Phase I Study of Elraglusib (9-ING-41), a Glycogen Synthase Kinase-3β Inhibitor, as Monotherapy or Combined with Chemotherapy in Patients with Advanced Malignancies

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ABSTRACT

Purpose: The safety, pharmacokinetics, and efficacy of elraglusib, a glycogen synthase kinase-3β (GSK-3β) small-molecule inhibitor, as monotherapy or combined with chemotherapy, in patients with relapsed or refractory solid tumors or hematologic malignancies was studied.

Patients and Methods: Elraglusib (intravenously twice weekly in 3-week cycles) monotherapy dose escalation was followed by dose escalation with eight chemotherapy regimens (gemcitabine, doxorubicin, lomustine, carboplatin, irinotecan, gemcitabine/nab-paclitaxel, paclitaxel/carboplatin, and pemetrexed/carboplatin) in patients previously exposed to the same chemotherapy.

Results: Patients received monotherapy ($n = 67$) or combination therapy ($n = 171$) elraglusib doses 1 to 15 mg/kg twice weekly. The initial recommended phase II dose (RP2D) of elraglusib was 15 mg/kg twice weekly and was defined, without dose-limiting toxicity observation, due to fluid volumes necessary for drug administration. The RP2D was subsequently reduced to 9.3 mg/kg once weekly to reduce elraglusib-associated central/peripheral vascular access catheter blockages. Other common elraglusib-related adverse events (AE) included transient visual changes and fatigue. Grade ≥3 treatment-emergent AEs occurred in 55.2% and 71.3% of patients on monotherapy and combination therapy, respectively. Part 1 monotherapy ($n = 62$) and part 2 combination ($n = 138$) patients were evaluable for response. In part 1, a patient with melanoma had a complete response, and a patient with acute T-cell leukemia/lymphoma had a partial response (PR). In part 2, seven PRs were observed, and the median progression-free survival and overall survival were 2.1 [95% confidence interval (CI), 2–2.6] and 6.9 (95% CI, 5.7–8.4) months, respectively.

Conclusions: Elraglusib had a favorable toxicity profile as monotherapy and combined with chemotherapy and was associated with clinical benefit supporting further clinical evaluation in combination with chemotherapy.

Introduction

Glycogen synthase kinase-3 (GSK-3) is a serine/threonine kinase that regulates metabolism, including glycogen biosynthesis (1). It has two ubiquitously-expressed and highly-conserved isoforms—GSK-3α and GSK-3β—with homologous but nonoverlapping substrates and functional effects (1, 2). Both forms are constitutively active under basal conditions, and genetic deficiency of one isoform cannot be compensated by the other (3). They affect substrates involved in the pathogenesis of several diseases, including cancer, pleural fibrosis, and immune, metabolic, and neurologic disorders (4–6).

GSK-3β plays an important role in tumor progression through the modulation of oncogenes and epithelial–mesenchymal transition (7, 8). The expression of GSK-3β promotes tumor growth and chemotherapy resistance in solid tumors through prosurvival effects on NFκB and c-Myc pathways, as well as TRAIL and p53-mediated apoptotic mechanisms (9–12). Preclinical *in vivo* studies demonstrated that GSK-3β inhibition leads to antitumor activity in a spectrum of human cancers (11, 13–17). These data indicated GSK-3β as a potential therapeutic target in human malignancies.

Elraglusib (9-ING-41) is a maleimide-based, reversible ATP-competitive inhibitor of GSK-3 enzymatic activity, with limited activity against other kinases. Elraglusib inhibits the activity of GSK-3α and GSK-3β with similar potency and was identified from a library of GSK-3 inhibitors optimized for high blood–brain barrier

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Translational Relevance

Glycogen synthase kinase-3β (GSK-3β) has a contextual role in tumor progression, oncogene expression, immune modulation, and chemotherapy resistance. Elraglusib (9-ING-41) is a small-molecule inhibitor of GSK-3β studied in an accelerated design first-in-human dose-escalation trial as an intravenous single agent and in combination with chemotherapy in patients with relapse after, or progression on, the same chemotherapy regimen. This combination design was based on preclinical studies indicating the ability of elraglusib to enhance the antitumor activity of these chemotherapy regimens in chemoresistant tumor models. Elraglusib demonstrated an acceptable safety profile, and tumor responses were observed with single agent and combination regimens. Transient low-grade visual changes are a GSK-3β inhibitor class effect. Both the twice-weekly intravenous schedule and limited solubility warrant the development of an oral dosage form, which may reduce the visual adverse events. The results of this study support further clinical evaluation of elraglusib in combination with chemotherapy.

penetration (14). Despite inhibiting both GSK-3 isoforms, elraglusib is thought to manifest its antitumor activity primarily through inhibition of GSK-3β based on its ability to phenocopy the effects of GSK-3β knockdown. Furthermore, at doses up to 50 mg/kg in rats and up to 20 mg/kg in dogs, elraglusib did not cause hepatotoxicity or osteogenesis associated with previous GSK-3 inhibitors (18, 19). Transient, reversible elraglusib-related lethargy, prostration, and ptosis [all possible central nervous system (CNS) effects] were observed in rats but not in dogs. The only drug-related toxicity in dogs was a dose-dependent, reversible microscopic testicular atrophy. Mortality and life-threatening or irreversible toxicities were absent in dogs.

GSK-3 inhibitors, including elraglusib, sensitized cancer cells to concomitant chemotherapy, potentiated the effect of cytotoxic agents, and exhibited activity in multiple animal models of chemotherapy-refractory cancers across several tumor histologies, providing rationale for selecting the eight chemotherapy backbones as part of the combination therapy with elraglusib in this trial (8, 15, 16, 20–24). For example, elraglusib showed potent activity *in vivo* in models of pancreatic cancer and enhanced the antitumor effects of gemcitabine, nab-paclitaxel, and irinotecan. These effects were mediated by compromising DNA repair mechanisms and destabilizing the TopBP1/ATR/Chk1 pathway (22).

Preclinical data on the potential immune modulatory effects of elraglusib were demonstrated in colorectal cancer and melanoma (25–27). In human colorectal cancer cell lines, a decrease in VEGF and an increase in cancer cell death were observed through enhancement of natural killer and T-cell activity (26). Elraglusib prolonged survival in a microsatellite stable syngeneic colorectal cancer model, when given in combination with anti-PD-L1 (27). Tumors responding to the combination displayed higher infiltration of T cells, increased expression of granzyme B, and lower expression of TGFβ, indicative of tumor microenvironment modulation. In a B16 metastatic melanoma mouse model, elraglusib reduced expression of PD-1, TIGIT, and LAG-3, when given sequentially with anti-PD-1 antibodies, leading to shrinkage of CNS metastases (25). These results suggest that inhibition of GSK-3β regulates PD-L1 expression, increases tumor sensitivity to checkpoint inhibitors, and supports a potential immunomodulatory role for elraglusib. Other data indicated that the inhibition of GSK-3β may sensitize cancer cells to some standard chemotherapy (15, 22, 28). We thus hypothesized that elraglusib might overcome chemoresistance in human cancers.

Patients and Methods

Study design

This phase I study was an international, open-label, multicenter, nonrandomized trial (NCT03678883) evaluating the safety and tolerability of elraglusib as monotherapy (part 1) and in combination with eight chemotherapy regimens (part 2) in patients with relapsed and refractory malignancies (Supplementary Table S1). The study followed a standard "3+3" design in the dose-escalation phase. Part 2 of the study was initiated after the first three dose levels of single-agent elraglusib were deemed safe by the study Data and Safety Monitoring Board. Elraglusib was provided by Actuate Therapeutics.

Patients

Patients were enrolled at eight centers in the United States and one in the Netherlands. Eligible patients were ≥ 18 years old, with advanced solid or hematologic malignancies, and intolerant of or refractory to existing therapies. Additional eligibility criteria included an Eastern Cooperative Oncology Group performance status of 0 to 2 (later extended to 3) and adequate bone marrow, liver, and renal functions. Patients were required to have had previous treatment with the same chemotherapy agents used in combination with elraglusib during part 2 of the study with some exceptions in the lomustine and doxorubicin arms. Patients with primary CNS tumors and stable or slowly progressing brain metastases or leptomeningeal disease were eligible if on stable doses of anticonvulsants and steroids. Study representativeness is shown in Supplementary Table S2.

The study was conducted in accordance with Good Clinical Practice and the Declaration of Helsinki. The study protocol and informed consent form were reviewed and approved by the study center's Institutional Review Board or ethics committee. Written informed consent was obtained from all study participants.

Procedures

In part 1 of the study, patients received elraglusib at doses of 1, 2, 3.3, 5, 7, 9.3, 12.4, and 15 mg/kg on days 1 and 4 of each week in a 21-day cycle (Supplementary Table S3). Dose increments followed a modified Fibonacci sequence for percent increase. The starting dose and cycle duration were chosen based on animal toxicology studies evaluating elraglusib administered intravenously three times per week for 4 weeks at doses of up to 50 mg/kg in rats and 20 mg/kg in dogs. Using International Council for Harmonization S9 guidance and the highest non-severe toxic doses extrapolated to body surface area in animals, the human equivalent doses were calculated as 0.81 mg/kg (from the rat model) and 1.8 mg/kg (from the dog model). Considering the reduced dosing interval of twice weekly in humans, the selected starting dose was 1 mg/kg.

In part 2, patients received elraglusib at doses of 3.3, 5, 7, 9.3, 12.4, and 15 mg/kg on days 1 and 4 of each week in eight different chemotherapy arms (Supplementary Table S4). The concomitant chemotherapy agents included gemcitabine, doxorubicin, lomustine, carboplatin, irinotecan, gemcitabine plus nab-paclitaxel (GnP), paclitaxel plus carboplatin, and pemetrexed plus carboplatin (Supplementary Table S2). The starting dose for part 2 was set as the third dose level from part 1, unless emerging safety, pharmacokinetic, pharmacodynamic, and regimen administration data signaled to start at a lower dose level.

The standard "3+3" design was used in all cohorts until the MTD was determined or the highest protocol-defined dose level (15 mg/kg, based on volumes of fluid necessary for administration) completed enrollment without dose-limiting toxicities (DLT). Once a dose level from part 1 was determined safe, patients in part 2 could be escalated to that dose level beginning at the 3.3 mg/kg dose level. Treatment continued until disease progression and/or unacceptable toxicity. Doses of concomitant chemotherapy were adjusted for toxicity following each site's specific protocols.

In part 1, blood samples were collected to measure plasma elraglusib on days 1, 2, 4, 8, and 9 of cycles 1 and 2. In part 2, blood samples were collected to measure plasma elraglusib in the presence of concomitant chemotherapy concentrations on days 1, 4, and 8 of cycles 1 and 2. Blood samples were collected before drug administration, at the end of chemotherapy infusion, and 1 hour after completing elraglusib infusion. For patients receiving concomitant lomustine, blood samples were collected on days 1 and 8 of weeks 1 and 5 during cycle 1 before the oral dose of lomustine, before elraglusib infusion, and 1 hour after completing elraglusib infusion.

Study objectives and assessments

The primary objectives of the study were safety, tolerability, and the definitions of DLTs, MTDs, and recommended phase II study dose (RP2D) of elraglusib as monotherapy and in combination with chemotherapy regimens. All patients who received any dose of elraglusib were considered evaluable for toxicity, which was assessed according to the Common Terminology Criteria for Adverse Events v.4.03. Safety and tolerability were assessed by the number and incidence of treatment-emergent adverse events (TEAE) based on MedDRA-preferred terms. In addition, the type, frequency, severity, timing, duration, and relationship of TEAEs to elraglusib were analyzed.

A DLT was defined as a select hematologic and nonhematologic AE that occurred in the first 21 days of the first treatment cycle, unless a clear alternative explanation (e.g., related to underlying disease/progression) was available for the emergence of the AE. Hematologic AEs that qualified for consideration as a DLT consisted of grade 4 neutropenia lasting over 7 days, grade 3 febrile neutropenia [defined as absolute neutrophil count less than 1,000/mm^3 with a single temperature greater than 38.3°C (101°F) or a sustained temperature of 38°C (100.4°F) or greater for longer than 1 hour], grade 3 or above neutropenic infection, grade 3 or above thrombocytopenia with bleeding, and grade 4 thrombocytopenia defined as platelet count less than 25,000/mm^3. Nonhematologic AEs that qualified for consideration as a DLT included grade 3 or higher toxicities, except for grade 3 nausea, vomiting, or diarrhea that lasted <72 hours with adequate antiemetic and other supportive care; grade 3 electrolyte abnormality that lasted <72 hours, was clinically uncomplicated or asymptomatic, and resolved spontaneously or responded to conventional medical interventions; grade 3 or 4 amylase or lipase elevations without symptoms or clinical manifestations of pancreatitis; and grade 3 or higher infusion reaction, allergic reaction, or anaphylaxis. Other nonhematologic AEs eligible for the classification as a DLT were bilirubin levels of two or more times higher than the upper limit of normal (five or more times higher than the upper limit of normal in the setting of liver metastases or infiltration of malignant cells) unrelated to disease progression or other known cause and a treatment delay of more than 3 weeks for the next scheduled cycle due to persisting treatment-related toxicities. All DLTs needed to represent a clinically significant shift from baseline.

MTD was defined as the highest dose at which no more than 1 of 6 patients experienced a DLT, and at least 6 patients were treated with this dose. The RP2D was defined as the highest dose that did not exceed MTD.

Secondary endpoints included the assessment of antitumor activity by the RECIST, version 1.1 (RECISTv1.1) for solid tumors and Response Assessment in Neuro-Oncology (RANO) criteria for primary CNS tumors. The efficacy population included all patients who received one or more doses of elraglusib, had at least one postbaseline assessment of efficacy, or who discontinued study early due to progressive disease (PD). Clinical outcomes were reported by overall response rate (ORR), stable disease (SD), duration of response (DoR), progression-free survival (PFS), and overall survival (OS). ORR was defined as the percent of patients achieving complete response (CR) and partial response (PR). SD rate was defined as percent of patients achieving SD ≥ 4 cycles. One cycle of elraglusib monotherapy was defined as 21 days. Most chemotherapy combination cycles were 21 days except GnP, which was 28 days and lomustine, which was 12 weeks. SD in patients with primary brain tumors was defined using RANO criteria. DoR was defined as the time from documented tumor response to disease progression, PFS was defined as the time from study enrollment to objective tumor progression or death, and OS was defined as the time from study enrollment to death from any cause. For PFS, the earliest date from the end of treatment, end of study, or latest visit was used for patients without a date for tumor progression or death. OS was followed for at least 12 months.

Pharmacokinetic analyses

The pharmacokinetic population consisted of patients with at least one blood sample that provided evaluable pharmacokinetic data for elraglusib. Pharmacokinetic sampling was performed predose and up to 24 hours postdose for elraglusib monotherapy and up to 72 hours postdose for elraglusib in combination with chemotherapy. Supplementary Table S5 displays a schedule for the collection of blood samples in part 1. Plasma concentrations were used to calculate pharmacokinetic parameters and their ratios using standard noncompartmental analysis methods.

Cytokine analysis

Multiplex immunoassay

A custom R&D systems Human Premixed Multi-Analyte Kit (catalog no. LXSAHM-39, R&D Systems, Inc.) was run on a Luminex 200 Instrument (Luminex Corporation) according to the manufacturer's instructions. Human plasma levels of IL1α, IL1β, IL2, IL6, IL8, IL12(p70), IL17A, IFNγ, TNFα, MCP-1, MIP-1α, MIP-1β, and TGFβ were measured. All samples were centrifuged after being thawed and were diluted using a 2-fold dilution with Calibrator Diluent RD6-52. Patient samples were run in duplicate, and all analyte values were reported in picograms per milliliter (pg/mL).

TGFβ-1 assay

A TGFβ-1 Premixed Magnetic Luminex Performance Assay (catalog no. FCSTM17-1, R&D Systems, Inc.) was run on a Luminex 200 Instrument (Luminex Corporation) according to the manufacturer's instructions. Human plasma levels of TGFβ-1 were measured. All samples were centrifuged after being thawed and were diluted using a 15-fold dilution. Total TGFβ-1 was quantified by activating patient samples with 1N HCl for 10 minutes, neutralizing with 1.2N NaOH/0.5M HEPES, and then immediately assaying for TGF-β1. Patient samples were run in duplicate, and all analyte values were reported in pg/mL.

IHC assay for GSK-3β expression

Tumor tissues were collected from patients enrolled in both parts 1 and 2. The preferred tissue was fresh predose tissue, but an archival tissue was acceptable if fresh predose tissue was impossible to collect or unavailable. The collection of postdose tissue samples was encouraged but not mandatory. Tissues were analyzed for tumor content and the quality by hematoxylin and eosin (H&E) staining, and the tissues that passed the pathology review were analyzed for GSK-3β expression using IHC. GSK-3β IHC was performed on formalin-fixed, paraffin-embedded tumor sections using a method developed and validated by Mosaic Laboratories. The method involved a validated GSK-3β IHC assay, GSK-3β (rabbit clone D5C5Z), and an automated detection with Leica Bond RX (Leica Biosystems) using commercially available reagents.

GSK-3β expression was assessed for the staining intensity (ranging from 0 as negative to 3+ as strong) and the percentage of stained tumor cells. Additional analyses were performed to correlate GSK-3β expression with outcomes such as days on study, OS, tumor response, and DoR.

Statistical analysis

For parts 1 and 2, formal statistical hypothesis testing was not planned. In part 1, 30 to 50 patients were expected to be enrolled based on the 3+3 dose-escalation design, the observed safety profile, and the number of dose escalations required to meet MTD. In part 2, 12 to 18 patients were expected to be enrolled into each concomitant chemotherapy group based on the 3+3 dose-escalation design and observed safety profiles.

Descriptive statistics were used to analyze safety, efficacy, and pharmacodynamic and pharmacokinetic parameters. Categorical variables are summarized by frequency distributions (number and percentages of patients), while continuous variables are represented with mean, SD, median, minimum, and maximum. Kaplan–Meier methodology assessed time-to-event variables such as PFS and OS. For estimates, two-sided 95% confidence intervals (CI) were provided. All statistical analyses were performed using SAS software version 9.4 or higher.

Data availability

Actuate Therapeutics is committed to sharing access to patient-level data and supporting clinical documents from eligible studies with qualified external researchers. These requests are reviewed and approved on the basis of scientific merit. The data generated in this study are not publicly available due to patient privacy issues but are available upon reasonable request to Actuate. All data provided will be anonymized to adhere to all applicable privacy laws and regulations. Actuate Therapeutics will accept requests for sharing the data presented in this article and will accommodate requests under confidentiality with a proper description of the intended use of that data. These requests may be sent directly to Actuate at info@actuatetherapeutics.com. The sequencing data described in this publication are also not publicly available due to the risk of compromising patient privacy but are also available under confidentiality for any qualified request to info@actuatetherapeutics.com.

Results

Patient disposition and baseline characteristics

Between January 2019 and August 2021, 67 patients and 171 patients received at least one dose of elraglusib as monotherapy (part 1) or in combination with chemotherapy (part 2), respectively. In the study, eight dose levels were evaluated in 67 patients in part 1 and six dose levels in 171 patients during part 2. Supplementary Table S6 depicts the number of patients enrolled in each dose level in parts 1 and 2 and the reasons for study discontinuation. Supplementary Table S7 displays the enrollment and reasons for treatment discontinuation of 171 patients from part 2 across eight different cohorts with concomitant chemotherapy as follows: gemcitabine ($n = 36$), doxorubicin ($n = 10$), lomustine ($n = 14$), carboplatin ($n = 27$), irinotecan ($n = 34$), GnP ($n = 27$), paclitaxel plus carboplatin ($n = 17$), and pemetrexed plus carboplatin ($n = 6$).

Table 1. Patient demographics and baseline characteristics.

Characteristic	Part 1 ($N = 67$[a])	Part 2 ($N = 171$[a])
Age (years), mean (range)	59.4 (27–88)	58.9 (27–90)
Sex, *n* (%)		
Male	37 (55.2)	82 (48)
Female	30 (44.8)	89 (52)
Race, *n* (%)		
White	59 (88.1)	149 (87.1)
Black or African American	3 (4.5)	3 (1.8)
Asian	1 (1.5)	2 (1.2)
Other	4 (6)	17 (10)
Ethnicity, *n* (%)		
Hispanic or Latino	13 (19.4)	26 (15.2)
Not Hispanic or Latino	54 (80.6)	132 (77.2)
Not reported	NA	3 (1.8)
Unknown	NA	10 (5.8)
Number of prior chemotherapy regimens, median (range)	3 (1–13)	4 (1–15)
Prior therapies, *n* (%)		
Chemotherapy	51 (76.1)	166 (97.1)
Immunotherapy	23 (34.3)	35 (20.5)
Targeted therapy	16 (23.9)	23 (13.5)
Hormonal therapy	4 (6)	8 (4.7)
Investigational agent	12 (17.9)	27 (15.8)
Other	6 (9)	13 (7.6)
ECOG performance status, *n* (%)		
0	24 (35.8)	47 (27.5)
1	40 (59.7)	114 (66.7)
2	2 (3)	10 (5.8)
3	1 (1.5)	0 (0)

Abbreviations: ECOG, Eastern Cooperative Oncology Group; NA, not applicable.
[a]Two patients began treatment in part 1 (monotherapy) and transitioned to part 2 (combination chemotherapy) and are included in results for both study parts.

Patient demographic and baseline characteristics for both parts are displayed in **Table 1**. The median ages were 60 (part 1) and 61 years old (part 2), and 45% and 52% of patients were female. Patients received a median of 3 and 4 prior systemic chemotherapy regimens in the monotherapy and combination with chemotherapy groups, respectively. Solid tumors represented most treated cancers [$n = 60$ (89.6%) in part 1 and $n = 159$ (93%) in part 2]. The most common cancers were colorectal cancer, melanoma, and pancreatic cancer in part 1 (Supplementary Table S8) and pancreatic, colorectal, and lung cancers in part 2 (Supplementary Table S9).

The median duration of treatment with elraglusib was 39 days (range, 1–1,264 days) as monotherapy and 53 days (range, 1–1,040 days) in combination with chemotherapy agents. The median number of administered doses for elraglusib was 12 doses in part 1 and 15 doses in part 2. The primary reason for treatment discontinuation was disease recurrence or progression in part 1 ($n = 52$; 77.6%) and

Table 2. TEAEs of any grade reported in ≥20% of patients treated with elraglusib.

	Patients, *n* (%)			
	Elraglusib monotherapy part 1 (*N* = 67)		Elraglusib with chemotherapy part 2 (*N* = 171)	
Adverse event	**Any grade**	**Grade ≥3**	**Any grade**	**Grade ≥3**
Any TEAE	67 (100)	37 (55.2)	171 (100)	124 (72.5)
Serious TEAE	29 (43.3)	26 (38.8)	72 (42.1)	67 (39.2)
Leading to treatment discontinuation	6 (9)	4 (6)	36 (21.1)	30 (17.5)
Leading to death	5 (7.5)	5 (7.5)	18 (10.5)	18 (10.5)
TEAEs of any grade in ≥20% of patients				
Visual impairment	34 (50.7)	0	104 (60.8)	1 (0.6)
Fatigue	32 (47.8)	2 (3)	86 (50.3)	8 (4.7)
Nausea	25 (37.3)	1 (1.5)	77 (45)	3 (1.8)
Diarrhea	21 (31.3)	3 (4.5)	52 (30.4)	6 (3.5)
Anemia	17 (25.4)	4 (6)	80 (46.8)	43 (25.2)
Vomiting	17 (25.4)	1 (1.5)	47 (27.5)	5 (2.9)
Headache	16 (23.9)	0	36 (21.1)	1 (0.6)
Abdominal pain	12 (17.9)	3 (4.5)	38 (22.2)	6 (3.5)
Neutrophil count decrease	2 (3)	2 (3)	45 (26.3)	36 (21.1)
Platelet count decrease	1 (1.5)	0	50 (29.2)	27 (15.8)
White blood cell count decrease	Not reported	Not reported	42 (24.6)	28 (16.3)

Abbreviations: DLT, dose-limiting toxicity; TEAE, treatment-emergent adverse event.

part 2 ($n = 108$; 63.2%; Supplementary Table S7). In part 1, 6% of patients discontinued treatment due to AEs (0% attributable to elraglusib), and 21.1% discontinued treatment due to AEs in part 2 (1.2% attributable to elraglusib; **Table 2**; Supplementary Table S10). The primary reason for study discontinuation by patients in both parts 1 and 2 was the difficult logistics of a twice-weekly regimen, particularly in the context of the COVID-19 pandemic.

Safety

No DLTs related to elraglusib occurred in part 1. In part 2, DLTs of grade 2 decrease in appetite, grade 2 fatigue, and grade 2 malaise that delayed the next scheduled cycle by more than 3 weeks occurred in 1 patient treated with gemcitabine plus elraglusib in the 5 mg/kg cohort. These DLTs were categorized as possibly related to elraglusib and gemcitabine. None of the additional patients enrolled in that cohort ($N = 8$ in total) experienced a DLT and dose escalation was allowed to continue. Because no additional DLT was experienced in the other dosing cohorts, the MTD and RP2D initially was 15 mg/kg, the prespecified highest dose evaluated in both study parts.

In both parts, all patients experienced TEAEs (**Table 2**). Non-DLT grade 3 or higher TEAEs affected 55.2% of patients receiving elraglusib monotherapy and 72.5% of patients receiving elraglusib in combination with chemotherapy (**Table 2**; Supplementary Table S10). In part 2, grade 3 or higher decrease in neutrophil count/neutropenia occurred in 21.1% of patients ($n = 36$) and grade 3 anemia in 25.2% of patients ($n = 43$). Most grade 3 or higher TEAEs were attributed to concomitant chemotherapy [48.5% ($n = 83$) attributed to chemotherapy vs. 5.8% ($n = 10$) attributed to elraglusib; **Table 3**; Supplementary Table S10].

The most common TEAEs attributed to elraglusib were visual changes and fatigue across both study parts, and the majority of TEAEs that occurred in ≥ 20% of patients were grade 1 or 2 (**Table 2**). Visual changes affected 50.7% of patients ($n = 34/67$) receiving elraglusib monotherapy and 60.8% of patients ($n = 104/171$) receiving elraglusib with chemotherapy. Commonly reported symptoms were visual disturbances/changes and darkened vision, where patients described lights as brighter and skin tones darker. Most cases of visual changes were grade 1 or 2 except for grade 3 in one patient who received elraglusib

Table 3. Incidence of common TEAEs related to elraglusib versus concomitant chemotherapy.

	Adverse event	Part 2 (*N* = 171)	Grade 1	Grade 2	Grade 3	Grade 4
Related to elraglusib	Visual impairment, *n* (%)	102 (59.6)	93 (54.4)	8 (4.7)	1 (0.6)	0 (0)
	Fatigue, *n* (%)	34 (19.9)	8 (4.7)	26 (15.2)	0 (0)	0 (0)
	Infusion-related reaction, *n* (%)	17 (9.9)	3 (1.8)	13 (7.6)	1 (0.6)	0 (0)
	Nausea, *n* (%)	16 (9.4)	10 (5.8)	6 (3.5)	0 (0)	0 (0)
	Headache, *n* (%)	14 (8.2)	13 (7.6)	1 (0.6)	0 (0)	0 (0)
	Vascular access complication, *n* (%)	12 (7.0)	3 (1.8)	9 (5.3)	0 (0)	0 (0)
Related to concomitant chemotherapy	Fatigue, *n* (%)	69 (40.4)	17 (9.9)	46 (26.9)	6 (3.5)	0 (0)
	Anemia, *n* (%)	65 (38.0)	5 (2.9)	26 (15.2)	34 (19.9)	0 (0)
	Nausea, *n* (%)	57 (33.3)	32 (18.7)	23 (13.5)	2 (1.2)	0 (0)
	Platelet count decreased, *n* (%)	46 (26.9)	10 (5.8)	11 (6.4)	15 (8.8)	10 (5.8)
	Neutrophil count decreased, *n* (%)	44 (25.7)	1 (0.6)	8 (4.7)	14 (8.2)	21 (12.3)
	White blood cell count decreased, *n* (%)	41 (24.0)	4 (2.3)	9 (5.3)	16 (9.4)	12 (7.0)

15 mg/kg with pemetrexed and carboplatin in part 2. All cases of visual changes were transient, usually lasted less than 1 hour, completely resolved, and lacked any associated retinal, ocular, or systemic symptoms or findings. There were two cases of ataxia and gait disturbances (grade 1 and 2, respectively) possibly related to elraglusib.

Infusion reactions occurred in 4 patients (6%) in part 1 and 23 patients (13.5%) in part 2. Most infusion reactions were grade 1 or 2 except for a grade 3 infusion reaction in 2 patients from part 2. In those cases, one grade 3 infusion reaction was attributed to carboplatin while the other was probably related to elraglusib. All infusion reactions were reversed and responded to standard medical care, including acetaminophen, antihistamines, H2-blockers, and steroids.

Serious TEAEs were reported in 43.3% of patients receiving elraglusib monotherapy and 42.1% of patients receiving elraglusib with chemotherapy (**Table 2**). However, only 1 patient had serious TEAEs in part 1—grade 2 nausea, grade 2 vomiting, and grade 2 diarrhea—that were possibly related to elraglusib monotherapy, and the patient was hospitalized due to these TEAEs. In part 2, 5 patients experienced serious TEAEs possibly related to elraglusib: grade 3 transient visual disturbance, grade 2 blurred vision, grade 3 recurrent fall, and grade 3 infusion-related reaction. One patient had grade 3 diarrhea and grade 3 peripheral sensory neuropathy that were probably caused by the combination of elraglusib, paclitaxel, and carboplatin.

Elraglusib administration was associated with both peripheral and central venous line/catheter and/or device (ports) occlusions (**Table 3**), which necessitated device removal and replacement in some patients and was associated with pulmonary embolism in one patient. Initially some of these events were reported as procedures rather than AE, and their frequency as elraglusib-attributable AE was not appreciated until aggregate data became available. Elraglusib deposition within ports or at the catheter tip at removal was evident in some patients. Drug precipitation causing occlusions occurred in patients independent of institution, drug batch, site or time of manufacture, or type of line or venous access device used. Elraglusib-associated occlusions appear to have been reduced by decreasing the dose administered to 9.3 mg/kg once per week. Additional flushing of lines and ports beyond standard of care was also recommended as was the reduction of the elraglusib infusion solution concentration from 1.0 to 0.75 mg/mL.

Pharmacokinetics

The pharmacokinetic analysis included 57 patients in part 1 and 78 patients in part 2. The time to the maximum concentration of elraglusib when administered in combination with chemotherapy ranged between 0.5 and 2.2 hours. The mean terminal half-life ranged from 17.2 to 22.1 hours with monotherapy up to doses of 12.5 mg/kg and 13.1 to 28.1 hours in combination with chemotherapy. The maximum concentrations of elraglusib were dose proportional with monotherapy, and exposures measured by the AUC for 24 hours increased at approximately 20% greater rate than a dose-proportional change. The coadministration with carboplatin, gemcitabine, lomustine, or paclitaxel plus carboplatin increased concentrations of elraglusib during and following the infusion compared with the administration of elraglusib monotherapy (Supplementary Figs. S1–S8). Repeated dosing with carboplatin, gemcitabine, GnP, or paclitaxel plus carboplatin revealed a similar drug accumulation as elraglusib monotherapy. The repeated dosing of elraglusib with doxorubicin, irinotecan, or lomustine led to modest increases in elraglusib exposure.

Clinical activity/pharmacodynamics

Overall, 62 patients in part 1 and 138 patients in part 2 were part of the efficacy population and evaluable for response (**Table 4**). In the efficacy evaluable population, the ORR and SD were 3.2% and 38.7% for elraglusib monotherapy and 5.1% and 41.3% for combination therapies, respectively. The median PFS (mPFS) was 1.6 months (95% CI, 1.3–2.2) for elraglusib monotherapy (Supplementary Fig. S9A) and 2.1 months (95% CI, 2.0–2.6) for the combination of elraglusib and chemotherapy (Supplementary Fig. S10A). The median duration of treatment was 39 days for monotherapy and 53 days for combination therapies. The median OS was 7.7 months (95% CI, 5.1–9.7; Supplementary Fig. S9B) for elraglusib monotherapy, with 12-month survival of 19.4%. On the basis of observed data and compared with lower dose levels, elraglusib monotherapy at dose levels of 5 mg/kg and above trended toward longer OS (Supplementary Figs. S11 and S12).

Table 4. Best overall response of elraglusib as monotherapy and in combination with chemotherapy (efficacy population).

Outcome	Elraglusib monotherapy part 1 ($N = 62$)	Elraglusib with chemotherapy part 2 ($N = 138$)
Best overall response, *n* (%)		
Complete response	1 (1.6)	0 (0.0)
Partial response	1 (1.6)	7 (5.1)
Stable disease	24 (38.7)	57 (41.3)
Progressive disease	30 (48.4)	70 (50.7)
Not reported	6 (9.7)	4 (2.9)
PFS, median (95% CI), months[a]	1.6 (1.3–2.2)	2.1 (2.0–2.6)
OS, median (95% CI), months[a]	7.7 (5.1–9.7)	6.9 (5.7–8.4)

[a]Using Kaplan–Meier method.

The median OS was 6.9 months (95% CI, 5.7–8.9; Supplementary Fig. S10B) for the combination of elraglusib and chemotherapy, with a 12-month survival of 20.3%. Individual chemotherapy combinations were analyzed for mPFS and median OS (mOS) for the efficacy evaluable (Supplementary Table S11; Supplementary Fig. S13). The combinations of elraglusib with carboplatin single agent, elraglusib plus carboplatin and paclitaxel or pemetrexed have been combined for analysis.

In part 1, a patient with refractory BRAF V600E-mutated melanoma achieved CR (DoR of 40.9 months; Supplementary Fig. S14; ongoing at the time of data cutoff), and a patient with acute T-cell leukemia/lymphoma had PR (DoR of 7.6 months; Supplementary Figs. S15–S17). In part 2, PR was observed in 7 patients (5.1%). These consisted of 4 patients in the paclitaxel and carboplatin arm: cervical cancer (DoR of 8.7 months), endometrial cancer (DoR of 15 months), anal carcinoma (DoR of 1 month), and ovarian cancer (DoR of 5.7 months); 3 patients in other combination arms: gliosarcoma (lomustine arm; DoR of 3.9 months), pancreatic cancer (nab-paclitaxel and gemcitabine arm; DoR of 2.9 months), soft-tissue sarcoma (doxorubicin arm; DoR of 5.8 months). Two patients with PR experienced reductions in CA-125 levels (by 89% and 92%) after receiving elraglusib in combination with paclitaxel and carboplatin for ovarian cancer and endometrial carcinoma.

Cytokine analysis

Seven patients treated with single-agent elraglusib in part 1 had cytokines profiled at predose, and 4 hours, 24 hours, and day 8 postdose. The tumor types profiled were pancreatic ($n = 2$), melanoma (2), appendiceal (1), mucinous adenocarcinoma (1), and glioblastoma (1). Patient cytokine levels demonstrated changes across timepoints and were plotted by patient using absolute cytokine values measure as described under Patients and Methods (Supplementary Fig. S18).

Table 5. Positive staining for GSK-3β by the type of tissue sample from parts 1 and 2.

Sample	Positive for GSK-3β staining, *n* (%)
Part 1	
Archival sample ($N = 36$)	30 (83.3)
Fresh predose sample ($N = 32$)	19 (59.4)
Part 2	
Archival sample ($N = 67$)	56 (83.6)
Fresh predose sample ($N = 44$)	36 (81.8)

GSK-S3β expression

Fifty (74.6%) patients from part 1 and 96 (56.1%) patients from part 2 provided at least one sample of predose tumor tissues eligible for GSK-3β expression analysis. Fresh tissue samples were collected from 32 (47.8%) patients from part 1 and 44 (25.7%) patients from part 2, while archival samples were available for 36 (47.8%) patients from part 1 and 67 (39.2%) patients from part 2. Because the collection of postdose tissue samples was optional, only a few paired biopsies were available but were insufficient for the analysis.

Table 5 summarizes tumor tissues from parts 1 and 2 that were stained for GSK-3β expression. Positive GSK-3β immunostaining was observed in over 80% of the archival samples from parts 1 and 2 and the fresh tissue samples from part 2 and in 59.4% of the fresh tissue samples from part 1. The percent staining ranged from 5% to 100%, and the staining intensity varied from 1+ to 3+. To date, no correlations between GSK-3β expression and evaluated clinical outcomes have been observed.

Discussion

With elraglusib monotherapy, a patient with *BRAF V600K*-mutated melanoma refractory to checkpoint inhibitors (nivolumab, ipilimumab) and dabrafenib/trametinib achieved a durable CR by CT and complete metabolic response by PET that has lasted over 4.3 years and is ongoing. Ten other patients with refractory melanoma enrolled in the study, but none with V600K mutation. As this was the only patient with melanoma to achieve a CR, the emerging evidence of a unique molecular signature with decreased dependence on the MAPK pathway and strong inhibition of multiple antiapoptotic pathways of this subtype could in part explain the superior response (29). A patient with acute T-cell leukemia/lymphoma, who had progressed after IFN/chemotherapy, mogamulizumab and lenalidomide, showed disease control for 16.3 months (including a PR lasting 7.6 months). *Ex vivo* treatment with elraglusib of $CD8^+$ T cells derived from this patient enhanced the secretion of modulators of T-cell response including IFNγ, granzyme B, and TRAIL (30). The small sample size of each combination cohort in part 2 limited the interpretation of preliminary efficacy results and speculation of a possible sensitizing role of elraglusib upon reexposure to chemotherapy backbones. The ongoing randomized phase II study of gemcitabine plus nab-paclitaxel with versus without elraglusib will help address this question (NCT03678883).

Transient visual changes attributed to elraglusib were reported by more than half of treated patients with both monotherapy and combination regimens. Almost all visual changes were grade 1 or 2, dose dependent and all rapidly fully resolved without any associated retinal, ocular, or other findings. GSK-3β is present in Muller and photoreceptor cells in the retina and regulates glycogen synthesis as part of glycogen metabolism (31). Thus, elraglusib-associated transient visual changes may reflect inhibition of GSK-3β within photoreceptors. Transient visual changes were generally observed during the first few cycles of treatment only suggesting a desensitization to this adverse effect. Transient visual impairment was also reported in clinical studies with another GSK-3β inhibitor LY2090314 (32, 33).

Elraglusib administration was associated with both peripheral and central venous line/catheter and/or device (ports) occlusions in some patients, which necessitated device removal and replacement in some patients and was associated with pulmonary embolus in 1 patient. No patient or line/device-associated risk factors for this serious complication were identified. Elraglusib is a lipophilic drug formulated for intravenous administration in typical intravenous fluids such as saline or D5W at near its solubility limit and drug deposition within ports or catheter tips at time of removal was demonstrated. Elraglusib-associated occlusions appeared to have been reduced by dose reduction to 9.3 mg/kg (which allows the infusion solution to be diluted below its solubility limit to 0.25–0.75 mg/mL) intravenously once per week; this regimen is being used in the ongoing elraglusib study in patients with metastatic pancreatic cancer (NCT03678883).

The pharmacokinetic profile of elraglusib is distinct from other GSK-3β inhibitors. Elraglusib's terminal half-life ranges between 18 to 22 hours as monotherapy or 13 to 28 hours in combination with chemotherapy. This half-life is distinct from that of LY2090314 whose half-life was approximately 3 hours at the MTD (33). LY2090314 did not achieve sufficient plasma exposure to inhibit tumor cell growth based on IC_{50} data, which may have contributed to the lack of observed therapeutic benefit (32). Elraglusib achieved plasma exposures that exceeded its *in vitro* IC_{50} for >6 hours after a single administration at doses of 5 mg/kg or higher and approached 24 hours at doses of 9.3 mg/kg and higher (**Fig. 1**). Elraglusib exposures (AUC) are approximately dose proportional with the AUC∞ at 9.3 mg/kg being 2.1X.

The pharmacokinetic profile of elraglusib in most patients was not significantly altered by its addition to standard anticancer regimens that included carboplatin, doxorubicin, gemcitabine, lomustine, nab-paclitaxel, or paclitaxel. However, coadministration of elraglusib with select chemotherapy regimens increased the exposure to elraglusib after a single dose. While inconsistent data collection, handling, and processing may be a contributing factor, the increase in exposure of elraglusib after coadministration with lomustine or paclitaxel plus carboplatin could also be due to lomustine and paclitaxel being substrates for cytochrome P450 (CYP) enzymes (34, 35). Similarly, repeated dosing of elraglusib in combination with doxorubicin, irinotecan, or lomustine resulted in increased exposure of elraglusib, suggesting that elraglusib is competing for shared CYP isoforms with these agents (34, 36, 37).

Emerging data have credentialed GSK-3β as a novel immunomodulatory target, and elraglusib has shown the ability to regulate immune checkpoint expression and to enhance the activity of anti-PD-1 and anti-PD-L1 antibodies *in vivo* (25–27). In a melanoma mouse model, elraglusib, in combination with an anti-PD-1 mAb, displayed synergistic decreases in the expression of immune checkpoints PD-1, TIGIT, and LAG-3 (25). Sequential therapy of anti-PD-1 mAb followed by elraglusib was especially effective, suggesting that elraglusib enhances the effects of $CD8^+$ T cells after "priming" with anti-PD-1 mAbs. Huntington and colleagues showed that plasma from elraglusib-treated patients in part 1 of this study demonstrated reduced VEGF

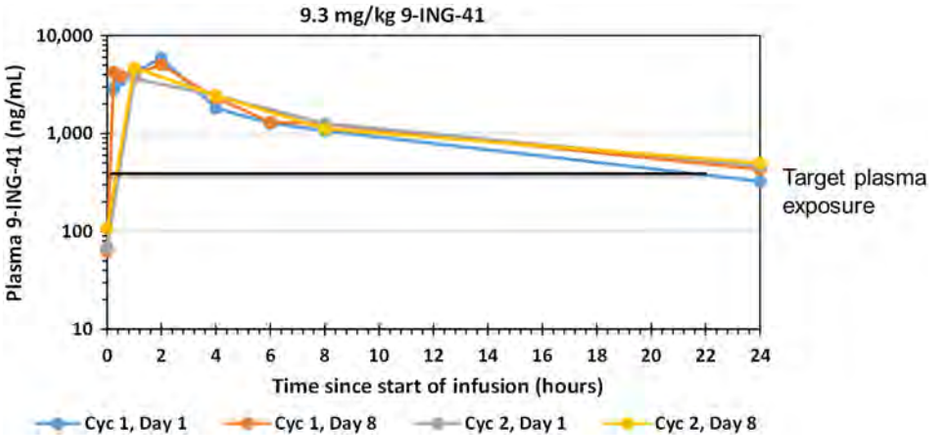

Figure 1.
Elraglusib plasma exposure at 9.3 mg/kg exceeds *in vitro* IC_{50} for up to 24 hours after a single administration. The *in vitro* IC_{50} is approximately 1–3 mmol/L represented by the line.

and BAFF and elevated IL1β, CCL22, and CCL4 concentrations correlated with longer survival (38). Using paired tumor biopsies from patients with colorectal cancer in part 1, Huntington and colleagues also showed that tumor-infiltrating immune cells had reduced expression of inhibitory immune checkpoints (VISTA, PD-1, PD-L2) and elevated expression of T-cell activation markers (CTLA-4, OX40L) after elraglusib treatment (38). Changes in plasma cytokine levels were observed even after a single dose of elraglusib across various tumor histologies (Supplementary Fig. S18), demonstrating that elraglusib treatment leads to immune regulation. In part 1, several patient populations received and failed anti-PD-1 treatment prior to being enrolled in 1801. For example, 11 patients with metastatic melanoma refractory to anti-PD-1 treatment were all enrolled into part 1 and received elraglusib as a single agent across a variety of dose levels. Many of these patients, including the durable CR (Supplementary Fig. S14), demonstrated reasonable mOS (10 months) despite the fact that these patients had received on average three prior lines of treatment. Clinically meaningful mOS > 12 months was also observed in patients with lung cancer treated in part 2 that had also been pretreated with checkpoint inhibitors (25). Thus, a focus for further studies on elraglusib in checkpoint inhibitory failures seems warranted.

Although over 80% of available predose tissue samples (except for about 60% of fresh tissue samples from part 1) were positive for GSK-3β expression in our study, further analyses revealed a lack of correlation between predose GSK-3β expression and clinical outcomes. These findings correspond to previous studies revealing inconsistent results for GSK-3β expression as a prognostic predictor for different types of cancer. In ovarian cancer, one study showed no correlation between GSK-3β expression and cancer severity, while another study revealed a shortened survival with higher expression of GSK-3β (39, 40). Furthermore, GSK-3β expression was associated with worse clinical outcomes in lung and bladder cancers but with improved outcomes in gastric cancer and squamous cell carcinoma of the tongue (17, 41–45). The correlation between predose GSK-3β expression and clinical outcomes is complex, and most likely depends on the type of cancer and GSK-3β context.

Conclusions

Elraglusib had activity as a monotherapy or in combination in patients with relapsed or refractory malignancies. Vascular access line occlusions and limited drug solubility need to be taken into account when designing future trials. The development of an oral preparation is ongoing. These data support further studies of elraglusib in patients with relapsed or refractory malignancies, including combinations with chemotherapy and immunotherapies.

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Note

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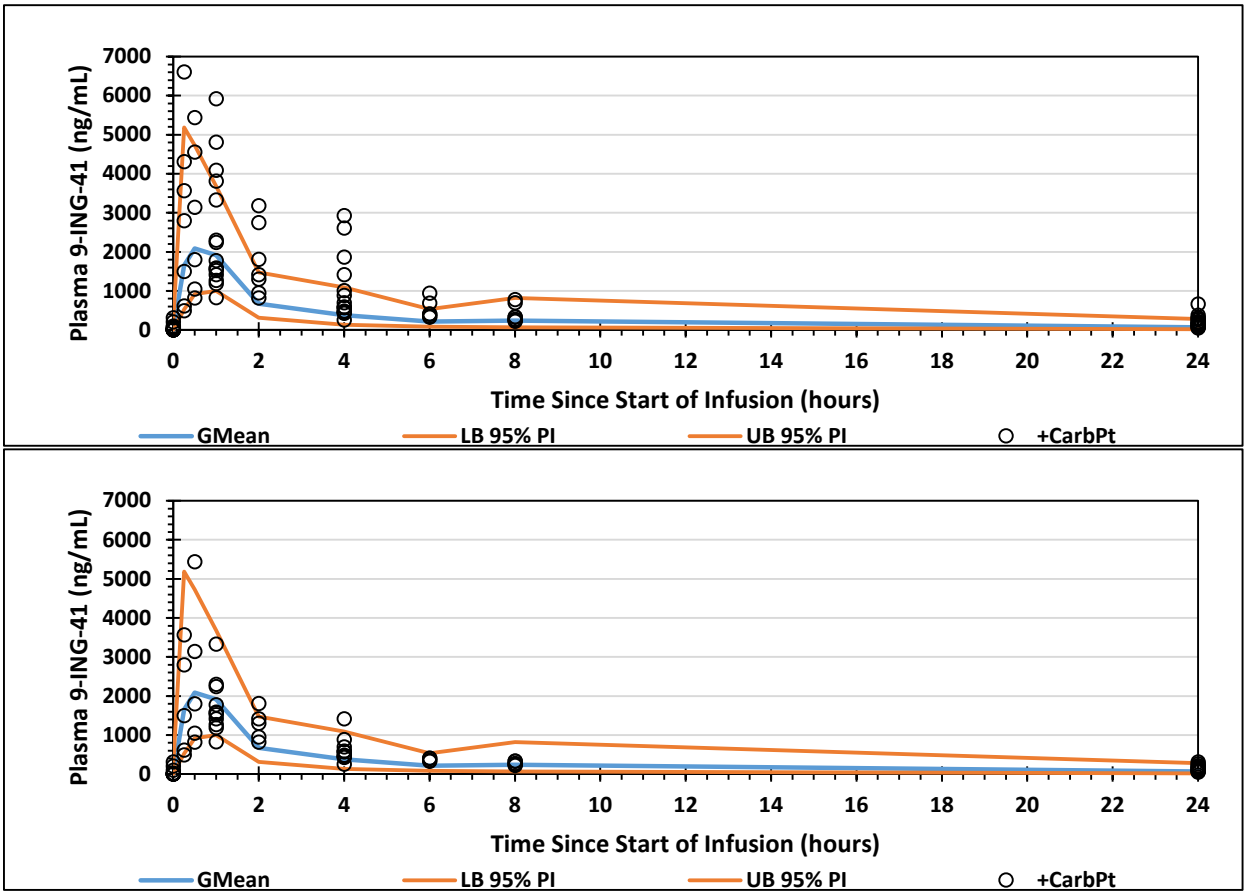
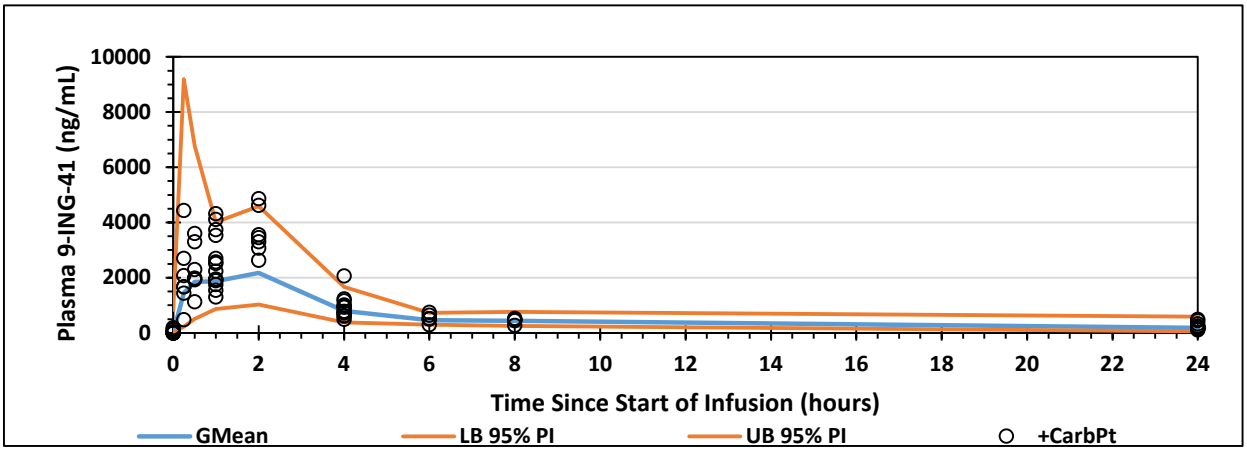
Figure S1. Plasma Elraglusib (9-ING-41) concentrations in 4 Patients after 3.3 mg/kg elraglusib, while receiving a carboplatin regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 3.3 mg/kg as a single agent. PK evaluable population.



Abbreviations: GMean, geometric mean; CarbPt, carboplatin; LB, lower bound; PI, prediction interval; UB, upper bound.

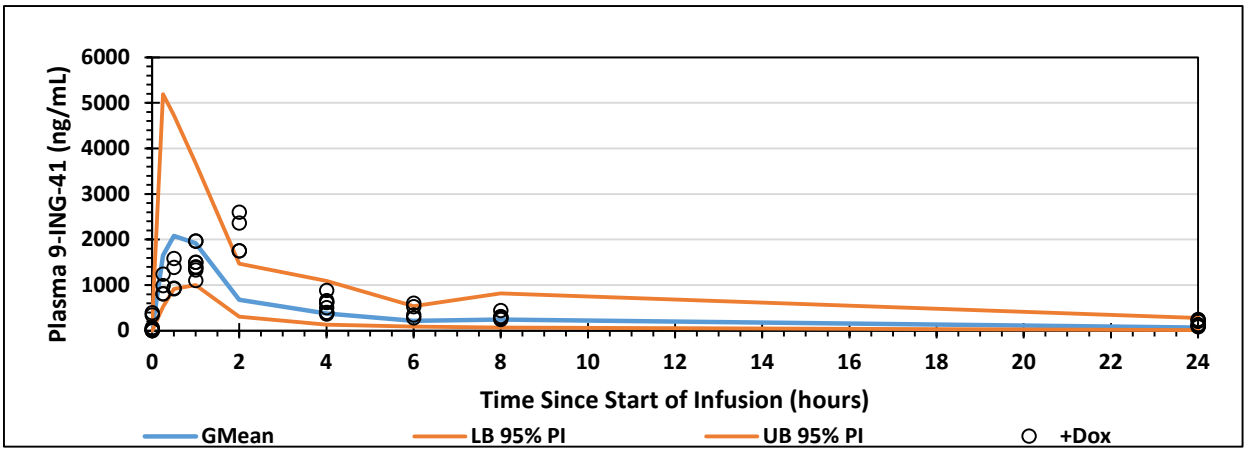
Note: Patient 07-011 was removed from the second figure to demonstrate that most unexpectedly higher concentrations were accounted for by a single patient.

Figure S2. Plasma elraglusib (9-ING-41) concentrations in 4 patients after 5.0 mg/kg elraglusib while receiving a carboplatin regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 5.0 mg/kg as a single agent. PK Evaluable Population



Abbreviations: GMean, geometric mean; CarbPt, carboplatin; LB, lower bound; PI, prediction interval; UB, upper bound

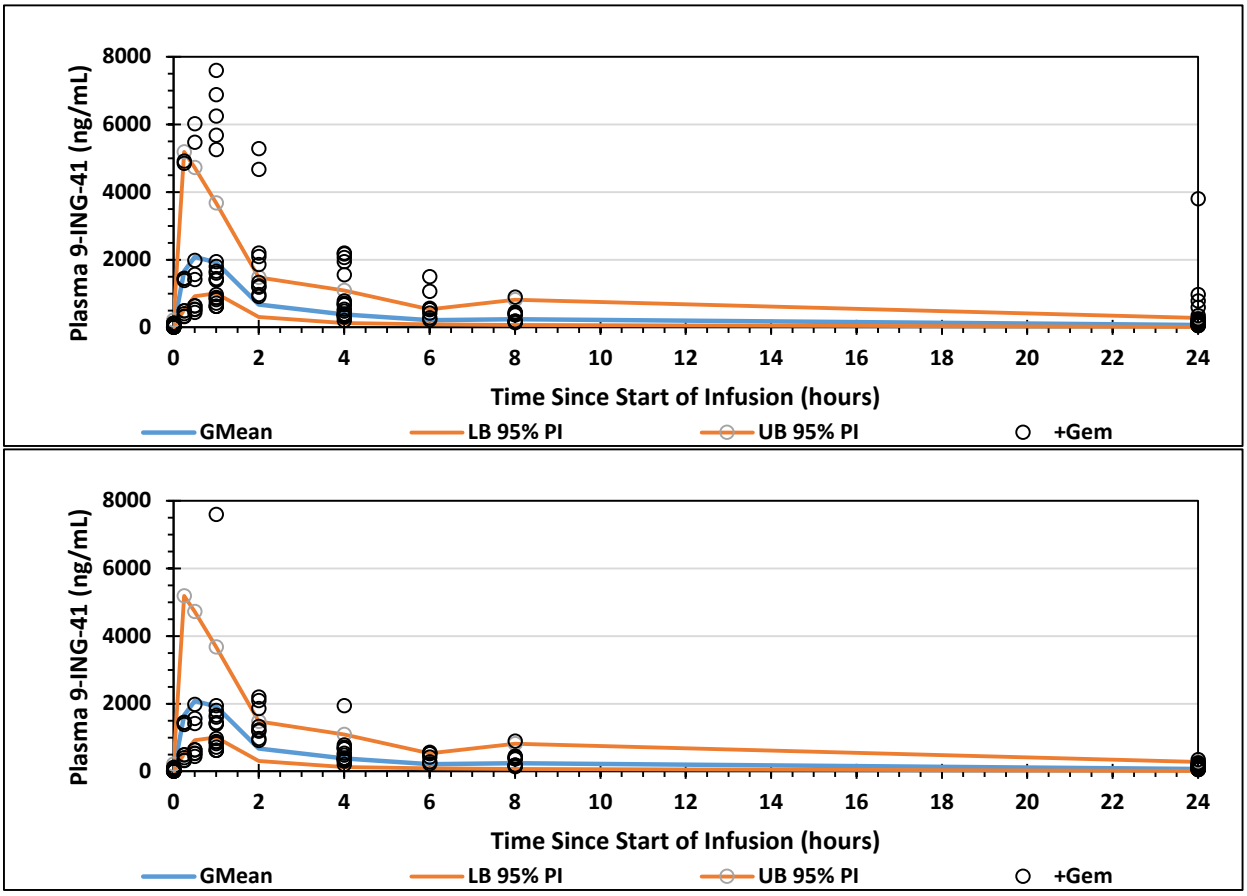
Figure S3. Plasma elraglusib (9-ING-41) concentrations in 2 patients after 3.3 mg/kg elraglusib while receiving a doxorubicin regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 5.0 mg/kg as a single agent. PK Evaluable Population.



Abbreviations: GMean, geometric mean; Dox, doxorubicin; LB, lower bound; PI, prediction interval; UB, upper bound

Note: most of the apparent deviations are likely related to the approximately 1-hour infusion used for monotherapy patients versus the approximately 2-hour infusion used with the doxorubicin combination regimen.

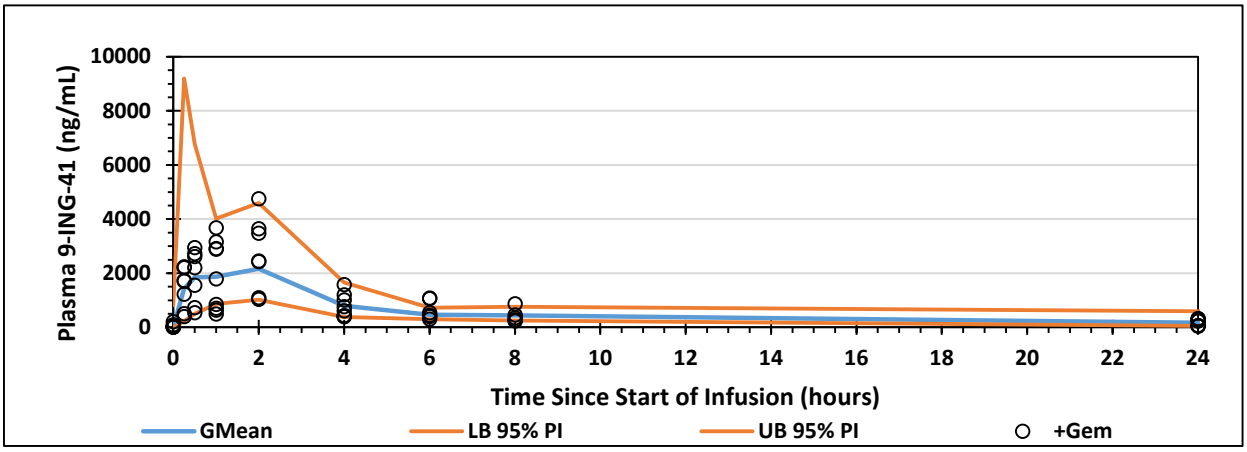
Figure S4. Plasma elraglusib (9-ING-41) concentrations in 4 patients after 3.3 mg/kg elraglusib while receiving a gemcitabine regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 3.3 mg/kg as a single agent. PK Evaluable Population.



Abbreviations: GMean, geometric mean; Gem, gemcitabine; LB, lower bound; PI, prediction interval; UB, upper bound

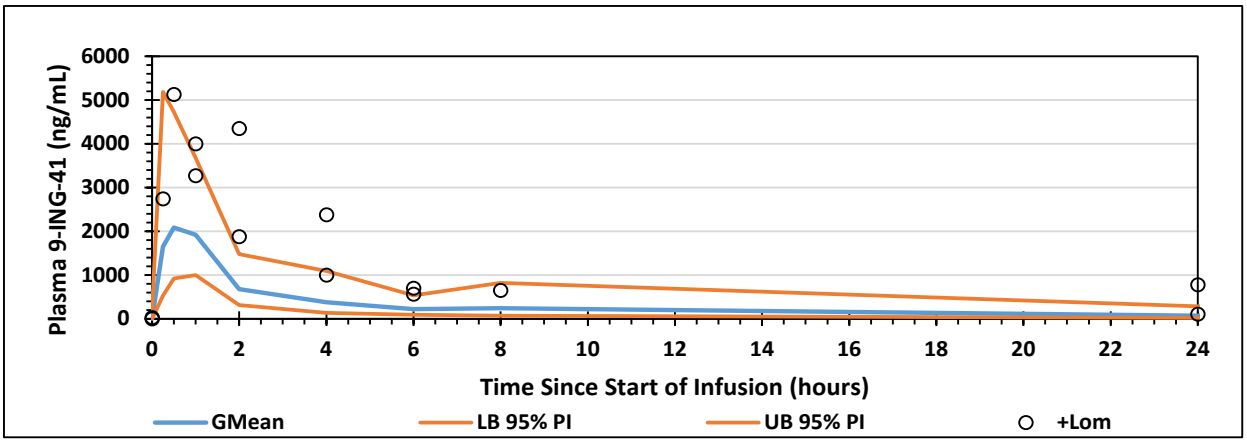
Note: Patient 07-010 was removed from the second figure to demonstrate that most unexpectedly higher concentrations were accounted for by a single patient.

Figure S5. Plasma elraglusib (9-ING-41) concentrations in 4 patients after 5 mg/kg elraglusib while receiving a gemcitabine regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 5 mg/kg as a single agent. PK Evaluable Population.



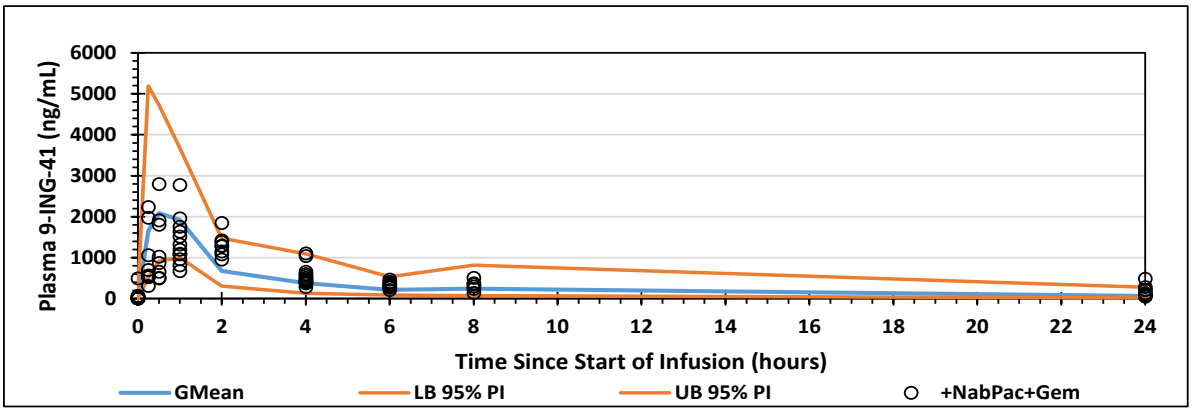
Abbreviations: GMean, geometric mean; Gem, gemcitabine; LB, lower bound; PI, prediction interval; UB, upper bound

Figure S6. Plasma elraglusib (9-ING-41) concentrations in 1 patient after 3.3 mg/kg elraglusib while receiving a lomustine regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 3.3 mg/kg as a single agent. PK Evaluable Population.



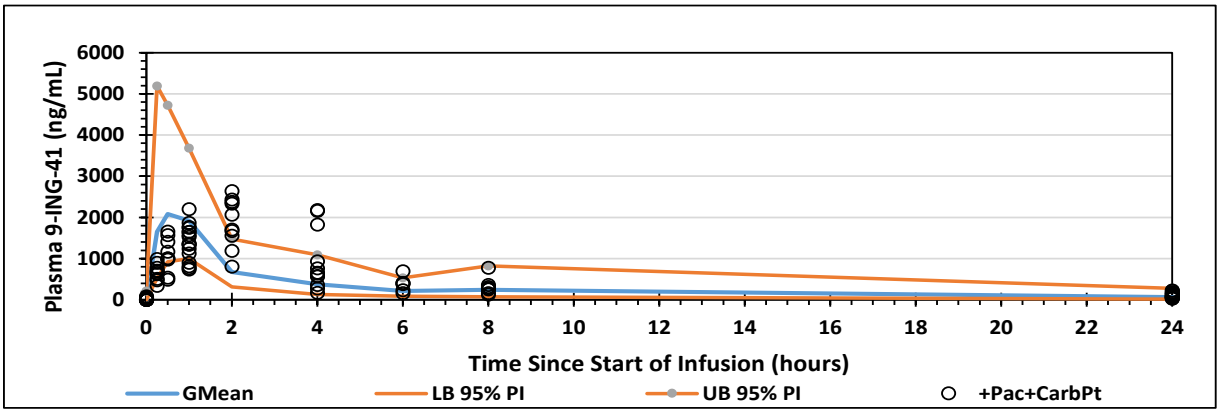
Abbreviations: GMean, geometric mean; Lom, lomustine; LB, lower bound; PI, prediction interval; UB, upper bound
Note: most of the apparent deviations are likely related to the approximately 1-hour infusion used for monotherapy patients versus the approximately 2-hour infusion used with the lomustine combination regimen.

Figure S7. Plasma elraglusib (9-ING-41) concentrations in 4 patients after 3.3 mg/kg elraglusib while receiving a nab-paclitaxel plus gemcitabine regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 3.3 mg/kg as a single agent. PK Evaluable Population.



Abbreviations: GMean, geometric mean; NabPac+Gem, nab-paclitaxel + gemcitabine; LB, lower bound; PI, prediction interval; UB, upper bound

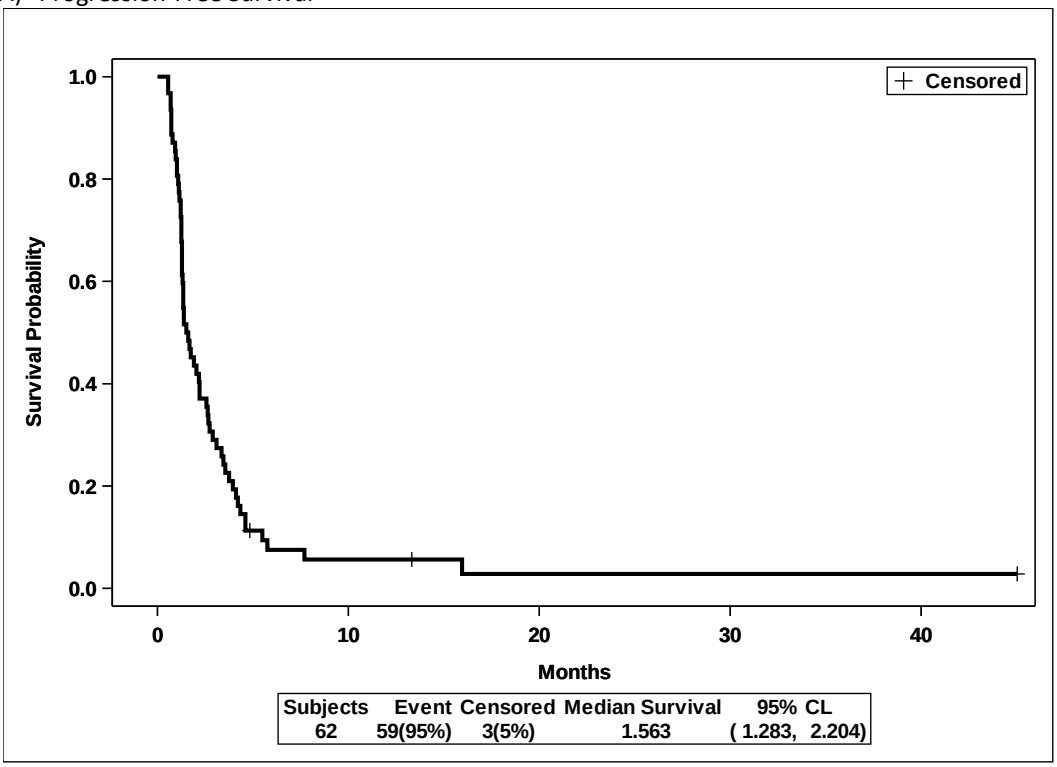
Figure S8. Plasma elraglusib (9-ING-41) concentrations in 6 patients after 3.3 mg/kg elraglusib while receiving paclitaxel plus carboplatin regimen, superimposed on geometric mean (± 95% PI) profile for elraglusib after 3.3 mg/kg as a single agent. PK Evaluable Population.



Abbreviations: GMean, geometric mean; Pac+CarbPt, paclitaxel + carboplatin; LB, lower bound; PI, prediction interval; UB, upper bound

Figure S9. Progression-Free Survival (A) and Overall Survival (B) with Elraglusib Monotherapy.

(A) Progression-Free Survival



(B) Overall Survival

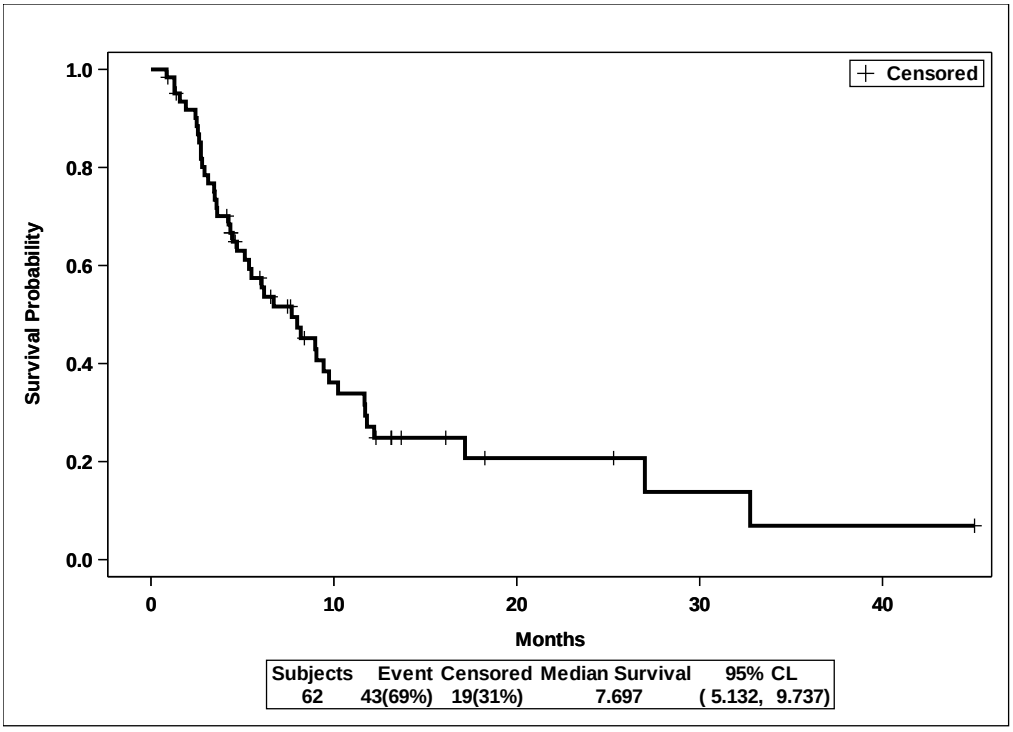
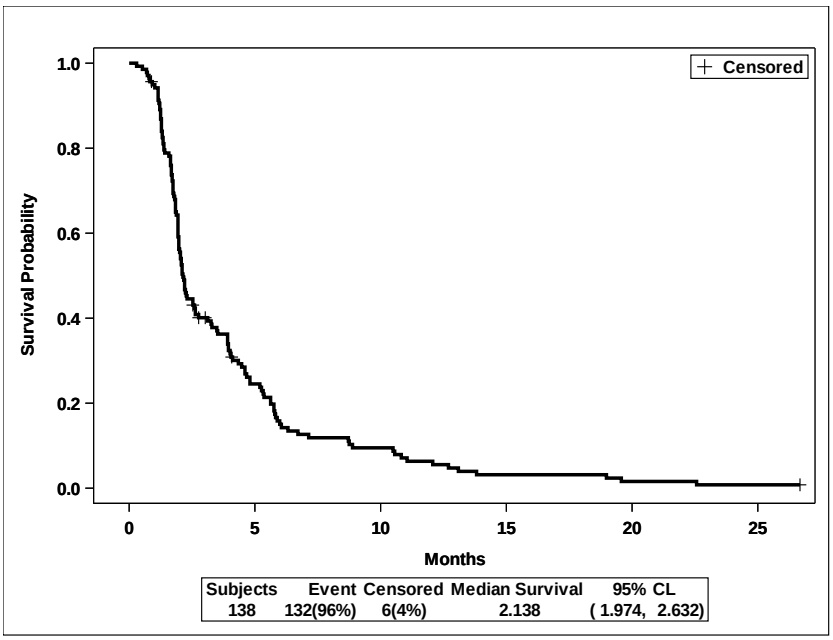


Figure S10. Progression-Free Survival (A) and Overall Survival (B) with Elraglusib in Combination with Chemotherapy.

(A) Progression-Free Survival



(B) Overall Survival

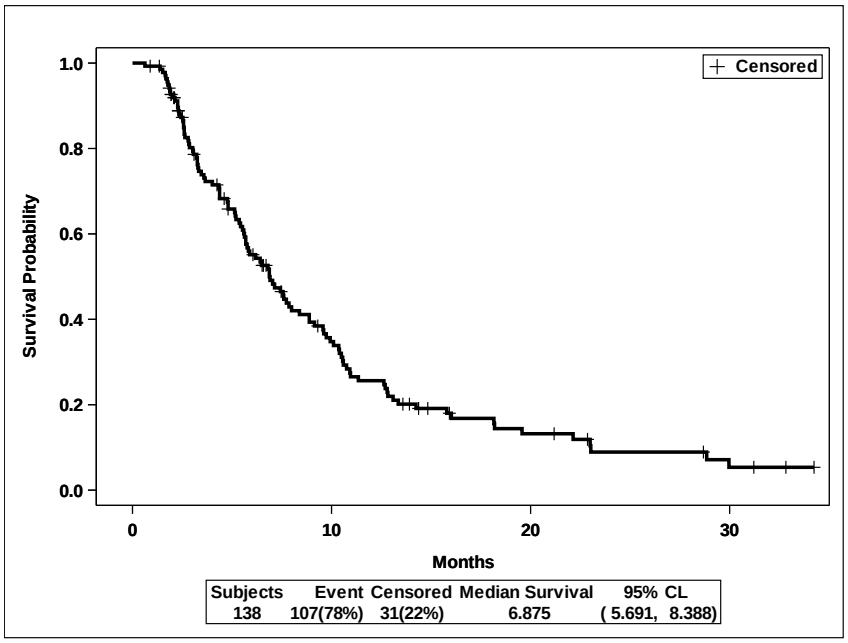


Figure S11. Days on Study by Elraglusib Dose Level in Part 1.

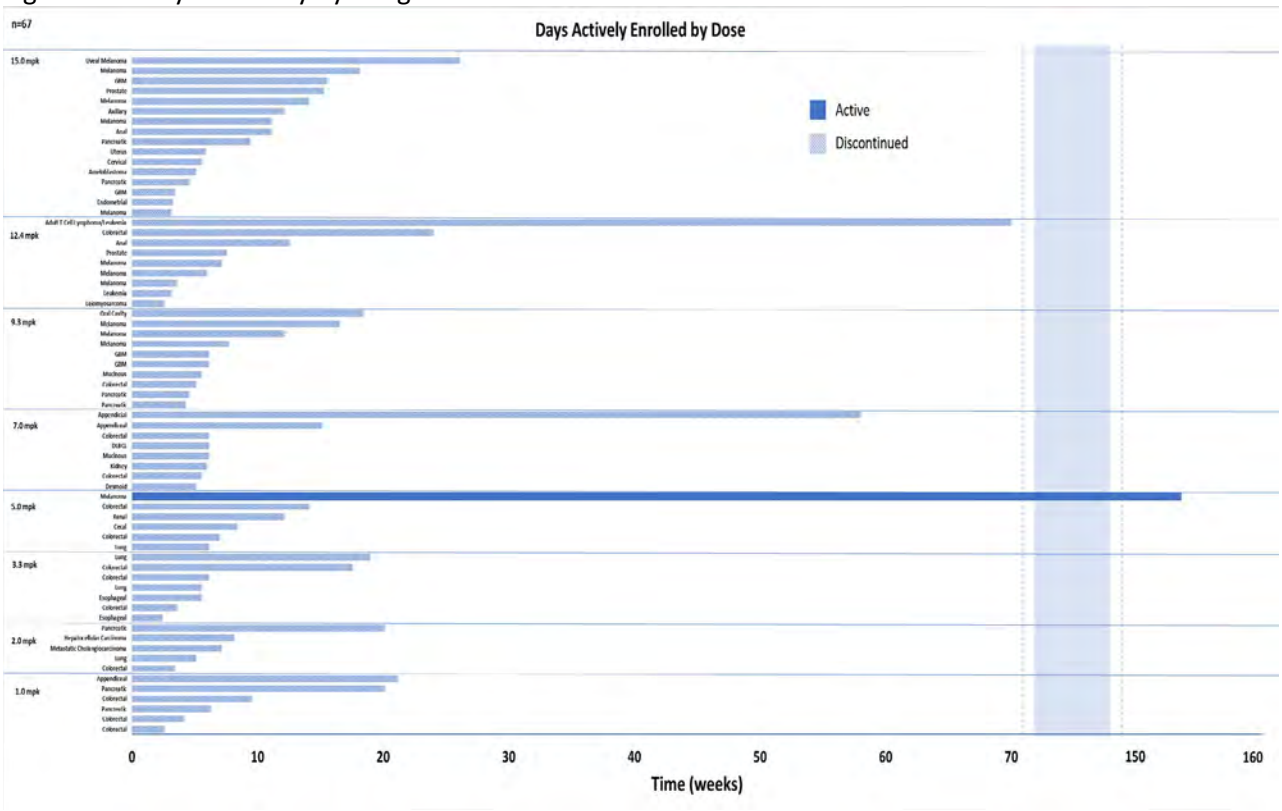


Figure S12. Overall survival by elraglusib dose level in part 1.

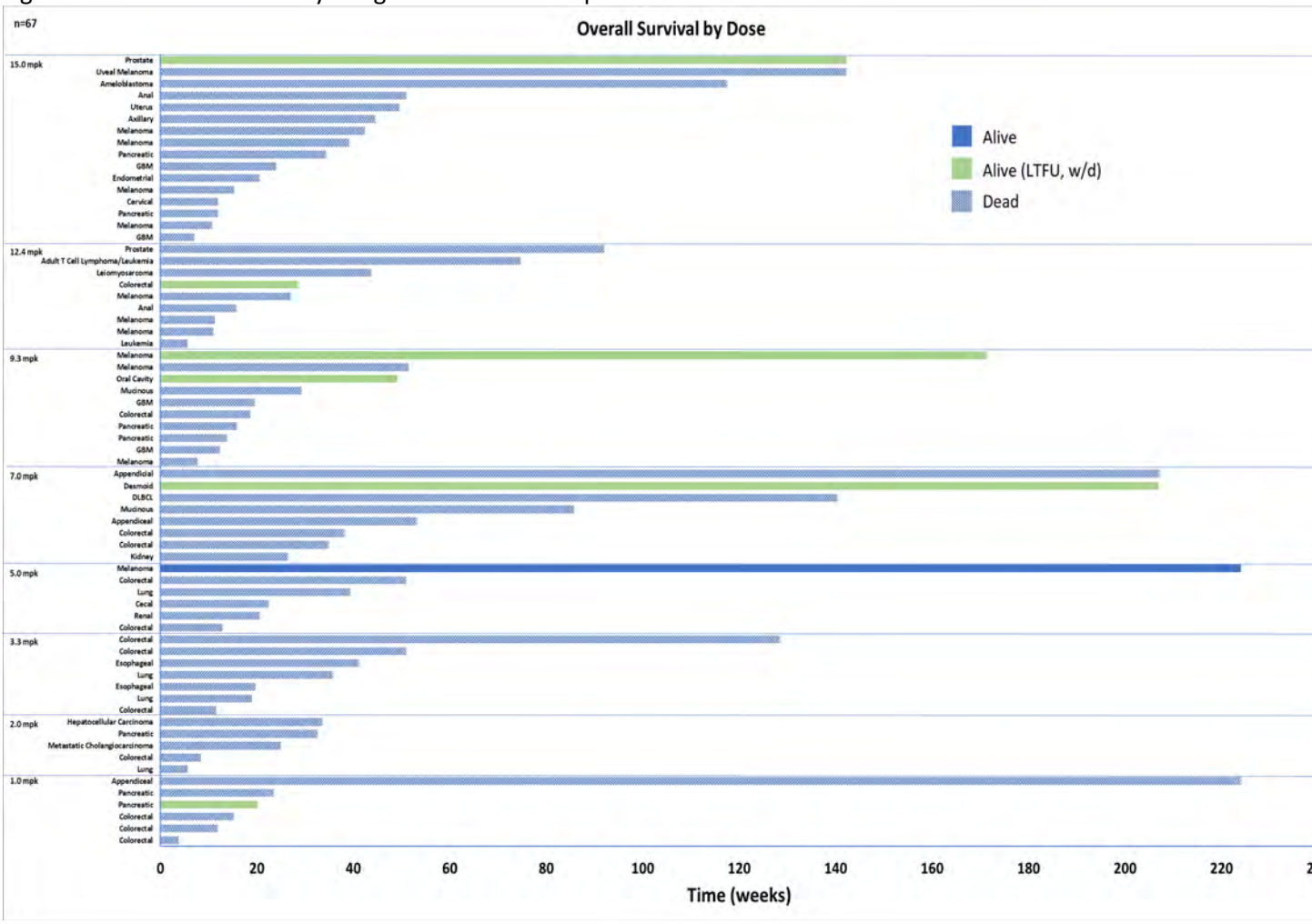
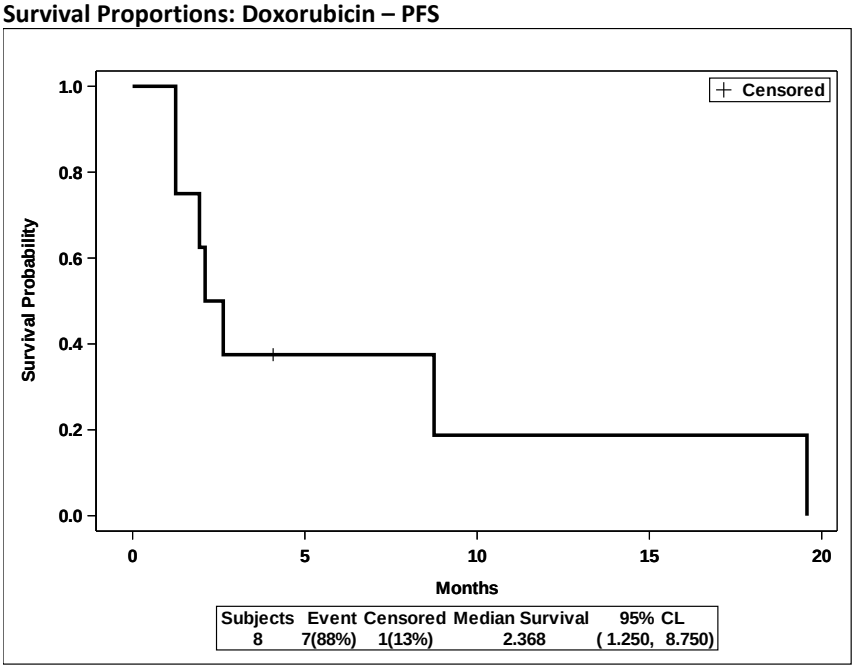


Figure S13. Median progression-free survival (PFS) and overall survival (OS) by combination treatment backbone in Part 2. Efficacy evaluable population.

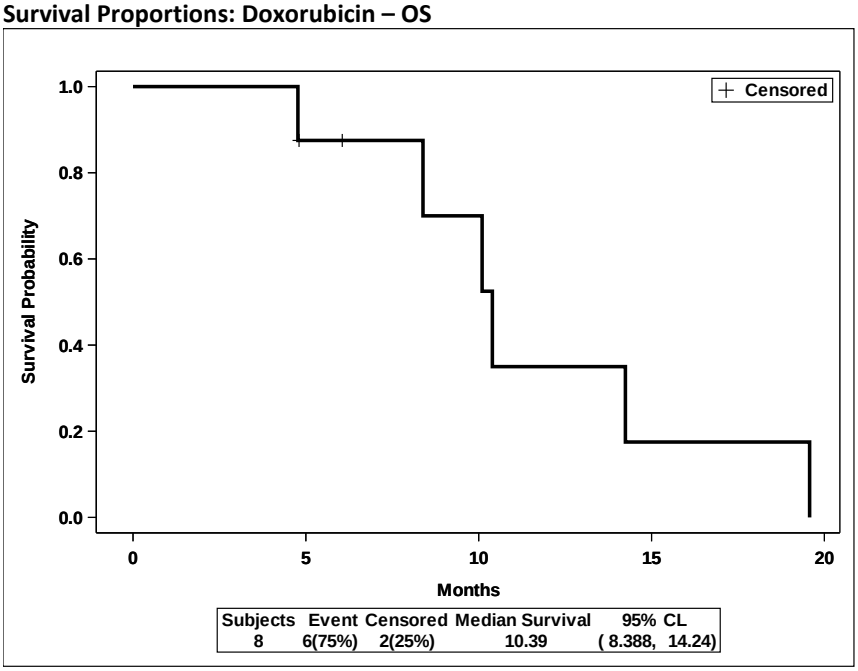
Chemotherapy backbone	mPFS (months)	mOS (months)
Doxorubicin	2.4	10.4
Irinotecan	2.1	6.9
Any carboplatin group	2.1	6.9
Gemcitabine/nab-paclitaxel	3.1	5.6
Lomustine	5.3	11.4

Abbreviations: mOS, median overall survival; mPFS, median progression-free survival

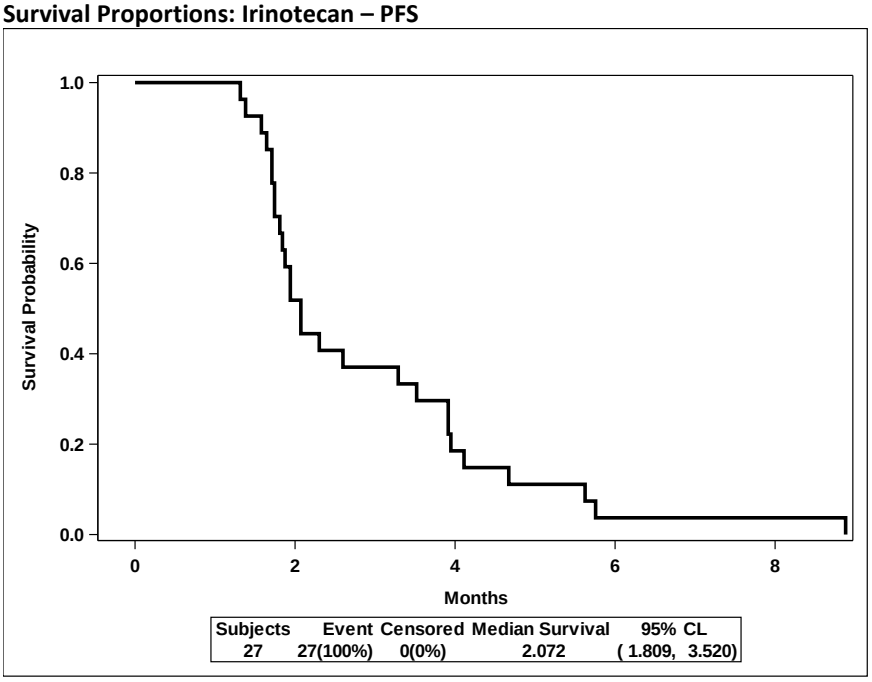
Survival Proportions: Doxorubicin – PFS



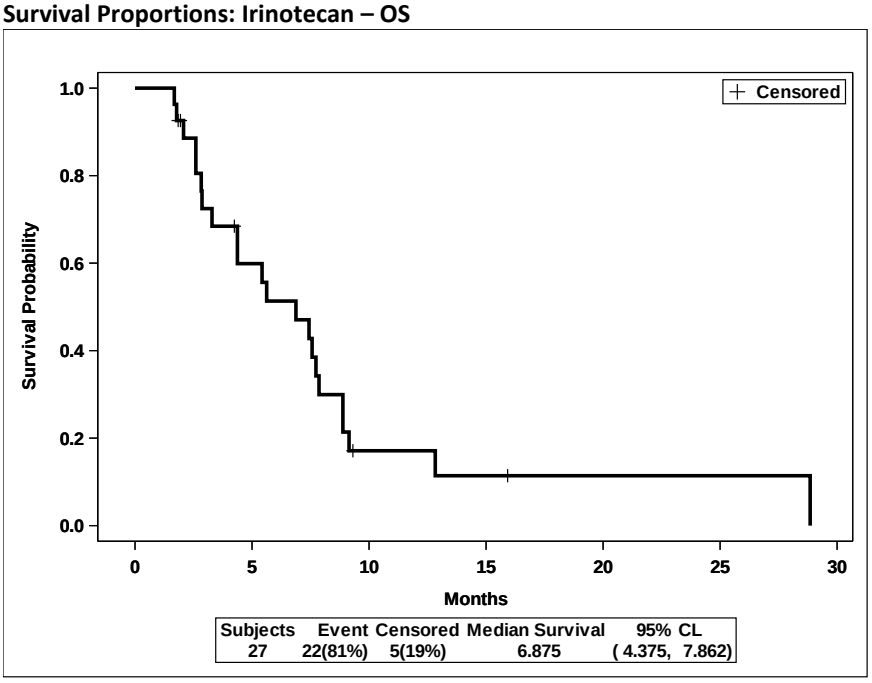
Survival Proportions: Doxorubicin – OS



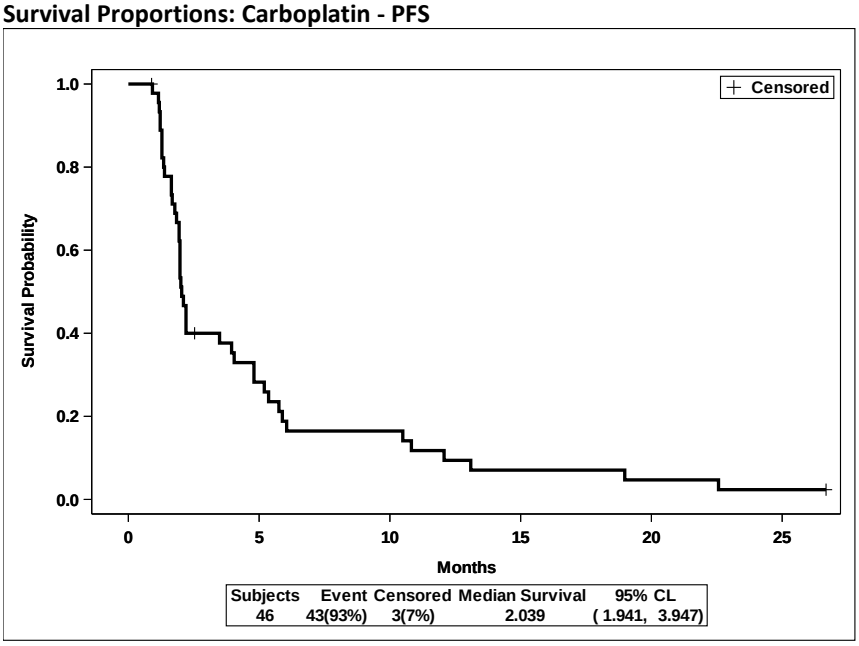
Survival Proportions: Irinotecan – PFS



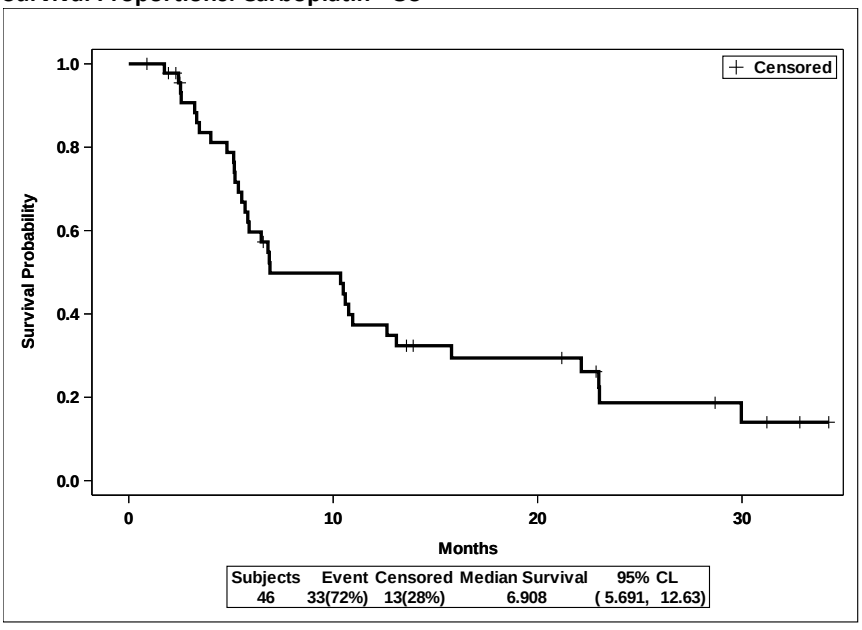
Survival Proportions: Irinotecan – OS



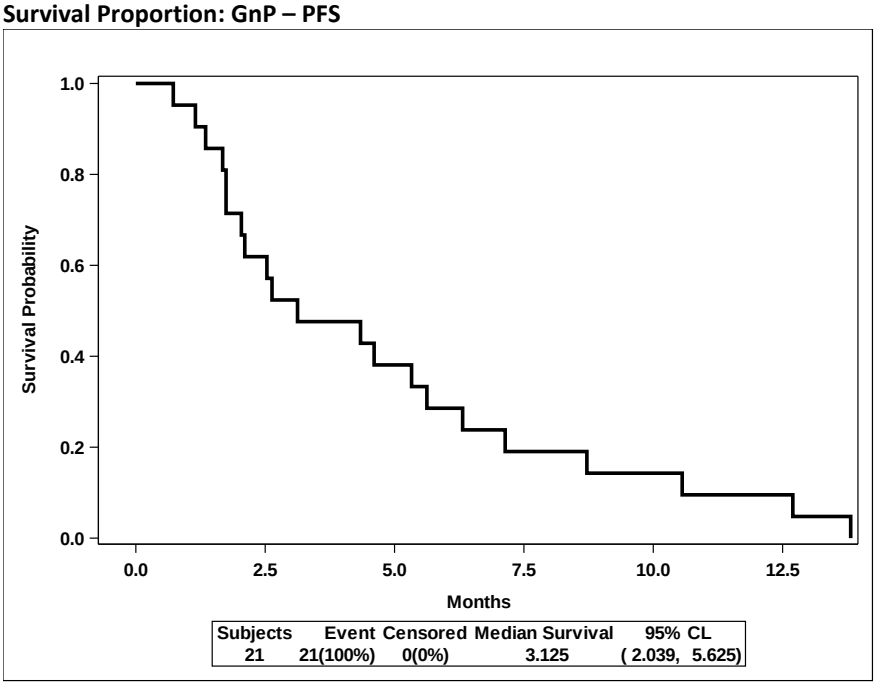
Survival Proportions: Carboplatin - PFS



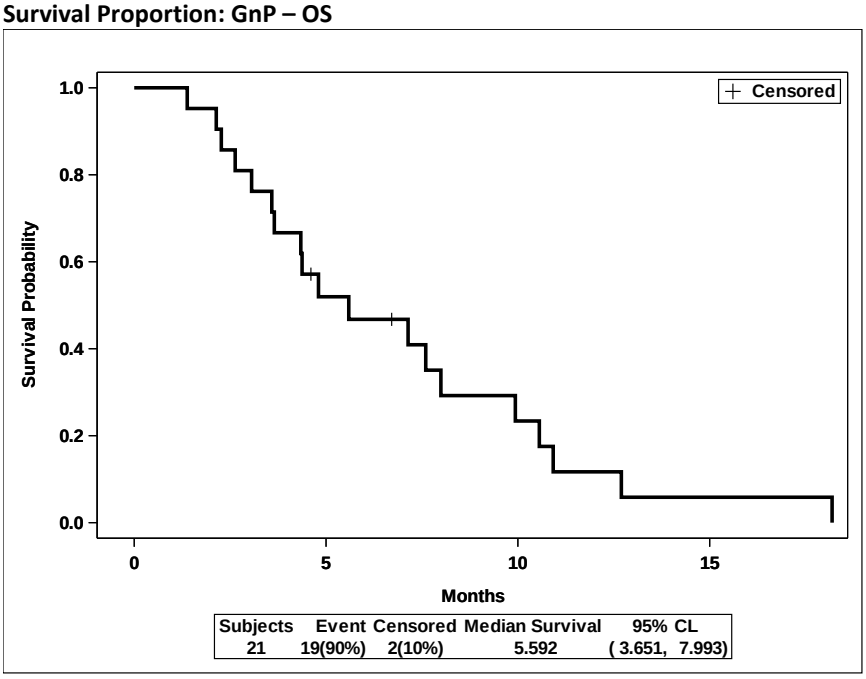
Survival Proportions: Carboplatin - OS



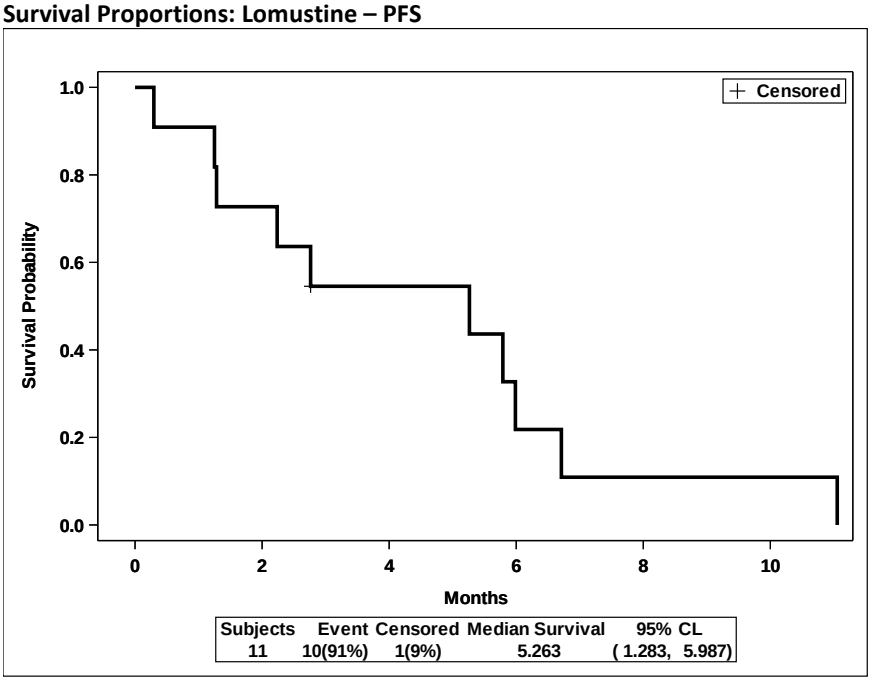
Survival Proportion: GnP – PFS



Survival Proportion: GnP – OS



Survival Proportions: Lomustine – PFS



Survival Proportions: Lomustine - OS

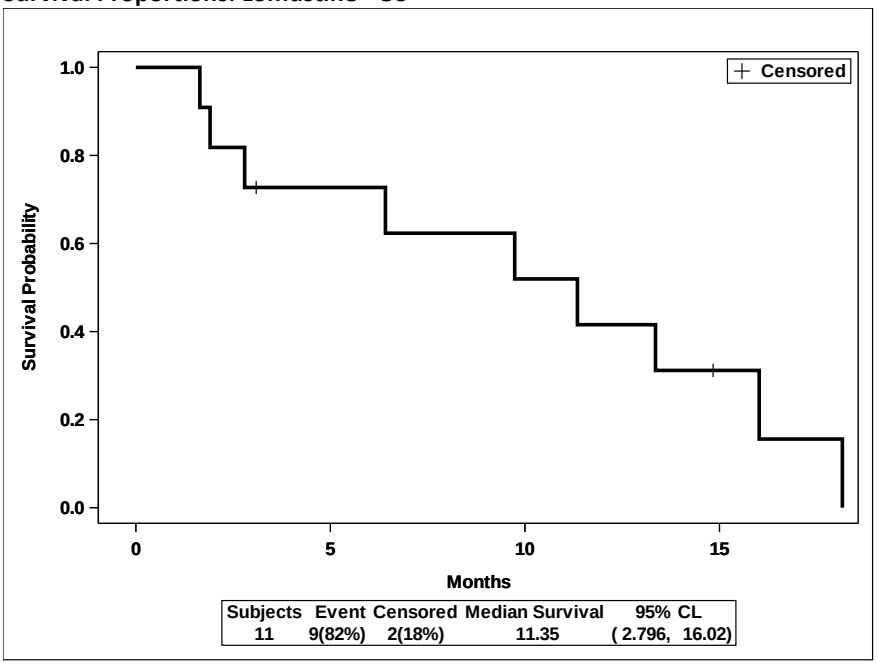
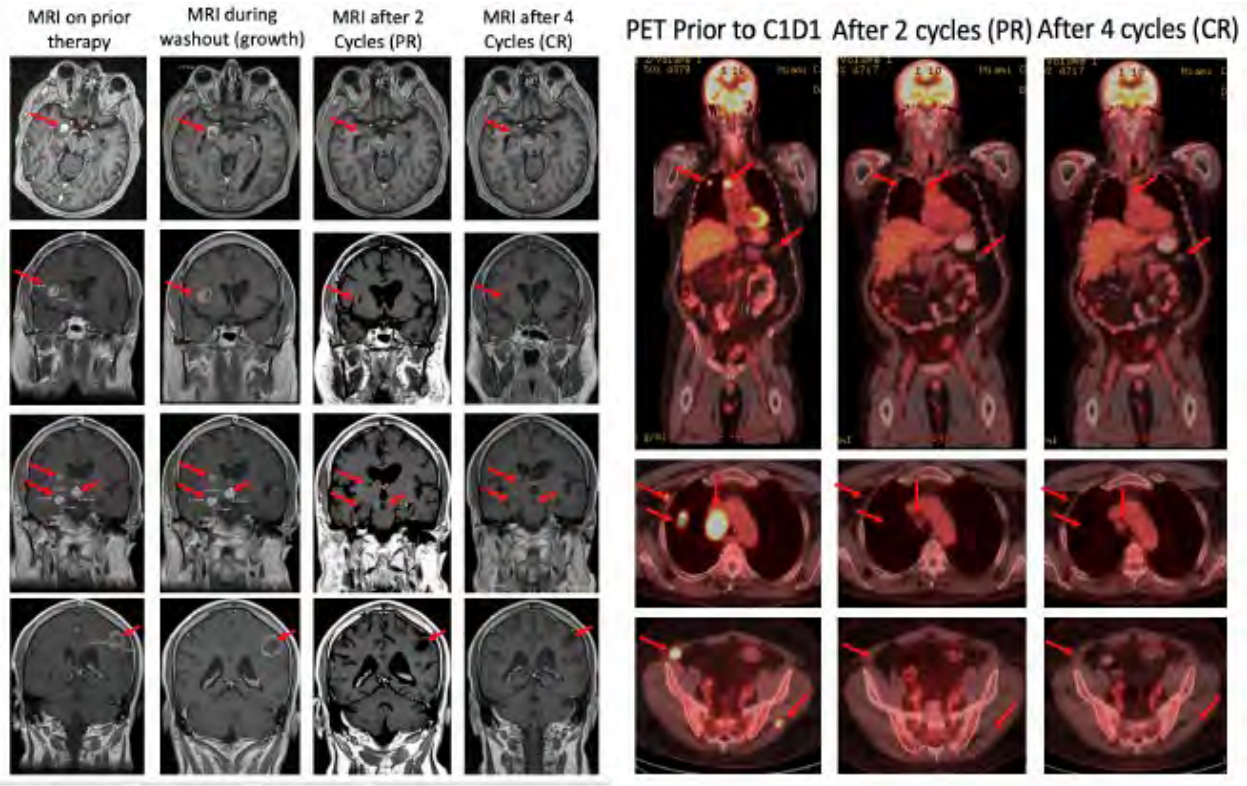


Figure S14. Complete response with elraglusib monotherapy in a patient with *BRAF*-mutated melanoma.



Abbreviations: C, cycle; CR, complete response; D, day; MRI, medical resonance imaging; PR, partial response.

Figure S15. Days on the study by cancer histology in Part 1.

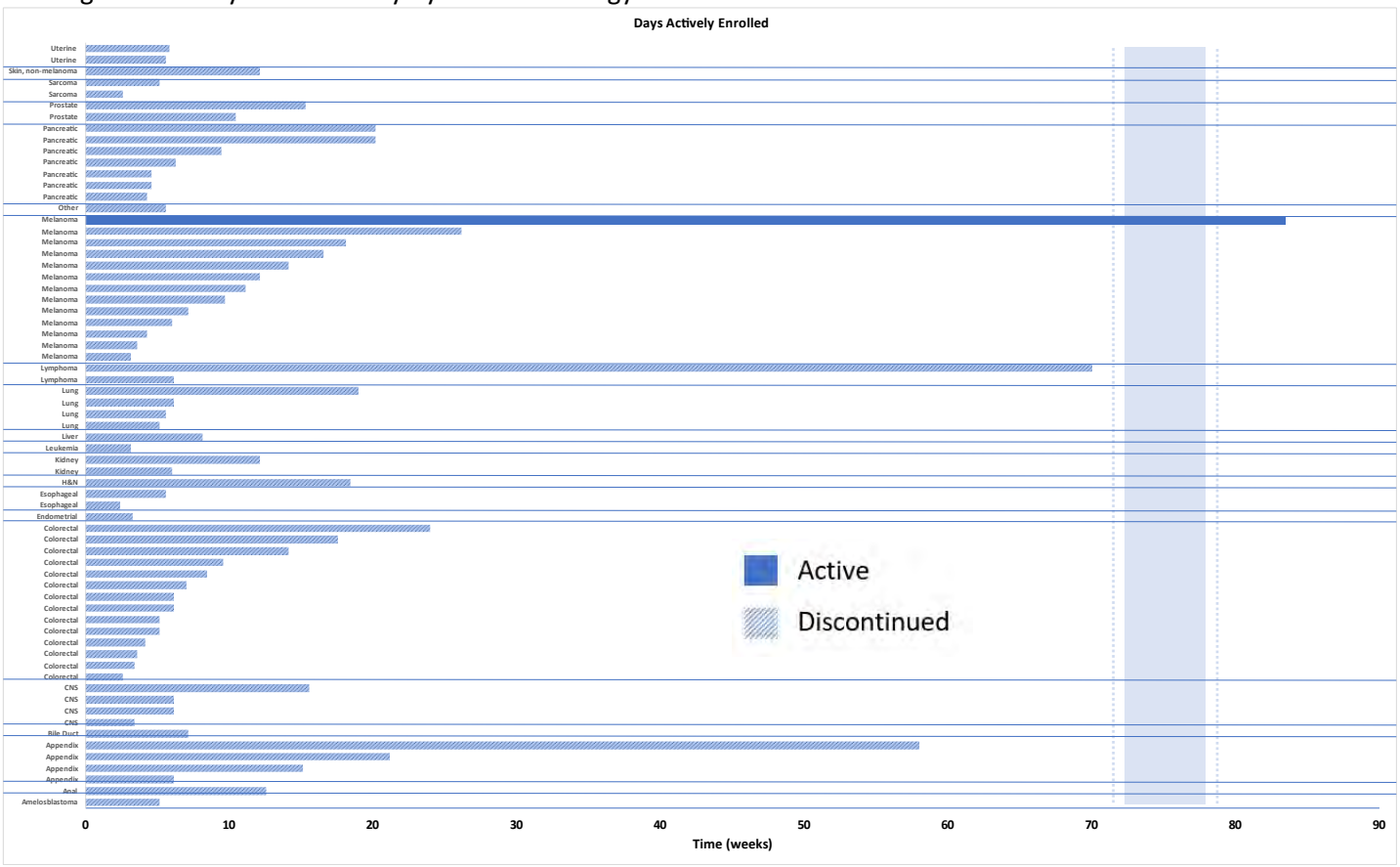


Figure S16. Overall survival by cancer histology in Part 1.

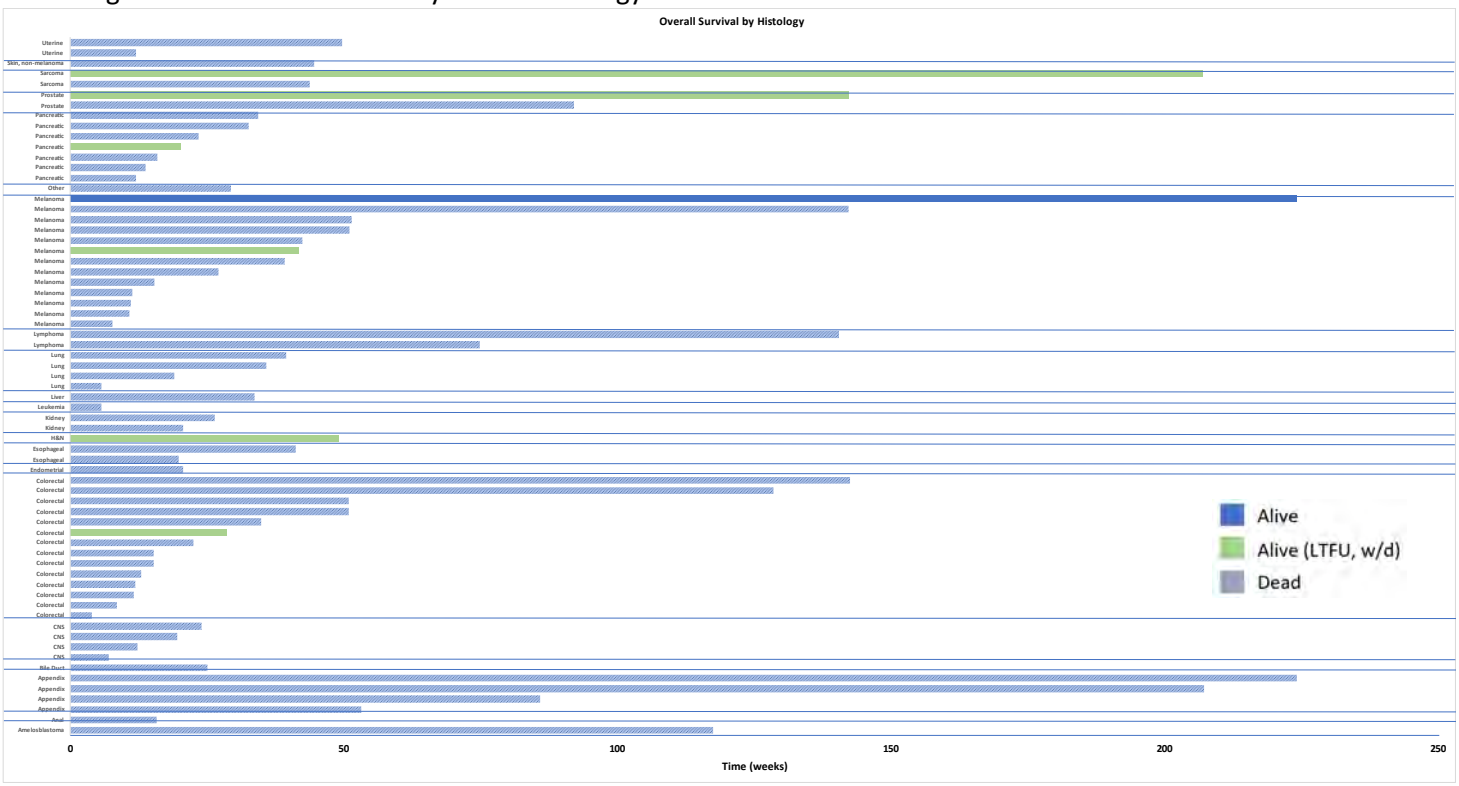


Figure S17. Best overall response by cancer histology in Part 1.

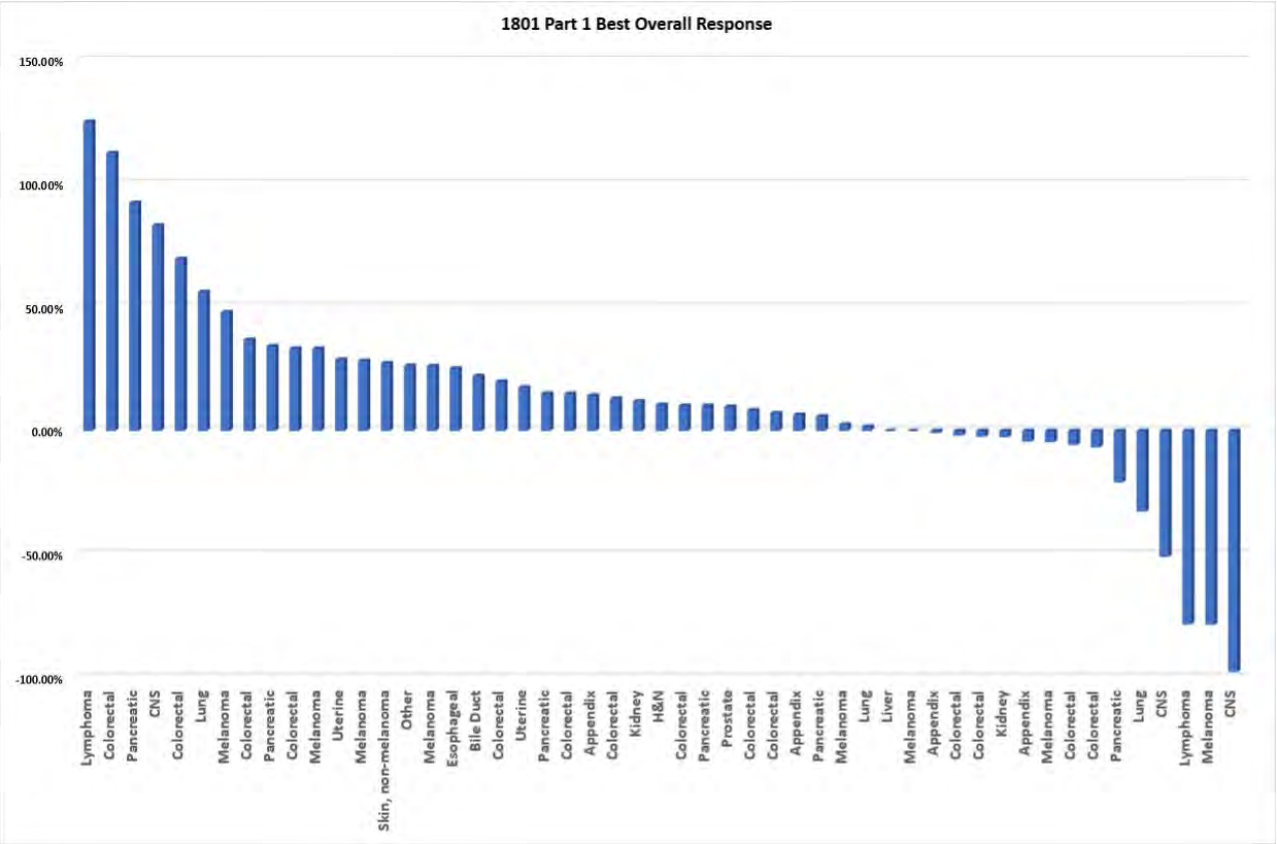


Figure S18. Cytokine responses in patients treated with elraglusib monotherapy A) C-reactive protein (CRP); B) interferons (IFN); C) interleukin (IL)-1alpha; D) IL-1beta; E) IL-2; F) IL-6; G) IL-8; H) IL-12p70; I) IL-17; J) monocyte chemoattractant protein (MCP)-1; K) macrophage inflammatory protein (MIP)-1apha; L) MIP-1beta; M) transforming growth factor(TGF)-beta; N) tumor necrosis factor (TNF)

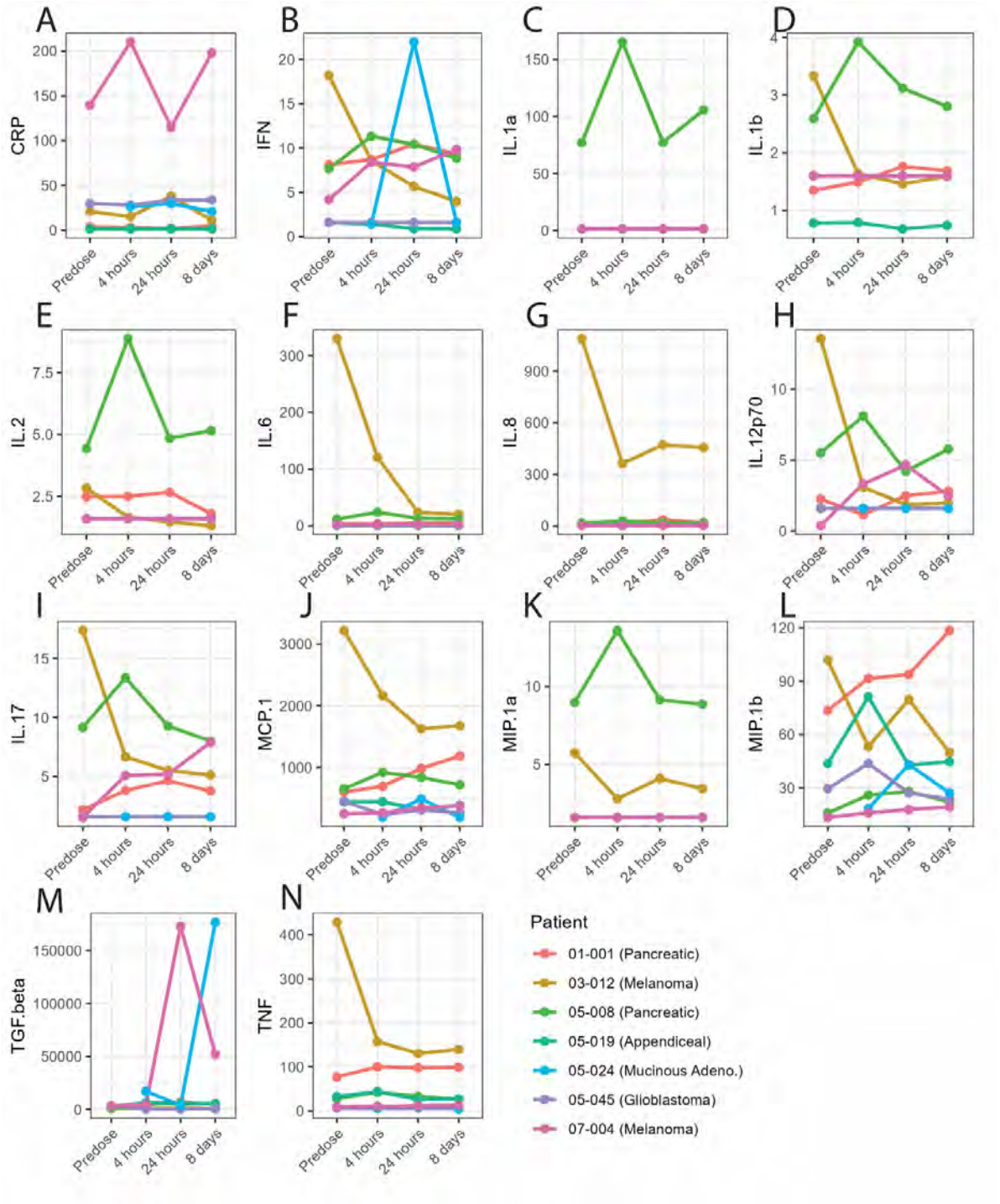


Table S1. Chemotherapy Regimens Used in Combination with Elraglusib in Part 2.

Chemotherapy Agent(s)	Regimen
Gemcitabine	Gemcitabine 1250 mg/m^2 as a 30-minute IV infusion on Days 1 and 8 of a 21-day cycle
Doxorubicin	Doxorubicin 75 mg/m^2, IV bolus on Day 1 of a 21-day cycle up to a maximum lifetime dose of 550 mg/m^2
Lomustine	Lomustine 30 mg/m^2 PO as a single dose weekly for 12 weeks
Carboplatin	Carboplatin AUC 6 IV over 1 hour on Day 1 of a 21-day cycle
Irinotecan	Irinotecan 350 mg/m^2 as a 90-minute IV infusion on Day 1 of a 21-day cycle
Nab-paclitaxel plus gemcitabine (GnP)	Nab-paclitaxel 125 mg/m^2 IV over 30-minutes immediately followed by gemcitabine 1000 mg/m^2 IV over 30-minutes on Days 1, 8 and 15 of a 28-day cycle
Paclitaxel plus carboplatin	Paclitaxel 175 mg/m^2 IV over 3 hours immediately followed by carboplatin AUC 6 IV over 1 hour on Day 1 of a 21-day cycle
Pemetrexed plus carboplatin	Pemetrexed 500 mg/m^2 IV over 10 minutes, followed 30 minutes later by carboplatin AUC 5 IV over 30 minutes, both administered on Day 1 of a 21-day cycle

Abbreviations: AUC, area under the curve; IV, intravenous; PO, by mouth.

Supplementary Table S2. Representativeness of Study Participants	
Cancer type(s)/subtype(s)/stage(s)/condition	Relapsed or refractory solid tumors or hematologic malignancies [16 cancer types in Part 1 (Table S8) and 26 different cancer types in Part 2 (Table S9)]
Considerations related to:	
Sex	Globally, an estimated 19.3 million incidences and 10 million deaths due to cancer were reported in GLOBOCAN 2020. Out of these total cases, approximately 51.4% were in men and 48.6% were in women. The most common cancers detected in men are lung (14.3%), prostate (14.1%), non-melanoma skin (7.2%), and stomach (7.1%) cancers. In females the frequently diagnosed cancers are breast (24.5%), lung (8.4%), and cervix (6.5%) cancers.(*)
Age	Worldwide, the median age of cancer onset of any cancer type (with the exception of skin cancers) is linked to the median age of the underlying population in that country and can vary by as much as a decade (e.g. breast cancer). Median age

	at onset and correlation with median age of the population in a particular country for different cancer-specific histologies are discussed in Bidoli et al. (2021) (**)
Race/ethnicity	Worldwide, diagnosis of cancer by race again correlates with the population of a particular country. In the US, cancer incidence per 100,000 is 437 for White; 335 for Hispanic; 427 for Black and 259 for Asian/Pacific Islander. However, there are large disparities in incidence of specific cancer types and deaths based on cancer types in the US. For example, the incidence of multiple myeloma is >2.2X higher and the death rate is 2X higher in Blacks than in Whites. Black men have a 1.7X higher incidence and >2X higher death rate from prostate cancer. Black women have a 1.4X higher rate of death from breast cancer. Hispanics have a 1.7X incidence and a 1.9X higher rate of death from stomach cancer than White. The Incidence and death from stomach cancer for Asians is 1.8X and 2X, respectively, compared to White (***)
Geography	This study enrolled patients in the US, Netherlands and Spain. Of these, >95% were enrolled in the US. US sites were distributed across the country and thus patients were enrolled from the Southwest, South, Midwest, Northwest, Northeast and Southeast.
Other considerations	The majority of patients enrolled in part 1 and part 2 were heavily pre-treated (Table 1) reflecting patients with advanced cancers participating in phase I trials. Patients received a median of 3 and 4 prior systemic chemotherapy regimens in the monotherapy (part 1) and combination with chemotherapy (part 2) groups. Patients were required to have had previous treatment with the same chemotherapy agents used in combination with elraglusib during part 2 of the study

	with some exceptions in the lomustine and doxorubicin arms.
	The median ages in this study were 60 (part 1) and 61 years old (part 2), and the study was nearly equally divided between male and female (55 and 45%, respectively in part 1; 48 and 52%, respectively, in part 2). Solid tumors represented the most treated cancers (n=60 [89.6%] in part 1 and n=159 [93%] in part 2). The most common cancers were colorectal cancer, melanoma, and pancreatic cancer in part 1 (Supplementary Table S8) and pancreatic, colorectal, and lung cancers in part 2 (Supplementary Table S9). In part 2, patients received elraglusib in combination with a prior chemotherapy regimen where the patient was considered not to have benefited, failed treatment, or had disease progression during treatment prior to enrolment in the 1801 study. This requirement is fairly unique to our study. Race was primarily White (88.1% in part 1 and 87.1% in part 2; of these, Hispanic comprised 19.4% of White in part 1 and 15.5% in part 2). Black or African American comprised 4.5 % of the study population in part 1 and 1.8% in part 2 and Asian comprised 1.5% and 1.2%, respectively. In general, the study population was not representative of the population demographics of patients typically treated at the sites that conducted this study or of the cancer types that comprised most of this trial. Further, this study does not represent global cancer population demographics.
Overall representativeness of this study	

*Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, Bray F. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA Cancer J Clin. 2021; 71(3):209-249. doi: 10.3322/caac.21660.

**Bidoli E, Lamaj E, Angelin T, Forgiarini O, De Santis E, Serraino D. Linearity of Age at Cancer Onset Worldwide: 25-Year Population-Based Cancer Registry Study. Cancers (Basel). 2021;13(21):5589. doi: 10.3390/cancers13215589.
***AACR Cancer Disparities Progress Report, 2022.

Table S3. Elraglusib Dose-Escalation Levels in Part 1.

Dose Level	Elraglusib Dose
1 (Starting Dose)	1.0 mg/kg
2	2.0 mg/kg (100% increase[a] from Level 1)
3	3.3 mg/kg (67% increase[a] from Level 2)
4	5.0 mg/kg (50% increase[a] from Level 3)
5	7.0 mg/kg (40% increase[a] from Level 4)
6	9.3 mg/kg (33% increase[a] from Level 5)
7	12.4 mg/kg (33% increase[a] from Level 6)
8	15 mg/kg (20% increase[a] from Level 7)

[a]% increase of preceding dose according to a modified Fibonacci sequence; intermediate doses could not be evaluated

Table S4. Elraglusib Dose-Escalation Levels in Part 2.

Dose Level	Elraglusib Dose
3	3.3 mg/kg (67% increase[a] from Level 2)
4	5.0 mg/kg (50% increase[a] from Level 3)
5	7.0 mg/kg (40% increase[a] from Level 4)
6	9.3 mg/kg (33% increase[a] from Level 5)
7	12.4 mg/kg (33% increase[a] from Level 6)
8	15 mg/kg (20% increase[a] from Level 7)

[a] % increase of preceding dose according to a modified Fibonacci sequence; intermediate doses could not be evaluated

Table S5. Pharmacokinetic Sampling Time Points in Part 1.

Treatment Cycle/Day	PK Time Points	Treatment Cycle/Day	PK Time Points
Cycle 1 Day 1	Pre-dose	Cycle 2 Day 1	
	0.25 hour (± 5 min) post-dose		
	0.5 hour (± 5 min) post-dose		Pre-dose
	1 hour (± 10 min) post-dose		1 hour (± 10 min) post-dose
	2 hours (± 10 min) post-dose		4 hours (± 30 min) post-dose
	4 hours (± 30 min) post-dose		8 hours (± 30 min) post-dose
	6 hours (± 30 min) post-dose		

Treatment Cycle/Day	PK Time Points	Treatment Cycle/Day	PK Time Points
	8 hours (± 30 min) post-dose		
Day 2	24 hours (1 ± hour) post first dose of study drug on Cycle 1 Day 1	Day 2	24 hours (1 ± hour) post first dose of study drug on Cycle 2 Day 1
Day 4	Pre-dose	Day 4	Pre-dose
Day 8	Pre-dose	Day 8	
	0.25 hour (± 5 min) post-dose		
	0.5 hour (± 5 min) post-dose		Pre-dose
	1 hour (± 10 min) post-dose		1 hour (± 10 min) post-dose
	2 hours (± 10 min) post-dose		4 hours (± 30 min) post-dose
	4 hours (± 30 min) post-dose		8 hours (± 30 min) post-dose
	6 hours (± 30 min) post-dose		
	8 hours (± 30 min) post-dose		
Day 9	24 hours (1 ± hour) post first dose of study drug on Cycle 1 Day 8	Day 9	24 hours (1 ± hour) post first dose of study drug on Cycle 2 Day 8

Abbreviations: PK, pharmacokinetics

Table S6. Elraglusib Dose Levels and Reasons for Study Discontinuation at Each Dose Level of Elraglusib in Parts 1 and 2.

Elraglusib dose level in mg/kg, n	Reason for study discontinuation, n (%)					
	Completed	Death	Lost to follow-up	Progressive disease	Withdrawal by patient	Other – hospice
Part 1						
1.0 (n=6)	1 (16.7)	3 (50.0)	0	1 (16.7)	1 (16.7)	0
2.0 (n=5)	0	3 (60.0)	0	0	2 (40.0)	0
3.3 (n=7)	1 (14.3)	6 (85.7)	0	0	0	0
5.0 (n=6[a])	0	4 (66.7)	0	0	1 (16.7)	0
7.0 (n=8)	2 (25.0)	4 (50.0)	0	0	2 (25.0)	0
9.3 (n=10)	0	8 (80.0)	2 (20.0)	0	0	0
12.4 (n=9)	1 (11.1)	6 (66.7)	0	0	2 (22.2)	0
15.0 (n=16)	2 (12.5)	10 (62.5)	2 (12.5)	0	2 (12.5)	0
Total (N=67)	**7 (10.4)**	**44 (65.7)**	**4 (6.0)**	**1 (1.5)**	**10 (14.9)**	**0**
Part 2						
3.3 (n=21)	2 (9.5)	16 (76.2)	0	1 (4.8)	2 (9.5)	0
5.0 (n=39)	2 (5.1)	30 (76.9)	2 (5.1)	0	5 (12.8)	0
7.0 (n=38)	2 (5.3)	27 (71.1)	1 (2.6)	0	8 (21.1)	0
9.3 (n=11)	1 (9.1)	7 (63.6)	0	0	2 (18.2)	0
12.4 (n=2)	2 (100.0)	0	0	0	0	0
15.0 (n=60)	2 (3.3)	39 (65.0)	4 (6.7)	0	12 (20.0)	1 (1.7)
Total (N=171)	**11 (6.4)**	**119 (69.6)**	**7 (4.1)**	**1 (0.6)**	**29 (20.0)**	**1 (0.6)**

[a]Includes one patient who continues to receive treatment.

Table S7. Reasons for Treatment Discontinuation by Concomitant Chemotherapy Regimens in Part 2.

Concomitant Chemotherapy Regimens	Reason for treatment discontinuation, n (%)						
	Progressive Disease	Adverse Event	Investigator discontinues treatment	Initiation of alternative anticancer therapy	Withdrawal by patient	Death	Other
Gemcitabine (n=36)	22 (61.1)	0	3 (8.3)	0	5 (13.9)	4 (11.1)	2 (5.6)
Doxorubicin (n=10)	6 (60.0)	0	1 (10.0)	1 (10.0)	2 (20.0)	0	0
Lomustine (n=14)	9 (64.3)	0	2 (14.3)	0	3 (21.4)	0	0
Carboplatin (n=27[a])	21 (77.8)	0	1 (3.7)	0	2 (7.4)	2 (7.4)	0
Irinotecan (n=34)	21 (61.8)	0	3 (8.8)	0	7 (20.6)	1 (2.9)	2 (5.9)
Nab-paclitaxel plus gemcitabine (n=27)	16 (59.3)	3 (11.1)	0	0	7 (25.9)	0	1 (3.7)
Paclitaxel plus carboplatin (n=17[a])	9 (52.9)	0	4 (23.5)	0	3 (17.6)	0	0
Pemetrexed plus carboplatin (n=6)	4 (66.7)	0	1 (16.7)	0	1 (16.7)	0	0
Total (N=171)	**108 (63.2)**	**3 (1.8)**	**15 (8.8)**	**1 (0.6)**	**30 (17.5)**	**7 (4.1)**	**5 (2.9)**

[a]Includes one patient who continues to receive treatment.

Table S8. Tumor Types by Elraglusib Dose Level in Part 1.

Tumor type	Number of Patients (%) by Elraglusib Dose Level (mg/kg)								
	1.0 (n=6)	2.0 (n=5)	3.3 (n=7)	5.0 (n=6[a])	7.0 (n=8)	9.3 (n=10)	12.4 (n=9)	15.0 (n=16)	Total (N=67)
Colorectal	3	1	3	3	2	1	2	0	15 (22.4%)
Melanoma	0	0	0	1	0	3	3	6	13 (19.4%)
Pancreas	2	1	0	0	0	2	0	2	7 (10.4%)
Lung	0	1	2	1	0	0	0	0	4 (6.0%)
Glioblastoma	0	0	0	0	0	2	0	2	4 (6.0%)
Appendix	1	0	0	0	2	0	0	0	3 (4.5%)
Head and Neck	0	0	0	0	0	1	0	2	3 (4.5%)
Leukemia/Lymphoma	0	0	0	0	1	0	2	0	3 (4.5%)
Cervix/Uterus/Endometrium	0	0	0	0	0	0	0	3	3 (4.5%)
Esophageal	0	0	2	0	0	0	0	0	2 (3.0%)
Liver	0	2	0	0	0	0	0	0	2 (3.0%)
Prostate	0	0	0	0	0	0	1	1	2 (3.0%)
Renal	0	0	0	1	1	0	0	0	2 (3.0%)
Unknown (mucinous)	0	0	0	0	1	1	0	0	2 (3.0%)
Desmoid tumor	0	0	0	0	1	0	0	0	1 (1.6%)
Sarcoma	0	0	0	0	0	0	1	0	1 (1.6%)

[a]Includes one patient who continues to receive treatment.

Table S9. Tumor Types by Elraglusib Dose Level in Part 2.

Tumor Type	Number of Patients (%) by Elraglusib Dose Level (mg/kg)						
	3.3 (n=21)	**5.0 (n=39)**	**7.0 (n=38)**	**9.3 (n=11)**	**12.4 (n=2)**	**15.0 (n=60)**	**Total (N=171)**
Adrenal Gland	0	1	0	0	0	0	1 (0.6%)
Anaplastic oligodendroglioma	0	1	0	0	0	0	1 (0.6%)
Astrocytoma	0	1	0	0	0	0	1 (0.6%)
Biliary Tract	0	1	1	0	0	0	2 (1.2%)
Breast	3	2	2	0	1	0	8 (4.7%)
Cervix/Uterus/Endometrium	0	1	2	1	0	3	7 (4.1%)
CNS	1	3	0	0	0	2	6 (3.5%)
Colorectal	1	3	10	0	1	9	24 (14.0%)
Endometrial	0	0	1	0	0	0	1 (0.6%)
Esophageal	1	1	1	0	0	2	5 (2.9%)
Fallopian Tube	0	0	0	0	0	1	1 (0.6%)
Gallbladder	0	0	1	0	0	1	2 (1.2%)
Glioblastoma	0	1	0	0	0	4	5 (2.9%)
Gliosarcoma	0	1	0	0	0	0	1 (0.6%)
Head and Neck	1	2	1	0	0	0	4 (2.3%)
Liposarcoma	0	0	1	0	0	0	1 (0.6%)
Liver	0	1	0	1	0	3	5 (2.9%)
Lung	1	2	4	2	0	2	11 (6.4%)
Melanoma	1	1	0	0	0	0	2 (1.2%)
Merkel Cell	1	0	0	0	0	0	1 (0.6%)
Mesothelioma	0	0	0	0	0	2	2 (1.2%)
Other	0	1	1	1	0	0	3 (1.8%)
Ovarian	2	0	1	2	0	5	10 (5.8%)
Pancreas	7	16	12	4	0	19	58 (33.9%)
Sarcoma	1	0	0	0	0	6	7 (4.1%)
Unknown	1	0	0	0	0	1	2 (1.2%)

Table S10. Treatment-Emergent Adverse Events Related to Elraglusib Versus Concomitant Chemotherapy in Part 2.

Category	Part 2 Total (N=171)	
	Related to Elraglusib	Related to Concomitant Chemotherapy
Total Number of Related TEAEs Reported	512	1199
Patients with Related TEAE	138 (80.7%)	152 (88.9%)
Patients with Serious Related TEAE	5 (2.9%)	22 (12.9%)
Patients Discontinued Due to Related TEAE	2 (1.2%)	5 (2.9%)
Patients with DLT Related to Study Drug	1 (0.6%)	1 (0.6%)
Patients with Grade 3 or 4 Related TEAE	10 (5.8%)	83 (48.5%)
Patient Deaths Related to Study Drug	0 (0.0%)	0 (0.0%)

Abbreviations: DLT, dose-limiting toxicity; TEAE, treatment-emergent adverse event