL-ONC1, blood samples (plasma and cells) are drawn at the time points found in *Section 15.0 Study Assessments and Procedures* for analysis of β-glucuronidase, lymphocyte subsets, cytokine analysis, neutralizing antibodies, etc.

CTC Analysis: Optional analysis of circulating tumor cells (CTC) may be included in any cohort. The number of patients in any cohorts to be tested for CTC will be at the discretion of the Sponsor. To assess CTCs, 2 × 10 mL of whole blood is drawn into specific tubes. Samples will be immediately shipped to the testing facility (e.g., ARUP) for analysis using the CellSearch system.

PHASE 1b: Whole blood samples for CTC analysis are collected based on the number of CTCs reported at baseline prior to the first GL-ONC1 infusion:

1. 5+ CTCs: Collect samples on W3/D17, W15/D101 and W24/D164.
2. 1-5 CTCs: Collect samples on W3D17. If this result is 5+ CTCs, continue sample collection on W15/D101 and W24/D164.
3. 0 CTCs: Do not collect any additional samples.

Phase 2 (Cohorts A, B): Regardless of baseline CTC count, fresh whole blood samples are collected prior to the first GL-ONC1 treatment (i.e., baseline) and then at W3 (14 days after date of first GL-ONC1 dose), W6, W15 and W24.

Phase 2 (Cohort C): CTC analyses are performed according to time points of treatment depending on whether time frame for chemo +/- bev is q3 weeks or q4 weeks. Regardless of baseline CTC count, samples are collected at baseline and at W3 (14 days after date of first GL-ONC1 dose) and W5 to W6. Then, for q3W schedule collect at W12 and W21, or for q4W collect at W17 and W29.

Phase 2 (Cohort D): Follow time lines above for Phase 1b until chemo ± bev is administered and then follow Phase 2 Cohort C time lines.

20.5 CA-125 GCIG Response Criteria

Blood for this test is drawn with the clinical labs to further analyze response and progression of ovarian cancer according to CA-125 levels at the time points listed in *Section 15.0 Study Assessments and Procedures*.

20.6 General Precautions

- All specimens should be handled as potentially hazardous in accordance with universal precautions for Biosafety Level-2 (BSL-2) containment procedures.
- All treatment-related waste from activities performed in the BSL-2 facility will be handled in accordance with biohazard requirements.
- The clinical specimen should be handled in a laminar air flow bio-safety hood.
- Personal protective equipment (PPE) such as a lab coat, goggles and disposable gloves should be worn and changed as needed; the sleeves of lab coat should be tightened during the experiment.
- All non-disposable labware shall be sterilized in an autoclave. It is preferred that disposal labware is used as much as possible.

20.7 Specimen Banking

Patient biological samples processed by Genelux Corporation will be maintained in a BSL-2 laboratory with controlled access by designated staff in locked freezers. Specimens will be stored indefinitely or until they are used up. If future use is denied or withdrawn by the patient (submitted in writing to the Investigator), best efforts will be made to stop any additional studies and to destroy the specimens. If advised that a patient has withdrawn consent for continued storage and future research use of biological sample(s), Genelux has a documented and witnessed process of destruction in place.

The Sponsor will be responsible for reviewing and approving requests for clinical specimens from potential research collaborators outside of Genelux. Collaborators will be required to complete an agreement (a Material Transfer Agreement or recharge agreement) that states specimens will only be released for use in disclosed research. Any data obtained from the use of clinical specimens will be the property of Genelux for publication and any licensing agreement which will be strictly adhered to.
The biological specimens, DNA, and their derivatives may have significant therapeutic or commercial value. The Informed Consent form contains this information and informs the patient that there is the potential for financial gain by Genelux or a collaborating researcher or entity.

The following information obtained from the patient's medical record may be provided to research collaborators when specimens are made available:

- Patient study assigned ID number;
- Diagnosis;
- Collection time in relation to study treatment;
- Clinical outcome – if available;
- Demographic data

21.0 ADVERSE EVENTS

21.1 Definitions

Adverse event (AE): An AE is any untoward medical occurrence in a patient receiving study treatment and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including a clinically significant abnormal laboratory finding), symptom, or disease temporally associated with the use of an experimental intervention, whether or not related to the intervention.

Adverse Reaction (AR): This is any adverse event caused by an investigational agent. Adverse reactions are a subset of suspected adverse reactions for which there is reason to conclude that the investigational agent caused the event.

Suspected Adverse Reaction (SAR): Any adverse event for which there is a reasonable possibility that the investigational agent caused the adverse event. For the purposes of IND safety reporting, 'reasonable possibility' means there is evidence to suggest a causal relationship between the drug and the adverse event. A suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

To clarify the meaning of 'reasonable possibility', the following examples for consideration would suggest a causal relationship between the drug and the adverse event.

- A single occurrence of an event that is uncommon and known to be strongly associated with drug exposure.
- One or more occurrences of an event that is not commonly associated with drug exposure but is otherwise uncommon in the population exposed to the drug.
- An aggregate analysis of specific events observed in a clinical trial (such as known consequences of the underlying disease or condition under investigation or other events that commonly occur more frequently in the drug treatment group than in a concurrent or historical control group.

The Investigator is required to evaluate the available evidence and make a judgment about the likelihood that the drug actually caused the adverse event.

Unexpected: Refers to adverse events or suspected adverse reactions which are considered 'unexpected' if the event/reaction is not listed in the Investigator Brochure or is not listed at the specificity or severity that has been observed. Unexpected also refers to adverse events or suspected adverse reactions that are mentioned in the Investigator Brochure as occurring with a class of drugs or as anticipated form the pharmacological properties of the drug but are not specifically mentioned as occurring with the particular drug under investigation. Adverse events that are anticipated to occur as part of the disease process are considered 'unexpected' because they would not be listed in the Investigator Brochure.

Adverse events can be spontaneously reported or elicited during open-ended questioning, examination, or evaluation of a patient. (In order to prevent reporting bias, patients should not be questioned regarding the specific occurrence of one or more AEs.)

21.2 Serious Adverse Events

A ***serious AE (SAE)*** is any untoward medical occurrence that at any dose:

- results in death
- is life-threatening (defined as an event in which the patient was at risk of death at the time of the

event; it does not refer to an event which hypothetically might have caused death if it were more severe)
- requires inpatient hospitalization or causes prolongation of existing hospitalization (see **NOTE** below)
- results in persistent or significant disability/incapacity
- is a congenital anomaly/birth defect
- is an important medical event (defined as a medical event(s) that may not be immediately life-threatening or result in death or hospitalization but, based upon appropriate medical and scientific judgment, may jeopardize the patient or may require intervention [e.g., medical, surgical] to prevent one of the other serious outcomes listed in the definition above.) Examples of such events include, but are not limited to, intensive treatment in an emergency room or at home for allergic bronchospasm; blood dyscrasias or convulsions that do not result in hospitalization.)
- Suspected transmission of an infectious agent (e.g., any organism, virus or infectious particle, pathogenic or non-pathogenic) via the study drug is an SAE.
- Complications due to pregnancy and overdose are handled as SAEs for data transmission purposes. All pregnancies, whether determined to be an SAE or not, will be captured in the safety database.

NOTE: The following hospitalizations are not considered SAEs:

- a visit to the emergency room or other hospital department < 24 hours, that does not result in admission (i.e., hospital admission occurring > 24 hours from reporting to emergency room or hospital) (unless considered 'important medical event' or event life threatening)
- elective surgery, planned prior to signing consent
- admissions as per protocol for a planned medical/surgical procedure
- routine health assessment requiring admission for baseline/trending of health status (e.g., routine colonoscopy)
- medical/surgical admission for purpose other than remedying ill health state and was planned prior to entry into the study. Appropriate documentation is required in these cases
- admission encountered for another life circumstance that carries no bearing on health status and requires no medical/surgical intervention (e.g., lack of housing, economic inadequacy, care-giver respite, family circumstances, administrative).

Progression of the malignancy under study (including signs and symptoms of progression), should not be reported as an AE/SAE in the following situations:

- In patients with measurable disease if disease progression has been confirmed by RECIST 1.1.
- In patients with no measurable disease if disease progression has been confirmed clinically, including evaluation by PET or PET CT scan with contrast, physical examination and CA-125 levels. **NOTE:** Disease progression would be immediately reported as individual safety reports only when there is evidence suggesting causal relationship between the study drug and the event (21 CFR 321.32(c)(5)).
- In this protocol, disease progression is captured in the context of efficacy assessment and recorded on the disease progression module of the eCRF.

21.3 Serious Adverse Event Collection and Reporting

All unexpected, serious adverse reactions will be reported to the FDA in accordance to 21 CFR 312.32. Following the patient's written consent to participate in the study, all SAEs, whether related or not related to study drug, must be collected and reported as specified below, including those thought to be associated with protocol-specified procedures.

- ALL PHASES & COHORTS: All SAEs that occur during the Screening Period that are related to screening procedures are collected and reported.
- PHASE 1b AND PHASE 2 COHORTS A & B: All SAEs that occur during the Treatment Period and the Post-treatment Period regardless of causality are reported. If a patient receives a second cycle of GL-ONC1, SAEs are reported for 30 days if determined to be attributed to GL-ONC1.
- PHASE 2 COHORTS C & D: SAEs regardless of causality are reported during the GL-ONC1 Treatment Period (Week 1) till before start of further chemotherapy ± bevacizumab. With the initiation of chemotherapy ± bevacizumab and through the 30-day post last treatment visit, only SAEs that are determined to have a degree of attribution to GL-ONC1 are reported.
- ALL PHASES & COHORTS: Only SAEs deemed to have a degree of attribution to GL-ONC1 treatment are reported during the Year 2 and Year 3 Long-term Follow-up Periods.

If a patient discontinues treatment early, collect SAE information for 30 days after the last GL-ONC1 dose or for 30 days after the last chemotherapy ± bevacizumab treatment if registered into Phase 2 Cohorts C or D.

An SAE report should be completed for any event where doubt exists regarding its status of seriousness. SAEs must be recorded on the SAE page in the eCRF within the EDC. If the Investigator believes that an SAE is not related to study drug but is potentially related to the conditions of the study (such as withdrawal of previous therapy, or a complication of a study procedure), the relationship should be specified in the narrative section of the SAE eCRF. If applicable, pregnancies are recorded on the pregnancy eCRF pages.

SAEs and pregnancies must be reported to Worldwide Clinical Trials (WCT) Pharmacovigilance (PV) team within 24 hours via EDC in accordance with the reporting specifications listed above. If EDC is unavailable, SAEs or pregnancies are recorded on the paper back-up forms and sent to WCT's PV team by email to drugsafety@worldwide.com or fax to 1-866-387-5539**.** If only limited information is initially available, follow-up reports are required. (**NOTE**: Follow-up SAE reports should include the same adverse event term(s) initially reported.)

If an ongoing SAE changes in its intensity or relationship to study drug, or if new information becomes available, a follow-up SAE report should be sent within 24 hours to the CRO using the same procedure for transmitting the initial SAE report.

All SAEs should be followed to resolution or stabilization.

The following GL-ONC1 adverse reactions also require expedited real-time reporting to the FDA **regardless of attribution**:

- All Grade 3 or greater study drug reactions;
- All events related to systemic or localized occurrence of vaccinia infection;
- All Grade 2 or greater allergic reactions.

21.4 Routine Reporting Requirements

Each year by the end of December, the Sponsor will file with the FDA and other Investigators the *GL-ONC1 Development Safety Update Report* (DSUR), including study information from the GL-ONC1-015 trial. A copy of this report will be provided to the Investigator for clinical site essential study documents. The December reporting period is based on the Development International Birth Date (DIBD) for the first authorization of GL-ONC1 to be investigated in a human clinical trial. This authorization was given by the United Kingdom's Medicines and Healthcare products Regulatory Agency (MHRA) on November 1, 2007.

21.5 Non-serious Adverse Events

A *non-serious adverse event* is an AE not classified as a SAE.

21.6 Non-serious Adverse Event Collection and Reporting

The collection of non-serious AE information should begin after initiation of GL-ONC1 treatment. Non-serious AEs should be followed to resolution or stabilization or reported as SAEs if they subsequently qualify under the SAE definitions. Follow-up is also required for non-serious AEs that cause interruption or discontinuation of investigational drug, or those that are present at the end of study treatment, as appropriate. All identified non-serious AEs are recorded and described on the *Adverse Event Log* electronic CRF (eCRF). Completion of supplemental CRFs may be requested for AEs and/or laboratory abnormalities that are reported/identified during the course of the study. The Investigator or designee should monitor all AEs that are documented as ongoing at each patient visit so a stop date can be obtained when applicable.

PHASE 1b AND PHASE 2 COHORTS A & B: Toxicity assessments (i.e., all non-serious and serious adverse events) will be continuous during the Treatment Period and Post-treatment Period. If a patient receives a second cycle of GL-ONC1, AEs/SAEs are reported for 30 days if determined to be attributed to GL-ONC1.

PHASE 2 COHORT C: All non-serious adverse events (i.e., regardless of causality to GL-ONC1) are collected during the GL-ONC1 Treatment Week (Week 1) through the end of Week 5. Beginning at Week 6 with initiation of chemotherapy ± bevacizumab, only non-serious adverse events determined to have causality to GL-ONC1 will be reported during the Post-GL-ONC1 Treatment Period (i.e., q3w group from Weeks 6 to 59 and q4w group from Weeks 6 to 65) and at the 30-day post-treatment visit.

PHASE 2 COHORT D: Adverse event reporting will follow the time lines and requirements listed above for the Phase 1b and Phase 2 Cohorts A & B until chemotherapy ± bevacizumab is administered after which specifics listed above for the Phase 2 Cohort C are followed.

21.7 Laboratory Test Abnormalities

Clinical laboratory results that are outside of the normal ranges which are deemed clinically significant by the Investigator are reported on the non-serious *Adverse Event Log* eCRF and/or *SAE Report Form*, as appropriate:

- Any laboratory test result that is clinically significant or meets the definition of an SAE.
- Any laboratory abnormality that required the patient to have investigational drug discontinued or interrupted.
- Any laboratory abnormality that required the patient to receive specific corrective therapy/medication.
- The abnormality suggests a disease and/or organ toxicity that is new or has worsened from baseline.
- The abnormality is of a degree that requires additional active management (e.g., change of dose, discontinuation of the drug, close observation, more frequent follow-up assessments, or further diagnostic investigation).

However, at the Investigator's discretion, any out-of-normal range lab parameter may be reported as an AE.

It is expected that wherever possible, the clinical, rather than the laboratory term is used by the reporting Investigator (e.g., anemia versus low hemoglobin value).

If more than one clinically significant lab result is found on a lab test (e.g., hematology), the Investigator should determine if an underlying condition is the reportable as an AE/SAE instead of the individual lab parameters (e.g., anemia instead of decrease in red blood cell count, hematocrit, reticulocyte and hemoglobin levels).

Elevated CA 125: As an elevated CA 125 tumor marker is common in women diagnosed with ovarian cancer, reporting CA 125 as clinically significant is not required unless the Investigator determines that it should be reported as such. This practice will reduce the over reporting of this tumor marker as AEs.

21.8 Pregnancy

If, following initiation of the investigational product, it is subsequently discovered that a patient is pregnant, or may have been pregnant at the time of investigational product exposure, including during at least 6 months after product administration, further treatment with the study drug or other study treatments in this protocol is discontinued in an appropriate manner for the specific treatment agent. Protocol-required procedures for study discontinuation and follow-up must be performed on the patient unless contraindicated by pregnancy (e.g., imaging studies). Other appropriate pregnancy follow-up procedures should be considered, if indicated.

The Investigator must immediately notify the Genelux's Medical Monitor and CRO Pharmacovigilance team of this event and complete the *Pregnancy Report* eCRFs, or if EDC is not available, complete and forward a *Pregnancy Notification Form* to the CRO Pharmacovigilance team upon discovery, but no later than 24 hours, and in accordance with SAE reporting procedures.

Follow-up information regarding the course of the pregnancy, including perinatal and neonatal outcome and, where applicable, offspring information must be reported on the *Pregnancy Outcome* eCRF or paper CRF.

21.9 Overdose

An overdose is defined as the accidental or intentional administration of any dose of a product that is considered both excessive and medically important. All occurrences of overdose must be reported as an SAE. In this clinical protocol, all treatments are administered by the Investigator of qualified, trained designees in the clinic.

21.10 Other Safety Considerations

Any significant worsening noted during interim or final physical examinations, and any other potential safety assessments, whether or not these procedures are required by the protocol, should also be recorded as a non-serious or serious AE, as appropriate, and reported accordingly.

21.11 Deaths

ALL PHASES AND ALL COHORTS: Deaths that occur during the GL-ONC1 Treatment Period and through Week 5 of the GL-ONC1 Post-treatment Period (i.e., within 30 days of the last GL-ONC1 infusion) require expedited safety reporting to the designated Pharmacovigilance Team. Safety reporting requirements between the Pharmacovigilance Team, Genelux, the Medical Monitor and the Investigator and/or Sub-investigator will follow the procedures established between the Sponsor and CRO.

Deaths that occur during the GL-ONC1 Post-treatment Period (i.e., Phase 1b and Phase 2 Cohorts A & B: Week 6 to Week 48 Post-treatment Period; Phase 2 Cohort C for q3w Week 6 to 59 & at 30-day post-last treatment; q4w Week 6 to Week 65 and at 30-day post-last treatment and Phase 2 Cohort D at

intervals listed for Phase 1b and Phase 2 Cohorts A & B until initiation of chemo ± bev follow Phase 2 Cohort C time points), and during the Year 2 and Year 3 Long-term Follow-up Period (all phases and cohorts) which have no attribution to the investigational drug are promptly recorded on the *Death Report* eCRF documenting primary, and if applicable, secondary cause(s) of death. If during these study periods the Investigator feels a patient's death may have a degree of attribution to GL-ONC1 treatment, a patient's death is recorded on the *SAE eCRF* or emailed/faxed to the Pharmacovigilance team as an expedited safety report.

21.12 Severity

All adverse events will be graded according to the *NCI Common Terminology Criteria for Adverse Events* (CTCAE) version 4.03. The CTCAE v4.03 is available at https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm. If an AE is not listed in the CTCAE, the Investigator will use his/her best medical judgment to determine the toxicity grade.

If no CTCAE grading is available, the severity of an AE can be graded as follows:

- *Mild (Grade 1):* the event causes discomfort without disruption of normal daily activities.
- *Moderate (Grade 2):* the event causes discomfort that affects normal daily activities.
- *Severe (Grade 3):* the event makes the patient unable to perform normal daily activities or significantly affects his/her clinical status.
- *Life-threatening (Grade 4):* the patient was at risk of death at the time of the event.
- *Fatal (Grade 5):* the event caused death.

21.13 Relationship

Attribution categories for adverse events in relationship to GL-ONC1 therapy are as follows:

Not Related

- The adverse event or suspected adverse reaction is not related if exposure to the study drug has not occurred, or
- The occurrence of the adverse event or suspected adverse reaction is not reasonably related in time, or
- The adverse event or suspected adverse reaction is considered unlikely to be related to use of the study drug (i.e., there are no facts/evidence or arguments to suggest a causal relationship).

Unlikely

- An adverse event or suspected adverse reaction whose time relationship to study drug administration makes a causal connection improbable, but which could be plausibly explained by underlying disease or other drugs or chemicals.

Possibly Related

- The study drug administration and the adverse event or suspected adverse reaction are reasonably related in time and
- The adverse event or suspected adverse reaction could be explained equally well by factors or causes other than exposure to the study drug.

Probably Related

- The study drug administration and the adverse event or suspected adverse reaction are reasonably related in time and
- The adverse event or suspected adverse reaction is more likely explained by exposure to the study drug than by other factors or causes.

Definitely Related

- The study drug administration and the adverse event or suspected adverse reaction are related in time, and
- The adverse event or suspected adverse reaction is related by exposure to the study drug than by other factors or causes.

21.14 Expected adverse events for the class of vaccinia virus

The following adverse events are considered expected toxicities for this class of compound: chills, nausea, vomiting, flu-like symptoms (fever, myalgia, and arthralgia), headache, rhinitis, diarrhea, rash, thrombocytopenia and reduced appetite. See *Appendix 6: Summary of Vaccinia-related Adverse Events* for a summary of vaccinia-related adverse events.

Any adverse event term that falls within the reactions noted for vaccinia virus which is not listed below in the expected adverse events for GL-ONC1, will be considered as 'unexpected' for the purposes of AE/SAE reporting requirements for GL-ONC1.

21.15 Expected adverse events for GL-ONC1

Based on currently known safety data from a phase I clinical trial using GL-ONC1, the following expected reactions, similar to cold or flu symptoms, are provided below.

Systemic effects and local injection symptoms

Systemic effects	Local Infusion
• fever • rigors/chills • nausea • fatigue/lethargy • sweating • rash/ulceration • myalgia • lymph node swelling • asthenia • headache • nausea • vomiting • high or low blood pressure	• redness • injection site reaction • swelling • pustule formation • pain • rash/ulceration

22.0 PHASE 1B STATISTICAL CONSIDERATIONS AND SAMPLE SIZE

This study is descriptive in nature. The sample size is based on clinical and regulatory considerations and has no formal statistical basis. The anticipated sample size is 12 to 18 patients. Descriptive statistics

will be used to summarize all baseline patient characteristics and changes of the efficacy variables. All patients who receive GL-ONC1 will be included in the safety analysis. Incidence of serious and non-serious adverse events, including those that are dose limiting, will be tabulated by system organ and preferred term. Patients, who have at a minimum one imaging time point following the initiation of treatment, will be assessed for efficacy objectives.

23.0 PHASE 2 COHORTS A, B & C STATISTICAL CONSIDERATIONS AND SAMPLE SIZE

Data (see *Section 2.6 Overview of Clinical Trial Experience with Oncolytic Viruses*, updated summary for **GL-ONC1-015** trial) from the Phase 1b part of this trial have demonstrated clinically significant results in this heavily pretreated patient population, including (1) evidence of anti-tumor activities, e.g., stabilized and/or reduced CA-125 tumor biomarker, tumor shrinkage by RECIST 1.1 (including objective response), reduction in circulating tumor cells (CTC), and encouraging Disease Control Rate (DCR = OR + SD≥15 weeks) = 55 % in 6/11 evaluable pts (4 in Ch1, 2 in Ch2). The Phase 2 portion of this trial is to further investigate anti-tumor response of GL-ONC1 monotherapy or combination therapy to observe trend of clinical benefits in a larger number of patients. PFS, ORR, CBR, and continued safety evaluation will be documented. Patients from corresponding cohorts at equivalent dose levels in the Phase1b and 2 portions will be evaluated together. To evaluate PFS as compared to historical data or to a patient's own treatment history, for a study with 80% power at 1-sided level of significance of 10%, we anticipate 21 evaluable patients (6 from Phase 1b, and 15 from Phase 2; up to 26 patients total to account for any non-evaluable patients). Therefore, the proposed number of patients in Phase 2 (Cohorts A & B) at each dose level is 20.

The study analysis is designed to evaluate ORR with 90% power using a 1-sided level of significance of 5%. The proposed number of subjects in added cohort (Ch C) is up to 35, with 28 evaluable.

Published data from the AURELIA Phase 3 study in subjects with platinum-resistant recurrent ovarian cancer with ≤ 2 prior lines of therapy and treated with bevacizumab and chemotherapy report an ORR = 27.3% by RECIST; and ORR = 31.8% by GCIG CA-125 criteria. Data from the VIRO-15 study will be evaluated in the context of these results, even though the VIRO-15 trial has enrolled subjects with significantly more prior lines of therapy (median ≥ 5). The statistical analysis is designed to provide evidence of a response that is either likely to be similar to that seen in Aurelia, or potentially a more robust improvement of ORR above the AURELIA trial, e.g., a doubling of ORR by RECIST from 27% to 54% (Using a Simon 2-stage design, Stage 1: Immediately proceed to stage 2 when reach 5 or more responders in up to 15 evaluable subjects; Stage 2: Enroll up to an additional 13 subjects; 12 or more responders of the 28 evaluable). Part of the 28 evaluable subjects could be from Chs A & B if they received further chemo +/- bev after treatment with GL-ONC1. If Chs A and/or B subjects are included together with Ch C subjects, the number of evaluable subjects needed to be enrolled into Ch C could be fewer than 28. The ORR for subjects in Chs A & B will be evaluated independently from Ch C, regardless of whether it is considered acceptable to include Chs A & B subjects with those in Ch C.

24.0 PHASE 2 COHORT D STATISTICAL CONSIDERATIONS AND SAMPLE SIZE

Cohort D is designed to evaluate ORR as one of the Primary Objectives with 90% power using a 1-sided level of significance of 5%. Using a Simon 2-stage design, Stage 1: Immediately proceed to Stage 2 when reach 5 or more responders in up to 15 evaluable subjects. Therefore, the initial number of subjects in Ch D is 15 (expandable to an additional 13 subjects in Stage 2, with 28 subjects total).

25.0 ETHICAL CONSIDERATIONS

25.1 Good Clinical Practice

This study will be conducted in accordance with Good Clinical Practice (GCP), as defined by the International Conference on Harmonization (ICH) and in accordance with the United States Code of Federal Regulations, Title 21, Part 50 (21 CFR 50) and 21 CRF 312.

The study will be conducted in compliance with the protocol. The protocol, and any amendments, as well as the patient informed consent form will receive Institutional Review Board (IRB) approval prior to initiation of the study or use in the study for amendments.

All potential serious breaches must be reported to Genelux immediately. A serious breach is a breach of the conditions and principles of GCP in connection with the study or the protocol, which is likely to affect, to a significant degree, the safety or physical or mental integrity of the patients of the study or the scientific value of the study.

Study personnel involved in conducting this study will be qualified by education, training, and experience to perform his/her respective task(s).

This study will not use the services of study personnel where sanctions have been invoked or where there has been scientific misconduct or fraud (e.g., loss of medical licensure, debarment).

25.2 Institutional Review Board Committee

Before study initiation, the Investigator must have written and dated approval from the IRB for the protocol, consent form, patient recruitment materials/process (e.g., advertisements), if applicable, and any other written information to be provided to patients. The IRB will be provided the *GL-ONC1 Investigator's Brochure* as well as any subsequent updates, as applicable.

The IRB will also receive reports, updates and other information (e.g., expedited safety reports, amendments, administrative letters, and annual Development Safety Update Report (DSUR) reports) according to IRB's requirements.

25.3 Informed Consent

Investigators must ensure that patients, or, in those situations where consent cannot be given by patients, their legally acceptable representatives, are clearly and fully informed about the purpose, potential risks, and other critical issues regarding clinical studies in which they volunteer to participate. Medical procedures or tests that are conducted as standard of care in a medical practice prior to obtaining signed informed consent, and which would have occurred regardless of whether a patient is considered for participation in this clinical trial, can be used for screening purposes (e.g., physical examination, vital signs, clinical blood draws, medical assessment of a patient's health and performance status, imaging scans, etc.). Such medical procedures/tests must occur within the study specified screening windows.

Genelux will provide the Investigator with an appropriate sample informed consent form which will include all elements required by FDA, ICH, GCP and applicable regulatory requirements. The sample informed consent form will adhere to the ethical principles that have an origin in the Declaration of Helsinki.

Investigators must:

- Provide a copy of the consent form and written information about the study in the language in which the patient is most proficient prior to clinical study participation. The language must be non-technical and easily understood.
- Allow time necessary for patient or patient's legally acceptable representative to inquire about the details of the study.
- Obtain an informed consent signed and personally dated by the patient or the patient's legally acceptable representative and by the person who conducted the informed consent discussion. The patient will be asked to write the time she signed informed consent.
- Obtain the IRB's written approval of the written informed consent form and any other information to be provided to the patients, prior to the beginning of the study, and after any revisions are completed for new information.
- Revise the informed consent whenever important new information becomes available that is relevant to the patient's consent. The Investigator, or a person designated by the Investigator, should fully inform the patient of all pertinent aspects of the study, and of any new information relevant to the patient's willingness to continue participation in the study. This communication should be documented in the patient's medical record.

The consent form must also include a statement that Genelux, any authorized agents of Genelux, as well as regulatory authorities, will have direct access to patient records. Patients should be provided with a form for review and signature in compliance with the Health Insurance Portability and Accountability Act (HIPAA), and a Patient's Bill of Rights (if applicable for the state in which the Investigator conducts the trial). The State of Florida and Florida Hospital have a *Patient Bill of Rights and Responsibility as a Hospital Patient.*

The patient must also be informed about the nature of the study to the extent compatible with the patients' understanding, and should she become capable, personally sign and date the consent form as soon as possible. The explicit wish of a patient unable to give his or her written consent, who is capable of forming an opinion and assessing this information to refuse participation in, or to be withdrawn from, the clinical study at any time should be considered by the Investigator.

The rights, safety, and well-being of the study patients are the most important considerations and should prevail over interests of science and society.

25.4 Institutional Biosafety Committees

The National Institutes of Health (NIH) Office of Biotechnology Activities) provides oversight for use and containment of GL-ONC1 at the Biosafety Level-2 (BSL-2). Thus, applicable study documents and the *GL-ONC1 Investigator's Brochure* will be provided to the IBC for initial review and approval of the study, as well as annual progress reports, safety reports and applicable updates during the trial and at trial closure.

26.0 STUDY MANAGEMENT

26.1 Conflict of Interest

Any Investigator/Sub-investigator (including family members) who has a conflict of interest with this study (patent ownership, royalties, or financial gain greater than the minimum allowable by their institution, etc.) must report conflicts on the Sponsor's *Financial Disclosure Form* for review.

26.2 Compliance

Compliance with the Protocol and Protocol Revisions

The study shall be conducted as described in this approved protocol. All revisions to the protocol must be discussed with, and be prepared by, the Sponsor. The Investigator should not implement any deviation or change to the protocol without prior review by the Sponsor, and documented approval from the IRB of an amendment, except where necessary to eliminate an immediate hazard(s) to study patients.

If a deviation or change to a protocol is implemented to eliminate an immediate hazard(s) prior to obtaining IRB approval, as soon as possible the deviation or change is submitted to:

- IRB for review and approval;
- Genelux;
- Regulatory Authority(ies), if required by local regulations.

Documentation of approval signed by the chairperson or designee of the IRB, if applicable, must be sent to Genelux.

If an amendment substantially alters the study design or increases the potential risk to the patient, (1) the consent form must be revised and submitted to the IRB for review and approval; (2) the revised form must be used to obtain consent from patients currently enrolled in the study if they are affected by the amendment; and (3) the new form must be used to obtain consent from new patients prior to enrollment.

If the revision is an administrative letter, Investigators must inform his/her IRB.

26.3 Amendments to the Protocol

Should amendments to the protocol be required, the amendments will be originated and documented by the Sponsor. It should also be noted that when an amendment to the protocol substantially alters the study design or the potential risk to the patient, a revised consent form might be required, as noted above. The written amendment, and if required, the amended consent form, are sent by the Investigator or designee to the IRB and IBC, if applicable, for approval prior to implementation.

26.4 Monitoring

Representatives of the CRO and/or from the Sponsor must be allowed to visit site location(s) periodically to assess patient safety, data quality and study integrity. While on-site, the study monitor will review study records for direct comparison with source documents, discuss the conduct of the study with the Investigator, and verify that the facilities remain acceptable. Certain eCRF pages and/or electronic files may serve as the source documents (e.g., physical exam, clinical lab results). In addition, the study may be evaluated by independent auditors selected by Genelux as well as government inspectors who must be allowed access to eCRFs, source documents, essential documents, other study files, and study facilities. Genelux will notify the Investigator and designee's of the outcome from an independent audit, but the audit reports will be kept confidential.

The Investigator must notify Genelux promptly of any inspections scheduled by regulatory authorities, and promptly forward copies of inspection reports to Genelux.

26.5 Investigational Site Training

Genelux will provide quality investigational staff training prior to study initiation. Training topics will include, but are not limited to: GCP, AE reporting, study details and procedures, background on the development of GL-ONC1 for human clinical trial investigation, study drug handling and preparation requirements, electronic CRFs, study documentation, informed consent, and enrollment of WOCBP.

26.6 Patient Data Protection

In accordance with the *Health Information Portability and Accountability Act* (HIPAA), patients who have provided written informed consent must also sign a patient authorization form to release medical information to the Sponsor and designated affiliates, and allow regulatory authorities, and IRB/IBC access to patient's medical information relevant to the study.

26.7 Source Data, Source Documents and Good Documentation Practices

The International Conference on Harmonisation (ICH) Good Clinical Practice (GCP) E6 1.51 defines source data as:

All information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents (original records or certified copies).

ICH E6 1.52 defines source documents as:

Original documents, data and records (e.g., hospital records, clinical and office charts, laboratory notes, memoranda, patients' diaries or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate copies, microfiches, photographic negatives, microfilm or magnetic media, X-rays, patient files, and records kept at the pharmacy, at the laboratories and at medico-technical departments involved in the clinical trial).

Good Documentation Practices

The FDA and the European Medicines Agency (EMA) provide key attributes for good documentation practices to employ in the conduct of clinical trials. The degree to which the data fulfills the data quality criteria establishes acceptability of the data. It also determines the degree of excellence of the data quality. Qualities like consistency, credibility and corroboration help establish data integrity along with the data quality. The Investigator is responsible to ensure that all designees assigned to this clinical trial follow the criteria listed below:

- Attributable: It should be clear who has documented the data.
- Legible: Readable and signatures identifiable.
- Contemporaneous: The information should be documented in the correct time frame along with the flow of events. If a clinical observation cannot be entered when made, chronology should be recorded. Acceptable amount of delay should be defined and justified.
- Original: Original, if not original should be exact copy; the first record made by the appropriate person. The Investigator should have the original source document.
- Accurate: Accurate, consistent and real representation of facts.
- Enduring: Long-lasting and durable.
- Available and accessible: Easily available for review of treating physicians and during audits/inspections. The documents should be retrievable in reasonable time.
- Complete: Complete till that point in time.
- Consistent: Demonstrate the required attributes consistently.

- Credible: Based on real and reliable facts.
- Corroborated: The data should be backed up by evidence.

26.8 Records Retention

The Investigator must retain all study records and source documents for the maximum period required by applicable regulations and guidelines, or institution procedures, or for the period specified by the Sponsor, whichever is longer. The Investigator must contact Genelux prior to destroying any records associated with the study to allow Genelux a chance to obtain study records for long-term archiving. Genelux will notify the Investigator when the study records are no longer needed.

If the Investigator withdraws from the study (e.g., relocation, retirement), the records shall be transferred to a mutually agreed upon designee (e.g., another Investigator or Genelux). Notice of such transfer is given in writing to Genelux.

26.9 Study Drug Records

It is the responsibility of the Investigator to ensure that a current disposition record of investigational product (those supplied by the Sponsor) is maintained at each study site where study drug is inventoried and dispensed. Records or logs must comply with applicable regulations and guidelines and should include:

- amount received and placed in storage area;
- amount currently in storage area;
- batch number;
- amount dispensed to each patient, including unique patient identifiers;
- amount transferred to another area/site for dispensing or storage;
- amount destroyed at study site, if applicable;
- amount returned to the Sponsor;
- dates and initials of person responsible for study drug dispensing and accountability, as per the Delegation of Authority Form.

The Sponsor will provide forms to facilitate inventory control if the investigational site does not have an established system that meets these requirements.

26.10 Electronic Case Report Forms

An Investigator is required to prepare and maintain adequate and accurate case histories designed to record all observations and other data pertinent to the investigation on each individual treated or entered in the investigation (i.e., screen fail). Data reported on the eCRF that are derived from source documents must be consistent with the source documents or the discrepancies must be explained and corrected, as applicable.

Electronic CRFs will be prepared for all data collection fields except for fields specific to SAEs and pregnancy, which will be reported on the paper *Serious Adverse Event Report Form* and the *Pregnancy Surveillance form*, respectively. Spaces may be left blank only in those circumstances permitted by study-specific CRF completion guidelines provided by the Sponsor.

The confidentiality of records that could identify patients must be protected, respecting the privacy and confidentiality rules in accordance with the applicable regulatory requirement(s).

The Investigator will maintain a signature sheet to document signatures and initials of all persons authorized to make entries and/or corrections on eCRFs. The Investigator should review completed

eCRF, including any paper SAE/pregnancy CRFs, in a timely manner. For electronic CRFs, review and approval/signature is completed electronically through the electronic data capture tool. Any changes and corrections to eCRFs will be captured in an audit trail in the electronic data capture (EDC).

Each individual electronically completing/signing the electronic CRFs (eCRFs) must meet CRO training requirements and must only access the CRO electronic data capture tool using the unique user account provided by the CRO following user training. User accounts are not to be shared or reassigned to other individuals.

26.11 Publications

The data collected during this study are confidential and proprietary to the Sponsor. Any publications, posters or abstracts arising from this study require approval by the Sponsor prior to publication or presentation, and must adhere to the Sponsor's publication requirements as set forth in the approved clinical trial agreement (CTA). All draft publications, including posters, abstracts or detailed summaries of any proposed presentations, must be submitted to the Sponsor at the earliest practicable time for review, but at any event not less than 30 days before submission or presentation unless otherwise set forth in the CTA. The Sponsor shall have the right to delete any confidential or proprietary information contained in any proposed presentation, poster or abstract and may delay publication for up to 60 days for purposes of filing a patent application.

27.0 REFERENCES

1. Ascierto ML, Worscheck A, Yu Z, Adams S, Reinboth J, Chen NG, Pos Z, Roychoudhuri R, Di Pasquale G, Bedognetti D, Uccellini L, Rossano F, Ascierto PA, Stroncek DF, Restifo NP, Wang E, Szalay AA, Marincola FM. Premissivity of the NCI-60 cancer cell lines to oncolytic vaccine virus GLV-1H68. *BMC Cancer* 2011;11:451.
2. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L,Dodd L, Kaplan R, Lacombe D, Verweij J. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur J Can* 2009;45:228-47.
3. Wolchok JD, Hoos A, O'Day S, *et. al.* Guidelines for the evaluation of immune therapy activity in solid tumors: Immune-related response criteria. *Clin Cancer Res* 2009;15:7412-20.
4. Rustin GJS, Vergote I, Eisenhauer E, Pujade-Lauraine E, Quinn M, Thigpen T, de Bois A, Kristensen G, Jakobsen A, Sagae S, Greven K, Parmar M, Friedlander M, Cervantes A, Vermorken J. Definitions of response and progression in ovarian cancer clinical trials incorporating RECIST 1.1 and CA 125 agreed by the Gynecological Cancer Intergroup (GCIG). *Internat J Gyn Can* 2011;21(2):419-23.
5. Cohen SJ, Punt CJ, Iannotti N, Saidman BH, Sabbath KD, Gabrail NY, Picus J, Morse M, Mitchell E, Miller MC, Doyel GV, Tissing H, Terstappen LW, Meropol NJ. Relationship of circulating tumor cells to tumor response, progression- free survival, and overall survival in patients with metastatic colorectal cancer. *J Clin Oncol* 2008;26(19):3213-21.
6. de Bono JS, Scher HI, Montgomery RB, Parker C, Miller MC, Tissing H, Doyle GV, Terstappen LW, Pienta KJ, Raghavan D. Circulating tumor cells predict survival benefit from treatment in metastatic castration-resistant prostate cancer. *Clin Cancer Res* 2008;14(19):6302-09.
7. Cristofanilli M, Budd GT, Ellis MJ, Stopeck A, Matera J, Miller MC, Reuben JM, Doyle GV, Allard WJ, Terstappen LW, Hayes DF. Circulating tumor cells, disease progression, and survival in metastatic breast cancer. *N Engl J Med* 2004;351(8):781-91.
8. Krebs GM, Hou JM, Ward TH, Blackhall FH, Dive C. Circulating tumour cells: their utility in cancer management and predicting outcomes. *Ther Adv Med Oncol* 2010;2(6):351-65.
9. De Giorgi U, Valero V, Rohren E, Dawood S, Ueno NT, Miller MC, Doyle GV, Jackson S, Andreopoulou E, Handy BC, Reuben JM, Fritsche HA, Macapinlac HA, Hortobagyi GN, Cristonfanilli

M. Circulating tumor cells and [18F]fluorodeoxyglucose positron emission tomography/computed tomography for outcome prediction in metastatic breast cancer. *J Clin Oncol* 2009;27(20):3303-11.
10. Budd GT, Cristofanilli M, Ellis MJ. Circulating tumor cells versus imaging—predicting overall survival in metastatic breast cancer. *Clin Cancer Res* 2006;12:6403.
11. Cohen SJ, Punt CJ, Iannotti N, Saidman BH, Sabbath KD, Gabrail NY, Picus J, Morse MA, Mittchell E, Miller MC, Doyle GV, Tissing H, Terstappen LW, Meropol NJ. Prognostic significance of circulating tumor cells in patients with metastatic colorectal cancer. *Ann Oncol* 2009;20(7):1223-29.
12. Krebs MG, Sloane R, Priest L, Lancashire L, Hou JM, Greystoke A, Ward TH, Ferraldeschi R, Hughes A, Clack G, Ranson M, Dive C, Blackhall FH. Evaluation and prognostic significance of circulating tumor cells in patients with non small cell lung cancer (NSCLC). *J Clin Oncol* 2011;29(12):1556-63.
13. Mandrekar SJ, Qi Y, Hillman SL, Allen Ziegler KL, Reuter NF, Rowland KM Jr, Kuross SA, Marks RS, Schild SE, Adjei AA. Endpoints in phase II trials for advanced non-small lung cancer. *J Thorac Oncol* 2010;5:3-9.
14. Cui L, Kwong J, Wang CC. Prognostic value of circulating tumor cells and disseminated tumor cells in patients with ovarian cancer: a systemic review and meta-analysis. *J Ovarian Res* 2015:8:398.
15. Poveda A, Kaye SB, McCormack R, Wang S, Parekh T, Ricci D, Lebedinsky CA, Tercero JC, Zintl P, Monk BJ. Circulating tumor cells predict progression free survival and overall survival in patients with relapsed/recurrent advanced ovarian cancer. *Gyn Oncol* 2011;122(3):567-72.
16. Pujade-Lauraine E, Hilpert F, Weber B, Reuss A, Poveda A, Kristensen G, Sorio R, Vergote I, Witteveen P, Bamias A, Pereira D, Wimberger P, Oaknin A, Raza Mirza M, Follana P, Bollag D, Ray-Coquard I. Bevacizumab combined with chemotherapy for platinum-resistant recurrent ovarian cancer: The AURELIA open-label randomized phase III trial. *J Clin Onco* 2014;13:1302-08.
17. Parato KA, Senger D, Forsyth PA, Bell JC. Recent progress in the battle between oncolytic viruses and tumours. *Nat Rev Cancer* 2005;5(12):965-76.
18. Breitbach CJ, De Silva NS, Falls TJ, Aladl U, Evgin L, Paterson J, Sun YY, Roy DG, Rintoul J, Daneshmand M, Parato K, Stanford MM, Lichty BD, Fenster A, Kirn D, Atkins H, Bell JC. Targeting tumor vasculature with an oncolytic virus. *Mol Ther.* 2011;19(5):886-94.
19. Zhang Q, Yu YA, Wang E, Chen N, Danner RL, Munson PJ, Marincola FM, Szalay AA. Eradication of solid human breast tumors in nude mice with an intravenously injected light-emitting oncolytic vaccinia virus. *Cancer Res.* 2007;67(20):10038-46.
20. Kelly KJ, Brader P, Woo Y, Li S, Chen N, Yu YA, Szalay AA, Fong Y. Real-time intraoperative detection of melanoma lymph node metastases using recombinant vaccinia virus GLV-1h68 in an immunocompetent animal model. *Int J Cancer* 2009;124(4):911-8.
21. Gentschev I., Stritzker J, Hofmann E, Weibel S, Yu YA, Chen N, Zahange Q, Bullerdiek J, Nolte I, Szalay AA. Use of an oncolytic vaccinia virus for the treatment of canine breast cancer in nude mice: preclinical development of a therapeutic agent. *Cancer Gene Ther.* 2009;16(4): 320-8.
22. Gentschev I, Adelfinger M, Josupeit R, Rudolph S, Ehrig K, Donat U, Weibel S, Chen NG, Yu YA, Zhang Q, Heisig M, Thamm D, Stritzker J, Macneill A, Szalay AA. Preclinical evaluation of oncolytic vaccinia virus for therapy of canine soft tissue sarcoma. *PLoS One* 2012;7(5):e37239.
23. Kawa A, Arakawa S. The effect of attenuated vaccinia virus AS strain on multiple myeloma; a case report. *Jpn J Exp Med.* 1987;57(1):79-80.
24. Arakawa S Jr, Hamami G, Umezo K, Kamidono S, Ishigami J, Arakawa S. Clinical trial of attenuated vaccinia virus AS strain in the treatment of advanced adenocarcinoma. Report on two cases. *J Cancer Res Clin Oncol.* 1987;113(1):95-8.
25. Hansen RM, Libnoch JA. Remission of chronic lymphatic leukemia after smallpox vaccination. *Arch Intern Med.* 1978;138(7):1137-8.
26. Yettra M. Remission of chronic lymphocytic leukemia after smallpox vaccination. *Arch Intern Med.* 1979;139(5):603.
27. Lane JM, Ruben FL, Neff JM, Millar JD. Complications of smallpox vaccination, 1968: results of ten statewide surveys. *J Infect Dis* 1970;122:303-309.
28. Lane JM, Ruben FL, Neff JM, Millar JD. Complications of smallpox vaccinations, 1968: national surveillance in the United States. *New Engl J Med* 1969;281:1201-1208.
29. Mastrangelo MJ, Lattime EC. Virotherapy clinical trials for regional disease: in situ immune

modulation using recombinant poxvirus vectors. *Cancer Gene Ther.* 2002;9(12):1013-21.

30. Gomella LG, Mastrangelo MJ, McCue PA, Maguire HC JR, Mulholland SG, Lattime EC. Phase I study of intravesical vaccinia virus as a vector for gene therapy of bladder cancer. *J Urol*. 2001;166(4):1291-95.
31. Mastrangelo MJ, Maguire HC, Lattime EC. Intralesional vaccinia/GM-CSF recombinant virus in the treatment of metastatic melanoma. *Adv Exp Med Biol*. 2000;465:391-400.
32. Mastrangelo MJ, Maguire HC Jr, Eisenlohr LC, Laughlin CE, Monken CE, McCue PA, Kovatich AJ, Lattime EC. Intratumoral recombinant GM-CSF-encoding virus as gene therapy in patients with cutaneous melanoma. *Cancer Gene Ther*. 1999;6(5):409-22.
33. Liu TC, Hwang T, Park BH, Bell J, Kirn DH. The targeted oncolytic poxvirus JX-594 demonstrates antitumoral, antivascular, and anti-HBV activities in patients with hepatocellular carcinoma. *Mol Ther. 2008*;16(9):1637-42.
34. Park BH, Hwang T, Liu TC, Sze DY, Kim JS, Kwon HC, Oh SY, Han SY, Yoon JH, Hong SH, Moon A, Speth K, Park C, Ahn YJ, Daneshmand M, Rhee BG, Pinedo HM, Bell JC, Kirn DH. Use of a targeted oncolytic poxvirus, JX-594, in patients with refractory primary or metastatic liver cancer: a phase I trial. *Lancet Oncol*. 2008;9(6):533-42.
35. Ikutani M, Yanagibashi T, Ogasawara M, Tsuneyama K, Yamamoto S, Hattori Y, Kouro T, Itakura A, Nagai Y, Takaki S, Takatsu K. Identification of innate IL-5-producing cells and their role in lung eosinophil regulation and antitumor immunity. *J Immunol* 2012;188(2):703-13.
36. Formentinit A, Braun P, Fricke H, Link KH, Henne-Bruns D, Kornmann M. Expression of interleukin-4 and interleukin-13 and their receptors in colorectal cancer. *Int. J. Colorectal Dis* 2012;27:1369-76.
37. Meyerhoff RR, Chi-Fu JY, Speicher PJ, Gulack BC, Hartwig MG, D'Amico TA, Harpole DH, Berry MF. Impact of mesothelioma histologic subtype on outcomes in the surveillance, epidemiology, and end results database. *J Surg Res* 2015;196(1):23-32.
38. Ang KK, Harris J, Wheeler R, Weber R, Rosenthal DI, Nguyen-Tân PF, Westra WH, Chung CH, Jordan RC, Lu C, Kim H, Axelrod R, Silverman CC, Redmond KP, Gillison ML. Human papillomavirus and survival of patients with oropharyngeal cancer. *N Engl J Med* 2010;363(1): 24-35).
39. Buller 1985. Vaccinia viruses as vectors for vaccine antigens. Quiinnan GV (Ed.), New York: Elsevier:37-4631.
40. Jacquet P, Sugarbaker PH. Clinical research methodologies in diagnosis and staging of patients with peritoneal carcinomatosis. 1996. In: *Peritoneal Carcinomatosis: Principles of Management,* P. H. Sugarbaker, Ed., pp. 359–374, Kluwer Academic, Boston, Mass, USA.
41. Galanis E, Hartmann LC, Cliby WA, Long HJ, Peethambaram PP, Barrette BA, Kaur JS, Haluska PJ Jr, Aderca I, Zollman PJ, Sloan JA, Keeney G, Atherton PJ, Podratz KC, Dowdy SC, Stanhope CR, Wilson TO, Federspiel MJ, Peng KW, Russell SJ. Phase I trial of intraperitoneal administration of an oncolytic measles virus strain engineered to express carcinoembryonic antigen for recurrent ovarian cancer. *Cancer* Res. 2010;70(3):875-82.
42. Pujade-Lauraine *et al*., J Clin Oncol. 2014;32(13):1302-8; Colombo *et al*., Crit Rev Oncol Hematol. 2016;97:335-48.
43. Larson EL, 1992, 1993, and 1994 Guidelines Committee. APIC guideline for hand washing and hand antisepsis in health-care settings. *Am J Infect Control* 1995;23:251-69.
44. Dixon CW. *Smallpox.* London, England: Churchill, 1962.
45. Henderson DA, Inglesby TV, Bartlett JG, Ascher MS, Eitzen E, Jahrling PB, Hauer J, Layton M, McDade J, Osterholm MT, O'Toole T, Parker G, Perl T, Russell PK, Tonat K. Smallpox as a biological weapon: medical and public health management. Working Group on Civilian Biodefense. *JAMA* 1999;281:2127-37.
46. Cooney EL, Collier AC, Greenberg PD, Coombs RW, Zarling J, Arditti DE, Hoffman MC, Hu SL, Corey L. Safety and immunological response to a recombinant vaccinia virus vaccine expressing HIV envelope glycoprotein. *Lancet* 1991; 337:567-72.

28.0 LIST OF ABBREVIATIONS

ADL	Activities of Daily Living
AE	Adverse event
ALT	Alanine aminotransferase (also known as SGPT)
ANC	Absolute neutrophil count
ASCO	American Society of Clinical Oncology
AST	Aspartate transaminase (also known as SGOT)
ATD	Accelerated Titration Design
ATE	Arterial thromboembolic event
AUC	Area under the curve
bev	bevacizumab
BMI	Body Mass Index
BOR	Best Overall Response
BP	Blood pressure
BSL-2	Biosafety Level 2
BSL	Bioservice Scientific Laboratories
C	Celsius
C-RP	C-reactive protein
CA-125	Cancer Antigen-125
CBI	Comparative Biosciences
CBR	Clinical Benefit Rate
CDC	Centers for Disease Control
CEF	Chicken embryo fibroblast
Ch	Cohort
Chs	Cohorts
CR	Complete Response
CrCl	Creatinine Clearance
CT	Computed Tomography
CTA	Clinical Trial Agreement
CTC	Circulating tumor cells
CTCAE	Common Terminology Criteria for Adverse Events
D	Day
DCR	Disease Control Rate
DHM	Drug Handling Manual
DIBD	Development International Birth Date
DLT(s)	Dose Limiting Toxicity(ies)
DNA	Deoxyribonucleic acid
DoR	Duration of Response
DTC	Disseminated tumor cells
ECOG	Eastern Cooperative Oncology Group
eCRF	Electronic Case Report Form
EDC	Electronic Data Capture
e.g.	for example
etc.	et cetera (and the rest)
FACT-O	Functional Assessment of Cancer Therapy-Ovarian
FDA	Food and Drug Administration
FSH	Follicle Stimulating Hormone
FTV	Function tumor volume
GC	gemcitabine + carboplatin
G-CSF	Granulocyte colony-stimulating factor
GCIG	Gynaecologic Cancer Intergroup

GCP	Good Clinical Practice
GFP	Green Fluorescent Protein
GGT	Gamma-glutamyltransferase
GI	Gastrointestinal
GLP	Good Laboratory Practices
GM-CSF	Granulocyte-macrophage colony-stimulating factor
GMP	Good Manufacturing Practices
HA	Hemagglutinin
HIV	Human immunodeficiency virus
hpi	Hours post infection
HR	Heart rate
hrs	hours
HRT	Hormone Replacement Therapy
IBC	Institutional Biosafety Committee
ICF	Informed Consent Form
ICH	International Congress on Harmonization (Good Clinical Practice)
IDS	Investigational Drug Services
i.e.	that is
IHC	Immunohistochemical
INN	International Nonproprietary Name
INR	International Normalized Ratio
ISF	Institutional Site File
IP	Intraperitoneal
IPL	Intrapleural
IRB	Institutional Review Board
irBOR	Immune-related Best Overall Response
irCR	Immune-related Complete Response
irPD	Immune-related Progressive Disease
irPR	Immune-related Partial Response
irRC	Immune-related Response Criteria
irSD	Immune-related Stable Disease
IV	Intravenous
kb	kilobytes
kD	kilo dalton
Kg	kilograms
LDH	Lactate dehydrogenase
LIVP	Lister strain from the Institute of Viral Preparations, Moscow
MedDRA	Medical Dictionary for Regulatory Activities
M	Meter
mg	Milligrams
MHRA	Medicines and Healthcare products Regulatory Agency
min	Minute(s)
mL	Milliliters
mm	millimeter
MMMT	Malignant Mixed Mullerian Tumor
MOI	Multiplicity of infection
mRNA	Messenger Ribonucleic acid
MSV	Master Seed Virus
MTD	Maximum Tolerated Dose
MDSCs	Myeloid-derived suppressor cells
NAb	Neutralizing antibodies
NCI	National Cancer Institute
NIH	National Institutes of Health

NYCBOH	New York City Board of Health
NYHA	New York Heart Association
OBA	Office of Biotechnology Activities
OC	Ovarian cancer
ORR	Overall Response Rate
OS	Overall Survival
PCI	Peritoneal Cancer Index
PD	Pharmacodynamic or Progressive Disease
PDGF	Platelet-derived Growth Factor
PFS	Progression-free Survival
pfu	Plaque forming unit
PK	Pharmacokinetics
PLD	Pegylated liposomal doxorubicin
PNI	Prognostic Nutritional Index
PPE	Personal protective equipment
PR	Partial Response
PRN	*pro re nata* (when necessary, as needed)
pts	Patients
PV	Pharmacovigilance
Q	Every (as in Q3Weeks)
QoL	Quality of Life
qPCR	Quantitative Polymerase Chain Reaction
RAC	Recombinant DNA Advisory Committee
RBC	Red blood cell
RECIST	Response Evaluation Criteria in Solid Tumors
RNA	Ribonucleic acid
ROC	Recurrent ovarian cancer
RT	Robust Take
SAE	Serious adverse effect
SRIC	Safety Run-in Cohort
SD	Stable Disease
SPD	Sum of Perpendicular Diameter
SUSAR	Suspected Unexpected Serious Adverse Reaction
TK	Thymidine kinase
TMF	Trial Master File
Tregs	Regulatory T cells
TTF	Time-to-Treatment Failure
ULN	Upper limit of normal
USAN	United States Adopted Name
VACV	Vaccinia virus
VEGF	Vascular Endothelial Growth Factor
VIG	Vaccinia Immune Globulin
VPA	Viral plaque assay
WCT	Worldwide Clinical Trials
WHO	World Health Organization
Wk	Week
WOCBP	Woman of child-bearing potential
WSV	Working Seed Virus

29.0 APPENDICES

29.1 Appendix 1. ECOG Performance Status Scales

Grade	Performance scale
0	Able to carry out all normal activity without restriction.
1	Restricted in physically strenuous activity but ambulatory and able to carry out light work.
2	Ambulatory and capable of all self-care but unable to carry out any work; up and about more than 50% of waking hours.
3	Capable of only limited self-care; confined to bed or chair more than 50% of waking hours.
4	Completely disabled; cannot carry on any self-care; totally confined to bed or chair.
5	Deceased.

As published in Am. J. Clin. Oncol.: *Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P.: Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Oncol 1982;5:649-55.*

29.2 Appendix 2. Immune-related Response Criteria

The Immune-related Response Criteria (irRC) was developed to address the shortcoming of the RECIST and WHO criteria with regards to the use of immunotherapeutic agents which induce an immune response that can subsequently increase tumor burden or new lesions may appear. Thus, Wolchok *et al.* (3) modified WHO criteria and developed the irRC for determination of response to treatment for immunotherapeutic agents. For this clinical trial, irRC will be used as an exploratory measure of disease response.

Antitumor response based on total measurable tumor burden: Only index and measurable new lesions are used to determine response.

Baseline Tumor Assessment: The sum of the products of the two largest perpendicular diameters (SPD) of all index lesions (5 lesions per organ up to 10 visceral lesions and 5 cutaneous index lesions) is calculated.

Subsequent Tumor Assessment: At each tumor assessment time point, the SPD of the index lesions and of new, measurable lesions (≥ 5 × 5 mm; up to 5 new lesions per organ: 5 cutaneous lesions and 10 visceral lesions) are added together to provide the total tumor burden:

$$\text{Tumor Burden} = SPD_{\text{index lesions}} + SPD_{\text{new, measurable lesions}}$$

Time Point Response Assessment using irRC: The percentage change in tumor burden per assessment time point is used for index and new measurable lesions. Decreases in tumor burden are assessed relative to baseline measurements.

irRC Overall Response

irRC overall response is defined below:

- Immune-related Complete Response (irCR) is the complete disappearance of all lesions whether measurable or not, and no new lesions. Confirmation by a repeat, consecutive assessment no less than 4 weeks from the date first documented irCR.
- Immune-related Partial Response (irPR) is the decrease in tumor burden ≥ 50% relative to baseline which is confirmed by a consecutive assessment at least 4 weeks after first documentation irPR.
- Immune-related Stable Disease (irSD) is determined when response does not meet the criteria for irCR or irPR, in absence of irPD. Confirmation of irSD is not required.
- Immune-related Progressive Disease (irPD) is the increase in tumor burden ≥ 25% relative to nadir (minimum recorded tumor burden). Confirmation is required by a repeat, consecutive assessment no less than 4 weeks from the date first documented irPD in the absence of rapid clinical deterioration. It is recommended that the confirmation scan be at the discretion of the Investigator as follow-up with observation alone may not be in the best interest of patients experiencing a rapid decline in performance status.

irRC Overall Responses			
Measurable Response	**Non-measurable Response**		**Overall Response**
Index & new measurable lesions (tumor burden), %[1]	**Non-index lesions**	**New non-measurable lesions**	**Using irRC**
↓ 100	Absent	Absent	irCR[2]
↓ 100	Stable	Any	irPR[2]
↓ 100	Unequivocal progression	Any	irPR[2]
↓ ≥ 50	Absent/Stable	Any	irPR[2]
↓ ≥ 50	Unequivocal progression	Any	irPR[2]
↓ ≥ 50 to < 25 ↑	Absent/Stable	Any	irSD
↓ ≥ 50 to < 25 ↑	Unequivocal progression	Any	irSD
≥ 25 ?	Any	Any	irPD[2]

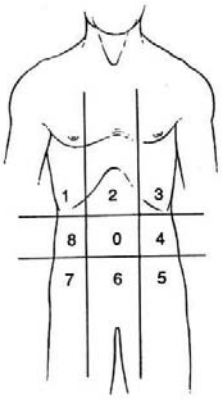
[1] Decreases assessed related to baseline, including measurable lesions only (>5 × 5 mm).
[2] Assuming response (irRC) and progression (irPD) are confirmed by a second, consecutive assessment at least 4 weeks apart.

29.3 Appendix 3. Sugarbaker Peritoneal Cancer Index

The Peritoneal Cancer Index (PCI) (40) was developed and validated as a peritoneal carcinomatosis assessment to determine at the time of surgical exploration the extent of disease in the abdomen and pelvis taking into consideration both the lesion size and distribution of peritoneal surface malignancy. The PCI quantitatively combines tumor distribution in 13 abdominopelvic regions with a lesion size score as shown below. To achieve the PCI score, the size of intraperitoneal nodules are assessed.

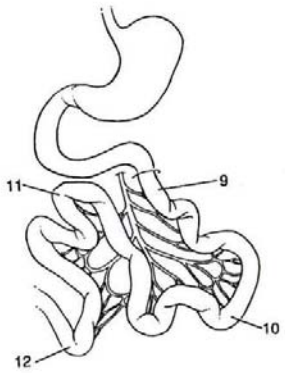
The lesion size (LS) score is evaluated after lysis of all adhesions and inspection of all parietal and visceral peritonea surfaces within the abdominopelvic regions defined below. If there is a confluence of tumor, the lesion size is scored as 3. The maximal score is 39 (13 × 3).

Peritoneal Cancer Index



Regions	Lesion Size
0 Central	____
1 Right Upper	____
2 Epigastrium	____
3 Left Upper	____
4 Left Flank	____
5 Left Lower	____
6 Pelvis	____
7 Right Lower	____
8 Right Flank	____
9 Upper Jejunum	____
10 Lower Jejunum	____
11 Upper Ileum	____
12 Lower Ileum	____
PCI	☐

Lesion Size Score
LS 0 No tumor seen
LS 1 Tumor up to 0.5 cm
LS 2 Tumor up to 5.0 cm
LS 3 Tumor > 5.0 cm or confluence



Regions	Anatomic Structures
0 Central	Midline abdominal incision; entire greater omentum; transverse colon
1 Right upper	Superior surface of the right lobe of the liver; undersurface of the right hemidiaphragm; right retro hepatic space
2 Epigastrium	Epigastric fat pad; left lobe of liver; lesser omentum; falciform ligament
3 Left upper	Undersurface of the left hemidiaphragm; spleen; tail of pancreas; anterior and posterior surfaces of stomach
4 Left flank	Descending colon; left abdominal gutter
5 Left lower	Pelvic sidewall lateral to the sigmoid colon; sigmoid colon
6 Pelvis	Female internal genitalia with ovaries, tubes and uterus; bladder, Douglas pouch, rectosigmoid colon
7 Right lower	Right pelvic sidewall; cecum, appendix
8 Right flank	Right abdominal gutter; ascending colon
9 Upper jejunum	For this clinical trial, these areas will not be assessed as patients with unresolved bowel obstruction, and extensive intra-abdominal adhesions and/or tumor involvement of the small bowel are not eligible for treatment under this clinical protocol
10 Lower jejunum	
11 Upper ileum	
12 Lower ileum	

29.4 Appendix 4. Safety Issues

There are two types of skin rashes to consider in patients treated with vaccinia virus:

Vaccinia-specific rashes: Formation of a papule, vesicle, ulcer, or crusted lesion, surrounded by an area of induration is usually results in a scar. Vaccinia virus can be cultured from these skin sites beginning at the time of development of a papule (occurring as early as 2 to 5 days after virotherapeutic treatment until the scab separates from the skin lesion.

Non-specific rashes: Common non-specific rashes associated with vaccinia virus include fine reticular maculopapular rashes, lymphangitic streaking, generalized urticaria, and broad, flat, roseola-like erythematous macules and patches. These rashes do not contain vaccinia virus and are believed to be caused by an immune response to vaccinia virus application. Erythematous or urticarial rashes can occur approximately 10 days after vaccinia virus application. The vaccinia-treated patient is usually afebrile, and the rash resolves spontaneously within 2 to 4 days. Non-specific rashes are usually self-limited. These persons appear well and benefit from simple supportive care measures (e.g., oral anti-antihistamine agents).

Infection Control Recommendations for Patients

For 2 days after treatment with GL-ONC1, patients are recommended to use the following home care precautions.

There is a low but potential risk of developing infection after treatment with GL-ONC1 which could be spread virus from GL-ONC1 to others. To reduce this risk further, patients are asked to contact the clinical team immediately if she develops new symptoms. GL-ONC1 is a vaccinia virus, and so patients, in theory, can spread virus from the injection sites, and any skin ulcers, acneiform pustules or rashes which develop to other parts of the body, or to other people through close physical contact. As patients may shed virus after GL-ONC1 treatment, the following precautions are recommended to patients to take around others. Patients will be advised that if blister-like skin lesions develop, that the virus may spread until the area heals or the scab falls off. Thorough hand hygiene is the most important way to prevent spreading the virus.

- To use a separate bathroom at home (if at all possible).
- To treat the toilet with bleach after each use (specifically, pouring ½ cup of bleach into the toilet and waiting 10 minutes before flushing, followed by another ½ cup of bleach when the water levels return to normal).
- To thoroughly wash their hands after using the bathroom.
- To use separate eating utensils. Wash them in a mix of 1 tablespoon of bleach with 1 gallon of water. After washing, soak for at least 1 minute in the bleach solution and air dry. To avoid direct contact with immunocompromised people, pregnant women and infants.
- To avoid close contact with people with skin diseases (eczema, atopic dermatitis and related diseases)

Staff who are pregnant or may be pregnant will not participate in the care of patients receiving GL-ONC1.

All sampling and sample analysis is done on Biosafety Level 2 (BSL-2). All sampling supplies that have been in contact with patients' specimens will be managed according to agreed local Institutional biosafety SOPs.

Throughout the conduct of the study, all personal will be informed and instructed by the Investigator and his/her designees to assure safety measures are known and adhered to. Safety recommendations will be updated if new data emerges.

Past experience with smallpox vaccines suggests that generalised rashes (erythematous, papulovesicular, urticarial, folliculitis, nonspecific) are not uncommon following smallpox vaccinations. These rashes are self-limited and require little or no therapy. No hospitalisation is required for vaccinees as noted by the Centers for Disease Control and Prevention (CDC) of the United State https://www.cdc.gov/mmwr/preview/mmwrhtml/rr5204a1.htm). In case a papule develops, which happens around 2 to 5 days after vaccination or virus infusion, the virus will be shed from the vaccination site or the infected skin lesion, until the lesion is re-epithelialized typically in about 14 to 21 days after vaccination or virus infusion (https://www.cdc.gov/mmwr/preview/mmwrhtml/rr5010a1.htm). Standard precaution should be taken to reduce the risk of accidental infection of other sites or of contact spread to other individuals (http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5010a1.htm).

These standard precautions, as outlined by CDC (http://www.cdc.gov/mmwr/pdf/rr/rr5010.pdf: *Vaccinia (Smallpox) Vaccine Recommendations of the Advisory Committee on Immunization Practices* (ACIP, 2001), are as follows:

PREVENTING CONTACT TRANSMISSION OF VACCINIA VIRUS

Vaccinia virus can be cultured from the site of primary vaccination beginning at the time of development of a papule (i.e., 2-5 days after vaccination) until the scab separates from the skin lesion (i.e., 14-21 days after vaccination). During that time, care must be taken to prevent spread of the virus to another area of the body or to another person by inadvertent contact. Thorough hand-hygiene with soap and water or disinfecting agents should be performed after direct contact with the site or materials that have come into contact with the site to remove virus from the hands and prevent accidental inoculation to other areas of the body (43). In addition, care should be taken to prevent contact of the site or contaminated materials from the site by unvaccinated persons. The vaccination site can be left uncovered, or it can be loosely covered with a porous bandage (e.g., gauze) until the scab has separated on its own to provide additional barrier protection against inadvertent inoculation. An occlusive bandage should not be routinely used because maceration of the site might occur. Bandages used to cover the vaccination site should be changed frequently (i.e., every 1 to 2 days) to prevent maceration of the vaccination site secondary to fluid buildup. Hypoallergenic tape should be used for persons who experience tape hypersensitivity. The vaccination site should be kept dry, although normal bathing can continue. No salves or ointments should be placed on the vaccination site. Contaminated bandages and, if possible, the vaccination site scab, after it has fallen off, should be placed in sealed plastic bags before disposal in the trash to further decrease the potential for inadvertent transmission of the live virus contained in the materials. Clothing or other cloth materials that have had contact with the site can be decontaminated with routine laundering in hot water with bleach (44,45).

Recently vaccinated health-care workers should avoid contact with unvaccinated patients, particularly those with immunodeficiencies, until the scab has separated from the skin at the vaccination site. However, if continued contact with unvaccinated patients is unavoidable, health-care workers can continue to have contact with patients, including those with immunodeficiencies, as long as the vaccination site is well-covered and thorough hand-hygiene is maintained. In this setting, a more occlusive dressing might be required. Semipermeable polyurethane dressings (e.g., Opsite®) are effective barriers to vaccinia and recombinant vaccinia viruses (46). However, exudates can accumulate beneath the dressing, and care must be taken to prevent viral contamination when the dressing is removed. In addition, accumulation of fluid beneath the dressing can increase the maceration of the vaccination site. Accumulation of exudates can be decreased by first covering the vaccination site with

dry gauze, then applying the dressing over the gauze. The dressing should also be changed at least once a day. To date, experience with this type of containment dressing has been limited to research protocols. The most critical measure in preventing inadvertent implantation and contact transmission from vaccinia vaccination is thorough hand-hygiene after changing the bandage or after any other contact with the vaccination site.

29.5 Appendix 5. New York Heart Association (NYHA) Functional Classification

In 1928 the New York Heart Association published a classification of patients with cardiac disease based on clinical severity and prognosis. This classification has been updated in seven subsequent editions of Nomenclature and Criteria for Diagnosis of Diseases of the Heart and Great Vessels (Little, Brown & Co.). The ninth edition, revised by the Criteria Committee of the American Heart Association, New York City Affiliate, was released March 4, 1994.

Grading is based on the individual physician's judgment. Functional capacity is an estimate of what the patient's heart will allow the patient to do and should not be influenced by the character of the structural lesions or an opinion as to treatment or prognosis. The objective assessment of a patient with cardiac disease who has not had specific tests of cardiac structure or function is classified as undetermined. Patients are rated in one of four categories based on how much she is limited during physical activity; the limitations/symptoms are in regards to normal breathing and varying degrees in shortness of breath and or angina pain. The classifications are summarized below.

NYHA Class	Functional Capacity	Objective Assessment
Class I	Patients with cardiac disease but without resulting limitation of physical activity. Ordinary physical activity does not cause undue fatigue, palpitation, dyspnea, or anginal pain.	**A.** No objective evidence of cardiovascular disease.
Class II	Patients with cardiac disease resulting in slight limitation of physical activity. They are comfortable at rest. Ordinary physical activity results in fatigue, palpitation, dyspnea, or anginal pain.	**B.** Objective evidence of minimal cardiovascular disease.
Class III	Patients with cardiac disease resulting in marked limitation of physical activity. They are comfortable at rest. Less than ordinary activity causes fatigue, palpitation, dyspnea, or anginal pain.	**C.** Objective evidence of moderately severe cardiovascular disease.
Class IV	Patients with cardiac disease resulting in inability to carry on any physical activity without discomfort. Symptoms of heart failure or the anginal syndrome may be present even at rest. If any physical activity is undertaken, discomfort is increased.	**D.** Objective evidence of severe cardiovascular disease.

As published in*: The Criteria Committee of the New York Heart Association*. *Nomenclature and Criteria for Diagnosis of Diseases of the Heart and Great Vessels*. 9th ed. Boston, Mass: Little, Brown & Co; 1994:253-256.

Examples:

- A patient with minimal or no symptoms but a large pressure gradient across the aortic valve or severe obstruction of the left main coronary artery is classified: Functional Capacity I, Objective Assessment D.
- A patient with a severe anginal syndrome but angiographically normal coronary arteries is classified: Functional Capacity IV, Objective Assessment A.
- A patient with acute myocardial infarction, shock, reduced cardiac output, and elevated pulmonary artery wedge pressure is classified: Functional Capacity IV, Objective Assessment D.
- A patient with mitral stenosis, moderate exertional dyspnea, and moderate reduction in mitral valve area is classified: Functional Capacity II or III, Objective Assessment C.

29.6 Appendix 6. Summary of Vaccinia-related Adverse Events

Adverse Event	Description	Risk factor or Predisposition	Treatment
Eczema vaccinatum (EV)	▪ High fever ▪ Generalized lymphadenopathy with extensive vesicular and pustular eruption ▪ Onset: concurrently or shortly after local vaccinial lesion in vaccinee, or in contacts, 5-19 days after suspected exposure ▪ Risk for secondary bacterial or fungal infections ▪ Virus recovered from lesions ▪ High mortality rate with poor prognosis	▪ History of eczema or atopic dermatitis irrespective of disease activity or severity ▪ Less frequently, persons without a history of dermatological conditions	▪ Prompt evaluation and diagnosis ▪ Infection-control precautions ▪ Might require multiple doses of vaccinia immune globulin (VIG) (cidofovir, second-line therapy. ▪ Hemodynamic support ▪ Volume and electrolyte repletion ▪ Observe for secondary skin infections
Progressive vaccinia (PV)	▪ Non-healing vaccination site ▪ Painless progressive (central) necrosis at the vaccination site ▪ Occasional metastatic lesions in skin, bones and viscera ▪ No inflammation initially ▪ Absence of inflammatory cells on histopathological examination ▪ Inflammation weeks later ▪ Bacterial infection might develop ▪ Differential diagnosis: severe bacterial infection, severe chickenpox, disseminated herpes simplex, and other necrotic conditions ▪ Prognosis: poor, despite therapy	▪ Humoral and cellular immunocompromise (e.g., malignancy, human immunodeficiency virus(HIV)/acquired immunodeficiency syndrome (AIDS), severe combined immunodeficiency syndrome (SCIDS) or hypogammaglobulinemia) ▪ Protective level of T-cell count or humoral immunity unknown	▪ Prompt evaluation and diagnosis ▪ Infection-control precautions ▪ Might require multiple doses of VIG (cidofovir second-line therapy) ▪ Surgical debridement of progressive necrotic lesions not proven useful
Postvaccinial encephalitis (PVE) or encephalomyelitis (PVEM)	▪ Diagnosis of exclusion ▪ Appears similar to post-infectious encephalomyelitis or toxic encephalopathy caused by other agents ▪ Abrupt onset of symptoms: fever, headache, malaise, lethargy, vomiting, meningeal signs, seizures, paralysis, drowsiness, altered mental status, or coma ▪ Age <2 (encephalopathy): cerebral vascular changes occurring 6-10 days post-vaccination ▪ Age ≥ 2 years (encephalomyelitis):	▪ Age <1 year	▪ Intensive supportive care ▪ Anticonvulsants as needed ▪ VIG not recommended ▪ Antiviral role unclear ▪ Use of modern imaging studies has not been evaluated

Adverse Event	Description	Risk factor or Predisposition	Treatment
Postvaccinial encephalitis (PVE) or encephalomyelitis (PVEM) (continued)	demyelinating changes occurring 11-15 days post-vaccination ▪ Cerebral spinal fluid (CSF): normal or nonspecific, monocytosis, lymphocytosis, or elevated protein ▪ Prognosis: mortality, 25%; neurological sequelae, 25%; complete recovery, 50%		
Fetal vaccinia (FV)	▪ Incidence: rare (<50 reported cases ▪ Route of transmission: unknown ▪ Outcomes: premature birth, fetal loss, high mortality ▪ Not associated with congenital anomalies	▪ Cases in all trimesters of pregnancy ▪ Greatest risk, third trimester	▪ Efficacy of VIG unknown ▪ Antivirals not recommended
Generalised vaccinia (GV)	▪ Maculopapular or vesicular rash ▪ Onset: 6-9 days post-vaccination] ▪ Nontoxic, with or without fever ▪ Differential diagnosis: erythema multiforme (EM), varicella, inadvertent inoculation, progressive vaccinia (PV) and smallpox	▪ Hematogenous spread ▪ Lesions contain vaccinia ▪ More serious among immunocompromised persons	▪ Usually self-limited in immunocompetent person ▪ Infection-control precautions ▪ VIG usually not indicated ▪ Anti-inflammatory medications ▪ Antipruritic medications ▪ Antivirals usually not indicated.
Inadvertent inoculation	▪ Most common complication ▪ Physical transfer of vaccinia virus from a vaccination site to second site on the vaccinee or to a close contact of vaccinee	▪ Manipulation of vaccination site ▪ Children aged <4 years ▪ Conditions that disrupt the epidermis (e.g., burns, severe acne or psoriasis)	▪ Usually self-limited ▪ Resulting in 3 weeks ▪ Infection-control precautions ▪ VIG if extensive body surface involved or severe ocular disease (cidofovir, second-line therapy.
Ocular vaccinia: Inadvertent periocular or ocular implantation with vaccinia virus. Can range from mild to severe	▪ Keratitis: Marginal infiltration for ulceration with or without stromal haze/infiltration ▪ Conjunctivitis: Hyperemia, edema, membranes, focal lesions, fever, lymphadeno-pathy ▪ Blepharitis: Lid pustules on or near the lid margin, edema, hyperemia, lymphadenopathy, cellullitis, fever	▪ Manipulation of vaccination site, followed by eye rubbing ▪ More likely with conditions that cause eye itching and scratching (conjunctivitis, corneal abrasion/ulceration	▪ Ophthalmologic consultation ▪ Certain ophthalmologists consider off-label topical antiviral medications ▪ Topical prophylactic antibacterial medications for keratitis ▪ VIG for severe blepharitis and blepharo-conjunctivitis (without keratitis)

Adverse Event	Description	Risk factor or Predisposition	Treatment
Ocular vaccinia: Inadvertent periocular or ocular implantation with vaccinia virus. Can range from mild to severe (continued)			▪ VIG not indicated for isolated keratitis ▪ VIG considered for keratitis with vision-threatening conditions ▪ VIG indicated for keratitis with life-threatening conditions that require VIG
Erythema multiforme (EM) and Stevens-Johnson Syndrome (SJS)	▪ Typical bull's eye (target) lesions ▪ Hypersensitivity reaction ▪ Pruritis ▪ Onset: 10 days post-vaccination ▪ Can progress to SJS	▪ No known risk factors	▪ Antipruritic medications ▪ VIG not indicated ▪ Hospitalization and supportive care for SJS ▪ Steroid use for SJS is controversial
Pyogenic infections of vaccination site	▪ Uncommon ▪ Onset: 5 days post-vaccination ▪ Fever not specific for bacterial infection ▪ Flatulence at vaccination site	▪ More frequent in children (touching vaccination site)	▪ Gram stain ▪ Bacterial culture ▪ Antibacterial medications, if clinically indicated ▪ No topical medications
Robust take (RT)	▪ > 7.5 cm with swelling, warmth and pain at vaccination site ▪ Fluctuant lymph nodes not expected ▪ Peak symptoms: 8-10 days post-vaccination ▪ Non-progressive ▪ Improvement in 24-72 hours	▪ Might be more likely among first-time vaccines	▪ Observation most important ▪ Antibacterial medications not indicated ▪ Rest affected limb ▪ Antipruritic medications ▪ Anti-inflammatory medications ▪ No salves for ointments
Tape adhesive reactions	▪ Sharply demarcated raised lines of erythema that correspond to adhesive placement ▪ Local pruritis ▪ No systemic illness	▪ Sensitivity to adhesives	▪ No salves, ointments, or topical/oral steroids ▪ Frequent bandage changes ▪ Periodic bandage removal

* See text for details

29.7 Appendix 7. Functional Assessment of Cancer Therapy Quality of Life Questionnaire

This validated quality of life questionnaire is used with authorization from the *Functional Assessment of Chronic Illness Therapy (FACIT) Measurement System* (www.facit.org), including the *Functional Assessment of Cancer Therapy* (FACT), owned and copyrighted by, and the intellectual property of, David Cella, Ph.D. (FACT-O English (Universal) copyrighted 1987, 1997).

Subject ID: _____ Date: _____

Below is a list of statements that other people with your illness have said are important. **Please circle or mark one number per line to indicate your response as it applies to the past 7 days.**

	PHYSICAL WELL-BEING	**Not at all**	**A little bit**	**Some-what**	**Quite a bit**	**Very much**
GP1	I have a lack of energy	0	1	2	3	4
GP2	I have nausea	0	1	2	3	4
GP3	Because of my physical condition, I have trouble meeting the needs of my family	0	1	2	3	4
GP4	I have pain	0	1	2	3	4
GP5	I am bothered by side effects of treatment	0	1	2	3	4
GP6	I feel ill	0	1	2	3	4
GP7	I am forced to spend time in bed	0	1	2	3	4

	SOCIAL/FAMILY WELL-BEING	**Not at all**	**A little bit**	**Some-what**	**Quite a bit**	**Very much**
GS1	I feel close to my friends	0	1	2	3	4
GS2	I get emotional support from my family	0	1	2	3	4

GS3	I get support from my friends	0	1	2	3	4
GS4	My family has accepted my illness	0	1	2	3	4
GS5	I am satisfied with family communication about my illness	0	1	2	3	4
GS6	I feel close to my partner (or the person who is my main support)	0	1	2	3	4
Q1	*Regardless of your current level of sexual activity, please answer the following question. If you prefer not to answer it, please mark this box ☐ and go to the next section.*					
GS7	I am satisfied with my sex life	0	1	2	3	4

Please circle or mark one number per line to indicate your response as it applies to the <u>past 7 days</u>.

	EMOTIONAL WELL-BEING	**Not at all**	**A little bit**	**Some-what**	**Quite a bit**	**Very much**
GE1	I feel sad	0	1	2	3	4
GE2	I am satisfied with how I am coping with my illness	0	1	2	3	4
GE3	I am losing hope in the fight against my illness	0	1	2	3	4
GE4	I feel nervous	0	1	2	3	4
GE5	I worry about dying	0	1	2	3	4
GE6	I worry that my condition will get worse	0	1	2	3	4

	FUNCTIONAL WELL-BEING	**Not at all**	**A little bit**	**Some-what**	**Quite a bit**	**Very much**
GF1	I am able to work (include work at home)	0	1	2	3	4
GF2	My work (include work at home) is fulfilling	0	1	2	3	4
GF3	I am able to enjoy life	0	1	2	3	4
GF4	I have accepted my illness	0	1	2	3	4
GF5	I am sleeping well	0	1	2	3	4
GF6	I am enjoying the things I usually do for fun	0	1	2	3	4
GF7	I am content with the quality of my life right now	0	1	2	3	4

Please circle or mark one number per line to indicate your response as it applies to the <u>past 7 days</u>.

	ADDITIONAL CONCERNS	**Not at all**	**A little bit**	**Some-what**	**Quite a bit**	**Very much**
O1	I have swelling in my stomach area	0	1	2	3	4
C2	I am losing weight	0	1	2	3	4
C3	I have control of my bowels	0	1	2	3	4
O2	I have been vomiting	0	1	2	3	4
B5	I am bothered by hair loss	0	1	2	3	4
C6	I have a good appetite	0	1	2	3	4
C7	I like the appearance of my body	0	1	2	3	4
BMT5	I am able to get around by myself	0	1	2	3	4
B9	I am able to feel like a woman	0	1	2	3	4
O3	I have cramps in my stomach area	0	1	2	3	4
BL4	I am interested in sex	0	1	2	3	4
BMT7	I have concerns about my ability to have children	0	1	2	3	4