Abbreviated Title: M7824/Gemcitabine for AAPC

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Title: A Phase IB/II Single-arm Study of M7824 (MSB0011359C) in Combination with Gemcitabine in Adults with Previously Treated Advanced Adenocarcinoma of the Pancreas.

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Investigational Agent:

Drug Name:	M7824
IND Number:	136852
Sponsor:	Center for Cancer Research, National Cancer Institute
Manufacturer:	EMD Serono

Commercial Agent: Gemcitabine

PRÉCIS

Background:

- M7824 is an investigational agent in phase IB/II clinical development with dual activity against TGFβ signaling (TGFβ ligand 'trap'; extracellular domain of human TGFβ receptor II) and immune checkpoint ligand inhibition (PD-L1 inhibition; avelumab, fully human IgG1 mAb directed against human PD-L1) with an acceptable toxicity profile and early signals of anti-cancer activity including in pancreas cancer.
- Gemcitabine (2′,2′-Difluordesoxycytidine) is a standard-of-care nucleoside analogue in pancreas cancer with immunomodulatory mechanisms of actions in pancreas cancer patients.
- Preclinical studies in autochthonous and syngeneic murine models have shown that TGFβ inhibition and PD-L1 inhibition cooperate with gemcitabine to achieve reduction of tumor growth and extension of survival induce anti-tumor immunity, and reprogram the immune landscape.

Objectives:

- To determine the safety and tolerability of M7824 in combination with gemcitabine in subjects with metastatic or locally advanced pancreas cancer.
- To determine best overall response (BOR) rate according to Response Evaluation Criteria (RECIST 1.1) in advanced pancreas cancer subjects.

Eligibility:

- Histologically confirmed diagnosis of adenocarcinoma of the pancreas.
- Patients must have progressed on prior chemotherapeutic regimen.
- Concurrent treatment with non-permitted drugs and other interventions, prior therapy with any antibody / drug targeting T cell co-regulatory proteins (immune checkpoints) such as anti-PD 1, anti PD-L1, or anti-cytotoxic T lymphocyte antigen-4 (CTLA 4 antibody) is not allowed.

Design:

- The proposed study is a phase IB/II study of M7824 in combination with gemcitabine in a safety run-in of 6-18 patients and, if safe and tolerated, will proceed to an expansion phase II cohort with a standard Simon 'Minimax design'.

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1 INTRODUCTION

1.1 Study Objectives:

1.1.1 Primary Objectives:

Phase IB

- To determine the safety and tolerability of M7824 in combination with gemcitabine in subjects with metastatic or locally advanced pancreas cancer.

Phase II

- To determine the best overall response (BOR) rate according to Response Evaluation Criteria (RECIST 1.1) in advanced pancreas cancer subjects.

1.1.2 Secondary Objectives:

- To assess progression-free survival (PFS) according to RECIST 1.1
- To characterize overall survival (OS)
- To assess the immune-related BOR (irBOR) using the immune-related RECIST (irRECIST)

1.1.3 Exploratory Objectives:

- Determine the impact of M7824 on tumor perfusion by Dynamic Contrast Enhanced MRI (DCE-MRI)
- To evaluate impact of genomic profile of advanced pancreatic cancers on clinical response to M7824
- To correlate circulating free tumor DNA (cftDNA) levels with clinical course of pancreatic cancer patients treated with M7824 in combination with gemcitabine
- To evaluate the intratumoral immunogenicity of M7824 in combination with gemcitabine
- To characterize plasma Cmax and Cmin of M7824 and gemcitabine when given in combination
- To measure subjective Health-related Quality of Life (HRQoL) affecting disease-specific symptoms and treatment-related concerns.

1.2 Background and Rationale

1.2.1 Pancreas cancer

Pancreas cancer is a life-threatening disease. AACR estimates deaths due to pancreas cancer to rank 2nd among all cancer-related deaths in the U.S. by 2030 [1]. Pancreas cancer is in ≥85% of cases diagnosed in the later stages of its progression (stage III and IV), resulting in poor prognosis with a median survival of 6 to 9 months from symptom onset and less than 6 months from diagnosis (**Figure 1**).[2]

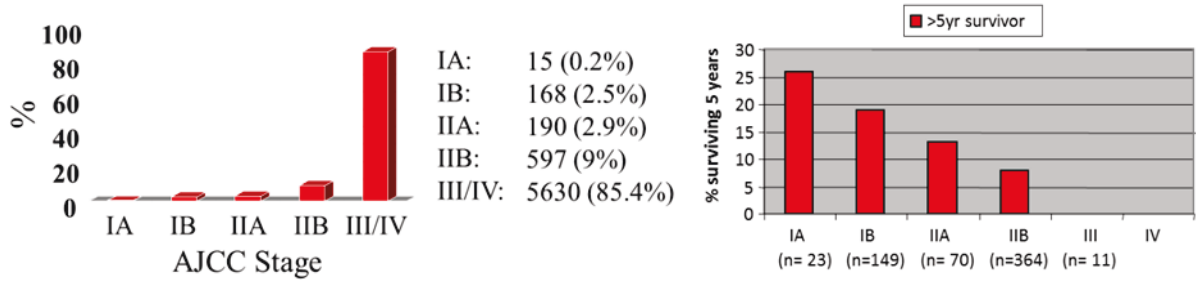


Figure 1 Percent patients at different AJCC stages at time of initial diagnosis of pancreas cancer (per 7th edition of American Joint Committee on Cancer, AJCC; left) of more than 6,000 pancreas cancer patients presenting between 2000 and 2005 to Memorial Sloan-Kettering Cancer Center, NY, and their corresponding 5-year survival rates (right) from MSKCC Pancreatic Cancer Registry.

There is no standardized second-line therapy in pancreas cancer. FDA has approved 5-FU-based (FOLFIRINOX) and gemcitabine-based (gemcitabine + abraxane [*nab*-paclitaxel]) regimens for 1st line therapy, but there is no evidence of clinical efficacy of the currently empirically chosen 2nd line regimens in randomized studies.[3, 4] FOLFIRINOX (a toxic triplet combination of oxaliplatin, irinotecan, and fluorouracil) extends median overall survival in patients with locally advanced or metastatic pancreatic cancer to merely 11.1 compared to 6.8 months with gemcitabine alone.[3] A similar marginal improvement in median overall survival from 6.7 months in the gemcitabine group to 8.5 months in the nab-paclitaxel in combination with gemcitabine group led to FDA approval of this combination in 2013.[4] New therapies are urgently needed.[5]

1.2.2 M7824, a bifunctional fusion protein targeting TGFβ signaling and the Programmed death-ligand 1 (PD-L1)

M7824 is a novel bifunctional fusion protein. It includes an antibody component (fully human IgG1 mAb) directed against human PD-L1 which is identical to avelumab, a PD-L1 immune checkpoint inhibitor recently approved for metastatic Merkel cell carcinoma and in clinical testing, either as monotherapy or in combination with standard of care or radiation therapy, for several solid organ malignancies. The second component of M7824 is a TGFβ-neutralizing trap component consisting of the extracellular domain of human TGFβ receptor II which efficiently binds TGFβ ligands TGFβ1, β2 and β3 which is fused to CH3-C terminus of the IgG via a flexible glycine-serine linker (**Figure 2**).

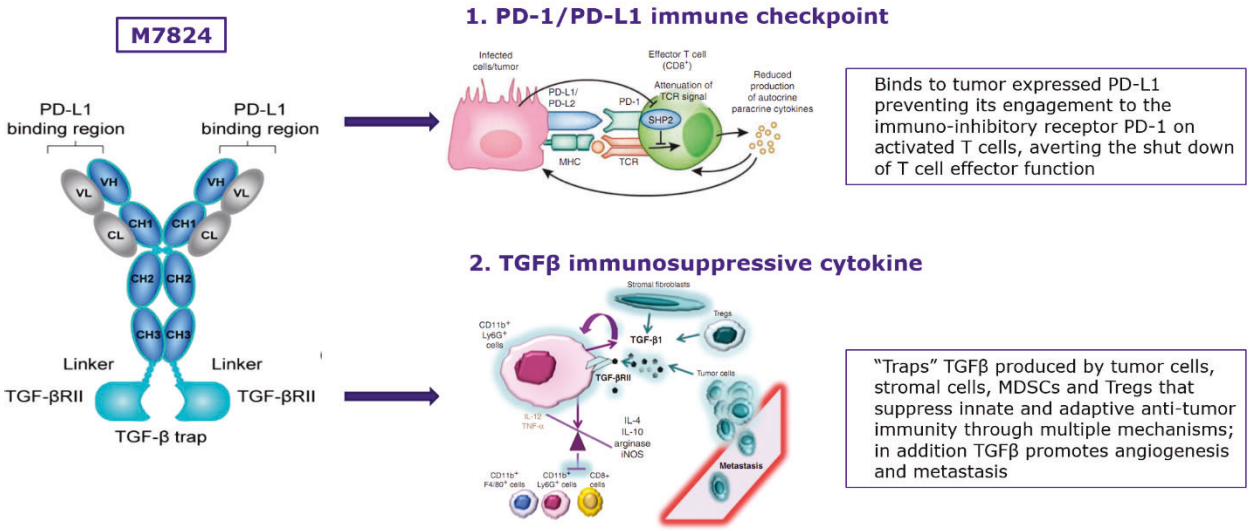


Figure 2 Anti-PD-L1/TGFβ Trap (M7824): Overcoming two major immunosuppressive mechanisms.

Dual targeting of the PD-L1 and TGFβ pathways represents a rational therapeutic strategy because these 2 key immune evasion pathways have independent and complementary immunosuppressive functions, thereby raising the possibility of synergistic antitumor activity.

1.2.3 Preclinical rational for the combination immunotherapy approach of dual TGFβ and PD-L1 inhibition and gemcitabine in pancreas cancer

Combination immunotherapy approaches are a promising new therapeutic strategy in pancreas and are currently under active investigation. In pancreatic cancer, one of the most clinically advanced combination immunotherapy strategies is combination vaccines in combination with metronomic cisplatin, given as an immunomodulator (as gemcitabine in combination with M7824 in this study) (Le DT). In a prespecified per-protocol analysis of patients who received at least three doses (two doses of Cy/GVAX plus one of CRS-207 or three of Cy/GVAX), overall survival was extended to 9.7 versus 4.6 months (arm A v B; HR, 0.53; P = .02) an effect similar to that of first line FOLFIRINOX combination therapy versus gemcitabine. Enhanced mesothelin-specific CD8 T-cell responses were associated with longer OS, regardless of treatment arm. Preclinically, a large number of promising combination immunotherapies have been identified which are awaiting clinical testing [6].

Targeting TGFβ signaling as part of combination immunotherapy is hereby particularly attractive as TGFβ ligands TGFβ1, 2, and 3 are (1) one of the most abundant cytokines in the tumor microenvironment and (2) exert immune suppressive function on a variety of immune effectors. Thus, TGFβ inhibition possibly affords reprogramming towards an anti-tumor environment on a number of different cellular levels not requiring the combination of several agents directed against individual immune cell populations (**Figure 3**).

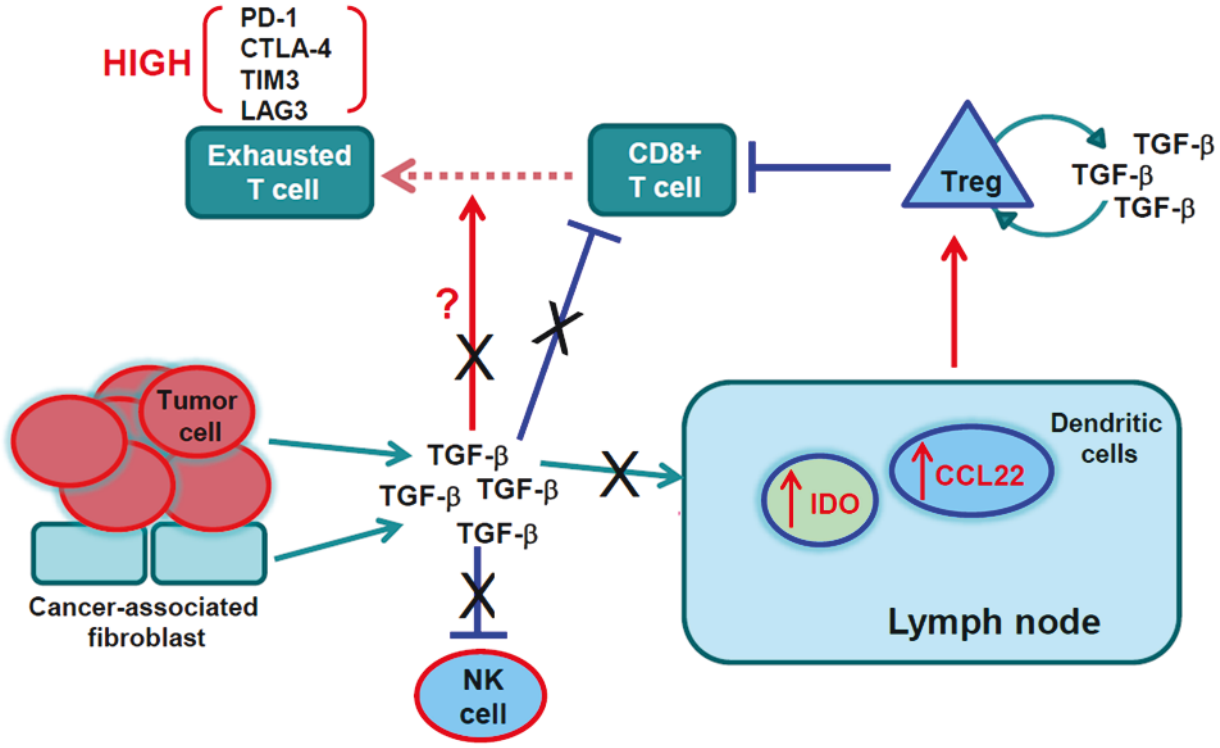


Figure 3 Pleiotropic effects of TGFβ signaling generates an immunotolerant tumor environment. Block of suppression of T cells and NK cells, block of activation of regulatory T cells (Treg) and exhaustion of effector T cells through TGFβ inhibition have therapeutic potential in novel immunotherapy strategies. (adapted from [7]).

Many types of cells in the tumor microenvironment produce TGFβ, including the tumor cells themselves, immature myeloid cells, regulatory T cells, and stromal fibroblasts; these cells collectively generate a large reservoir of TGFβ in the extracellular matrix. TGFβ signaling contributes to tumor progression by promoting metastasis, stimulating angiogenesis, and suppressing innate and adaptive antitumor immunity [8] As a broadly immunosuppressive factor, TGFβ directly down regulates the effector function of activated cytotoxic T cells and natural killer (NK) cells and potently induces the differentiation of naïve CD4+ T cells to the immunosuppressive regulatory T cells (Treg) phenotype.[9] In addition, TGFβ polarizes macrophages and neutrophils to a wound-healing phenotype (alternative activation of macrophages; M2 phenotype) that is associated with production of immunosuppressive cytokines [10] As a therapeutic strategy, neutralization of TGFβ activity has the potential to control tumor growth by restoring effective antitumor immunity, blocking metastasis, and inhibiting angiogenesis.

On the other hand, TGFβ has growth inhibitory effects on normal epithelial cells, functioning as a regulator of epithelial cell homeostasis, and it acts as a tumor suppressor during early carcinogenesis. As tumors progress toward malignancy, the growth inhibitory effects of TGFβ on the tumor are lost via mutation in one or more of the TGFβ pathway signaling components or through oncogenic reprogramming [6] Upon loss of sensitivity to TGFβ inhibition, the tumor continues to produce high levels of TGFβ, which then serve to promote tumor growth [6] The

TGFβ cytokine is overexpressed in various cancer types, including pancreas cancer, with correlation to tumor stage.[6, 7] High levels of TGFβ1 have also been found in autochthonous models of pancreas cancer.[7] A high level of TGF-β was found in serum of patients with pancreatic adenocarcinoma suggesting that TGF-β could possibly become a marker for monitoring disease activity.[11]

The programmed death 1 (PD-1) / PD-L1 axis is an important mechanism for tumor immune evasion.[12] Effector T cells chronically sensing antigen take on an exhausted phenotype are marked by PD-1 expression, and engage with tumor cells overexpressing PD-L1. Blocking the axis restores the effector function in these T cells. Additionally, in the tumor microenvironment, myeloid cells, macrophages, parenchymal cells, and T cells upregulate PD-L1. To date, single agent immune checkpoint inhibition in patients with advanced pancreatic cancers did not show objective response rates, indicating that a dual inhibition approach with PD-L1 inhibition in combination with gemcitabine, or PD-L1 inhibition with TGFβ neutralization, or – as suggested in this protocol with both might be necessary to overcome immune evasive cues of the pancreas cancer stroma and an attractive option to be investigated. [13]

1.2.4 Preclinical rationale for the combination of gemcitabine and PD-L1 inhibition in pancreas cancer

Immunologic studies in human pancreas cancer subjects have shown that gemcitabine has, in addition to its cytotoxic mechanism of action, weak immunomodulatory function by reducing peripheral MDSCs, T-regs, and TGFβ levels in pancreas cancer patients without affecting proliferative capacity of cytotoxic CD8+ T cells (**Figure 4**). When tested in the autochthonous Ras-driven Pdx-1-Cre; LSL-Kras$^{G12D/+}$; Ink4a(p16)/Arf(p19)$^{flox/flox}$ (KP16) model, gemcitabine given at sub-equivalent human doses increased intratumoral CD8+ T cells as well anti-tumor activity of CD8+ T when co-cultured with KP16 cancer cells (**Figure 4**). When combined with human equivalent doses of anti-PD-L1, gemcitabine in combination with PD-L1 blockade, as suggested in the proposed study (anti-PD-L1 component avelumab of M7828), extended survival compared to PD-L1 inhibition or gemcitabine alone.

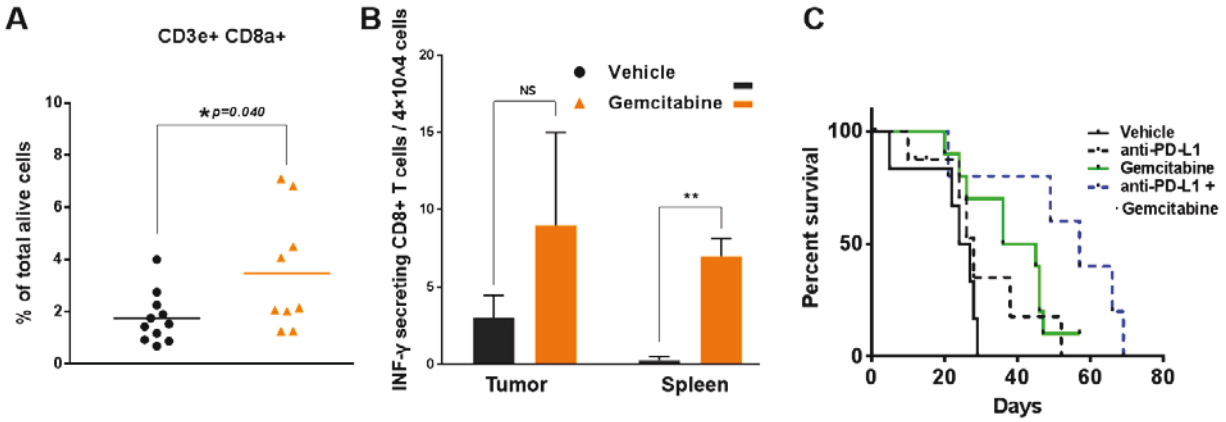


Figure 4 A Gemcitabine moderately increases infiltration of CD8+ T cells in murine pancreatic tumors and enhances in vitro anti-tumor responses of isolated CD8+ T cells. Flow cytometry profiles of KP16 tumors treated with vehicle (black) or 40mg/kg twice weekly gemcitabine (orange; animals N≥8) for 7 days. CD3+ CD8+ fraction gated on total alive cells. B Interferon gamma release measured by ELISpot assay of co-cultured CD8+ T cells isolated from tumors (left) and spleen (right) from

vehicle and gemcitabine-treated (40mg/kg twice weekly, animals harvested after 1 week). C Kaplan-Meier analysis of KP16 mice with confirmed ≥3mm pancreatic tumors randomized to vehicle, anti-PD-L1, 40mg/kg gemcitabine twice weekly, or anti-PD-L1 in combination with gemcitabine.

1.2.5 Preclinical rationale for the combination of gemcitabine and TGFβ inhibition

1.2.5.1 Preclinical rationale using the TGFβ receptor I small molecule inhibitor LY364947 in combination with gemcitabine

Due to the inherent immunogenicity of M7824 as a humanized antibody and extracellular human TGFβ receptor II component when administered to the fully immune competent autochthonous KP16 mice, initial studies with the TGFβ inhibitor LY364947 have been carried out to support the use of the dual TGFβ and PD-L1 inhibitor M7824 in combination with gemcitabine. Administration of M7824 to fully immune competent mice sensitizes the animals after 3 or 4 doses causing significant cytokine release and immune changes which carry the risk of false negative and positive readouts of studies in rodents with M7824.

LY364947, like M7824, acts as a TGFβ signaling inhibiting agent. LY364947 suppresses downstream TGFβ receptor signaling through binding to the TGFβ receptor I suppress phosphorylation and dimerization with TGFβ receptor II, SMAD activation and ultimately TGFβ-mediated gene transcription. M7824 shows similar end effector function like suppression of SMAD activation and TGFβ-mediated gene transcription, however its upstream mechanism is via trapping of free, unbound TGFβ ligands TGFβ1L, TGFβ2L, and TGFβ3L. We like to stipulate that with regard to effectiveness of TGFβ-mediated signal transduction, M7824 is likely to be the more efficacious agent due to

(1) More persistent and maintained elimination of trapped TGFβ ligands (as suggested by human PK/PD studies)

(2) Suppression of TGFβ escape signaling pathways including BMP and ALK receptor signaling. TGFβ ligands have been shown to stimulate receptor I's of ALK and BMP receptors which are part of the TGFβ superfamily. These pro-survival receptors are not inhibited by the TGFβ receptor I small molecule inhibitor LY364947 compared to M7824 implying more efficacious suppression of promiscuous TGFβ effects

Using the autochthonous Ras-driven Pdx-1-Cre; LSL-$Kras^{G12D/+}$; $Ink4a(p16)/Arf(p19)^{flox/flox}$ (KP16) model, TGFβ inhibition in combination with gemcitabine cooperatively

(1) Suppresses tumor growth and extends survival (**Figure 5**)

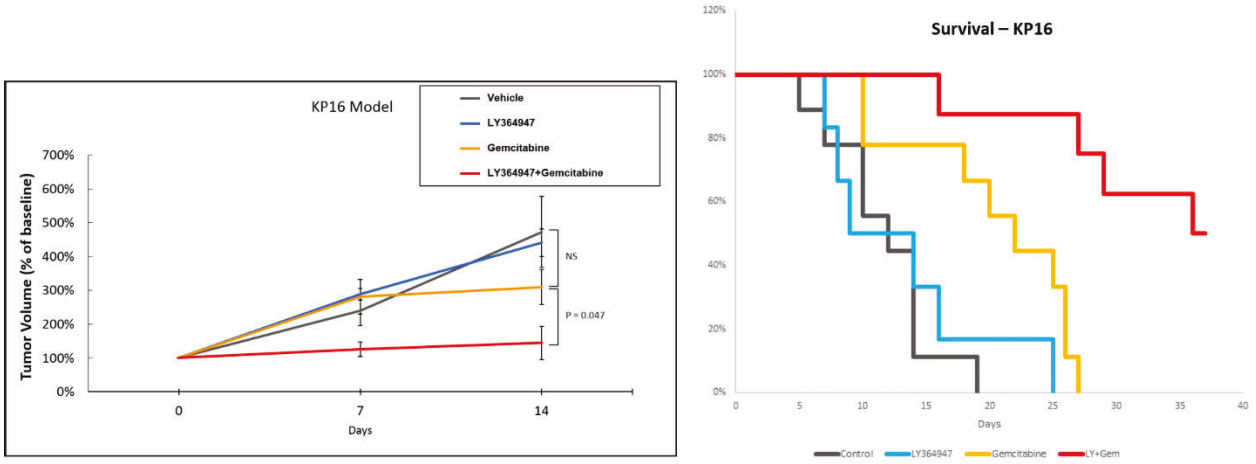


Figure 5 Left; suppression of tumor growth of pancreatic tumors after 14 days of treatment with vehicle (black), TGFβ receptor I inhibitor LY364947 (blue), gemcitabine (orange), or the combination of LY364947 and gemcitabine (in red). Volume analysis of tumor growth by 3D abdominal ultrasound for animals receiving indicated treatments (N=8). Right; Kaplan-Meier analysis of KP16 animals treated vehicle, LY364947, gemcitabine, or the combination of LY364947 and gemcitabine (p<0.001; log-rank; 2-tailed)

(2) Reduces immune evasive cues in the tumor microenvironment (**Figure 6**)

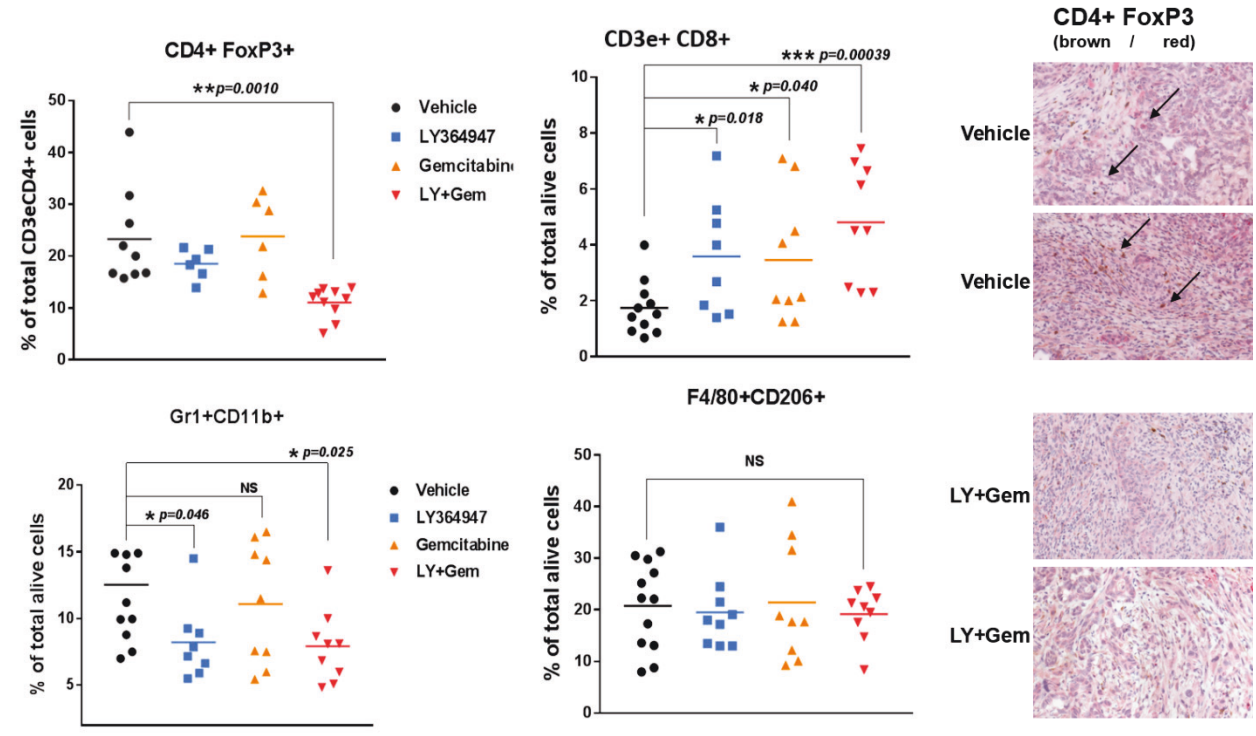


Figure 6 Flow cytometry analysis of immune cell populations from tumors in KP16 animals treated with vehicle (black), LY364947 (blue), gemcitabine (orange), or the combination of LY364947 and

gemcitabine (red) for 7 days prior to harvest and tumor digest. TβR-I blockade in combination with gemcitabine reduces CD4+FoxP3+ T regulatory cells (23.2% vs 11.0%; p=0.0010), increased intratumoral CD3e CD8a+ T cells (1.74% vs 4.81%; p=0.00039), reduced CD11b+Gr-1+ MDSCs (12.53% vs 7.91%; p=0.025) in murine pancreatic cancers with no impact on CD206+F4/80+Gr1- tumor associated macrophages (17.53% vs 16.92%; p=0.85), compared to vehicle-treated tumors in the examined cancers (t test; 2-tailed; * p<0.05; ** p<0.01). Right, immunohistochemically co-stain of anti-CD4 (brown) and FoxP3 (red), arrows indicate lymphocytes with positive anit-CD4 and nuclear FoxP3 stain.

(3) Increases in vitro anti-tumor immunity (**Figure 7**)

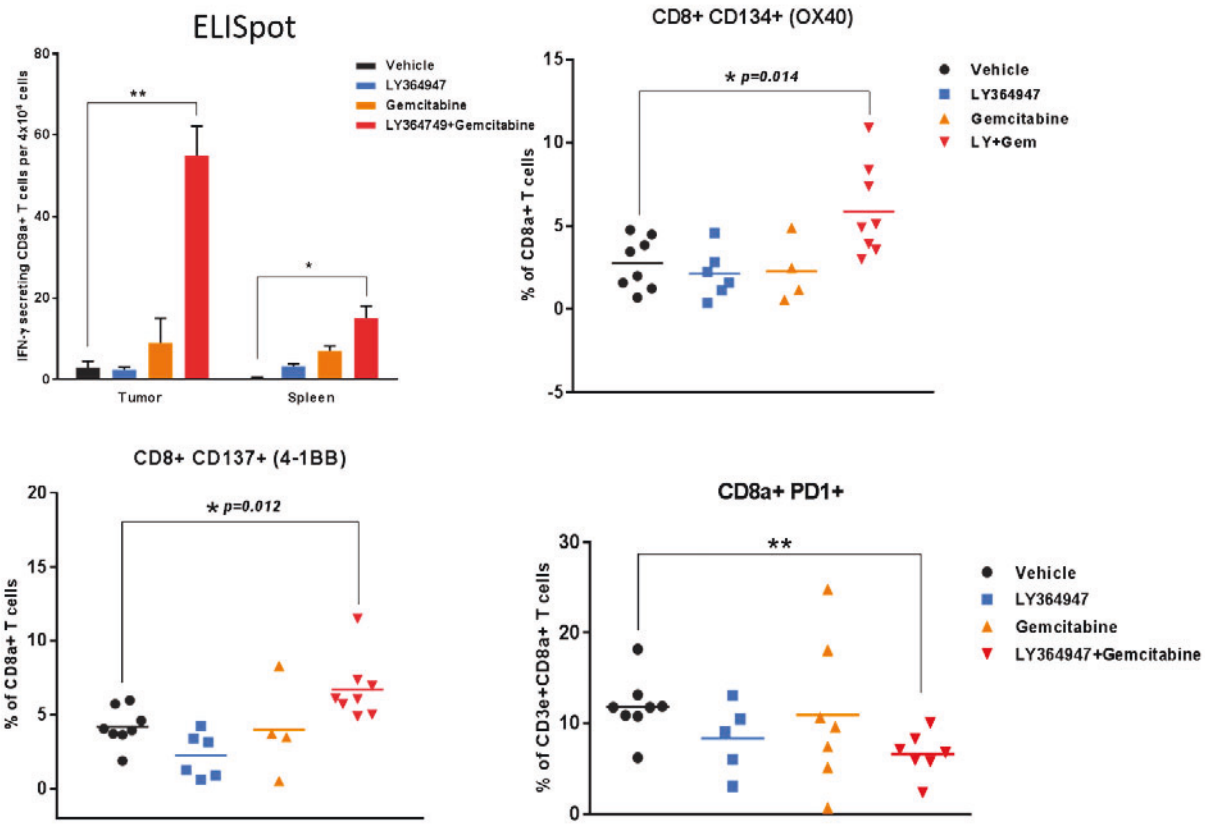


Figure 7 TGFβ blockade in combination with gemcitabine increases anti-tumor cellular immunity in murine pancreatic cancers. Left top, Induction of IFN-γ secretion by isolated intratumoral CD8a+ T cells from KP16 tumors treated with vehicle (black), LY364947 (blue), gemcitabine (orange), or the combination of LY364947 and gemcitabine (in red) following 20 hours co-culture by KP16 pancreatic cells and measured by ELISpot assay (N≥4), stimulation of splenic CD8a+ T cells shown on the right. Increased expression of T cell activation markers 4-1BB (CD137) (2.7% vs 5.9%; p=0.012) and OX40 (CD134) (4.2% vs 6.7%; p=0.014), and decreased PD-1 expression levels on CD8a+ (11.9% vs 6.6%; p=0.004) in LY364947 in combination with gemcitabine-treated tumors (t test; 2-tailed; * p<0.05; ** p<0.01).

M7824 with dual anti-PD-L1 and anti-TGFβ action might overcome cooperatively induced PD-L1 upregulation by TGFβ blockade and gemcitabine

In vitro and in vivo studies of LY364947 and gemcitabine show that both agents cooperatively upregulate PD-L1 on cancer cells, a possible mechanism limiting the combined anti-TGFβ in combination with gemcitabine approach (**Figure 8, Figure 9**). On human Panc1 cells treated for 24 hours with LY634947 and gemcitabine not affecting growth and not showing any additivity on in vitro drug response testing flow cytometry measured the fraction of PD-L1 positive cells as 0.56% in vehicle-treated, 1.25% in LY634947, 4.01% in gemcitabine-treated, and 8.55% in cells treated with both (**Figure 8**).

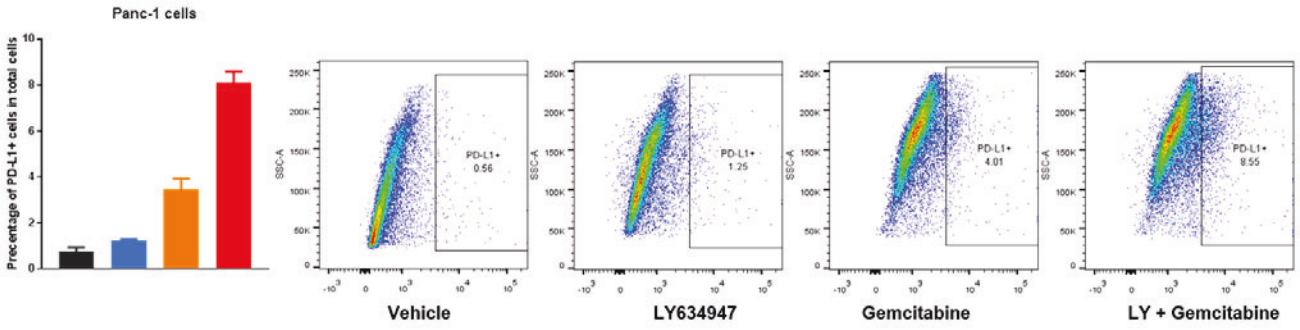


Figure 8 Cooperative upregulation of PD-L1 expression on human Panc1 pancreas cancer cells. Flow cytometry of PD-L1 expression of Panc1 cells treated with vehicle (black), 10µM LY634947, 1µM (Panc1) and 100nM (KP16) gemcitabine (orange), or the combination (in red). Previous drug response testing of gemcitabine in combination with 1, 10, and 100µM LY634947 showed no cooperative impact of LY634947 (LY) and gemcitabine on growth. Gemcitabine and LY634947 concentrations not affecting growth were selected for PD-L1 expression experiments.

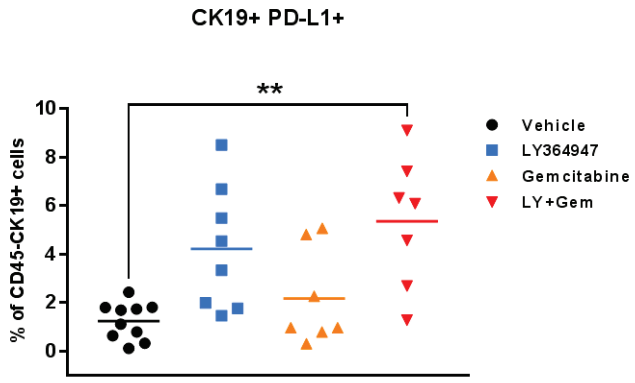


Figure 9 TGFβ blockade, in combination with gemcitabine, increases expression of the death receptor ligand expression on CK19-9 positive cancer cells in KP16 autochthonous pancreatic tumors.

That upregulation of PD-L1 is a resistance mechanism of combined TGFβ blockade and gemcitabine and targeting PD-L1, as suggested with the dual anti-PD-L1 TGFβ function of M7824 in this clinical protocol, might improve clinical outcome is shown in a survival experiment of transgenic KP16 animals treated with LY634947 plus gemcitabine versus LY634947 plus gemcitabine plus PD-L1 inhibition (**Figure 10**). Kaplan Meier analysis shows extension of survival with the triplet combination (purple curve) versus TGFβ inhibition plus gemcitabine (red

curve) suggesting that M7824 with dual PD-L1 and TGFβ inhibition might be superior to TGFβ inhibition alone.

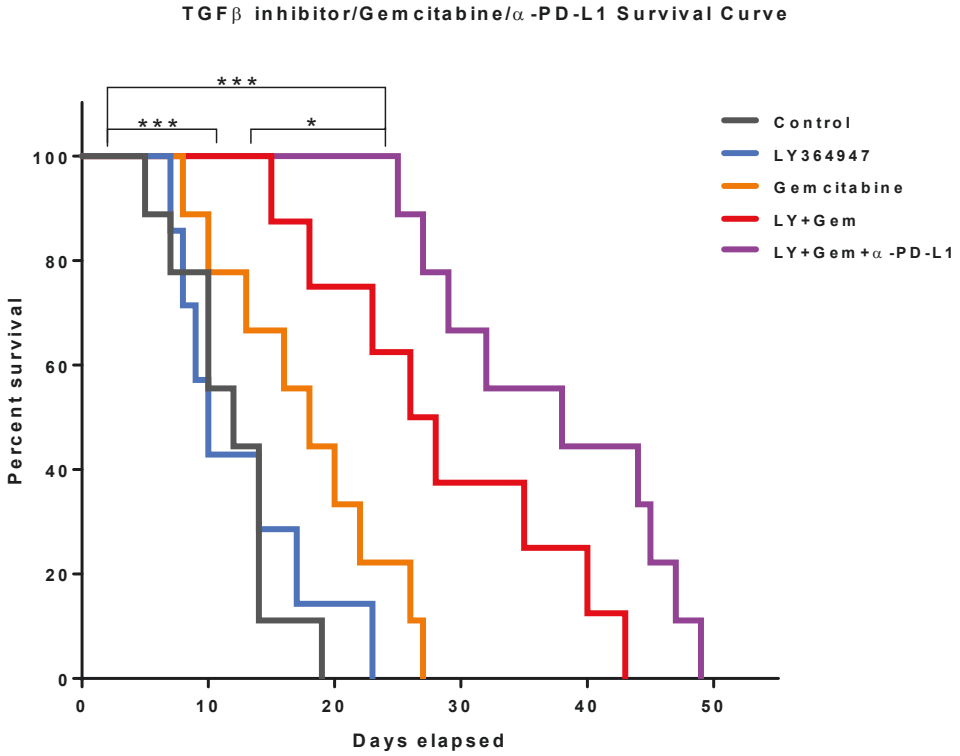


Figure 10 The addition of PD-L1 inhibition extends survival of KP16 mice treated with the TGFβ inhibitor LY634947 in combination with gemcitabine.

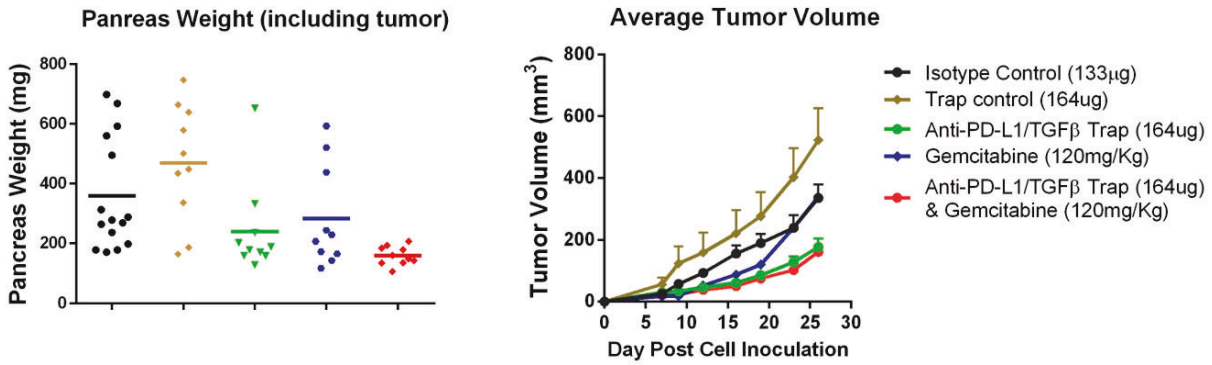
1.2.6 Preclinical rationale using M7824 in combination with gemcitabine

The inability to confidently administer M7824 for prolonged periods of time (beyond 10 – 14 days) due to the development of allogeneic immune responses against human sequences of M7824 has been a limiting factor in small animal studies with M7824. However, the following 3 preclinical experimental series are presented to support the combination of M7824 with gemcitabine:

Reduction of primary tumor weight in the murine C57B/L Panc02 model:

The C57B/L Panc02 model is a syngeneic orthotopic injection model of pancreas cancer inducing pancreatic tumors through the injection of KRAS-mutant murine Panc02 into the pancreas of immunocompetent C57B/L6 wild type mice. These tumors are populated by immune cell populations from the host as they grow. While main immune population like tumor-associated macrophages, MDSCs, and T-regs have been identified in the later stages of Panc02 tumors, there are differences to the immune landscape of autochthonous murine models of pancreas cancer or human pancreatic cytoarchitecture.

M7824, given on day 3, 5, and 7, in combination with gemcitabine reduced primary pancreatic tumor weight in this model with, in this model, no further cooperative effect on reduction of tumor growth compared to M7824 alone (**Figure 11**).



Isotype conrol	133 ug, i.v.	day 5, 7, 9
Trap control	164ug i.v.	day 5, 7, 9
Anti-PD-L1 (mut)/TGFb Trap	164ug i.v.	day 5, 7, 9
Gemcitabine	120 mg/kg, i.p.	day 3

Figure 11 M7824 in addition to gemcitabine reduces weights of Panc02 tumors (left). Animals were administered 3 doses of M7824 and 1 dose of gemcitabine (as indicated in table) and were harvested 28 days after start of treatment.

Contrary to above experiments in the autochthonous KP16 murine model of pancreas cancer, Panc02 mice were only given one single dose of gemcitabine, M7824 was due to its immunogenic toxicity limited to 3 injections, and tumors were allowed to grow back for an additional 14 days upon end of treatment with M7824 and gemcitabine. In addition, the known predominantly sarcomatoid histology of Panc02 tumors is considerably different from KP16 or KPC tumors, or human pancreas cancer specimens.

To assess cooperativity between M7824 and gemcitabine in a cancer model with an approved standard of care gemcitabine and with a greater epithelial tumor component than in the Panc02 model, M7824 and gemcitabine were also tested in the C57B/L6 MB49 preclinical bladder cancer model (**Figure 12**).

Improvement of clinical outcome in MB49 C57B/L6 mice treated with M7824 and gemcitabine compared to M7824 or gemcitabine monotherapy:

Immune competent C57BL6/J mice were subcutaneously inoculated with MB49 cells (1 x 10^6 per mouse) and when tumors reached 100mm^3 treated with either M7824 (492 µg, i.e., day 2, 5, 8), gemcitabine (120 mg/kg, day 0), or a combination of the two. Isotype control (400 µg, i.e., day 2, 5, 8) and/or PBS (0.2 mL, imp., day 0) were administered as controls. Tumor volume, overall survival, and body weight were assessed. Neither M7824 nor gemcitabine treatment alone significantly inhibited MB49 tumor growth relative to control treatment (day 16), whereas the combination of M7824 with gemcitabine significantly inhibited tumor growth compared with

M7824 ($p<0.001$, day 16) or gemcitabine ($p<0.001$) monotherapies (**Figure 12**). Furthermore, M7824 and gemcitabine combination therapy modestly extended the median survival of MB49 tumor-bearing mice relative to M7824 and gemcitabine monotherapy ($p<0.05$). Assessment of body weight indicated that all treatments were well tolerated, as no major decreases in body weight were observed throughout the observation period (data not shown).

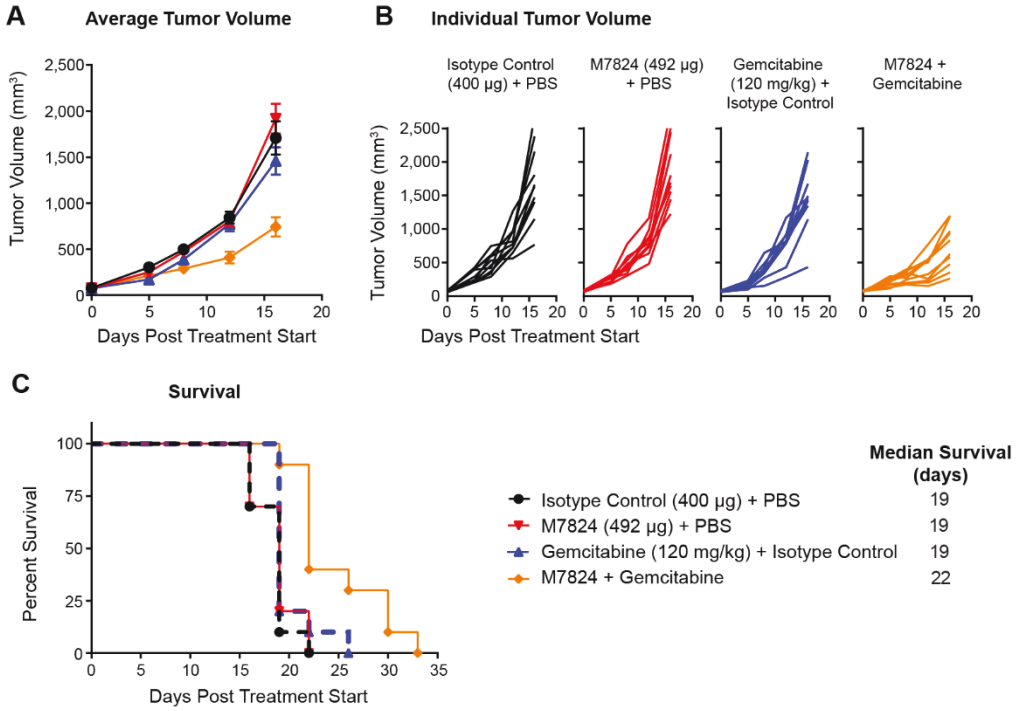


Figure 12 M7824 in cooperation with gemcitabine improves clinical outcome in the MB49 syngeneic bladder cancer model. A Reduction of tumor volume in animals treated with both M7824 and gemcitabine (in orange) compared to animals receiving monotherapy of M7824 (red) or gemcitabine (blue). Treatment schedules listed in table on the right. B Individual tumor volume measurements per animal per group. C Kaplan Meier analysis of treatment cohorts shows extension of survival in animals receiving combination treatment compared to M7824 and gemcitabine monotherapy.

M7824 induces additional anti-tumor perturbations in the tumor immune landscape compared to its PD-L1 inhibitor component avelumab alone:

Figure 4 has shown that gemcitabine cooperates with PD-L1 inhibition to extend survival of autochthonous KP16 pancreatic cancer. Comparing changes in the immune landscape of murine cancers upon treatment with avelumab, the PD-L1 component of M7824, with changes induced by M7824, PD-L1 inhibition through the avelumab fusion plus TGFβ inhibition, shows enhanced changes reprogramming the tumor microenvironment towards an anti-tumor immune milieu (**Table 1**). Most notably, treatment with the dual TGFβ-PD-L1 inhibitor M7824 had positive effects on the innate immune system (NK cells, MDSCs, DCs and M1 macrophages) that are not associated with PD-L1 blockade attesting to a second, cooperative anti-TGFβ function.

PD Response		Anti-PD-L1	M7824
Adaptive Immunity (CD8⁺ T cell response)	Proliferation	↑	↑
	Activation	↑	↑
	Degranulation	↑	↑
	PD-1 expression	↑	↑
	CXCR3 expression	↑	↑
	Eomes expression	↑	↑
	Tumor CD8⁺ T cells	↑	↑
	Tumor Tregs	↓	* ↑ ↓
Innate Immunity	Splenic MDSCs	NC	20% ↓
	Tumor MDSCs	10% ↓	50% ↓
	Splenic NK cells	NC	↑
	Tumor NK cells	NC	↑
	DC maturation	NC	↑
	Tumor M1/M2 ratio**	NC	↑

Table 1 The immuno-phenotypic signatures of the dual TGFβ-PD-L1 inhibitor M7824 compared to anti-PD-L1 (with avelumab) alone are distinct in tumor-bearing mice.

M7824 has also been combined with other chemotherapeutics in solid organ cancer models with the combination of M7824 with standard-of-care therapeutic agent(s) showing superior efficacy compared to individual M7824 or chemo monotherapy. These include:

The therapeutic potential of M7824 was explored in combination with various standard of-care therapies. In combination with the core components of the FOLFOX chemotherapy regimen (oxaliplatin, fluorouracil and oxaliplatin), an additive enhancement in efficacy was demonstrated against MC38 colorectal carcinoma. This finding is in line with results shown in **Figure 5** where blockade of TGFβ signaling inhibition, via the use of the TβRI small molecule inhibitor LY2109761, cooperates as shown with gemcitabine to suppress tumor growth and extend survival in genetically engineered animal models of pancreas cancer. In combination with fractionated, localized radiotherapy, a highly synergistic antitumor effect was achieved with only a single low dose of M7824 in the MC38 tumor model. An augmented antitumor effect was also observed when M7824 was combined with pazopanib in a mouse renal cell carcinoma model, and with anti-CTLA-4 in a B16 melanoma model. Immunological readouts from these combination studies consistently showed additive immunomodulatory effects that correlated with antitumor response as shown in **Figure 6** where TGFβ blockade and gemcitabine synergistically reprogram the immune landscape of KP16 pancreatic tumors towards an anti-tumor phenotype by, among others, cooperatively reducing T regulatory cells, MDSCs, or increasing intratumoral CD8+ T cells. Of note, TGFβ signaling inhibition in combination with gemcitabine not only decreases T regulatory cells, it also decreases their anti-tumor immune evasive function. **Figure 13** shows that intratumoral CD4+CD25+ T regulatory cells isolated from murine KP16 pancreatic tumors treated with TGFβ inhibition and gemcitabine impair upon co-culture anti-tumor activity of CD8+ T cells

significantly less than T regulatory cells isolated from vehicle-treated tumors. Overall, above combination treatment data suggest that M7824 can be explored as a combination partner in different clinical settings for the treatment of cancer patients as proposed in this study.

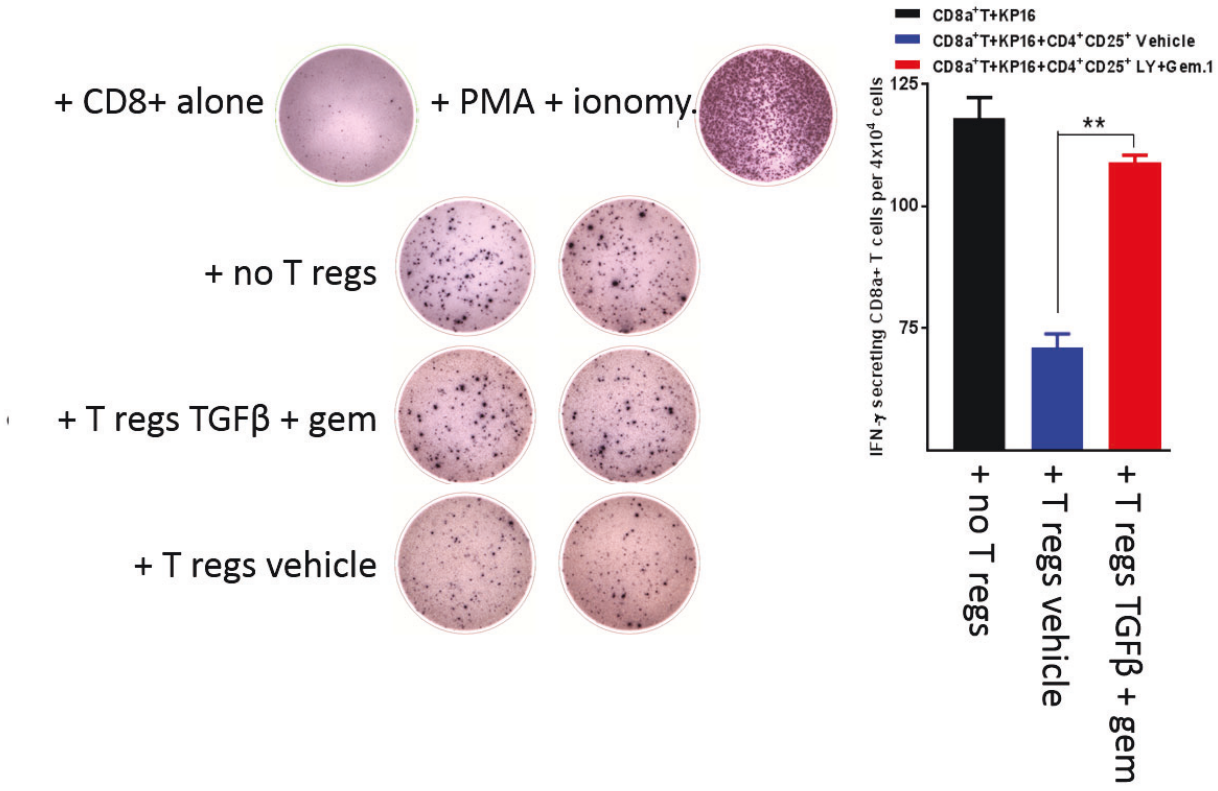
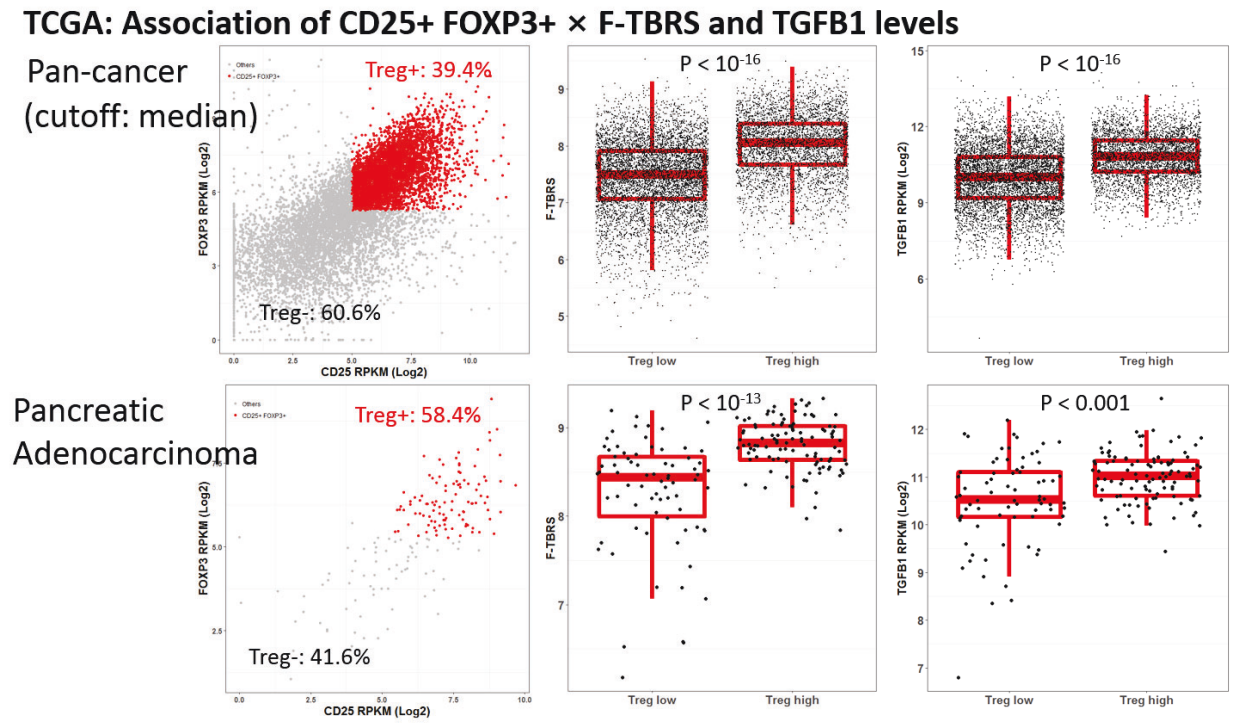


Figure 13 CD4+CD25+ intratumoral T regulatory cells isolated from murine pancreatic tumors treated for 7 days with LY634947 in combination with gemcitabine have reduced anti-tumor efficacy compared to T regulatory cells isolated from vehicle-treated tumors. ELISpot assay of CD8+ T cells and KP16 cancer cells after addition of no T regulatory cells (black), 10,000 CD4+CD25+ cells from vehicle treated tumors (blue), and 10,000 from LY2109761-combination with gemcitabine-treated tumors (red) (representative ELISpots of three groups on the left; summary of N=3 mice per group, right).

To show that T regulatory cell and myeloid immune populations cooperatively affected by TGFβ inhibition and gemcitabine in above studies and by the dual TGFβ-PD-L1 inhibitor are connected to TGFβ signaling and TGFβ levels in human samples, we correlated in clinical specimens from The Cancer Genome Atlas (TCGA) and the International Cancer Genome Consortium (ICGC) data sets expression levels of a previously validated TGFβ response signature (TGRS) as well as TGFβ1 gene expression levels with expression levels of

- T regulatory cell marker combination FoxP3 and CD25 (T regulatory cells)
- Monocyte/macrophage-lineage cell marker combination CD11b and CD33 (**Figure 14**).

There was a strong association ($p<10^{16}$) between TGFbeta-dependency measured by high expression levels of the TGFβ Response Signature (TBRS), and to a lesser degree TGFβ1 ligand levels, and expression levels of the immune cell population-selective markers $CD25^{high}$ $FoxP3^{high}$ (T regulatory cells) $CD11b^{high}$ $CD33^{high}$ (human MDSC markers) both in all cancers and in pancreatic cancer gene expression sets. These findings suggest that intratumoral TGFβ signaling is connected to T regulatory cell and myeloid immune cell population levels and that these immune cell populations are indeed regulated by TGFβ which supports above findings of altered Treg and MDSC cell fractions upon TGFβ inhibition.



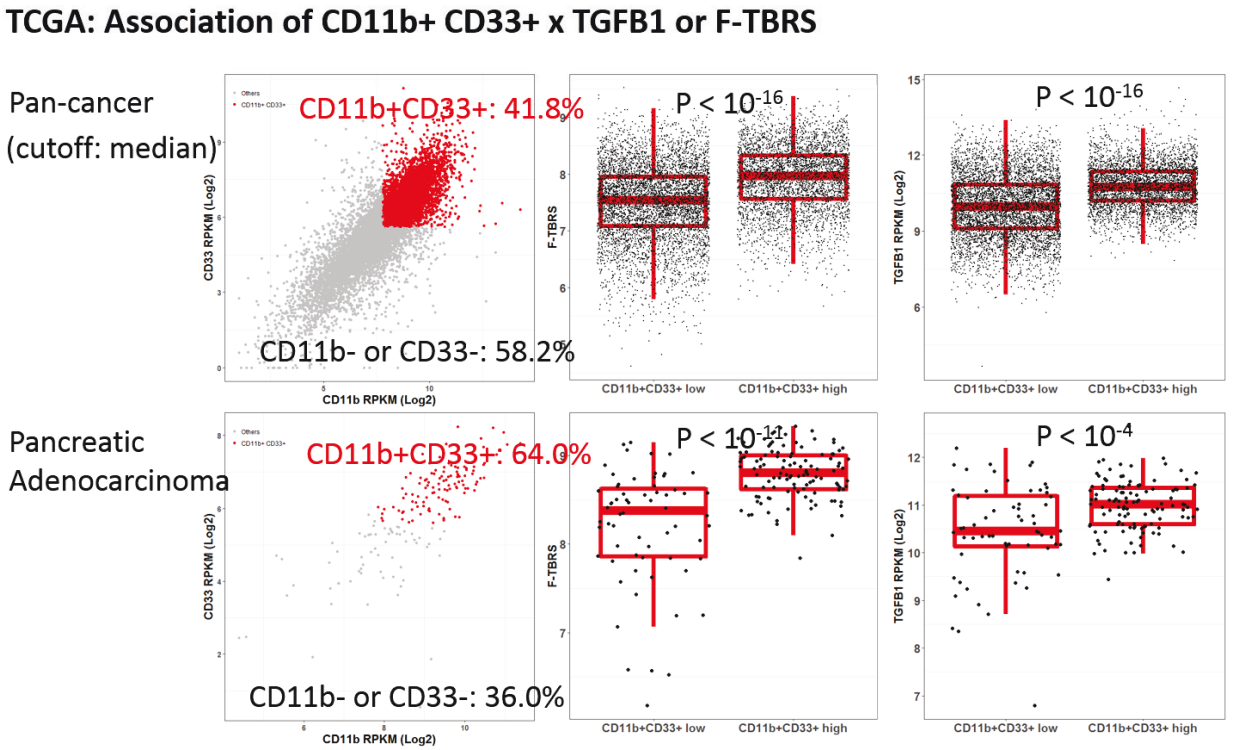


Figure 14 Association of expression levels of T regulatory-specific markers CD25, FoxP3 (top) and myeloid-selective markers CD11b, CD33 (bottom) with TGFβ Response Signature (TBRS; originally derived from large cancer collection; as a measure of 'dependent' on TGFβ biology) and TGFβ ligand 1 expression (TGFB1) in TCGA and ICGC Pan-Cancer and pancreas cancer datasets. Cutoff of

CD25high FoxP3high and CD11bhigh CD33high was defined by the median of all observations for each marker across all included cancer samples

In summary, TGFβ signaling inhibition in combination with gemcitabine induces immune-mediated cooperative mechanisms resulting in improved anti-tumor activity, suppression of tumor growth, and improved survival. These results attest to the immunomodulatory function of gemcitabine in pancreas cancer, an observation also made in human pancreas cancer patients (pancreas cancer patients treated with gemcitabine have been shown to have decreased MDSC, decreased T regulatory cell levels, increased T effector / T-reg ratios, and reduced TGFβ ligand 1 levels [13]), and justify the pursuit of a combination immune therapy approach of TGFβ signaling inhibition and gemcitabine.

1.2.7 Safety of combined TGFβ and PD-L1 inhibition by the dual TGFβ-PD-L1 inhibitor M7824

Targeting both, PD-1 / PD-L1 and TGFβ signaling axis, is attractive as an antitumor approach. A recent report found that blockade of TGFβ signaling in T cells or deletion of TGFβ1 from T cells in a mouse model led to diminished PD-1 expression in tumor-infiltrating CD8+ T cells.[14] Concomitant PD-1 and TGFβ blockade can restore pro-inflammatory cytokines.[15] These findings are consistent with our data generated in the transgenic pancreatic cancer KP16 mouse model showing decreased PD-1 expression on CD8+ T cells and that additional PD-L1 blockade is able to extend survival when added to TGFbeta inhibition and gemcitabine. Similarly, in a murine model of hepatocellular carcinoma, TGFβ appeared to increase the expression of PD-L1 in dendritic cells, which in turn promoted T-cell apoptosis and increased percentage of CD25+, Foxp3+ T regulatory cells.[16] Additionally, higher levels of circulating myeloid-derived suppressor cells (MDSCs), a significant source of TGFβ, are associated with failure to respond to anti-PD-1 therapy. Finally, the addition of PD-L1 inhibition to TGFβ blockade in combination with gemcitabine as shown in **Figure 10**, extends survival in the autochthonous KP16 pancreas cancer model.

Similarly, the dual TGFβ-PD-L1 inhibitor M7824 has been shown in syngeneic preclinical breast cancer models to enhances antitumor activity and prolongs survival above the effect of either the anti-PD-L1 antibody, avelumab, or TGFβRII control alone. Tumor re-challenge experiments in cured mice show durable protective immunity. In vivo studies showed that the antitumor effects were mediated by CD8+ T cells, as in above described KP16 animal experiments, and NK cells. CD8+ T-cell tumor infiltrates were observed and, overall, the CD8+ response was associated with long term protective immunity. Importantly, in the syngeneic MC38 model, M7824 showed significantly better efficacy than the combination of avelumab plus TGFβ Trap control, supporting the rationale of combining the 2 active moieties in 1 molecule.
Thus, given the emerging picture for PD-1 / PD-L1 class, in which responses are apparent but with room for increase in effect size, it is assumed that co-targeting a complementary immune modulation step will improve tumor response and it is expected that the anti-PD-L1 function of M7824 suppresses the immune evasive signal of increased PD-L1 expression on cancer cells and hence enhance efficacy of the TGFβ inhibition plus gemcitabine combination approach.
A brief summary of safety experience with the PD-1 inhibitors nivolumab (Opdivo®) and pembrolizumab (Keytruda®) is given here, based on prescribing information (refer to current label information for updated information). For pembrolizumab the section on Warnings and

Precautions includes adverse reactions of immune-mediated pneumonitis (2.9%), immune-mediated colitis (1%), immune-mediated hepatitis (0.5%), immune-mediated hypophysitis (0.5%), renal failure (0.5%) and immune-mediated nephritis (0.7%), immune-mediated hyperthyroidism (1.2%) and hypothyroidism (8.3%), and a variety of other immune-mediated adverse reactions occurring in less than 1% of patients. In addition, a warning for embryofetal toxicity is provided. For nivolumab the section on Warning and Precautions includes adverse reactions of immune-mediated pneumonitis (2.2%) with fatal immune-mediated pneumonitis in 0.9% (5/574), immune-medicated colitis (2.2%), immune-mediated hepatitis (1.1%), immune-mediated nephritis and renal dysfunction (0.7%), immune-mediated hyperthyroidism (3%) and hypothyroidism (8%) and a variety of other immune-mediated adverse reactions occurring in less than 1% of patients. In addition, a warning for embryofetal toxicity is provided.

Safety experience with various TGFβ targeting agents described in the literature suggests no overlapping immune-related profile with compounds of the anti-PD-1 / anti-PD-L1 class. In Phase I trials, the experience with a molecule with a highly similar mechanism to the M7824 TGFβ trap moiety, the anti-TGFβ-1 and 3 antibody fresolimumab, showed no dose limiting toxicity up to 15 mg/kg and no immune related events.[17] There were no DLTs and the only major AEs were skin lesions, mainly keratoacanthomas, some with atypical features, one event of squamous cell carcinoma, plus hyperkeratosis of the skin. Immune events were not reported. A syndrome known as Ferguson-Smith disease is caused by mutations in TGFβ is associated with the formation of keratoacanthomas, similar to the findings described for fresolimumab.[18] Therefore, it is plausible that skin tumors observed during fresolimumab treatment may be related to TGFβ inhibition. A neutralizing antibody against TGFβ-1, TβM1, was well tolerated when studied as high as 240 mg with diarrhea as the only DLT event. Notably, one event of low Hgb was observed in the high dose group. This is notable since the only preclinical finding associated with M7824 was decreased Hgb. Trabedersen, an antisense oligonucleotide that inhibits TGFβ2 expression, was associated with thrombocytopenia that was moderate.[19]

In line with above early clinical experience of similar anti-TGFβ agents, the toxicity profile of M7824 is predominantly benign and highly comparable to that of avelumab; in the recently released results of the phase I study NCT02517398 ‘Phase 1 Trial of M7824, a Bifunctional Fusion Protein Targeting PD-L1 and TGFβ, in Advanced Solid Tumors’ with dose escalation cohorts of 1, 3, 10, and 20mg/kg (=exceeding the 1,200mg flat starting dose in this study) M7824 given 2-weekly, the MTD was not reached: Data suggested overall good tolerance. One out of 16 patients developed a keratoacanthoma which could be related to the TGFβ inhibition mechanism of M7824. There was no grade 4-5 TEAEs. The only DLT observed was colitis. Overall, the MTD was not exceeded at doses up to 20 mg/kg (for further details, please also see Section **1.2.8**. Pharmacokinetic / Pharmacodynamic including human phase I findings of M7824).

On the other hand, toxicological findings with M7824 in cynomolgus monkey indicated a decrease in red blood cells (24% compared with the control group values) and related parameters (Hgb and hematocrit). These decreases were reversible during the recovery period; thus, anemia is considered a potential risk of the compound. Due to the involvement of TGFβ in repair of skin and other tissue injuries, alterations in wound healing or repair of tissue damage is considered a potential risk. Overall, evidence suggests non-overlapping toxicity profiles for anti-PD-L1 and anti-TGFβ agent classes. There is a theoretical potential of irAEs that would be the consequence

of a double blockade of negative regulatory loops of the immune system; however, that was not seen in the several hundred of patients treated with 1,200mg M7824 to date.

For full preclinical toxicology and in vivo pharmacology findings of M7824 including in mice and non-human primates, please refer to the latest version of the Investigator Brochure of M7824.

1.2.8 Pharmacokinetic / Pharmacodynamic profile of M7824 including human phase I findings

Preclinical PK data and PK / PharmDyn analysis for M7824 is available from mice and cynomolgus monkeys. The single-dose data in monkey shows non-linearity between the low doses of 0.8 and 4 mg/kg versus the high dose of 20 mg/kg, which had reduced clearance, suggesting a saturable component. The TGFβ1 binding was assessed and showed suppression for prolonged periods beyond drug exposure at all dose levels; however, baseline data contained data below lower limit of quantification and therefore these data were not considered relevant for PK/PharmDyn projections. The fusion protein appeared stable since there was no evidence of intralinker cleavage. Mouse PK / PharmDyn models for tumor and peripheral blood (CD3+ splenic) PD-L1 occupancy were also generated.

A brief summary of PK and PK / PharmDyn is as follows:

- The predicted human terminal half-life ($t_{1/2}$) for M7824 is approximately 6 days
- Simulations predict that a 1 mg/kg dose will provide an average exposure of approximately 7 μg/mL in humans
- Based on PK / PharmDyn modeling and human projections:
  - At a human dose of 0.1 mg/kg, 95% PD-L1 total occupancy at maximum serum concentration observed post-dose (C_{max}) is expected in PBMCs, providing approximately 60% total occupancy in tumor
  - At a human dose of 1.0 mg/kg, more than 95% of PD-L1 total occupancy at C_{max} is expected in PBMCs and tumor
  - At a human dose of 1.0 mg/kg, 20% effect is projected in tumor regression in the PK / PharmDyn model compared with 95% at 7.5 mg/kg
- Human projections indicate a dose of 7.5 mg/kg and higher (range 4 to 20 mg/kg), on a bi-weekly schedule is needed to achieve full efficacy based on a mouse tumor model in which complete tumor regressions were observed
- Anti-drug antibodies were observed in some animals; however, the impact on PK or PK / PharmDyn is not known. Preclinical antidrug antibody formation is not predictive of human antibodies.

M7824 effectively engaged PD-L1 and reduced levels of all three TGFβ ligands by several orders of magnitude to undetectable levels. PD-L1 engagement and suppression of TGFβ ligand levels was maintained throughout the 14-day dosing interval (**Figure 15**). In the recently released initial phase I dose escalation cohort of M7824, 12 subjects had enrolled in the dose-escalation part of the study with 3 subjects treated at each dose level of M7824: 1, 3, 10, and 20 mg/kg, once every 2 weeks. Median age was 56 years (range: 34 to 78). All had ECOG PS 0 or 1, with a median of 4 prior therapies (range: 2 to 7).

No subject experienced a DLT and no subject was discontinued from treatment for drug-related treatment-emergent adverse events (TEAEs). No subject was reported with a TEAE with a Preferred Term of infusion-related reaction; 3 subjects were reported with TEAEs that were classified as "infusion-related reactions", with the Preferred Terms of abdominal pain, pyrexia, and back pain.

Overall, 5 of 12 subjects (41.7%) had drug-related TEAEs. All drug related TEAEs were Grade 1 or 2: mouth hemorrhage (2 subjects, 16.7%), dry mouth, epistaxis, influenza-like illness, maculopapular rash, pruritis (1 subject, 8.3% each).

To date, 1 subject with pancreatic cancer (3 mg/kg cohort) has been reported with a confirmed PR according to RECIST (ongoing at 6 months). In addition, SD has been reported 5 subjects (41.7%), including 1 subject in the 1 mg/kg cohort, and 2 subjects in each of the 3 and 10 mg/kg cohorts. Five subjects (41.7%) were reported with a BOR of PD. At the time of the data cutoff, 8 subjects (66.7%) remained on treatment.

All subjects from the first 3 dose levels were evaluable for PK analysis showing at 3 and 10 mg/kg dose-linear pharmacokinetics and an approximate $t_{1/2}$ of 150 hours (6.25 days). Subjects from the 1, 3, 10, and 20 mg/kg dose levels were evaluable for PD-L1 target occupancy.

At 3 mg/kg and above, PD-L1 target occupancy was high throughout the entire dosing interval (from Cmax through Cmin). All subjects from the first 3 dose levels were evaluable for TGFβ1, 2, 3 plasma concentrations. After M7824 infusion, TGFβ1 and 3 concentrations were undetectable in all subjects at all doses and remained suppressed throughout the dosing interval. TGFβ2 suppression was also evident in all subjects but its degree was dependent on dose and drug concentrations. These data indicate that full pharmacological activity of both modes of action of M7824 is generally achieved in peripheral blood at the dose of 3 mg/kg.

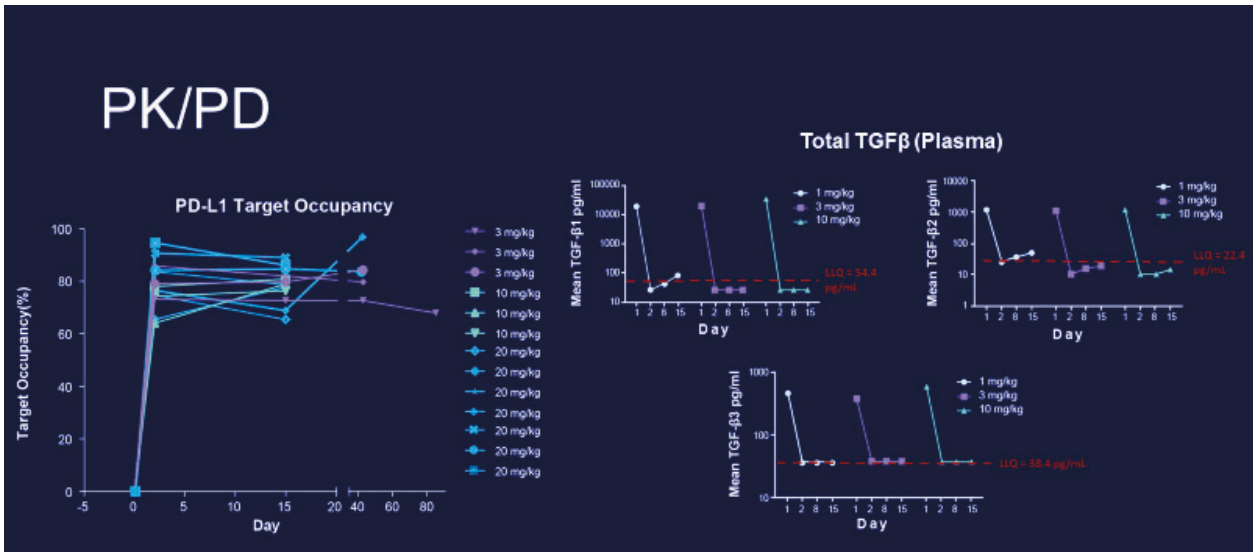


Figure 15 PD-L1 target occupancy and reduction of TGFβ ligands in human subjects treated on the 'Phase 1 Trial of M7824, a Bifunctional Fusion Protein Targeting PD-L1 and TGFβ' (NCT02517398). Patients from the 3, 10, and 20 mg/kg dose levels were evaluable for PD-L1 target occupancy by PBMC assay, which reached saturating levels rapidly peripherally throughout the entire dosing interval (right). Plasma concentrations of TGFβ1, β2, and β3 were substantially reduced, if not undetectable, after M7824 infusion for the entire dosing period (right).

1.2.9 Summary of Clinical Findings of M7824 including Adverse Events (AEs) in Pancreas Cancer Patients to date

As of March 21st, 2017, at least N=36 patients with a diagnosis have received at least one, or more, doses of 1,200mg of M7824. **Table 2** lists both drug-related and not-related AEs for items in which > 5% of patients experience the event, or if the event was Grade 3 or higher. Overall, the drug was well tolerated at the 1,200mg dose level with a typical AE profile for an advanced cancer patient cohort. Of note, anemia was (6 out 36; 16.7%) was the most grade 3 or greater toxicity. The only other grade 3 toxicities occurring in more than 10 percent of study subjects were fatigue (4 out of 36; 11.1%) and abdominal discomfort (4 out 36; 11.1%). There were 5 subjects with ≥grade 3 events listed as related to M7824. These include G4: lipase increased (1 event), and G3: anemia (2), colitis (1), diarrhea (1), ALT increase (1).

Table 2 Adverse Events (AEs) in pancreatic cancer cohort patients on EMR200647-001 dosed with 1,200mg M7824 (SMC 380 Expansion cut-off 21 Mar2017, First dose before 21 Feb 2017; N=36). Both drug-related and not drug-related AEs for items in which > 5% of patients experience the event, or if the event was Grade 3 or higher, are listed.

Primary System Organ Class	Any Grade	Grade >=3	Grade >=4	Grade 5
Preferred Term	**n (%)**	**n (%)**	**n (%)**	**n (%)**
Anemia	8 (22.2)	6 (16.7)	1 (2.8)	0
Leukocytosis	3 (8.3)	3 (8.3)	0	0
Palpitations	2 (5.6)	0	0	0
Abdominal Distention	4 (11.1)	0	0	0
Abdominal Pain	9 (25.0)	4 (11.1)	0	0
Abdominal Pain (upper)	2 (5.6)	0	0	0
Ascites	4 (11.1)	0	0	0
Constipation	9 (25.0)	2 (5.6)	0	0
Diarrhea	6 (16.7)	1 (2.8)	0	0
Dysphagia	2 (5.6)	0	0	0
GI Hemorrhage	2 (5.6)	2 (5.6)	0	0
Gingival Bleeding	2 (5.6)	0	0	0
Melena	2 (5.6)	0	0	0
Nausea	9 (25.0)	0	0	0
Upper GI hemorrhage	1 (2.8)	1 (2.8)	0	0
Vomiting	7 (19.4)	0	0	0
Chills	3 (8.3)	0	0	0
Fatigue	13 (36.1)	4 (11.1)	0	0
Malaise	2 (5.6)	0	0	0
Peripheral Edema	5 (13.9)	0	0	0
Pyrexia	9 (25.0)	0	0	0

Bile duct obstruction	1 (2.8)	1 (2.8)	0	0
Cholangitis	2 (5.6)	1 (2.8)	0	0
Biliary Tract Infection	1 (2.8)	1 (2.8)	0	0
Pneumonia	2 (5.6)	1 (2.8)	0	0
Sepsis	1 (2.8)	1 (2.8)	1 (2.8)	0
UTI	3 (8.3)	3 (8.3)	0	0
ALT increased	3 (8.3)	0	0	0
AST increased	5 (13.9)	0	0	0
Blood Alk Phos Increased	1 (2.8)	1 (2.8)	0	0
Blood Bilirubin Increased	3 (8.3)	2 (5.6)	1 (2.8)	0
GGT Increased	1 (2.8)	1 (2.8)	0	0
Lipase increased	1 (2.8)	1 (2.8)	1 (2.8)	0
Weight decreased	3 (8.3)	0	0	0
Decreased Appetite	9 (25.0)	2 (5.6)	0	0
Hypercalcemia	1 (2.8)	1 (2.8)	0	0
Hypokalemia	2 (5.6)	1 (2.8)	0	0
Hyponatremia	2 (5.6)	2 (5.6)	1 (2.8)	0
Hypophosphatemia	1 (2.8)	1 (2.8)	0	0
Arthralgia	2 (5.6)	0	0	0
Musculoskeletal Pain	2 (5.6)	0	0	0
Cancer Pain	1 (2.8)	1 (2.8)	0	0
Keratoacanthoma	2 (5.6)	0	0	0
Malignant Neoplasm Progression	1 (2.8)	1 (2.8)	0	0
Malignant Pleural Effusion	3 (8.3)	1 (2.8)	0	0
Pancreatic Carcinoma	1 (2.8)	1 (2.8)	1 (2.8)	1 (2.8)
Dizziness	4 (13.9)	3 (8.3)	1 (2.8)	1 (2.8)
Embolic Stroke	1 (2.8)	1 (2.8)	1 (2.8)	1 (2.8)
Headache	3 (8.3)	0	0	0
Peripheral Motor Neuropathy	1 (2.8)	1 (2.8)	0	0
Somnolence	1 (2.8)	1 (2.8)	0	0
Anxiety	1 (2.8)	1 (2.8)	0	0
Cough	2 (5.6)	0	0	0
Dyspnea	4 (11.1)	0	0	0
Hiccups	2 (5.6)	1 (2.8)	0	0
Nasal Congestion	2 (5.6)	0	0	0
Pleural Effusion	2 (5.6)	0	0	0
Pulmonary Embolism	2 (5.6)	2 (5.6)	0	0
Respiratory Failure	1 (2.8)	1 (2.8)	1 (2.8)	0
Pruritis	3 (8.3)	0	0	0
Rash	4 (11.1)	0	0	0
Hypertension	2 (5.6)	1 (2.8)	0	0

Preliminary clinical efficacy signal of the N=36 pancreatic cancer M7824 monotherapy cohort (provided by EMD Serono): Preliminarily, additional clinical activity has been seen with objective responses and prolonged disease stabilization. However, the cohort is being actively reviewed and remains dynamic.

In addition, after discussion among NCI investigators on multiple protocols using M7824, multiple bleeding events ranging from low grade gingival bleeding and epistaxis to more serious hemoptysis, GI bleeding and hematuria have been observed. Some of these events can be attributed to bleeding events related to cancer directly and others bleeding events can be attributed to colitis or cystitis which is a known toxicity of anti-PD-L1 agents including M7824. However, there remains the possibility that M7824 may increase the overall risk of bleeding in ways that may not be directly related to direct tumor bleeding or inflammatory bleeding events described with checkpoint inhibitors like M7824. It is hypothesized that this possible increased bleeding risk may be due to TGF beta inhibition which has an effect on angiogenesis; bleeding has also been observed in patients receiving M7824 and may be drug-related (e.g., gum bleeding, nose bleeds, coughing up blood, blood in their urine, or blood in the stool). Accordingly, patients will be notified of the same possible risk in the informed consent document for this study.

1.2.10 Rationale for the Clinical Trial

The administration of M7824 in combination with gemcitabine to subjects with advanced pancreatic tumors who have progressed on 1st-line systemic chemotherapy is justified by the following:

- PD-L1 inhibition, one of the dual components of M7824, in combination with gemcitabine cooperatively extends survival compared to PD-L1 inhibition or gemcitabine monotherapy alone in autochthonous murine pancreatic cancer models
- TGFβ blockade, the second component of M7824, in combination with gemcitabine cooperatively inhibits tumor growth and extends survival compared to TGFβ inhibitor or gemcitabine monotherapy alone in autochthonous murine pancreatic cancer models
- Upregulation of PD-L1 upon TGFβ inhibition is a mechanism of resistance to optimal efficacy of TGFβ blockade in combination with gemcitabine, and the addition of PD-L1 inhibition to TGFβ blockade and gemcitabine further extends survival ('triple immunotherapy combination')
- Pancreatic tumor weights are further reduced by the addition of gemcitabine compared to M7824 therapy alone in the syngeneic Panc02 pancreatic cancer mouse model
- M7824 either in combination with gemcitabine, or in combination with other systemic chemotherapy agents, additively reduces tumor growth compared to respective monotherapies in other syngeneic mouse models (MB49 bladder cancer, MC38 colon cancer)
- Safety experience with TGFβ-targeting agents described in the literature suggests no overlapping immune-related adverse profile with the PD-1/PD-L1 class or with gemcitabine.
- With the exception of anemia, M7824 and gemcitabine have non-overlapping toxicity profiles
- Clinical experience with M7824 to-date at the proposed 1,200mg flat dose level in pancreas cancer patients demonstrates an acceptable safety profile and as monotherapy clinical antitumor activity in multiple solid tumor types including pancreas cancer.

1.2.11 Rationale for M7824 Dose Levels with special consideration of overlapping toxicities with gemcitabine

The rationale for the starting dose of M7824 of 1,200mg is based on the clinical safety of this dose in multiple phase I expansion cohorts of M7824 including in pancreas cancer, the preclinical margin considerations to the NOAEL of the pivotal safety assessments, the PK/PD findings of the phase I study showing highly effective TGFβ suppression and near complete PD-L1 occupancy, as well as safety experience of avelumab including safety evaluation of avelumab leading to regulatory approval of avelumab for metastatic Merkel cell and urothelial cancer in 2017 (https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/761049s000lbl.pdf).

The pivotal preclinical safety assessment of M7824 in non-human primate showed no adverse, effects except a trend of reversible red blood cell reduction with associated parameters (Hgb and hematocrit) and with no dose dependency. The NOAEL was set at the highest dose, 140 mg/kg administered weekly. Based on allometric scaling, the human equivalent of 140 mg/kg in monkey is 45 mg/kg/week or 90 mg/kg every 2 weeks (3,150mg/week or 6,300mg flat dose levels for 70kg individual every two weeks), as calculated by standard guidance (Food and Drug Administration [FDA] 2005), and is more than 5-fold higher than the chosen 1,200mg flat dose. In this regard, it should be noted that cross-species biology of TGFβ is expected to be highly similar between human, mouse, and cynomolgus primate since TGFβ is an extremely highly conserved gene with the mouse sequence for TGFβ1 differing by only one amino acid and the cynomolgus monkey predicted to be 100% identical to human. For TGFβ3, the amino acid sequence is predicted to be 100% identical across mouse, human, and cynomolgus primate.

With regard to the PD-L1 component of M7824, it can be noted for the selected 1,200mg flat starting dose that PD-L1 inhibition by avelumab (the PD-L1 component of M7824) has resulted in expected immune features with a currently acceptable safety profile that continues to be assessed (refer to the current avelumab IB and https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/761049s000lbl.pdf). In dose escalation, avelumab did not achieve an MTD up to 20 mg/kg (1,400mg in 70kg individual), with only 1 DLT observed at the highest dose (refer to the current avelumab IB). To date, safety experience has been collected in more than 550 subjects with solid tumors exposed to the 10 mg/kg dose, as described further in this section. The exposure of M7824 will likely be similar at the same dose strength since the molarity for PD-L1 binding is approximately 20% less of M7824 but the half-life is predicted to be approximately 20% to 25% more.

One of the potential concerns of TGFβ ligand-binding agents has been the unknown therapeutic window between tumor-selective effects in the microenvironment of cancer and systemic toxicities. Binding of M7824 to free TGFβ in the periphery is projected to be substantial since TGFβ levels are very low, approximately 2 ng/mL.[20] Human PK simulations suggest that a 1 mg/kg dose would provide an average exposure of approximately 7 µg/mL, approximately 200 to 400 fold higher than the EC_{50} or IC_{50} for TGFβ binding (17.9 ng/mL) and interference (38.3 ng/mL) assays. However, for this molecule, which binds TGFβ at 1:1 molar ranges, EC_{50} and IC_{50} cannot be regarded as a fixed affinity constant and is therefore difficult to estimate in human dosing. At a dose of 0.1 mg/kg on a biweekly dose schedule, the C_{trough} is projected to be 150 ng/mL. However, from a safety standpoint, binding of TGFβ in the periphery is of little consequence since circulating, free TGFβ is unrelated to its functional tissue activity. TGFβ cytokines function not as circulating endocrines, but predominantly as autocrine and paracrine in

the local environment including tumor environment.[8] This lack of interference with essential tissue homeostasis regulated by TGFβ is corroborated by the lack of drug-related SAEs in the 36 pancreas cancer patients treated who received at least 1 dose of 1,200mg M7824 to date. While detailed intratumoral TGFβ levels and PK / PDdyn measures of treated subjects are awaited, optimal effects of M7824 -mediated TGFβ inhibition at 7.5 mg/kg (range 4 to 20 mg/kg based on 30% variability between subjects) is predicted to reach effective tumor growth inhibition of approximately 90%. Taken together the clinical safety data and preclinical NOAEL determinations in monkey (up to predicted 6,300mg in a highly-conserved target), 1,200mg is a reasonable starting dose from a safety perspective which offers anti-tumor efficacy.

Thus, together with the benign nature of preclinical and clinical safety assessments for M7824 other than Hgb changes overall suggesting a low likelihood for systemic immune synergies or overlapping toxicities between the PD-L1 and TGFβ pathways, and gemcitabine, based on the to date

- preclinical and clinical safety of M7824 administered at the 1,200mg dose level, and
- non-overlapping toxicity profile with gemcitabine

dose-adjustments or discontinuation of drug(s) for each drug – M7824 and gemcitabine - will be made / carried out separately based on the observed toxicity profile.

1.2.12 Rationale for Gemcitabine Dose Selection and Safety Measures

To account for possible gemcitabine toxicity exacerbated through the combination with M7824, the following two safety measures have been taken. The standard dose level of gemcitabine is 1,000 mg/m^2 over 30 minutes once weekly for the first 7 weeks, then one week rest, then once weekly for 3 weeks of each 28-day cycle.

Over the past decade there has been increased efforts to bring advances in pharmacogenomics to the bedside. **Figure 16** shows steps involved in catalyzing gemcitabine transport, intracellular activation, and metabolism. Previous retrospective studies have shown an association of nucleotide polymorphisms (SNP) of drug metabolic genes with toxicity of 2',2'-difluoro 2'-deoxycytidine (gemcitabine)-based chemoradiotherapy and possibly clinical outcome including hematologic toxicity and overall survival.

Gemcitabine enters the cell via facilitated transport of nucleoside transporters and becomes activated through ATP-requiring phosphorylation by deoxycytidine kinase (dCK) to the active tri-phosphatide metabolite dFdCTP in a rate-limiting step. The primary elimination route of gemcitabine is deamination by cytidine deaminase (CDA) to its main metabolite 2',2'-difluorodeoxyuridine (dFdU) as the critical enzyme controlling gemcitabine levels.

Due to this critical role of CDA in gemcitabine metabolism, pharmacogenomic studies exploring associations of small nucleotide polymorphisms (SNPs) in gemcitabine-metabolizing genes with clinical outcome have both in pancreas cancer and NSCLC focused on CDA and here on the CDA Lys^{27}Gln genotype (rs2072671, CDA 79A>C polymorphism; K27Q). For other polymorphisms, either in CDA or other gemcitabine-metabolizing genes, available clinical correlative studies are much rarer and evidence less robust. There are also large ethnic differences in SNP frequencies between Caucasian and Asian populations described.

In the predominantly Caucasian pancreas cancer patient population and based on analysis of samples from the large phase III randomized RTOG 9704 study, rates of CDA Lys^{27}Gln

polymorphism genotyping showed the homozygous wild type (Lys/Lys) was found in 39.5%, whereas the heterozygous (Lys/Gln) and homozygous variant (Gln/Gln) was found in 50.2% and 10.3%. [21] There was a statistically significant increased risk for severe hematological toxicity (≥grade 3) in particular for the homozygous CDA $Lys^{27}Gln/Gln$ variant (odds ratio (OR)=16.7; p=0.01) but not for non-hematological toxicities or survival. Importantly, this association was specific for the gemcitabine-treated patients and not seen in the cohort which received 5-FU. There have been other studies with similar, albeit less strong and in part conflicting, findings reported. In a retrospective analysis of 154 patients with potentially resectable pancreas cancer investigators from the MD Anderson Cancer Center examined 17 SNPs in gemcitabine-metabolizing genes and impact on survival and toxicity. CDA C111T, and not CDA 79C>A (K27Q), was associated with neutropenic toxicity. [22] in a small study of 31 patients with NSCLC, patients carrying the nonsynonymous CDA SNP 79A >C (CDA K27Q) had a 21% lower gemcitabine clearance as compared to wild-type patients, but the risk for chemotherapy-associated neutropenia (61% vs. 32%, P = 0.07) and severe neutropenia (17% vs. 5%, P = 0.26) failed to reach statistical significance what might have been due to the small sample size. [23] The mean lower CDA activity of CDA 79A>C carriers was later confirmed in healthy subjects. [24] Of note, in Japanese cancer patients, of the two examined CDA haplotypes harboring the non-synonymous SNPs CDA 79A>C ($Lys^{27}Gln$) and 208G>A ($Ala^{70}Thr$) variants, 208G>A ($Ala^{70}Thr$) was associated with higher drug-related toxicities but CDA 79A>C ($Lys^{27}Gln$) was not. [25] The recently released European FFCD-1004 Clinical Trial 'Impact of Cytidine Deaminase Activity on Clinical Outcome in Gemcitabine-Monotherapy Treated Patients' from the Fédération Francophone de Cancérologie Digestive aimed to prospectively examine impact of CDA activity affected by SNP in 128 patients with resected pancreatic cancer.[26] Surprisingly, only 5 out of 120 (4.65) of patients were found to have a CDA polymorphism and the study ultimately lacked statistical power to detect on impact on gemcitabine toxicity. Finally, both 'meta-analyses' conducted on the subject of CDA polymorphisms and outcome of gemcitabine to date, did demonstrate a moderate impact of CDA 79A >C (CDA K27Q) polymorphism on hematological toxicity in gemcitabine-treated patients quoting:

- The homozygote mutant CDA $Lys^{27}Gln/Gln$ genotype was associated with an OR of 3.25 (p=0.021)[27], and
- patients with the variant CDA 79C>A allele experienced more grade ≥3 leukopenia (OR=2.933, 95% CI 1.357-6.605) and more severe neutropenia (OR=1.313, 95% CI 0.157-10.981) [28]

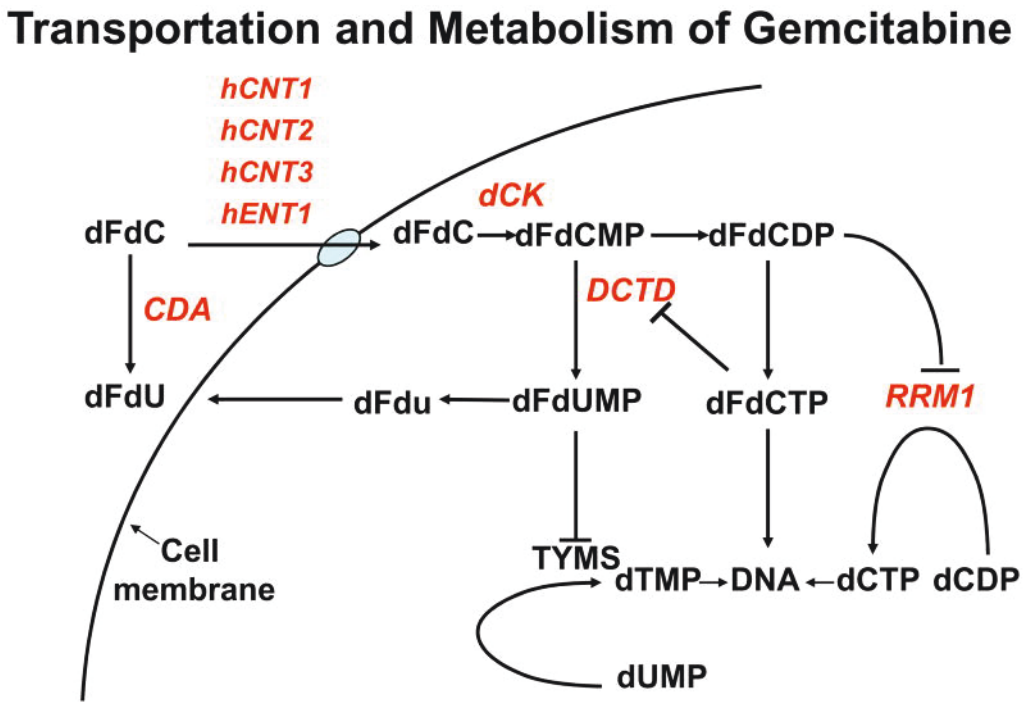


Figure 16 Key gemcitabine (dFdC) transportation and metabolism steps. Enzymes known to be affected by polymorphism highlighted in red (from Okazaki T et al, 2010).

Thus, to increase safety of patients treated with gemcitabine in combination of M7824, and in particular to account for any enhanced gemcitabine-driven toxicities due to the combination with gemcitabine, patients will be genotyped prior to study treatment with the Drug Metabolizing Enzymes and Transporter (DMET Plus) genotyping platform (Affymetrix) (patients will be co-signed onto NCI 16-C-0076 "Prospective Screening for Patient Specific Genotypes and Phenotypes that Influence Drug Dosing and Trial Selection in Cancer Patients", PI: Dr. William D. Figg). Depending on the genotyping results (turnaround time 1 – 2 weeks), patients will be stratified into low and high gemcitabine toxicity risk groups; administered starting dose of gemcitabine (Day 15) will be adjusted according to cytidine deaminase (CDA; the major enzyme responsible for gemcitabine clearance) rs2072671, CDA 79A>C polymorphism status:

- Wild type including synonymous variants of both alleles and heterozygous, monoallelic rs2072671, CDA 79A>C polymorphism (K27Q) will receive 100% of standard gemcitabine dose (1,000mg/m^2 weekly)
- Homozygous, biallelic rs2072671, CDA 79A>C polymorphisms (K27Q) will receive 60% of standard gemcitabine dose (600mg/m^2 weekly)

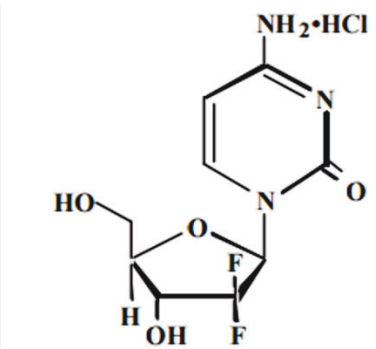
Upon Day 43, start of cycle #5, after the initial 4 doses of gemcitabine, patients who have received less than 100% of the standard gemcitabine dose (initially tested positive for either heterozygous or homozygous CDA variants) and who have not shown any gemcitabine-related toxicity will continue with standard (1,000mg/m^2) gemcitabine dose.

DMET samples will be drawn under NCI 16-C-0076 and sent to the Clinical Pharmacology Program for processing and analysis at Baseline. This analysis will be performed by the CLIA

Molecular Diagnostics Laboratory, Frederick, MD (CLIA certificate 21D0947274) through Dr. William D. Figg's Laboratory, CCR/NCI.

1.2.13 Gemcitabine in Pancreas Cancer

Gemcitabine HCl is a nucleoside analogue that exhibits antitumor activity. Gemcitabine HCl is 2´-deoxy-2´,2´-difluorocytidine monohydrochloride (β-isomer). The structural formula is as follows:



The empirical formula for gemcitabine HCl is $C_9H_{11}F_2N_3O_4$ • HCl. It has a molecular weight of 299.66. Gemcitabine HCl is a white to off-white solid. It is soluble in water, slightly soluble in methanol, and practically insoluble in ethanol and polar organic solvents. The clinical formulation is supplied in a sterile form for intravenous use only. Vials of Gemzar contain either 200 mg or 1 g of gemcitabine HCl (expressed as free base) formulated with mannitol (200 mg or 1 g, respectively) and sodium acetate (12.5 mg or 62.5 mg, respectively) as a sterile lyophilized powder. Hydrochloric acid and/or sodium hydroxide may have been added for pH adjustment.

Clinical Pharmacokinetics
Gemcitabine exhibits cell phase specificity, primarily killing cells undergoing DNA synthesis (S-phase) and also blocking the progression of cells through the G1/S-phase boundary. Gemcitabine is metabolized intracellularly by nucleoside kinases to the active diphosphate (dFdCDP) and triphosphate (dFdCTP) nucleosides. The cytotoxic effect of gemcitabine is attributed to a combination of two actions of the diphosphate and the triphosphate nucleosides, which leads to inhibition of DNA synthesis. First, gemcitabine diphosphate inhibits ribonucleotide reductase, which is responsible for catalyzing the reactions that generate the deoxy nucleoside triphosphates for DNA synthesis. Inhibition of this enzyme by the diphosphate nucleoside causes a reduction in the concentrations of deoxynucleotides, including dCTP. Second, gemcitabine triphosphate competes with dCTP for incorporation into DNA. The reduction in the intracellular concentration of dCTP (by the action of the diphosphate) enhances the incorporation of gemcitabine triphosphate into DNA (self-potentiation). After the gemcitabine nucleotide is incorporated into DNA, only one additional nucleotide is added to the growing DNA strands. After this addition, there is inhibition of further DNA synthesis. DNA polymerase epsilon is unable to remove the gemcitabine nucleotide and repair the growing DNA strands (masked chain termination).
(from https://www.accessdata.fda.gov/drugsatfda_docs/label/2014/020509s077lbl.pdf).

Human Pharmacokinetics
Gemcitabine disposition was studied in 5 patients who received a single 1,000 mg/m2 /30 minute infusion of radiolabeled drug

(https://www.accessdata.fda.gov/drugsatfda_docs/label/2014/020509s077lbl.pdf). Within one (1) week, 92% to 98% of the dose was recovered, almost entirely in the urine. Gemcitabine (<10%) and the inactive uracil metabolite, 2´-deoxy-2´,2´-difluorouridine (dFdU), accounted for 99% of the excreted dose. The metabolite dFdU is also found in plasma. Gemcitabine plasma protein binding is negligible. The pharmacokinetics of gemcitabine were examined in 353 patients, about 2/3 men, with various solid tumors. Pharmacokinetic parameters were derived using data from patients treated for varying durations of therapy given weekly with periodic rest weeks and using both short infusions (<70 minutes) and long infusions (70 to 285 minutes). The total Gemzar dose

Table 3: PK parameters of gemcitabine in human subjects.

Gemcitabine Clearance and Half-Life for the "Typical" Patient

Age	Clearance Men ($L/hr/m^2$)	Clearance Women ($L/hr/m^2$)	Half-Life[a] Men (min)	Half-Life[a] Women (min)
29	92.2	69.4	42	49
45	75.7	57.0	48	57
65	55.1	41.5	61	73
79	40.7	30.7	79	94

[a] Half-life for patients receiving a short infusion (<70 min).

varied from 500 to 3,600 mg/m2. Gemcitabine pharmacokinetics are linear and are described by a 2-compartment model. Population pharmacokinetic analyses of combined single and multiple dose studies showed that the volume of distribution of gemcitabine was significantly influenced by duration of infusion and gender. Clearance was affected by age and gender. Differences in either clearance or volume of distribution based on patient characteristics or the duration of infusion result in changes in half-life and plasma concentrations. **Table 3** shows plasma clearance and half-life of gemcitabine following short infusions for typical patients by age and gender. Gemcitabine half-life for short infusions ranged from 32 to 94 minutes, and the value for long infusions varied from 245 to 638 minutes, depending on age and gender, reflecting a greatly increased volume of distribution with longer infusions. The lower clearance in women and the elderly results in higher concentrations of gemcitabine for any given dose. The volume of distribution was increased with infusion length. Volume of distribution of gemcitabine was 50 L/m2 following infusions lasting <70 minutes, indicating that gemcitabine, after short infusions, is not extensively distributed into tissues. For long infusions, the volume of distribution rose to 370 L/m2, reflecting slow equilibration of gemcitabine within the tissue compartment. The maximum plasma concentrations of dFdU (inactive metabolite) were achieved up to 30 minutes after discontinuation of the infusions and the metabolite is excreted in urine without undergoing further biotransformation. The metabolite did not accumulate with weekly dosing, but its elimination is dependent on renal excretion, and could accumulate with decreased renal function. The effects of significant renal or hepatic insufficiency on the disposition of gemcitabine have not been assessed. The active metabolite, gemcitabine triphosphate, can be extracted from peripheral blood mononuclear cells. The half-life of the terminal phase for gemcitabine triphosphate from mononuclear cells ranges from 1.7 to 19.4 hours.

Data from 2 clinical trials evaluated the use of Gemzar in patients with locally advanced or metastatic pancreatic cancer.[13, 29] The first trial compared Gemzar to 5-Fluorouracil (5-FU) in patients who had received no prior chemotherapy. A second trial studied the use of Gemzar in pancreatic cancer patients previously treated with 5-FU or a 5-FU-containing regimen. In both studies, the first cycle of Gemzar was administered intravenously at a dose of 1,000 mg/m2 over 30 minutes once weekly for up to 7 weeks (or until toxicity necessitated holding a dose) followed by a week of rest from treatment with Gemzar. Subsequent cycles consisted of injections once weekly for 3 consecutive weeks out of every 4 weeks. The primary efficacy parameter in these studies was "clinical benefit response," which is a measure of clinical improvement based on analgesic consumption, pain intensity, performance status, and weight change. Definitions for improvement in these variables were formulated prospectively during the design of the 2 trials. A patient was considered a clinical benefit responder if either:

I. the patient showed a ≥50% reduction in pain intensity (Memorial Pain Assessment Card) or analgesic consumption, or a 20-point or greater improvement in performance status (Karnofsky Performance Scale) for a period of at least 4 consecutive weeks, without showing any sustained worsening in any of the other parameters. Sustained worsening was defined as 4 consecutive weeks with either any increase in pain intensity or analgesic consumption or a 20-point decrease in performance status occurring during the first 12 weeks of therapy. OR:

II. the patient was stable on all of the aforementioned parameters, and showed a marked, sustained weight gain (≥7% increase maintained for ≥4 weeks) not due to fluid accumulation. The first study was a multi-center (17 sites in US and Canada),

III. prospective, single-blinded, two-arm, randomized, comparison of Gemzar and 5-FU in patients with locally advanced or metastatic pancreatic cancer who had received no prior treatment with chemotherapy. 5-FU was administered intravenously at a weekly dose of 600 mg/m2 for 30 minutes. The results from this randomized trial are shown in **Table 4**. Patients treated with Gemzar had statistically significant increases in clinical benefit response, survival, and time to disease progression compared to 5-FU.[13] The Kaplan-Meier curve for survival is shown in **Figure 17**. No confirmed objective tumor responses were observed with either treatment.

Table 4 Randomized trial of gemcitabine versus 5-fluorouracil in pancreas cancer (Burris HA, 1997)

Gemzar Versus 5-FU in Pancreatic Cancer

	Gemzar	5-FU	
Number of patients Male Female	63 34 29	63 34 29	
Median age Range	62 years 37 to 79	61 years 36 to 77	
Stage IV disease	71.4%	76.2%	
Baseline KPS[a] ≤70	69.8%	68.3%	
Clinical benefit response	22.2% (N[c]=14)	4.8% (N=3)	p=0.004
Survival Median 6-month probability[b] 9-month probability[b] 1-year probability[b] Range 95% C.I. of the median	 5.7 months (N=30) 46% (N=14) 24% (N=9) 18% 0.2 to 18.6 months 4.7 to 6.9 months	 4.2 months (N=19) 29% (N=4) 5% (N=2) 2% 0.4 to 15.1+ months 3.1 to 5.1 months	p=0.0009
Time to Disease Progression Median Range 95% C.I. of the median	 2.1 months 0.1+ to 9.4 months 1.9 to 3.4 months	 0.9 months 0.1 to 12.0+ months 0.9 to 1.1 months	p=0.0013

[a] Karnofsky Performance Status.
[b] Kaplan-Meier estimates.
[c] N=number of patients.
+ No progression at last visit; remains alive.
The p-value for clinical benefit response was calculated using the two-sided test for difference in binomial proportions. All other p-values were calculated using the Logrank test for difference in overall time to an event.

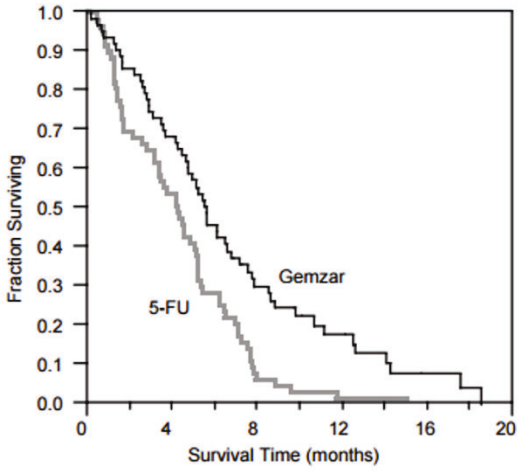


Figure 17 Overall survival by Kaplan-Meier analysis of Gemcitabine vs 5-FU systemic chemotherapy as first-line treatment in advanced pancreas cancer patients.

Clinical benefit response was achieved by 14 patients treated with Gemzar and 3 patients treated with 5-FU. One patient on the Gemzar arm showed improvement in all 3 primary parameters (pain intensity, analgesic consumption, and performance status). Eleven patients on the Gemzar arm and

2 patients on the 5-FU arm showed improvement in analgesic consumption and/or pain intensity with stable performance status. Two patients on the Gemzar arm showed improvement in analgesic consumption or pain intensity with improvement in performance status. One patient on the 5-FU arm was stable with regard to pain intensity and analgesic consumption with improvement in performance status. No patient on either arm achieved a clinical benefit response based on weight gain.

1.2.14 Clinical safety data of gemcitabine in combination with TGFβ inhibition to date.

To date limited data on TGFβ signaling inhibition combined with cytotoxic chemotherapy are available. The largest clinical experience of TGFβ inhibition in combination with systemic chemotherapy includes gemcitabine and comes from the TGFβ receptor I small molecule inhibitor galunisertib (LY2157299 monohydrate), a small molecule inhibitor of transforming growth factor-beta signaling pathway in clinical development for various solid organ malignancies including pancreatic cancer. In a phase IB study, when combined with gemcitabine, the combination (Phase I) had the expected manageable toxicity of gemcitabine in the form of myelosuppression with no increased immune-related adverse events.[30] The following large randomized phase II study (gemcitabine + galunisertib (GG) v gemcitabine + placebo (GP)) confirmed the safety of the phase I dose of galunisertib (LY2157299 monohydrate) in combination with standard dose gemcitabine (1,000mg/m2, as proposed in this study) with the majority of grade 3 and 4 AEs related to study drug and no additional toxicities synergistically induced through the combination.

The most frequent CTC grade 3/4 adverse events possibly related to study treatment (GG vs GP) were anemia (7.8% vs 13.5%), neutropenia (32.0% vs 26.9%) and thrombocytopenia (7.8% vs 9.6%) and are consistent with a gemcitabine-driven toxicity profile of the combination with no additive or new toxicity with the addition of TGFβ inhibition.

The efficacy evaluation of the study showed no statistically significant difference between the two groups, combination with the immune checkpoint inhibitor durvalumab is currently tested in clinical trials (NCT02734160; A Study of Galunisertib (LY2157299) and Durvalumab (MEDI4736) in Participants with Metastatic Pancreatic Cancer).

1.2.15 Clinical safety data of gemcitabine in combination with PD-L1 inhibition to date.

With the recognition of immunomodulatory activity of certain chemotherapeutic agents (as an extension of the concept of metronomic chemotherapy, that more cytotoxicity is not always warranted), there is a current large interest combining immune checkpoint inhibitors with systemic chemotherapy. By causing apoptotic cell death of cancer cells, chemotherapy can be immunogenic by stimulating anticancer immune effectors directly or mitigating immunosuppressive mechanisms.[31] Systemic chemotherapy may stimulate Immunosurveillance by antigenicity, immunogenicity or susceptibility.[32] Antigenicity is the result of increasing the expression and/or presentation of tumor-associated antigens on the cell surface of cancer cells. There are currently not less than 6 trials registered on clinicaltrials.gov combining gemcitabine with various immune checkpoint inhibitors. Several phase IB studies have shown safety of the combined immune checkpoint inhibitor and chemotherapy approach. As examples are listed:

Pembrolizumab, in combination with gemcitabine, gemcitabine and docetaxel, gemcitabine and nab-paclitaxel, or gemcitabine and vinorelbine or irinotecan (I) (Pembro Plus study) which showed the safety of combination with transaminitis, cytopenia’s, rash, diarrhea, fatigue, nausea and vomiting being the most common mild toxicities and efficacy signals in several tumor types

including pancreas cancer.[33] More importantly, the recently released results of the KEYNOTE-021 study in NSCLC show that overall toxicities in patients in the pembrolizumab plus chemotherapy group was similar between groups (23 [39%] of 59 patients in the pembrolizumab plus chemotherapy group and 16 [26%] of 62 in the chemotherapy alone group).[34] The most common grade 3 or worse treatment-related adverse events in the pembrolizumab plus chemotherapy group were anemia (seven [12%] of 59) and decreased neutrophil count (three [5%]); an additional six events each occurred in two (3%) for acute kidney injury, decreased lymphocyte count, fatigue, neutropenia, and sepsis, and thrombocytopenia which were also not different between the groups.[34]

Avelumab, the PD-L1 parent component of M7824, is currently in combination with standard chemotherapy in third line gastric cancer (JAVELIN Gastric 300), standard of care chemoradiotherapy (SoC CRT) in advanced head and neck carcinomas (JAVELIN Head and Neck 100), as well as other solid organ malignancies without the emerge of synergistic toxicities between avelumab and the cytotoxic chemotherapy [EMD Serono]

1.2.16 Health-related Quality of Life

It is conceivable that clinical benefit of the tested combined M7824 gemcitabine approach is limited to improvement in QoL measures of treated patients. For example, one of the initial studies of gemcitabine showed 'only' improvement in pain scores and QoL parameters but no change in traditional efficacy readouts response or survival. To evaluate the impact of M7824 in combination with gemcitabine on patients' pain scores and treatment-related reported symptoms scores of symptom subsets of the FACT-Hep (**Appendix B**) will be longitudinally completed before and during the treatment.

The 'Functional Assessment of Cancer Therapy' with the hepatobiliary and pancreatic specific module (FACT-Hep)' is a 45-item self-report instrument developed specifically to measure HRQoL in patients with hepatobiliary cancer (i.e. liver, bile duct and pancreatic cancers).[35] It assesses symptoms and other HRQoL concerns across four dimensions (physical well-being, social/family well-being, emotional well-being and functional well-being together with an 18-item disease-specific hepatobiliary cancer subscale including back and stomach pain, gastrointestinal symptoms, anorexia, weight loss and jaundice in patients with hepatobiliary cancers. It was developed by a process of item generation based on input from patients with hepatobiliary cancers, item reduction based on clinician input, scale construction and reliability/validity testing. Respondents of FACT-based questionnaires rate each item using a five-point Likert-type scale ranging from 0 (not at all) to 4 (very much).

1.2.17 Justification of Research Studies

1.2.17.1 Dynamic Contrast Enhanced MRI (DCE-MRI):

As a possible stromal modulator 'breaking the barrier of the dense microenvironment of pancreatic tumors and enabling increased drug delivery into these tumors', the impact of M7824 onto tumoral perfusion measured by Dynamic Contrast Enhanced MRI (DCE-MRI) will be compared pre-start of M7824 and after first dose M7824. Previous preclinical work (data not shown) has measured increased perfusion by DCE-MRI of murine autochthonous pancreatic tumor after pre-treatment with two doses of the TGFβ receptor I small molecule inhibitor LY364947. To evaluate of radiologic assessments via DCE-MRI can function as a reliable biomarker for response to M7824, either after monotherapy, or after M7824 in combination with gemcitabine, baseline perfusion

measures of a target lesion will be compared by serial DCE-MRI at time points designated in Study Calendar (Section **3.7**)

1.2.17.2 Somatic genomic profile of pancreatic cancers derived from specimens:

There is strong preclinical evidence, including from autochthonous models of pancreas cancer, that the underlying genotype of pancreatic cancers might govern bot, response to TGFβ as well as immune check point blockade. In particular, variants affecting the TGFβ receptor - SMAD axis, have been cited as potentially protective against an initial pro-tumor effect of TGFβ inhibition due to the tumor suppressive function of TGFβ in the epithelial cancer compartment. Thus, early correlations between genotype and clinical outcome of patients aim to (1) improve future selection of patients for the dual TGFβ 'trap' PD-L1 blockade in combination with gemcitabine approach as well as (2) improve understanding of the mechanism of action of M7824 in pancreas cancer.

1.2.17.3 Serum cytokines and serum TGFβ levels

One of the least elaborate ways to confirm that the administered M7824 is active and is engaging its target, is to measure suppression of TGFβ levels (via 'trapping') and induced changes in the cytokine profile systemically. Following TGFβ and cytokine levels longitudinally across treatment course will allow detection of lack of M7824 activity and can trigger additional investigations (ADA development) into lack of efficacy.

1.2.17.4 ADA

Anti-Drug Antibody development is an accepted mechanism of loss of efficacy administered human monoclonal antibodies. Measuring titers will ensure that lack of efficacy of M7824 is not due to ADA development.

1.2.17.5 Evaluate the intratumoral immunogenicity of M7824 in combination with gemcitabine on pre- and on-treatment biopsies:

Preclinical studies show that TGFβ inhibition and gemcitabine cooperate immunologically to induce anti-tumor responses reducing immune evasive cues of the intratumoral immune milieu and enhancing T cell-mediated anti-tumor responses. The flow cytometry studies on pre- and on-treatment tumor biopsies aim to (1) investigate immunogenic cooperativity of M7824 in combination with gemcitabine in human tumors, (2) identify an immunologic landscape likely to response to the pursued combination approach, and (3) aid to develop longitudinal predictive biomarkers for later improved patient selection (correlation with immune cell changes pre- and on-treatment and clinical outcome).

1.2.17.6 Rationale for circulating free tumor DNA measures:

Measures of copies of genomic tumor DNA released into the circulation has been shown to be an accurate measure of tumor burden in patients with solid organ cancers and to have value to follow response to anti-cancer treatment both for the early identification of responders as well as patients to recur after remission. Including longitudinal measurements from baseline and on-treatment of circulating free tumor DNA into explorative objectives examines the value of identifying responders early and the development of this test as a predictive biomarker for the tested M7824 gemcitabine combination treatment.

1.2.17.7 Rationale for abbreviated PK studies M7824 and gemcitabine:

Abbreviated PK measurements of both M7824 and gemcitabine will be taken to exclude that M7824 and gemcitabine pharmacology are interdependent (M7824 plasma Cmax and Cmin levels will be compared to PK measures taken during M7824 1,200mg flat dose monotherapy expansion cohorts, gemcitabine plasma Cmax and Cmin levels will be correlated to initial DMET genotyping and compared between the two gemcitabine toxicity risk groups). Gemcitabine PKs will be used to adjust gemcitabine schedule during the study, patients in the high-risk group who have not shown any toxicity and whose PKs don't show any elevated Cmax and Cmin levels can have their doses increased to the standard 1,000 mg/m^2 level.

1.2.18 Amendment B

With this amendment we want to update DLT definition. Originally, we inadvertently left out DLT definitions and did not exclude adverse events with clear evidence of an alternative causality or expected and shortly resolved.

As a result, there were DLTs, that in the opinion of the PI, are not related to study treatment or expected and can be excluded but were not mentioned as exclusions.

So, adhering to the current DLT definition in the protocol capturing 'Any DLT . . . (Section **3.1.1**), we have 2 DLTs and per protocol need to go to lower dose of study drug M7824. However, we are reluctant to do this, as (1) the combination treatment is generally well tolerated in the 6 patients on study so far, and (2) there are some early signs of clinical efficacy (no patient came off study yet due to progression of disease, there are 2 biochemical responses) with original dose level 0 (1,200mg flat dose M7824) and we do not want to risk the potential benefit with lowering of M7824 dose unnecessarily, in particular in a disease like pancreatic cancer where drug delivery and drug penetration is a major issue.

In several hundreds of patients treated with the standard 1,200mg flat dose, many of these patients treated at the Clinical Center, M7824 was found to have a very favorable side-effect profile nearly identical to approved immune checkpoint inhibitors like pembrolizumab or nivolamab. While we want to capture any possible additive toxicity signal when combining M7824 and gemcitabine, we also do not want to unnecessarily lower M7824 dose levels in a disease where the lack of sufficient intratumoral exposure levels due to the uniqueness of pancreas cancer (high interstitial pressures, poor vascularization) is a major impediment to the efficacy of approved cytotoxic therapies.

While we have not seen any formal PRs yet (for two patients also too early), none of the pancreas cancer patients treated so far have shown despite widely metastatic disease radiologic progression of disease (all stable disease).

There have been two large drops (>50%) in tumor marker levels occurring after >2 months of therapy (which we consider consistent with the mechanism of immunotherapy).

Dose level -1 (500 mg of M7824) potentially will deprive patients of efficacious doses in a disease where drug penetration and low intratumoral drug levels are a significant impediment of therapeutic efficacy and should only occur if the new drug combination is found not safe at the respective dose level.

With this amendment we want to omit current patients from the DLT analysis and repeat the entire safety run-in with 6 new study subjects under the new DLT criteria.

2 ELIGBILITY ASSESSMENT AND ENROLLMENT

2.1 Eligibility Criteria

2.1.1 Inclusion Criteria

- Patient must be able to understand and willing to sign a written informed consent document
- Age ≥18 years. Because no dosing or adverse event data are currently available on the use of M7824 in combination with gemcitabine in patients <18 years of age, children are excluded from this study, but will be eligible for future pediatric trials
- Histologically or cytologically proven pancreatic adenocarcinoma (subjects with endocrine or acinar pancreatic carcinoma are not eligible).
- Patients must have disease that is not amenable to potentially curative resection.
- Subjects must have progressed on or after standard first-line systemic chemotherapy.
- ECOG performance status of 0 to 1 (**Appendix A)**
- Must have evaluable or measurable disease per RECIST 1.1. See Section **6.3** for the evaluation of measurable disease.
- Adequate hematological function defined by:
  - white blood cell (WBC) count ≥ 3×10^9/L
  - with absolute neutrophil count (ANC) ≥ 1.5×10^9/L,
  - lymphocyte count ≥ 0.5×10^9/L,
  - platelet count ≥120×10^9/L, and
  - Hgb ≥9 g/ dL (more than 48 hours post-completion of blood transfusion))
- Adequate hepatic function defined by:
  - a total bilirubin level ≤ 1.5 × ULN,
  - an AST level≤2.5×ULN,
  - ALT level ≤2.5 ×ULN.
- Adequate renal function defined by:
  - Creatinine up to 1.5-times upper institutional limits OR creatinine clearance (CrCl) >50 mL/min/1.73 m^2 OR within normal as predicted by the Cockcroft-Gault formula:

$$\text{CrCl} = \frac{(140 - \text{age (y)}) \times (\text{weight in kg}) \times (0.85, \text{if female}) \times 1.73\ m^2}{72 \times \text{Serum Creatinine (mg/dL)} \times \text{pt's BSA } (m^2)}$$

- The effects of the study treatment on the developing human fetus are unknown; thus, women of childbearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) within 28 days prior to study entry, for the duration of study participation and up to 120 days after the last dose of the drug. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately.

2.1.2 Exclusion Criteria

- Patients who are receiving any other investigational agents
- Prior therapy with any antibody / drug targeting T cell coregulatory proteins (immune checkpoints) such as anti-PD-1, anti-PD-L1, or anti-CTLA-4 antibody.
- Anticancer treatment within designated period before enrollment including:
  - minor surgical procedure (such as biliary stenting) within 14 days
  - major surgical procedure or radiation treatment within 28 days
  - chemotherapy or experimental drug treatment with published half-life known to be 72 hours within 14 days
  - experimental drug treatment with unpublished or half-life greater than 72 hours within 28 days
  - radiotherapy for measurable lesions delivered in a normal organ-sparing technique within 21 days (except for palliative radiotherapy)
- Concurrent treatment with non-permitted drugs including herbal remedies with immunostimulating properties (for example, mistletoe extract) or known to potentially interfere with major organ function (for example, hypericin) (see Section **4.2**).
- Previous malignant disease (other than the target malignancy to be investigated in this trial) within the last 3 years. Subjects with a history of cervical carcinoma in situ, superficial or non-invasive bladder cancer, or basal cell or squamous cell carcinoma in situ previously treated with curative intent are NOT excluded.
- Rapidly progressive disease which, in the opinion of the Investigator, may predispose to inability to tolerate treatment or trial procedures.
- Subjects with active central nervous system (CNS) metastases causing clinical symptoms or metastases that require therapeutic intervention are excluded. Subjects with a history of treated CNS metastases (by surgery or radiation therapy) are not eligible unless they have fully recovered from treatment, demonstrated either no clinical or radiographic progression for at least 2 months, and do not require continued steroid therapy. Subjects with CNS metastases incidentally detected during Screening which do not cause clinical symptoms and for which standard of care suggests no therapeutic intervention is needed are eligible.
- Receipt of any organ transplantation, including allogeneic stem-cell transplantation, except transplants that do not require immunosuppression (e.g., corneal transplant, hair transplant)
- Significant acute or chronic infections including tuberculosis (history of exposure or history of positive tuberculosis test; plus, presence of clinical symptoms, physical or radiographic findings)
- Active autoimmune disease that might deteriorate when receiving an immunostimulatory agent with the exceptions:
  - diabetes type I, vitiligo, alopecia, psoriasis, hypo- or hyperthyroid disease not requiring immunosuppressive treatment are eligible;

- subjects requiring hormone replacement with corticosteroids are eligible if the steroids are administered only for the purpose of hormonal replacement and at doses ≤ 10 mg of prednisone or glucocorticoid equivalent per day;
- administration of steroids for other conditions through a route known to result in a minimal systemic exposure (topical, intranasal, intra-ocular, or inhalation) is acceptable.

- Known severe hypersensitivity reactions to monoclonal antibodies (Grade ≥3 NCI-CTCAE v5.0), any history of anaphylaxis or history of uncontrolled asthma.
- Known severe hypersensitivity to gemcitabine.
- Female patients who are pregnant or breastfeeding. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with M7824 in combination with gemcitabine, breastfeeding should be discontinued.
- Known alcohol or drug abuse.
- Clinically significant cardiovascular / cerebrovascular disease as follows: cerebral vascular accident / stroke (< 6 months prior to enrollment), myocardial infarction (< 6 months prior to enrollment), unstable angina, congestive heart failure (New York Heart Association Classification Class ≥ II), or serious cardiac arrhythmia.
- Clinically relevant diseases (for example, inflammatory bowel disease) and / or uncontrolled medical conditions, which, in the opinion of the Investigator, might impair the subject's tolerance or ability to participate in the trial.
- Vaccine administration of live attenuated vaccines within 28 days of enrollment.
- Patients with known contrast allergies requiring pre-medication with steroids.
- HIV, HCV, HBV positive patients on antiviral drugs are excluded due to the absence of previous experience with concurrent use of antiviral medications and the investigational drug product to be evaluated in the current study and possible for adverse pharmacokinetic and/or pharmacodynamic interactions.
- Known inherited bleeding disorder and/or history of bleeding diathesis such as vWF deficiency.

2.1.3 Recruitment Strategies

The study will be posted on the CCR website and on clinicaltrials.gov.

2.2 Screening Evaluation

Within 28 days prior to enrollment unless otherwise noted below:

- Complete medical history and physical examination, including height, weight, vital signs, EKG, and ECOG performance status.
- Laboratory Evaluation
  - Hematological profile: CBC with differential and platelet count;

- Biochemical profile: electrolytes, BUN, creatinine, AST, ALT, total bilirubin, calcium, phosphorus, albumin, magnesium, uric acid;
- TB testing (if clinically indicated);
- Serum or urine pregnancy test for female participants of childbearing age (in the absence of prior hysterectomy) (7 days prior to enrollment);

- CT of chest, abdomen and pelvis (or MRI abdomen);
- Tumor evaluation / tumor measurements;
- A brain CT / MRI scan if clinically indicated;
- Histologic or cytologic confirmation (at any time point prior to enrollment). If there is no available documentation, biopsy will be performed to confirm the diagnosis.

2.3 Registration Procedures

Authorized staff must register an eligible candidate with NCI Central Registration Office (CRO) within 24 hours of signing consent. A registration Eligibility Checklist from the web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) must be completed and sent via encrypted email to: NCI Central Registration Office ncicentralregistration-l@mail.nih.gov. After confirmation of eligibility at Central Registration Office, CRO staff will call pharmacy to advise them of the acceptance of the patient on the protocol prior to the release of any investigational agents. Verification of Registration will be forwarded electronically via e-mail to the research team. A recorder is available during non-working hours.

2.3.1 Treatment Assignment Procedures (**For registration purposes only**):

Cohorts

Number	Name	Description
1	Phase IB	Subjects with pancreas cancer enrolled to M7824 de-escalation dose levels.
2	Phase II	Subjects with pancreas cancer enrolled at the RP2D of M7824 after the RP2D is established

Arms

Number	Name	Description
1	Arm 1	Gemcitabine (dose based on genetic testing results) + de-escalating dose of M7824
2	Arm 2	Gemcitabine (dose based on genetic testing results) + RP2D of M7824

Arm assignment

Subjects in Cohort 1 will be directly assigned to Arm 1.

Subjects in Cohort 2 will be directly assigned to Arm 2.

2.4 Baseline Evaluation

Tests done at screening do not need to be repeated on baseline if performed in designated time frame prior to start of M7824 infusion.

Within 28 days prior to first dose:

- CT of chest, abdomen and pelvis (or MRI abdomen)
- Tumor evaluation / measurements
- Optional tumor research biopsy (only in patients enrolled onto the phase II)
- Research blood for DMET genotyping (see section **5.3**)
- Research Perfusion evaluation – DCE-MRI
- Research Quality of Life assessment (subject-reported outcomes / symptom severity assessments the 4th version of the 'Functional Assessment of Cancer Therapy with the hepatobiliary and pancreatic specific module (FACT-Hep **Appendix B**). (For English speaking subjects only).

3 STUDY IMPLEMENTATION

3.1 Study Design

This is an open label Phase IB/II trial, accruing initially one cohort to determine the safety and recommended Phase II dose (RP2D) of M7824 in combination with gemcitabine (Phase IB); and to examine the safety and efficacy of the M7824 in combination with gemcitabine in the following cohort (Phase II).

Up to 2 dose levels of M7824 will be tested in the safety run-in cohort IB with up to 12 subjects enrolled. Once a recommended phase 2 dose has been determined, up to 20 subjects will be evaluated at that dose level in the Phase II cohort, inclusive of those patients treated at the RP2D during the safety run-in.

Patients will receive treatment in cycles consisting of 14 (+/- 3) days.

Administration of M7824 will be every 2 weeks by IV infusion starting on day 1 of cycle 1. The starting M7824 dose is 1,200 mg.

Gemcitabine will be administered IV once a week starting on day 1 of cycle 2 for 4 weeks (cycles 2-3). Then, once weekly for 3 weeks with one week without gemcitabine starting on day 1 of cycle 4 (cycles 4-13).

Gemcitabine will be discontinued after 6 months of total gemcitabine therapy.

After gemcitabine discontinuation, treatment with M7824 will continue until patient meets off treatment criteria (Section **3.8.1)**.

In all patients, gemcitabine dosage will be adjusted according to DMET genotyping and risk profiling results; patients with low risk (wild type including synonymous variants of both alleles and heterozygous, monoallelic rs2072671, CDA 79A>C polymorphism (K27Q)) or high risk (homozygous, biallelic rs2072671, CDA 79A>C polymorphisms (K27Q)).

Patients with low risk will be assigned to standard dosage of 1,000 mg/m^2 per dose.

Patients with high risk will be started on 600 mg/m^2 per dose for first 4 doses of gemcitabine. After 4 doses of gemcitabine, patients in this category will continue on 600 mg/m^2 per dose or standard 1,000 mg/m^2 per dose (see Section **3.3.2**).

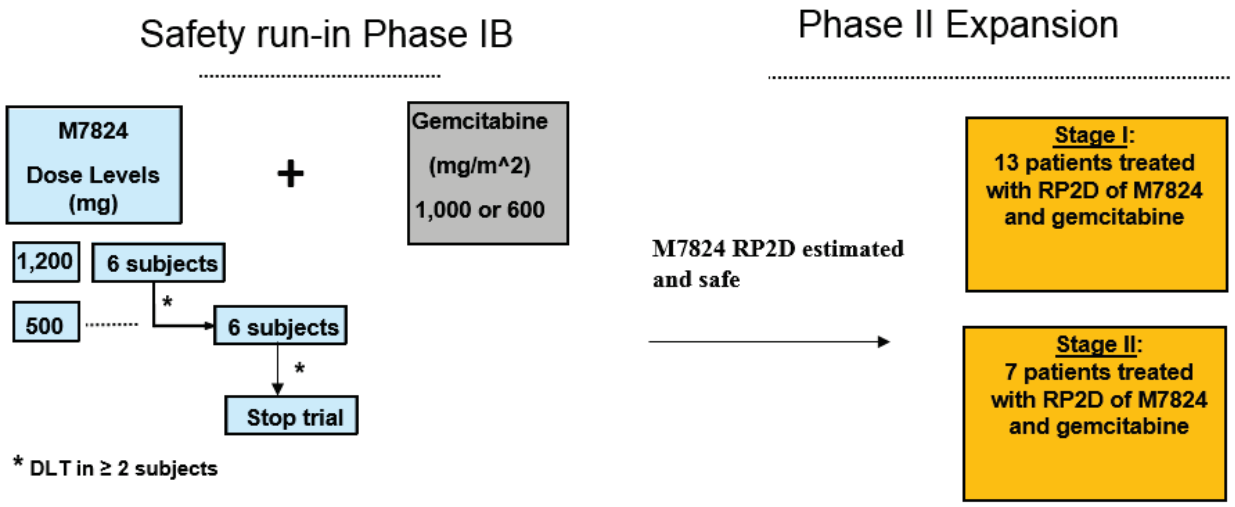


Figure 18 Design of phase IB/II study of M7824 in combination with gemcitabine in patients with advanced pancreas cancer.

3.1.1 Dose Limiting Toxicity

The DLT period is two cycles, 28 days.

DLT is defined as follows:

- Febrile neutropenia of any grade
- Any Grade ≥ 3 adverse event (AE), occurring during the DLT evaluation period, **except** for those listed below:
  - Adverse event for which there is a clear evidence of an alternative causality (i.e., other than M7824 or the study combination)
  - Grade 3 or 4 neutropenia if lasts < 1 cycle (14 days)
  - Grade 3 thrombocytopenia if lasts < 1 cycle (14 days)
  - Any Grade 3 autoimmune thyroid-related toxicity that clinically resolve to ≤ Grade 2 within 7 days of initiating therapy
  - Grade 3 diarrhea or skin toxicity that resolves to Grade ≤ 1 in less than 7 days after medical management (e.g., immunosuppressant treatment) has been initiated
  - Tumor flare phenomenon defined as local pain, irritation, or rash localized at sites of known or suspected tumor.

- Transient (≤ 24 hours) Grade 3 flu-like symptoms or fever, which is controlled with medical management.
- Transient (≤ 48 hours) Grade 3 fatigue, local reactions, headache, nausea, emesis that resolves to ≤ Grade 1 or Baseline grade.
- Any isolated and single Grade 3 liver function test abnormality in the form of transaminase, alkaline phosphatase, or bilirubin elevation with NO other associated laboratory (or other) abnormality that is not associated with symptoms or clinical manifestations of hepatitis or any immune-related event and which has resolved to Grade ≤ 1 within the subsequent cycle (14 days)
- Any isolated and single Grade 3 amylase or lipase abnormality with no other associated laboratory (or other) abnormality that is not associated with symptoms or clinical manifestations of pancreatitis and which has resolved to Grade ≤ 1 within the subsequent cycle (14 days)
- Any isolated and single Grade 3 Hgb decrease (< 8.0 g/dL) that is clinically manageable with blood transfusions or erythroid growth factor and has resolved to Grade 2 not requiring blood transfusions or erythroid growth factor therapy with the subsequent cycle (14 days).
- Grade 3 infusion-related reactions resolving within 6 hours from the end of infusion and controlled with medical management.
- Skin lesion related to TGFβ inhibition (i.e. keratoacanthoma, squamous cell carcinoma) that is local and can be resected with negative resection margins,
- Grade 3 skin toxicity that resolves to ≤ Grade 1 in less than 7 days after medical management (eg. immunosuppressant treatment, adequate and optimal anti diarrhea therapy) has been initiated.
- Single laboratory values out of normal range that are assessed as unrelated to study treatment according to the investigator, do not have any clinical correlate, and resolve to ≤ Grade 1 within 7 days with adequate medical management

3.1.2 Phase IB Safety Run-In (Dose de-escalation)

Following Amendment B, participants will be enrolled to 2 dose levels as follows:

The first 3 participants will be enrolled on DL0 and if 0/3 or 1/3 have a DLT during DLT period, the next group of 3 participants will be enrolled. If no more than 1/6 experience a DLT during DLT period, accrual may proceed in the phase II expansion cohort and DL0 will be the RP2D of M7824.

If 2 or more in 3-6 subjects at DL0 have a DLT during DLT period, then subsequent subjects will enroll at DL-1.

If 0/3 or 1/3 have a DLT at DL-1 during DLT period, then the next group of 3 participants will be enrolled at DL-1. If no more than 1/6 experience a DLT, then accrual may proceed to the phase II expansion cohort and DL-1 will be the RP2D of M7824.

If 2 or more in 3-6 subjects at DL-1 have a DLT during DLT period, then , no further attempts at completing the safety run-in will be conducted and the protocol will be closed to further accrual.

The dose de-escalation scheme in the safety run-in will use the following dose levels (**Table 5**):

Table 5 Dose modification in safety run-in of the study (Phase IB).

Dose level	M7824, mg, IV every 2 weeks	Gemcitabine, mg/m^2, IV every week* Dose assigned according to polymorphism testing
DL 0	1,200	Assigned dose
DL -1	500	Assigned dose
*Starting in cycle 2 (day 15) gemcitabine will be administered weekly for 4 weeks, then 3 weeks on 1 week off for no more than 6 months total of gemcitabine therapy		

Every subject in each dose level group of the safety run-in will be observed for at least 7 days after first dose of M7824 before the subsequent subject can be treated.

Subjects who do not complete the DLT observation period for reasons other than a DLT will be replaced and not included in the evaluation.

Once a total of 6 subjects have been treated with ≤1 patient having DLT and 28 days of observation have passed (completion of DLT period) after the first treatment of the 6th subject, patients can be accrued to the phase II expansion cohort.

The recommended phase 2 dose (RP2D) is the dose level of M7824 at which ≤1 individual experienced a DLT during the first 2 cycles of treatment (28 days).

In case of DLT patients will be taken off study treatment (M7824 and gemcitabine)

3.1.2.1 M7824

Intra-patient M7824 dose de-escalation is not permitted for subjects enrolled in the safety run-in cohort. Subject enrolled onto safety run-in cohorts who have either had a DLT or completed the DLT evaluation period will be part of the DLT evaluation of the respective phase I dose level.

3.1.2.2 Gemcitabine

During Phase I portion of this trial, gemcitabine starting dose levels will be adjusted according to results of polymorphism testing of the CDA gene using the DMET platform of the Clinical Pharmacology Program (as per NCI 16-C-0076) (See Section **3.3.2**).

If patients develop side effects related to gemcitabine, dose modifications according to label are recommended:

Dose modifications dosage adjustment is based upon the degree of hematologic toxicity experienced by the patient. Clearance in women and the elderly is reduced and women are less able to progress to subsequent cycles. Patients will be monitored prior to each dose with a complete blood count (CBC), including differential and platelet count. If marrow suppression is detected, gemcitabine therapy should be modified or suspended according to the guidelines in Section **3.4.2.** These subjects will be part of the DLT evaluation of the respective M7824 dose level they were accrued to. Failure to resume gemcitabine after a hold of ≥2 week will be analyzed as a DLT within respective dose level group.

For patients receiving 600 mg/m^2 gemcitabine per dose gemcitabine dosage will not be reduced and gemcitabine will be withheld.

3.1.3 Phase II Cohort

The efficacy part of the study will be conducted with the M7824 dose level found to be safe in combination with gemcitabine during phase IB. Once a recommended phase 2 dose of M7824 has been determined, up to 20 subjects will be evaluated at that dose level, inclusive of those patients treated at the RP2D during the safety run-in.

Enrollment into phase II part of the study can commence after DLT period of 6th subject of respective DLT cohort is complete.

Gemcitabine dosage will be assigned per Section **3.3.2**

During first stage of Phase II 13 patients will be enrolled. If 0 of 13 patients respond, then no further patients will be enrolled. If 1 or more of the first 13 evaluable patients enrolled have a response (PR or CR), then during second stage accrual will continue until a total of 20 evaluable patients have been enrolled. Since it may take several weeks for a response to be identified, a pause in the accrual of 6 weeks will be made before accruing to the second stage.

3.2 Study Stopping Rules

For safety reasons, the protocol will be temporarily halted until an expedited safety report is sent to and reviewed by the FDA and the SAE has been evaluated by the investigators for either of the following events attributable to treatment regimen occurring within 30 days of receiving investigational agents:

- One occurrence of grade 5 toxicity.
- Two occurrences of grade 4 toxicity.

3.3 Drug Administration

3.3.1 M7824.

M7824 will be administered as a 1-hour (-10 minutes / +20 minutes) IV infusion on Day 1 of each cycle (14 days) before gemcitabine.

M7824 is administered as an intravenous (IV) infusion via a peripheral OR central vascular access device (VAD). Confirm patient has a titanium port before accessing the Central VAD. A 0.2-micron polyethersulfone (PES) in-line filter is mandatory for administration.

In order to mitigate potential infusion-related reactions, premedication with an antihistamine and with acetaminophen (for example, 25-50 mg diphenhydramine po and 500-1,000 mg acetaminophen po or IV equivalent) approximately 60 to 120 minutes prior to each dose of M7824 is mandatory for the first 2 infusions and is optional and at the discretion of the Investigator after the second infusion. If Grade $\geq$ 2 infusion reactions are seen during the first two infusions, premedication should not be stopped. Steroids as premedication are not permitted.

As with all monoclonal antibody therapies, there is a risk of allergic reaction including anaphylactic shock. M7824 should be administered in a setting that allows for immediate access to an intensive care unit or equivalent environment and administration of therapy for anaphylaxis, such as the ability to implement immediate resuscitation measures. Steroids (dexamethasone 10

mg), epinephrine (1:1,000 dilution), allergy medications (IV antihistamines), bronchodilators, or equivalents, and oxygen should be available for immediate access.

Patients must be observed for 2 hours after the first M7824 dose. If no reactions are observed, the patients need to be monitored for only 30 minutes after subsequent doses.

If an allergic reaction occurs, the subject must be treated according to the best available medical practice. Please see the guidelines for handling of infusion-related reaction in **Table 6**.

Investigators should also monitor subjects closely for potential irAEs, which may become manifest after several weeks of treatment. Such events may consist of persistent rash, diarrhea and colitis, autoimmune hepatitis, arthritis, glomerulonephritis, cardiomyopathy, or uveitis and other inflammatory eye conditions.

For vital signs measurements see Study Calendar (Section **3.7)**.

3.3.2 Gemcitabine

Gemcitabine starting dose levels will be adjusted according to results of polymorphism testing of the CDA gene using the DMET platform of the Clinical Pharmacology Program (as per NCI 16-C-0076):

DMT testing - CDA polymorphism	Gemcitabine, mg/m^2, IV, first 4 doses	Gemcitabine, mg/m^2, IV following 4 doses
Wild type including synonymous mutation or heterozygous, monoallelic rs2072671, CDA 79A>C polymorphism (K27Q)	1,000	1,000
Homozygous	600	1,000 if no ≥3 grade toxicity and gemcitabine Cmax measures <30µg/mL
		600 if no ≥3 grade toxicity or ≥3 grade toxicity resolved within 2 weeks and gemcitabine Cmax measures ≥30µg/mL
		Discontinue gemcitabine if ≥3 grade toxicity is not resolved within 2 weeks

On day 1 of cycles 2, 3, 4 and 5 gemcitabine infusion will immediately commence upon completion of M7824 infusions as gemcitabine PK must be collected. On all other days when both drugs are given on the same day, gemcitabine may be administered anytime on this day after M7824

infusion. In case of local infusion reactions, contralateral extremities or other access routes should be used for gemcitabine infusions.

Gemcitabine will be given starting on Cycle 2 at the assigned dosage over 30 (+/- 10) minutes once weekly for the first consecutive 4 weeks (**Figure 19**). Starting on Cycle 4 once weekly for 3 weeks with one week without gemcitabine.

Gemcitabine will be discontinued after cycle 13 (6 months total of gemcitabine treatment).

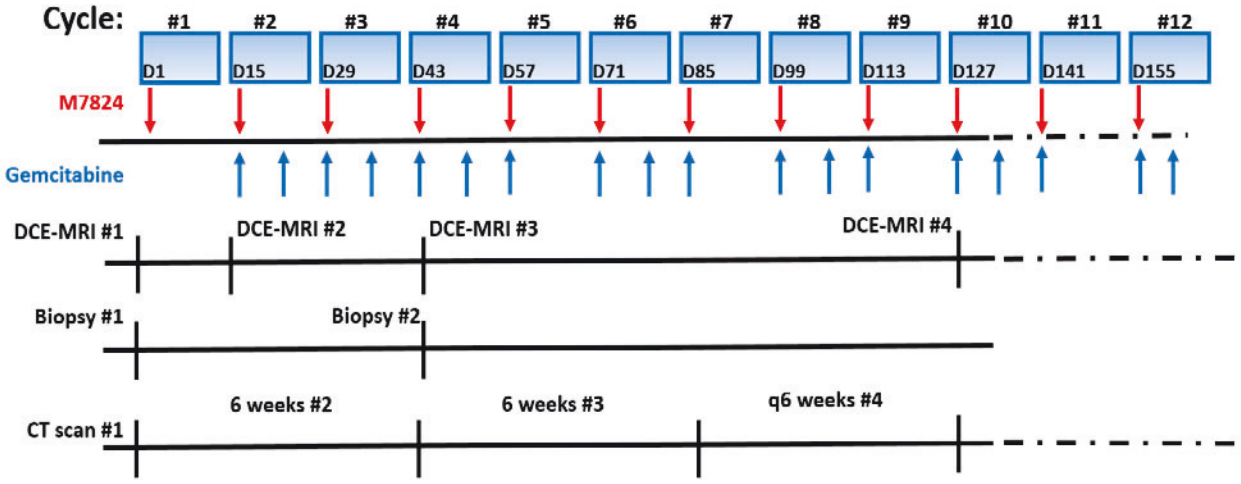


Figure 19 Schedule of drug administration of M7824 and gemcitabine and assessments on treatment including DCE-MRI and tumor biopsies.

3.4 Dose modifications:

3.4.1 M7824

During Phase IB dose modification of M7824 are not allowed.

During Phase II modification of M7824 will proceed as described in this section.

3.4.1.1 Adverse Drug Reactions (ADRs) Requiring Treatment Discontinuation

Certain ADRs, defined as an AE assessed as related to M7824 by the Investigator, may require dose interruption, or permanent treatment discontinuation of M7824. For certain ADRs assessed to be immune-related, **Table 7**, Management of Immune-Related Adverse Events criteria may supersede this section. These criteria may allow the subject to continue study if medically indicated.

Any Grade 4 ADRs require permanent treatment discontinuation except for single laboratory values out of normal range that do not have any clinical correlate and resolve to Grade ≤ 1 or Baseline grade within 7 days with adequate medical management.

Any Grade 3 ADRs require treatment discontinuation except for any of the following:

- Transient (≤ 6 hours) Grade 3 flu-like symptoms or fever, which is controlled with medical management.
- Transient (≤ 24 hours) Grade 3 fatigue, local reactions, headache, nausea, emesis that resolves to ≤ Grade 1 or Baseline grade.
- Tumor flare phenomenon defined as local pain, irritation, or rash localized at sites of known or suspected tumor.
- Any single Grade ≥ 3 drug-related transaminase, alkaline phosphatase, or bilirubin abnormality with no other associated laboratory (or other) abnormality that is not associated with symptoms or clinical manifestations of hepatitis. If the liver function abnormality not associated with symptoms or clinical manifestations of hepatitis has not resolved to Grade ≤ 1 within the subsequent 2 cycles (28 days), the subject should permanently discontinue treatment with M7824.
- Any single Grade ≥ 3 drug-related amylase or lipase abnormality with no other associated laboratory (or other) abnormality that is not associated with symptoms or clinical manifestations of pancreatitis. If the amylase or lipase abnormality not associated with symptoms or clinical manifestations of pancreatitis has not resolved to Grade ≤ 1 within the subsequent 2 cycles (28 days), the subject should permanently discontinue treatment with M7824.
- Grade 3 Hgb decrease (< 8.0 g/dL) that is clinically manageable with blood transfusions or erythroid growth factor use does not require treatment discontinuation.
- Increases in Eastern Cooperative Oncology Group performance status (ECOG PS) ≥ 3 that resolves to ≤ 2 by Day 1 of the next cycle (infusions should not be given if the ECOG PS is ≥ 3 on the day of planned M7824 administration and should be delayed until ECOG PS ≤ 2).
- Keratoacanthoma and squamous cell carcinoma of the skin. Any suspicious skin lesion should be biopsied and be surgically removed and a dermatological consult obtained.
- Grade 3 or 4 symptomatic endocrinopathies (e.g., thyroiditis or hypophysitis), treatment should be delayed, and treatment started according to (**Table 7**). If condition improves to Grade 1, treatment may be resumed. If ≥ 2 consecutive doses are missed, discontinuation of M7824 should be considered.
- Other immune-related ADRs, see Immune-Related Adverse Events (**Table 7**)

3.4.1.2 Adverse Drug Reactions Requiring Management:

Any Grade 2 ADR should be managed as follows:

- If a Grade 2 ADR resolves to Grade ≤ 1 by the last day of the current cycle, treatment may continue.
- If a Grade 2 ADR does not resolve to Grade ≤ 1 by the last day of the current cycle but it is manageable and / or not clinically relevant, it is possible the infusion will be given on the following cycle. If at the end of the following cycle, the event has not resolved to Grade 1, permanently discontinuing of treatment with M7824 should be considered.

- Upon the second occurrence of the same Grade 2 ADR in the same subject (except for fatigue and hormone insufficiencies that can be managed by replacement therapy), permanently discontinuing treatment with M7824 should be considered.
- Infusion-related reactions and hypersensitivity reactions (Grades 1 to 4) should be handled according to the Guidelines provided in **Table 6**.

3.4.1.3 Infusion-related Reactions

Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion.

Symptoms:

- Fever
- Chills
- Rigors
- Diaphoresis
- Headache

Management: (please see **Table 6**)

Table 6 Treatment Guidelines for Symptoms of Infusion-related Reactions Caused by M7824

NCI-CTCAE Grade	Infusion of M7824	Treatment	Premedication at subsequent dosing
Grade 1 – mild Mild transient reaction; infusion interruption not indicated; intervention not indicated.	Decrease the M7824 infusion rate by 50% and monitor closely for any worsening. The total infusion time for M7824 should not exceed 120 minutes	Increase monitoring of vital signs as medically indicated until the subject is medically stable.	None
Grade 2 – moderate Therapy or infusion interruption indicated but responds promptly to symptomatic treatment (for example, antihistamines, NSAIDs, narcotics, IV fluids); prophylactic	Stop M7824 infusion. If symptoms resolve within one hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g., from 100 mL/hr. to 50 mL/hr.). Otherwise dosing will be held until symptoms resolve and the subject	**Monitor symptoms.** Therapy include but is not limited to: IV fluids Antihistamines NSAIDS Acetaminophen Opioids Increase monitoring of vital signs as medically indicated until the	Subject to be pre-medicated 1.5h (± 30 minutes) prior to infusion of M7824 with: Diphenhydramine 50 mg po (or equivalent dose of antihistamine). Acetaminophen 500-1,000 mg PO (or equivalent

NCI-CTCAE Grade	Infusion of M7824	Treatment	Premedication at subsequent dosing
medications indicated for ≤ 24 hours.	should be pre-medicated for the next scheduled dose.	subject is medically stable. **Subjects who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further trial treatment administration.**	dose of antipyretic).
Grade 3 • Grade 3: Prolonged (for example, not rapidly responsive to symptomatic medication and / or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalization indicated for clinical sequelae.	• Stop the M7824 infusion immediately and disconnect infusion tubing from the subject. If resolved within 6 hours, infusion may be restarted by discretion of PI	Therapy includes but is not limited to: IV fluids Antihistamines NSAIDS Acetaminophen Opioids Oxygen Pressors Corticosteroids Epinephrine Increase monitoring of vital signs as medically indicated until the subject is medically stable. Hospitalization may be indicated. **Subjects who develop Grade 3 toxicity despite adequate premedication should be permanently discontinued from further trial**	Subject to be pre-medicated 1.5h (± 30 minutes) prior to infusion of M7824 with: Diphenhydramine 50 mg po (or equivalent dose of antihistamine). Acetaminophen 500-1,000 mg PO (or equivalent dose of antipyretic).

NCI-CTCAE Grade	Infusion of M7824	Treatment	Premedication at subsequent dosing
		treatment administration.	
• Grade 4: Life-threatening consequences; urgent intervention indicated.	• Subjects have to be withdrawn immediately from M7824 treatment and must not receive any further M7824 treatment	Therapy includes but is not limited to: IV fluids Antihistamines NSAIDS Acetaminophen Opioids Oxygen Pressors Corticosteroids Epinephrine Increase monitoring of vital signs as medically indicated until the subject is medically stable. Hospitalization may be indicated.	No subsequent dosing

NSAIDs=nonsteroidal anti-inflammatory drugs.

Additional Modifications for Subjects with Grade 2 Infusion-related Reactions

If, in the event of a Grade 2 infusion-related reaction that does not improve or worsens after implementation of the modifications indicated in **Table 6** (including reducing the infusion rate by 50%), the Investigator may consider treatment with corticosteroids and the infusion of M7824 should be stopped for that day. At the next infusion, the Investigator may consider the addition of H2-blocker antihistamines (for example, famotidine or ranitidine), in addition to premedication, for select subjects. However, prophylactic steroids are NOT permitted. If the subject has a second infusion-related reaction Grade ≥ 2 on the slower infusion rate, with or without the addition of further medication to premedication, the infusion should be stopped and the subject removed from M7824 treatment.

3.4.1.4 Severe Hypersensitivity Reactions and Flu-like Symptoms

If a hypersensitivity reaction occurs, the subject must be treated according to the best available medical practice including ACLS guidelines.

Subjects should be instructed to report any delayed reactions to the Investigator immediately.

A. Symptoms

- Impaired airway
- Decreased oxygen saturation (< 92%)
- Confusion
- Lethargy
- Hypotension
- Pale / clammy skin
- Cyanosis

B. Management

- Epinephrine injection and IV dexamethasone
- Patient should be placed on cardiac, blood pressure, heart rate, and oxygen saturation monitor immediately
- Alert intensive care unit for possible transfer if required

For prophylaxis of flu-like symptoms, a NSAID, for example, ibuprofen 400 mg or comparable NSAID dose, may be administered 2 hours before and 8 hours after the start of each dose of M7824 IV infusion.

3.4.1.5 Immune-Related Adverse Events

Since inhibition of PD-L1 and TGFβ signaling stimulates the immune system, irAEs may occur. Treatment of irAEs is mainly dependent upon severity (NCI-CTCAE grade):

- Grade 1 to 2: treat symptomatically or with moderate dose steroids, more frequent monitoring
- Grade 1 to 2 (persistent): manage similar to high grade AE (Grade 3 to 4)
- Grade 3 to 4: treat with high dose corticosteroids

Treatment of irAEs should follow guidelines set forth in **Table 7** below:

Table 7 Management of Immune-Related Adverse Events

Gastrointestinal irAEs		
Severity of Diarrhea/Colitis (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Diarrhea: < 4 stools/day over Baseline	Continue M7824 therapy Symptomatic treatment (e.g. loperamide)	Close monitoring for worsening symptoms Educate subject to report worsening immediately

Colitis: asymptomatic		If worsens: Treat as Grade 2, 3 or 4.
Grade 2 Diarrhea: 4 to 6 stools per day over Baseline; IV fluids indicated < 24 hours; not interfering with ADL Colitis: abdominal pain; blood in stool	Withhold M7824 therapy Symptomatic treatment	If improves to Grade ≤ 1: Resume M7824 therapy If persists > 5-7 days or recurs: Treat as Grade 3 or 4.
Grade 3 to 4 Diarrhea (Grade 3): ≥ 7 stools per day over Baseline; incontinence; IV fluids ≥ 24 h; interfering with ADL Colitis (Grade 3): severe abdominal pain, medical intervention indicated, peritoneal signs Grade 4: life-threatening, perforation	Withhold M7824 for Grade 3. Permanently discontinue M7824 for Grade 4 or recurrent Grade 3. 1.0 to 2.0 mg/kg/day prednisone IV or equivalent Add prophylactic antibiotics for opportunistic infections Consider lower endoscopy	If improves: Continue steroids until Grade ≤ 1, then taper over at least 1 month; resume M7824 therapy following steroids taper (for initial Grade 3). If worsens, persists > 3 to 5 days, or recurs after improvement: Add infliximab 5mg/kg (if no contraindication). Note: infliximab should not be used in cases of perforation or sepsis.
Dermatological irAEs		
Grade of Rash (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 to 2 Covering ≤ 30% body surface area	Continue M7824 therapy Symptomatic therapy (for example, antihistamines, topical steroids)	If persists > 1 to 2 weeks or recurs: Withhold M7824 therapy Consider skin biopsy Consider 0.5-1.0 mg/kg/day prednisone or equivalent. Once improving, taper steroids over at least 1 month, consider prophylactic antibiotics for opportunistic infections, and resume M7824 therapy following steroids taper. If worsens: Treat as Grade 3 to 4.
Grade 3 to 4 Grade 3: Covering > 30% body surface area; Grade 4: Life threatening consequences	Withhold M7824 for Grade 3. Permanently discontinue for Grade 4 or recurrent Grade 3. Consider skin biopsy	If improves to Grade ≤ 1: Taper steroids over at least 1 month; resume M7824 therapy following steroids taper (for initial Grade 3).

	Dermatology consult 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections	
Pulmonary irAEs		
Grade of Pneumonitis (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Radiographic changes only	Consider withholding M7824 therapy Monitor for symptoms every 2 to 3 days Consider Pulmonary and Infectious Disease consults	Re-assess at least every 3 weeks If worsens: Treat as Grade 2 or Grade 3 to 4.
Grade 2 Mild to moderate new symptoms	Withhold M7824 therapy Pulmonary and Infectious Disease consults Monitor symptoms daily; consider hospitalization 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consider bronchoscopy, lung biopsy	Re-assess every 1 to 3 days If improves: When symptoms return to Grade ≤ 1, taper steroids over at least 1 month, and then resume M7824 therapy following steroids taper If not improving after 2 weeks or worsening: Treat as Grade 3 to 4.
Grade 3 to 4 Grade 3: Severe new symptoms; New/worsening hypoxia; Grade 4: Life-threatening	Permanently discontinue M7824 therapy. Hospitalize. Pulmonary and Infectious Disease consults. 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consider bronchoscopy, lung biopsy	If improves to Grade ≤ 1: Taper steroids over at least 1 month If not improving after 48 hours or worsening: Add additional immunosuppression (for example, infliximab, cyclophosphamide, IV immunoglobulin, or mycophenolate mofetil)
Hepatic irAEs		
Grade of Liver Test Elevation (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Grade 1 AST or ALT > ULN to 3.0 x ULN and/or Total bilirubin > ULN to 1.5 x ULN	Continue M7824 therapy	Continue liver function monitoring If worsens: Treat as Grade 2 or 3 to 4.

Grade 2 AST or ALT > 3.0 to ≤ 5 x ULN and/or total bilirubin > 1.5 to ≤ 3 x ULN	Withhold M7824 therapy Increase frequency of monitoring to every 3 days.	If returns to Grade ≤ 1: Resume routine monitoring; resume M7824 therapy. If elevation persists > 5 to 7 days or worsens: Treat as Grade 3 to 4.
Grade 3 to 4 AST or ALT > 5 x ULN and/or total bilirubin > 3 x ULN	Permanently discontinue M7824 therapy Increase frequency of monitoring to every 1 to 2 days 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Consult gastroenterologist/ hepatologist Consider obtaining MRI/CT scan of liver and liver biopsy if clinically warranted	If returns to Grade ≤ 1: Taper steroids over at least 1 month If does not improve in > 3 to 5 days, worsens or rebounds: Add mycophenolate mofetil 1 gram (g) twice daily If no response within an additional 3 to 5 days, consider other immunosuppressants per local guidelines.
Renal irAEs		
Grade of Creatinine Increased (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 1 Creatinine increased > ULN to 1.5 x ULN	Continue M7824 therapy	Continue renal function monitoring If worsens: Treat as Grade 2 to 3 or 4.
Grade 2 to 3 Creatinine increased > 1.5 and ≤ 6 x ULN	Withhold M7824 therapy Increase frequency of monitoring to every 3 days 1.0 to 2.0 mg/kg/day prednisone or equivalent. Add prophylactic antibiotics for opportunistic infections Consider renal biopsy	If returns to Grade ≤1: Taper steroids over at least 1 month, and resume M7824 therapy following steroids taper. If worsens: Treat as Grade 4.
Grade 4 Creatinine increased > 6 x ULN	Permanently discontinue M7824 therapy Monitor creatinine daily 1.0 to 2.0 mg/kg/day prednisone or equivalent. Add prophylactic antibiotics for opportunistic infections Consider renal biopsy Nephrology consult	If returns to Grade ≤1: Taper steroids over at least 1 month.

Cardiac irAEs		
Myocarditis	**Initial Management**	**Follow-up Management**
New onset of cardiac signs or symptoms and / or new laboratory cardiac biomarker elevations (e.g. troponin, CK-MB, BNP) or cardiac imaging abnormalities suggestive of myocarditis.	Withhold M7824 therapy. Hospitalize. In the presence of life threatening cardiac decompensation, consider transfer to a facility experienced in advanced heart failure and arrhythmia management. Cardiology consult to establish etiology and rule-out immune-mediated myocarditis. Guideline based supportive treatment as per cardiology consult. * Consider myocardial biopsy if recommended per cardiology consult.	If symptoms improve and immune-mediated etiology is ruled out, re-start M7824 therapy. If symptoms do not improve/worsen, viral myocarditis is excluded, and immune-mediated etiology is suspected or confirmed following cardiology consult, manage as immune-mediated myocarditis.
Immune-mediated myocarditis	Permanently discontinue M7824. Guideline based supportive treatment as appropriate as per cardiology consult. * 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections.	Once improving, taper steroids over at least 1 month. If no improvement or worsening, consider additional immunosuppressants (e.g. azathioprine, cyclosporine A).
*Local guidelines, or e.g. ESC or AHA guidelines ESC guidelines website: https://www.escardio.org/Guidelines/Clinical-Practice-Guidelines AHA guidelines website: http://professional.heart.org/professional/GuidelinesStatements/searchresults.jsp?q=&y=&t=1001		
Endocrine irAEs		
Endocrine Disorder	**Initial Management**	**Follow-up Management**
Grade 1 or Grade 2 endocrinopathies (hypothyroidism, hyperthyroidism, adrenal insufficiency, type I diabetes mellitus)	Continue M7824 therapy Endocrinology consult if needed Start thyroid hormone replacement therapy (for hypothyroidism), anti-thyroid treatment (for hyperthyroidism), corticosteroids (for adrenal insufficiency) or insulin (for Type I diabetes mellitus) as appropriate. Rule-out secondary endocrinopathies (i.e. hypopituitarism / hypophysitis)	Continue hormone replacement/suppression and monitoring of endocrine function as appropriate.

Grade 3 or Grade 4 endocrinopathies (hypothyroidism, hyperthyroidism, adrenal insufficiency, type I diabetes mellitus)	Withhold M7824 therapy Consider hospitalization Endocrinology consult Start thyroid hormone replacement therapy (for hypothyroidism), anti-thyroid treatment (for hyperthyroidism), corticosteroids (for adrenal insufficiency) or insulin (for type I diabetes mellitus) as appropriate. Rule-out secondary endocrinopathies (i.e. hypopituitarism / hypophysitis)	Resume M7824 once symptoms and/or laboratory tests improve to Grade ≤ 1 (with or without hormone replacement/suppression). Continue hormone replacement/suppression and monitoring of endocrine function as appropriate.
Hypopituitarism/Hypophysitis (secondary endocrinopathies)	If secondary thyroid and/or adrenal insufficiency is confirmed (i.e. subnormal serum FT4 with inappropriately low TSH and/or low serum cortisol with inappropriately low ACTH): • Refer to endocrinologist for dynamic testing as indicated and measurement of other hormones (FSH, LH, GH/IGF-1, PRL, testosterone in men, estrogens in women) • Hormone replacement/suppressive therapy as appropriate • Perform pituitary MRI and visual field examination as indicated **If hypophysitis confirmed:** • Continue M7824 if mild symptoms with normal MRI. Repeat the MRI in 1 month • Withhold M7824 if moderate, severe or life-threatening symptoms of hypophysitis and/or abnormal MRI. Consider hospitalization. Initiate corticosteroids (1 to 2 mg/kg/day prednisone or equivalent) followed by corticosteroids taper during at least 1 month. • Add prophylactic antibiotics for opportunistic infections.	Resume M7824 once symptoms and hormone tests improve to Grade ≤ 1 (with or without hormone replacement). In addition, for hypophysitis with abnormal MRI, resume M7824 only once shrinkage of the pituitary gland on MRI/CT scan is documented. Continue hormone replacement/suppression therapy as appropriate.

Other irAEs (not described above)		
Grade of other irAEs (NCI-CTCAE v5)	**Initial Management**	**Follow-up Management**
Grade 2 or Grade 3 clinical signs or symptoms suggestive of a potential irAE	Withhold M7824 therapy pending clinical investigation	If irAE is ruled out, manage as appropriate according to the diagnosis and consider re-starting M7824 therapy If irAE is confirmed, treat as Grade 2 or 3 irAE.
Grade 2 irAE or first occurrence of Grade 3 irAE	Withhold M7824 therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Specialty consult as appropriate	If improves to Grade ≤ 1: Taper steroids over at least 1 month and resume M7824 therapy following steroids taper.
Recurrence of same Grade 3 irAEs	Permanently discontinue M7824 therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent Add prophylactic antibiotics for opportunistic infections Specialty consult as appropriate	If improves to Grade ≤ 1: Taper steroids over at least 1 month.
Grade 4	Permanently discontinue M7824 therapy 1.0 to 2.0 mg/kg/day prednisone or equivalent and/or other immunosuppressant as needed Add prophylactic antibiotics for opportunistic infections Specialty consult.	If improves to Grade ≤ 1: Taper steroids over at least 1 month
Requirement for 10 mg per day or greater prednisone or equivalent for more than 12 weeks for reasons other than hormonal replacement for adrenal insufficiency **Persistent Grade 2 or 3 irAE lasting 12 weeks or longer**	Permanently discontinue M7824 therapy Specialty consult	

3.4.1.6 Anemia

- If after the 28-Day-DLT period the subject experiences a Grade 3 Hgb decrease (< 8.0 g/dL) that is clinically manageable with blood transfusions or erythroid growth factor use, this does not require treatment discontinuation

- If Hgb < 7 g/dL, the Investigator should consider blood transfusion
- In case of any Hgb < 8 g/dL, the Investigator should use discretion to initiate anemia work up, including Coombs, haptoglobin, indirect bilirubin and peripheral smear, and prothrombin time (PT), activated partial thromboplastin time (aPTT), international normalized ratio (INR); Hgb, red blood cells, and hematocrit are to be closely monitored
- If a subject experiences significant anemia of Hgb <9.0 g/dl, then the amount of blood to be drawn may be reduced by not taking blood for soluble factors, TGFβ, circulating free tumor DNA, and PK studies. In particular, on M7824 infusion Days 1, 15, 29, and 43 (cycles #1 – 4) with pre- and post-infusion PK measurements total blood draw volume may be as high as 24.5 mL on day of infusion. CBC counts will be monitored closely to detect development of anemia and expeditiously restrict blood draws. The decision to reduce the time points for these biomarkers will be taken by the Principal Investigator. This will be documented. Blood will continue to be taken as scheduled for safety analyses and ADAs.

3.4.1.7 Rash with Hyperkeratosis / Keratoacanthoma / Squamous Cell Carcinoma of the Skin

Monitoring will include skin assessments every 4 weeks with biopsy of suspicious lesions. Dermatological consults should be requested as needed.

3.4.1.8 Alterations in Wound Healing or Tissue Damage Repair

Management should be discussed on a case-by-case basis. Dermatological consults should be requested as needed.

3.4.1.9 Dose Interruptions for Adverse Events not Related to Study Drug

In case of Grade 3 and Grade 4 AEs not study drug related, the study treatment may be interrupted based on the Investigator assessment and the subject will be medically treated for the event.

If the AE reduces to a lower tolerable grade the study treatment might be resumed in the subsequent cycle. If the AE remains the same despite the medical treatment until the next treatment (second cycle after the AE occurred) a consideration of a possible extension of the dose interruption for up to 3 additional cycles is allowed.

If upon the resumed study treatment, the subject experiences the same AE, permanent withdrawal from the study treatment should be considered.

Grade 3 and 4 laboratory abnormalities that do not have clinical significance and are not related to study drugs do not require dose interruption.

3.4.2 Gemcitabine

Toxicities aligned with gemcitabine toxicity profile will lead to dose reduction of the nucleoside analogue (of the selected starting dose according to DMET genotyping; please see **Table 8** below).

Table 8 Recommended Dose Reductions for Gemcitabine for Myelosuppression in Pancreatic Cancer and Non-Small Cell Lung Cancer

Absolute granulocyte count, x10^6/L		**Platelet count, x10^6/L**	**% of selected starting dose**

≥1,000	And	≥100,000	100%
500-999	Or	50,000-99,999	75%
<500	Or	<50,000	Hold, restart gemcitabine after recover to Grade 1 at 50% of dose

If Grade ≥3 hematologic toxicity possibly related to gemcitabine, restart gemcitabine after recover to Grade 1 at 50% of the dose.

Dose Modifications for Non-Hematologic Adverse Reactions

Permanently discontinue gemcitabine for any of the following:

- Unexplained dyspnea or other evidence of severe pulmonary toxicity
- Severe hepatic toxicity
- Hemolytic-uremic syndrome
- Capillary leak syndrome
- Posterior reversible encephalopathy syndrome

Withhold gemcitabine or reduce dose to 50% for other severe (Grade 3 or 4) non-hematological toxicity until resolved. For patients receiving 600mg/m^2 gemcitabine dose will not be reduced and gemcitabine will be withheld.

No dose modifications are recommended for alopecia, nausea, or vomiting.

3.5 Assessments on Treatment

In this trial, the treatment with M7824 will continue until patient meets off treatment criteria (Section **3.8.1**). In the case of PD, subjects will continue treatment and have confirmatory scans 4 weeks after the initial scan documenting PD. In case of confirmation of PD per RECIST patient will come off treatment.

For subjects who achieve a PR or CR on M7824 gemcitabine combination therapy and then subsequently develop disease progression after stopping therapy, due to reasons other than PD (AE which later resolve), 1 re-initiation course of treatment at the same dose and schedule of M7824 only is allowed at the discretion of the Investigator. Gemcitabine will not be administered again. The Investigator will need to confirm that the benefit of re-initiating treatment outweighs risks of toxicities involved, such as that which led to initial treatment discontinuation. In order to be eligible for re-treatment, the subject must not have experienced any toxicity that led to permanent treatment discontinuation of the initial M7824 therapy. Prior to re-initiation of the study treatment, malignant disease needs to be radiologically re-staged to assess all known sites of the disease and to establish a new baseline for subsequent tumor measurements.

Relevant safety laboratory samples must be drawn and results available and verified prior to re-initiating treatment. Patients should be reconsented at this point. Subjects who re-initiate treatment will stay on study and will be treated and monitored according to the Study Calendar **3.7**. During re-treatment no research procedures will be done.

A time window of up to 3 days before or 1 day after a scheduled visit day (-3 / +1 days) will be permitted for all study procedures. In addition, the tumor evaluation has a tumor assessment

visiting time window (repeat imaging) of 5 days prior to dosing (-5 days). Furthermore, if any Screening/Baseline procedures are conducted within 3 days prior to Day 1 of Cycle 1 the assessments scheduled on Cycle 1, Day 1 do not need to be repeated.

3.5.1 End-of-Treatment Visit

All subjects should undergo an End-of-Treatment visit after discontinuation of M7824 for any reason. This visit should be performed on the day or within 7 days after the decision to discontinue trial treatment but before any new antineoplastic therapy is started (if possible). If it is known to the Investigator at the time of the End of-Treatment visit that the subject will start new treatment within 28 days of last treatment or they will be unable to return within 28 days of last treatment, assessments associated with the 28-Day Safety Follow-up visit may be conducted at the End-of-Treatment visit. The discontinuation visit consists of assessments indicated in Study Calendar, Section **3.7**.

3.5.2 Post-Treatment Follow-up

3.5.2.1 28-Day Safety Follow-up Visit

A Safety Follow-up visit is scheduled 4 weeks (28 ± 5 days) after the last administration of M7824 but before any new therapy is started, if possible, whichever occurs earlier. The 28-Day Safety Follow-up visit will comprise a full assessment for safety, immunogenicity, and tumor response as appropriate, which will include assessments indicated in Study Calendar, Section **3.7.**

3.5.2.2 Long-term Follow-up

All SAEs ongoing at the 28-Day Safety Follow-up visit must be monitored and followed up by the Investigator until stabilization or until the outcome is known, unless the subject is documented as "lost to follow-up." In addition, all trial drug-related SAEs occurring after 28-Day Safety Follow-up visit and ongoing at the Safety Follow-up visit have to be followed up in the same manner.

Subjects without PD at the 28-Day Safety Follow-up visit will be followed till disease progression (CT / MRI scans every 12 weeks with the first assessment 12 weeks after the previous tumor assessment until PD).

Subjects with PD after the 28-Day Safety Follow-up visit, will be followed quarterly (± 14 days) for survival (including assessment of any further anticancer therapy) by phone call or e-mail for 1 year. Survival follow up will continue every 6 months after that.

3.6 Questionnaires

The rationale for questionnaires is discussed in Section **1.2.16** and timing is specified in Study Calendar **3.7.** The average time to complete these instruments is 20 minutes. For English speaking subjects only.

3.7 Study Calendar

Procedure	**Screening**	**Baseline**	**Cycles [1] (14 days each cycle)**								**EOT visit[20]**	**28 Days FU[20]**	**Long Term FU[19,20]**
			1	**2**	**3**	**4**	**5**	**6**	**7**	**8-N**			
M7824 [2]			X	X	X	X	X	X	X	X			
Gemcitabine [3]				X	X	X	X	X	X	X			
NIH Advance Directives Form [4]		X											
Medical History	X												
Confirmation of Pathology	X												
Height	X												
Physical exam, weight, ECOG [5]	X		X	X	X	X	X	X	X	X	X	X	
Vital signs [23]	X		X	X	X	X	X	X	X	X	X	X	
TB testing (if clinically indicated)	X												
EKG	X		X/X[6]									X	
CBC with differential [7]	X		X	X	X	X	X	X	X	X	X	X	
Biochemical profile [7, 8]	X		X	X	X	X	X	X	X	X	X	X	

Procedure	**Screening**	**Baseline**	**Cycles [1] (14 days each cycle)**								**EOT visit[20]**	**28 Days FU[20]**	**Long Term FU[19,20]**
			1	**2**	**3**	**4**	**5**	**6**	**7**	**8-N**			
Amylase / lipase			X	X		X			X	X[9]	X	X	
Urinalysis			X	X		X			X	X[9]	X	X	
Pregnancy testing (urine or serum)	X		X	X		X		X		X[10]		X	
PT, INR, aPTT, fibrinogen			X									X	
T4 and TSH			X	X		X			X	X[9]		X	
Tumor evaluation (CT Scan / MRI)[11]	X	X				X			X	X		X	X
Brain CT/MRI [12]	X												
Concomitant Medications			X	X	X	X	X	X	X	X	X	X	
Adverse events			X	X	X	X	X	X	X	X	X	X	X
Research blood for PK sampling – M7824[13]			X/X	X	X/X			X			X		
Research blood for PK sampling – gemcitabine [14]				X	X	X	X						
Research blood for ADA sampling [15]			X		X			X			X		

Procedure	**Screening**	**Baseline**	**Cycles [1] (14 days each cycle)**								**EOT visit[20]**	**28 Days FU[20]**	**Long Term FU[19,20]**
			1	**2**	**3**	**4**	**5**	**6**	**7**	**8-N**			
Research blood for Cytokines [15]			X	X	X		X		X				
Research blood for TGFβ 1, 2 and 3 [15]			X	X	X								
CEA, CA19-9, CA-125			X		X		X		X	X[16]	X	X[16]	
Research blood for Circulating free tumor DNA (cftDNA)			X	X	X		X		X		X	X	
Research blood for DMET pharmacogenomic screening[22]		X											
Research Tumor biopsy [17]		X				X							
Research Perfusion evaluation – DCE-MRI [21]		X		X		X				X	X		
FACT-Hep [18]		X				X				X		X	
Phone call or e-mail for survival/new cancer treatments every 3 months for 1 year, every 6 months after that													X

[1] Cycle indicated procedures will be performed on Day 1 of each cycle. Cycle length is 2 weeks (+/- 3 days). If any Screening/Baseline procedures are conducted within 3 days prior to Day 1 of Cycle 1, the assessments scheduled on Day 1 of Cycle 1 do not need to be repeated.

[2] M7824 will be administered as a 1 hour (-10 minutes / +20 minutes) IV infusion on Day 1 of each cycle before gemcitabine when both drugs are given on the same day.

[3] Gemcitabine will be given starting on Day 1 of Cycle 2 at assigned dose over 30 (+/- 10) minutes once weekly for the 4 weeks (cycles 2-3)). Starting on Cycle 4 once weekly for 3 weeks with one week without gemcitabine. Gemcitabine will be discontinued after Cycle 13 (after a total of 6 months of gemcitabine therapy).

[4] As indicated in section **10.3**, all subjects will be offered the opportunity to complete an NIH advanced directives form. This should be done preferably at baseline but can be done at any time during the study as long as the capacity to do so is retained. The completion of the form is strongly recommended, but is not required

[5] Assessed prior to trial treatment. Eye signs and symptoms and assessment of skin should be included into physical exam on baseline and following visits. If clinically indicated, ophthalmology and dermatology consults should be ordered.

[6] X/X means before and after infusion of M7824. Within 2 hours prior to M7824 infusion and within 2 hours after completion of M7824.

[7] Must be drawn and reviewed within 72 hours prior to drug administration. During screening within 28 days of enrollment.

[8] Biochemical profile: electrolytes, BUN, creatinine, AST, ALT, total bilirubin, calcium, phosphorus, albumin, magnesium, uric acid.

[9] Every 6 weeks

[10] Every 4 weeks

[11] CT or PET of chest, abdomen and pelvis (or MRI abdomen) with tumor measurements. For the purposes of this study, patients should be re-evaluated for response every 6 weeks. If a scan identifies objective response or PD, a confirmatory scan should be obtained 4 weeks following initial documentation of objective response or PD. Allowed time window is 5 days prior to dosing (-5 days). For subjects continuing treatment beyond 12-month tumor evaluations should take place every 12 weeks.

Subjects without PD at the 28-Day Safety Follow-up visit will be followed till disease progression (CT / MRI scans every 12 weeks with the first assessment 12 weeks after the previous tumor assessment until PD)

[12] Brain CT/MRI scan (either, with contrast preferred) only if clinically indicated at Screening (in case of neurological symptoms, previous history of brain metastases, or at the discretion of the Investigator).

[13] X/X means before and after infusion at Cycles 1 and 3. Pre-M7824 PK levels to be taken within 45 minutes prior M7824 infusion. Post-M7824 PK levels to be taken within 45 minutes after completion of M7824 infusion / as close as possible to completion of following gemcitabine infusion. Within 45 minutes prior to infusion at Cycles 2, 6 and EOT.

[14] Blood samples for the determination of gemcitabine (GEM) plasma levels will be obtained just prior to drug administration, end of infusion (EOI), and 0.5hr, 1hr, 2hr, and 4hr post EOI. These samples will be collected for the doses received on Day 1 of Cycles 2, 3, 4 and 5. Blood draws will be permitted to have a window of +/- 5 minutes to accommodate logistical challenges.

[15] To be taken within 30 minutes prior to start of M7824 infusion.

[16] On Cycles 8-13 every 6 weeks and then every 12 weeks.

[17] Optional Research biopsies might be performed at baseline and within 7 days after the Day 1 Cycle 4 M7824 administration.

[18] Subject-reported outcomes / symptom severity assessments 4th version of the 'Functional Assessment of Cancer Therapy with the hepatobiliary and pancreatic specific module (FACT-Hep) will be completed at Baseline, after cycle 3, 7 and every 12 weeks after start of treatment. By English speaking subjects only.

[19] All SAEs ongoing at the 28-Day Safety Follow-up visit must be monitored and followed up by the Investigator until stabilization or until the outcome is known, unless the subject is documented as "lost to follow-up." In addition, all trial drug-related SAEs occurring after 28 Day Safety Follow up visit and ongoing at the Safety Follow-up visit should be followed up in the same manner.

[20] If subjects are unable or not willing to come to NIH after treatment discontinuation for FU visits, they will be followed quarterly (± 14 days) by phone call or e-mail for survival, adverse events and further tumor therapy

[21] At baseline, cycles 2, 4, 10 and EOT visit. Patients who have heart pacemakers, metal implants, or metal chips or clips in or around the eyeballs cannot be scanned with an MRI because of the risk that the magnet may move the metal in these areas. DCE-MRI in cycle 2 should be obtained before first gemcitabine infusion, DCE-MRI at baseline and in cycles 2, 4 and 10 are mandatory, at EOT visit it is per PI discretion. If DCE-MRI is not obtained in cycle 2, it may be obtained in cycle 3, with the following DCE-MRI studies moving to cycle 5 and 11.

[22] Prior to study treatment, patients (under NCI 16-C-0076) will provide one peripheral blood sample to analyze genomic DNA via the DMET Plus kit for the assessment of the patient's cytidine deaminase (CDA) genotype. Based on the CDA genotype, patients will be stratified into a specified dosing strategy (as per Section **3.1**).

[23] During treatment vital signs must be measured within 1 hour before and 40 minutes following M7824 infusions. In addition, at least one time during the infusion of M7824.

3.8 Criteria for Removal from Protocol Therapy and Off Study Criteria

3.8.1 Criteria for Removal from Protocol Therapy

- Progressive disease. Subjects should continue treatment beyond the initial determination of PD, through their next tumor assessment, which in this case should be in 4 weeks if:
  - There are no new Grade 2 or greater symptoms or significant worsening of existing symptoms.
  - There is no increase in ECOG.
  - In the opinion of the Investigator, the subject does not require new anticancer therapy.
- Occurrence of DLT (see section **3.1.1**)
- Excessive toxicity (see section **3.4**)
- Necessity to administer a non-permitted concomitant drug
- PI discretion
- Positive pregnancy test

3.8.2 Off -Study Criteria

- Death
- Patient request to be withdrawn from study
- PI discretion
- PI decision to end the study

3.8.3 Off Protocol Therapy and Off-Study Procedure

Authorized staff must notify Central Registration Office (CRO) when a subject is taken off protocol therapy and when a subject is taken off-study. A Participant Status Updates Form from the web site (http://home.ccr.cancer.gov/intra/eligibility/welcome.htm) main page must be completed and sent via encrypted email to: NCI Central Registration Office ncicentralregistration-l@mail.nih.gov.

4 CONCOMITANT MEDICATIONS / MEASURES

4.1 Permitted Medicines

Any medications (other than those excluded by the clinical trial protocol) that are considered necessary to protect subject welfare and will not interfere with the trial medication may be given at the Investigator's discretion.

Palliative radiotherapy delivered in a normal organ-sparing technique may be administered during the trial. The assessment of PD will not be based on the necessity for palliative radiotherapy.

4.2 Prohibited Medicines

The following treatments must not be administered during the trial:

- Immunotherapy including interferons, immunosuppressive drugs (for example, chemotherapy or systemic corticosteroids except for short term treatment of allergic reactions, endocrine replacement therapy at low dose prednisone [≤ 10 mg daily] or glucocorticoid equivalent doses, or for the treatment of irAEs or other appropriate short term steroid use), or other experimental pharmaceutical products. Short term administration of systemic steroid or other immunosuppressant such as infliximab or mycophenolate (that is, for allergic reactions or the management of irAEs) is allowed. Steroids with no or minimal systemic effect (topical, inhalation) are allowed.
- Prophylactic use of corticosteroids for infusion related reactions is prohibited.
- Herbal remedies with immunostimulating properties (for example, mistletoe extract) or known to potentially interfere with major organ function (for example, hypericin).
- Any live attenuated vaccine therapies for the prevention of infectious disease. Administration of inactivated vaccines is allowed (for example, inactivated influenza vaccines).

If the administration of a non-permitted concomitant drug becomes necessary during the trial, the subject will be withdrawn from trial treatment.

5 BIOSPECIMEN COLLECTION

5.1. Correlative Studies for Research/Pharmacokinetic Studies

Correlative studies rationale is discussed in detail in Section **1.2.17** Justification of Research Studies of this protocol. In general, studies will address the effect on TGFβ concentrations and other soluble factors in plasma upon response to the study drugs, DCE-MRI as a measure if impact on perfusion upon stromal modulation by M7824 and the impact of the somatic genotype on response to M7824. We also will evaluate intratumoral alterations of the immune milieu / stroma of pre- versus on-treatment biopsies as well as follow copies of circulating free tumor DNA levels during treatment.

Test/assay	Volume (approx.)	Type of tube	Collection point	Location of specimen analysis
50-cancer gene panel	Tumor sample		Before treatment	Laboratory of Dr. M. Raffeld, Molecular Pathology
Multiplexed immunohistochemical evaluation of immune cell populations	Tumor sample		Before and after 7 weeks of treatment	Dr. Raffeld, Laboratory of Pathology NCI

Test/assay	**Volume (approx.)**	**Type of tube**	**Collection point**	**Location of specimen analysis**
M7824 Pharmacokinetics	Blood, 3.5 mL	Serum Separator Tubes (SST®) (gold capped)	See Study calendar **3.7**	EMD Serono * (Processed in Figg Lab)
Gemcitabine Pharmacokinetics	Blood, 4 mL	Sodium Heparin (green top) tube (pre-treated with THU)	See Study calendar **3.7**	Dr. Figg Lab
Cytokines by ELISA	Blood, 2x5 mL	Gold top SST	See Study calendar **3.7**	EMD Serono* (Processed in Figg Lab)
TGFβ 1, 2, 3 ligand concentrations by ELISA	Blood, 4.5 mL	Blue top CTAD	See Study calendar **3.7**	EMD Serono* (Processed in Figg Lab)
Circulating free tumor DNA (cftDNA)	Blood, 3.5 mL	EDTA (purple top) tubes	See Study calendar **3.7**	Dr. Raffeld, Laboratory of Pathology NCI (Processed in Figg Lab)
ADA by ELISA	Blood, 2.5 mL	Gold top SST	See Study calendar **3.7**	EMD Serono* (Processed in Figg Lab)
DMET pharmacogenomic screening	Plasma, 3.5 mL	EDTA (purple top) tubes	Baseline	Dr. Figg Lab

* Coded linked samples without key will be sent to EMD Serono:

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Samples will be sent for barcoding and initial storage to Blood Processing Core (BPC).

Patients will undergo blood and tissue sampling for research purposes on the time points outlined in the Study Calendar **3.7**.

5.1.1 Tumor and Tissue Collection

Tumor samples might be collected at baseline and within 7 days after the Cycle 4 Day 1 M7824 administration for flow cytometry and RNA evaluation.

Of any tumor biopsy, one sample will always be sent to Laboratory of Pathology for disease evaluation first (confirmation of metastatic pancreatic cancer). Leftover samples will be used for research.

It is preferable to not biopsy a target lesion; however, if only one lesion is amenable for biopsy, biopsy will be performed on this lesion.

Tissue processing: Fresh tumor tissue obtained from subjects for research purposes and not used for surgical pathology evaluation should undergo a tumor digest and used for flow cytometry to study immune cell populations. Other parts of obtained cancer tissues should either be snap frozen and used for later RNA extraction and fixed in 10% neutral buffered formalin (NBF) followed by paraffin-embedding or used for the generation of a tumor digest. Formalin substitutes are not suited as fixative.

Tissue storage: Fresh tumor tissue digest obtained from subjects for the evaluation of efficacy should be stored in defined cryopreservation medium containing 10% dimethyl sulfoxide [CryoStor® CS10].

Prioritization of limited sample of fresh tumor biopsy for research purposes: First priority: fresh sample for tumor digest and flow cytometry and RNASeq analysis; second priority: to formalin fix and generate a paraffin embedded (FFPE) tissue block; if the archival tumor containing FFPE tissue block cannot be provided, sections from the tumor biopsy used for FFPE should be provided which are freshly cut, 4 µm thick and mounted on positively-charged microscope slides. SuperFrost Plus glass slides are recommended for samples designated for IHC to reduce IHC assay failure. If the archival tumor block is available preferably 15 slides should be provided; if not possible a minimum of 5 slides is required.

5.1.2 Multiplexed immunohistochemical evaluation of immune cell population in tumor samples

Multiplexed immunohistochemical evaluation of immune cell population to examine the immune landscape for TGFβ signaling-dependent immune cell populations like T cells including T

regulatory cells, MDSCs, or tumor-associated macrophages and will be done in the Laboratory of Pathology.

5.1.3 Pharmacokinetics of M7824 and gemcitabine

Pharmacokinetic parameters are limited to Cmax (in µg/mL plasma), time to reach maximum concentration (tmax), and minimum serum concentration (Cmin; in µg/mL plasma).

Blood will be collected for PK assessments according to Study Calendar **3.7**.

Plasma M7824 PK measurements will be done at EMD Serono, plasma gemcitabine measurements in the Clinical Pharmacology Core (Dr. W. Figg).

5.1.3.1 M7824 Pharmacokinetics (EMD Serono)

Plasma M7824 PK measurements will be done at EMD Serono

5.1.3.2 Gemcitabine Pharmacokinetics (Figg Lab)

Blood samples for the determination of gemcitabine (GEM) plasma levels will be obtained from participating patients via 4 mL sodium heparin (green top) tube (BD, Franklin Lakes, NJ) collected according to Study Calendar **3.7**. Bioanalytical measurements will be conducted on an ultra HPLC-MS/MS system by the Clinical Pharmacology Program (CPP).

Because GEM is rapidly deactivated by cytidine deaminase (CD), green top tubes will be fortified with 100 uM tetrahydrouridine (THU). These tubes will be prepared in advance by the CPP and provided to the clinic. In advance of PK samples being drawn, please notify the CPP at least 24 hrs. prior to prepare fresh green top tubes with 100 uM THU.

This data will be used to monitor GEM plasma concentrations to assess any drug interactions with M7834 and/or correlations with pharmacogenomics, adverse events, and clinical response.

The samples will be placed immediately on wet ice and refrigerated. The date and exact time of each blood draw should be recorded on the sample tube and the PK sheet. Please page 102-11964 for immediate pick-up. Contact Dr. Figg's Clinical Pharmacology Program (Blood Processing Core) in Bldg. 10/5A09 at 301-402-3622 or 301-594-6131 with any questions.

Upon arrival in the Clinical Pharmacology Program, samples will be centrifuged and the plasma transferred into cryovials for storage at -80 C until the time of analysis. In addition, samples will be barcoded as described in Section **5.2.1**.

5.1.4 Soluble Factors Investigation

Blood will be collected to analyze soluble factors including cytokines and TGFβ 1, 2, 3 ligand concentrations according to Study Calendar **3.7**. The investigation will be done by EMD Serono using ELISA.

5.1.5 ADA Analysis.

Blood will be collected to analyze ADA according to Study Calendar **3.7**. Samples positive for ADAs will be re-analyzed to determine the titer. The investigation will be done by EMD Serono using ELISA.

5.2 Sample Storage, Tracking and Disposition

Samples will be ordered in CRIS and tracked through a Clinical Trial Data Management system. Should a CRIS screen not be available, the CRIS downtime procedures will be followed. All samples will be sent to Blood Processing Core (BPC) for processing and storage until they are distributed to Dr. Rudloff's lab sample analysis as described in the protocol. Samples will not be sent outside NIH without IRB notification and an executed MTA.

5.2.1 Samples Managed by Dr. Figg's Blood Processing Core (BPC)

5.2.1.1 BPC contact information

Please e-mail NCIBloodcore@mail.nih.gov at least 24 hours before transporting samples (the Friday before is preferred).

For sample pickup, page 102-11964.

For immediate help, call 240-760-6180 (main blood processing core number) or, if no answer, 240-760-6190 (main clinical pharmacology lab number).

For questions regarding sample processing, contact NCIBloodcore@mail.nih.gov.

5.2.2 Sample Data Collection

All samples sent to the Blood Processing Core (BPC) will be barcoded, with data entered and stored in the LABrador (aka LabSamples) utilized by the BPC. This is a secure program, with access to LABrador limited to defined Figg lab personnel, who are issued individual user accounts. Installation of LABrador is limited to computers specified by Dr. Figg. These computers all have a password restricted login screen. All Figg lab personnel with access to patient information annually complete the NIH online Protection of Human Subjects course.

LABrador creates a unique barcode ID for every sample and sample box, which cannot be traced back to patients without LABrador access. The data recorded for each sample includes the patient ID, name, trial name/protocol number, time drawn, cycle time point, dose, material type, as well as box and freezer location. Patient demographics associated with the clinical center patient number are provided in the system. For each sample, there are notes associated with the processing method (delay in sample processing, storage conditions on the ward, etc.).

Sample bar-codes are linked to patient demographics and limited clinical information. This information will only be provided to investigators listed on this protocol, via registered use of the LABrador. It is critical that the sample remains linked to patient information such as race, age, dates of diagnosis and death, and histological information about the tumor, in order to correlate genotype with these variables.

5.2.3 Sample Storage

Barcoded samples are stored in barcoded boxes in a locked freezer at either -20 or -80°C according to stability requirements. These freezers are located onsite in the BPC and offsite at NCI Frederick Central Repository Services in Frederick, MD. Visitors to the laboratory are required to be accompanied by laboratory staff at all times.

Access to stored clinical samples is restricted. Samples will be stored until requested by a researcher named on the protocol. All requests are monitored and tracked in LABrador. All researchers are required to sign a form stating that the samples are only to be used for research

purposes associated with this trial (as per the IRB approved protocol) and that any unused samples must be returned to the BPC. It is the responsibility of the NCI Principal Investigator to ensure that the samples requested are being used in a manner consistent with IRB approval.

Following completion of this study, samples will remain in storage as detailed above. Access to these samples will only be granted following IRB approval of an additional protocol, granting the rights to use the material.

If, at any time, a patient withdraws from the study and does not wish for their existing samples to be utilized, the individual must provide a written request. Following receipt of this request, the samples will be destroyed (or returned to the patient, if so requested), and reported as such to the IRB. Any samples lost (in transit or by a researcher) or destroyed due to unknown sample integrity (i.e. broken freezer allows for extensive sample thawing, etc.) will be reported as such to the IRB.

5.2.4 Procedures for Storage of Tissue Specimens in the Laboratory of Pathology

Tissues designated for clinical diagnostics are transported to the Laboratory of Pathology (LP) where they are examined grossly and relevant portions are fixed, embedded in paraffin and sectioned and stained for diagnostic interpretation. Unutilized excess tissues are stored for up to three months, in accordance with College of American Pathologists/Joint Commission on Accreditation of Healthcare Organizations (CAP/JCAHO) guidelines, and then discarded. Following completion of the diagnostic workup, the slides and tissue blocks are stored indefinitely in the LP's clinical archives. All specimens are catalogued and retrieved utilizing the clinical laboratory information systems, in accordance with CAP/JCAHO regulations. The use of any stored specimens for research purposes is only allowed when the appropriate IRB approval has been obtained. In some cases, this approval has been obtained via the original protocol on which the patient was enrolled.

5.2.5 Protocol Completion/Sample Destruction

All specimens obtained in the protocol are used as defined in the protocol. Any specimens that are remaining at the completion of the protocol will be stored in the conditions described in Section **5.2.1**. The study will remain open so long as sample or data analysis continues. Samples from consenting subjects will be stored until they are no longer of scientific value or if a subject withdraws consent for their continued use, at which time they will be destroyed. The PI will report any loss or destruction of samples to the NIH Intramural IRB as soon as he is made aware of such loss. If the patient withdraws consent the participant's data will be excluded from future distributions, but data that have already been distributed for approved research use will not be able to be retrieved.

The PI will report destroyed samples to the IRB if samples become unsalvageable because of environmental factors (ex. broken freezer or lack of dry ice in a shipping container) or if a patient withdraws consent. Samples will also be reported as lost if they are lost in transit between facilities or misplaced by a researcher. Freezer problems, lost samples or other problems associated with samples will also be reported to the NIH Intramural IRB and to the NCI Clinical Director.

5.3 Samples for Genetic/Genomic Analysis

5.3.1 Description of the scope of genetic/genomic analysis

- We will use baseline archival or fresh biopsy specimens for somatic tumor variant sequencing which will be performed by the CLIA-approved 50-cancer gene panel of the Molecular Pathology section (Dr. M. Raffeld lab).
- We plan to characterize cfDNA from blood at baseline and during treatment. All samples will be subjected to targeted sequencing. This will allow us to characterize somatic point mutations, and copy-number changes and will be performed in Dr. M. Raffeld's laboratory.
- DMET genotyping (DMET samples will be drawn under 16-C-0076 and sent to the Clinical Pharmacology Program for processing and analysis to determine polymorphisms in genes involved in gemcitabine metabolism). Prior to study enrollment, patients will enroll on NCI 16-C-0076 (Prospective Screening for Patient Specific Genotypes and Phenotypes that Influence Drug Dosing and Trial Selection in Cancer Patients, PI: William D. Figg). As per NCI 16-C-0076, one peripheral blood sample per patient will be collected in a EDTA (purple top) tube for pharmacogenetics studies to analyze the genomic DNA and assess genotype of the most relevant drug metabolizing enzymes and transporters (DMET). DNA will be analyzed on a DMET Plus (Affymatrix) genotyping platform that tests for 1,936 genetic variations in 225 drug disposition genes, including 47 CYP (phase I metabolism) genes, 13 non-CYP (phase I metabolism) genes, 78 phase II metabolizing genes (including UGTs), 63 transporters, 4 genes involved in facilitation of drug transporters, 9 genes involved in global regulation of drug metabolizing/transporting proteins, 4 drug binding proteins, and 4 drug targets. The samples will be sent to the NCI Frederick National Laboratory for Cancer Research (CLIA certificate 21D0947274) for analysis via the DMET Plus kit for the purpose of determining the patient's CDA genotype.

5.3.2 Privacy and Confidentiality of medical information/biological specimens

Fresh tumor samples will be stored in a minus 80-degree freezer. Initially the samples of each patient will be barcoded. At no time will patient's names be used on the blood and tissue samples. Sometimes, because a group collaboration or journal policy requires it, a subject's genetic data may be deposited in a database such as dbGaP. Although there is controlled access to such a database, such a submission carries theoretical risks of revealing the identity of the subject. This is discussed in the consent.

5.3.3 Management of Results

Subjects will be contacted if a clinically actionable gene variant is discovered. Clinically actionable findings for the purpose of this study are defined as disorders appearing in the American College of Medical Genetics and Genomics recommendations for the return of incidental findings that is current at the time of primary analysis. (A list of current guidelines is maintained on the CCR intranet: https://ccrod.cancer.gov/confluence/display/CCRCRO/Incidental+Findings+Lists). Subjects who still remain on the study will be contacted at this time with a request to provide a blood sample to be sent to a CLIA certified laboratory.

5.3.4 Genetic counseling

If the research findings are verified in the CLIA certified lab, the subject will be offered the opportunity to come to NIH (at our expense) to have genetic education and counseling with the NCI Genetics Branch to explain this result. If the subject does not want to come to NIH, a referral to a local genetic healthcare provider will be provided (at their expense). This is the only time

during the course of the study that incidental findings will be returned. No interrogations regarding clinically actionable findings will be made after the primary analysis.

6 DATA COLLECTION AND EVALUATION

6.1 Data Collection

The PI will be responsible for overseeing entry of data into an in-house password protected electronic system (C3D) and ensuring data accuracy, consistency and timeliness. The principal investigator, associate investigators/research nurses and/or data manager will assist with the data management efforts. All data obtained during the conduct of the protocol will be kept in secure network drives or in approved alternative sites that comply with NIH security standards. Primary and final analyzed data will have identifiers so that research data can be attributed to an individual human subject participant.

All adverse events, including clinically significant abnormal findings on laboratory evaluations, regardless of severity, will be followed until return to baseline or stabilization of event. Patients will be followed for adverse events for 28 days after removal from study treatment or until off-study, whichever comes first.

An abnormal laboratory value will be recorded in the database as an AE **only** if the laboratory abnormality is characterized by any of the following:

- Results in discontinuation from the study
- Is associated with clinical signs or symptoms
- Requires treatment or any other therapeutic intervention
- Is associated with death or another serious adverse event, including hospitalization.
- Is judged by the Investigator to be of significant clinical impact
- If any abnormal laboratory result is considered clinically significant, the investigator will provide details about the action taken with respect to the test drug and about the patient's outcome.

Grade 1 adverse events will not be collected

Hospitalization for social reason will not be considered an SAE.

6.1.1 Tumor data

The tumor disease information that will be documented and verified at the Screening visit for each subject includes:

- Detailed history of the tumor, including histopathological diagnosis, grading and staging in accordance with the Union Internationale Contre le Cancer Tumor Node Metastasis Classification **at diagnosis** (UICC TNM).
    - The M category (M0 or M1) of the tumor at the time of study entry, based on screening assessments
- All therapy used for prior treatment of the tumor (including surgery, radiotherapy and chemotherapy, immunotherapy).

- Any other conditions that were treated with chemotherapy, radiation therapy, or immunotherapy.

6.1.2 Concomitant Medications and Therapies

All concomitant medications taken by the subject during the trial, from the date of signature of informed consent are to be recorded in the appropriate section of the eCRF, noting the name, dose, duration and indication of each drug. Nondrug interventions (other than vitamins) and any changes to a concomitant medication or other intervention should also be recorded in the eCRF.

End of study procedures: Data will be stored according to HHS, FDA regulations and NIH Intramural Records Retention Schedule as applicable.

Loss or destruction of data: Should we become aware that a major breach in our plan to protect subject confidentiality and trial data has occurred, the IRB will be notified.

6.2 Data Sharing Plans

6.2.1 Human Data Sharing Plan

What data will be shared?

I will share human data generated in this research for future research as follows:

- Coded linked data in an NIH-funded or approved public repository.
- Coded linked data in BTRIS (automatic for activities in the Clinical Center)
- Coded linked or identified data with approved outside collaborators under appropriate agreements.

Data will be shared through*:*

- An NIH-funded or approved public repository: clinicaltrials.gov.
- BTRIS (automatic for activities in the Clinical Center)
- Approved outside collaborators under appropriate individual agreements.
- Publication and/or public presentations.

Data will be shared:

- Before publication.
- At the time of publication or shortly thereafter.

6.2.2 Genomic Data Sharing Plan

Unlinked genomic data will be deposited in public genomic databases such as dbGaP in compliance with the NIH Genomic Data Sharing Policy.

6.3 Response Criteria

For the purposes of this study, patients should be re-evaluated for response every 6 weeks for. In addition, if a scan identifies objective response, a confirmatory scan should be obtained 4 weeks following initial documentation of objective response. For subjects continuing treatment beyond 12 months tumor evaluations should take place every 12 weeks

Response and progression will be evaluated in this study using the new international criteria proposed by the revised Response Evaluation Criteria in Solid Tumors (RECIST) guideline (version 1.1) [[36, 37]] and Modified Immune-related response criteria (Section **6.4**). Changes in the largest diameter (unidimensional measurement) of the tumor lesions and the shortest diameter in the case of malignant lymph nodes are used in the RECIST criteria.

6.3.1 Disease Parameters

Measurable disease: Measurable lesions are defined as those that can be accurately measured in at least one dimension (longest diameter to be recorded) as:

- By chest x-ray: >20 mm;
- By CT scan:
    - Scan slice thickness 5 mm or under as >10 mm with CT scan
    - Scan slice thickness >5 mm: double the slice thickness
- With calipers on clinical exam: >10 mm.

All tumor measurements must be recorded in millimeters (or decimal fractions of centimeters).

Malignant lymph nodes. To be considered pathologically enlarged and measurable, a lymph node must be ≥15 mm in short axis when assessed by CT scan (CT scan slice thickness recommended to be no greater than 5 mm). At baseline and in follow-up, only the short axis will be measured and followed.

Non-measurable disease. All other lesions (or sites of disease), including small lesions (longest diameter <10 mm or pathological lymph nodes with ≥10 to <15 mm short axis), are considered non-measurable disease. Bone lesions, leptomeningeal disease, ascites, pleural/pericardial effusions, lymphangitis cutis/pulmonitis, inflammatory breast disease, and abdominal masses (not followed by CT or MRI), are considered as non-measurable.

Note: Cystic lesions that meet the criteria for radiographically defined simple cysts should not be considered as malignant lesions (neither measurable nor non-measurable) since they are, by definition, simple cysts.

'Cystic lesions' thought to represent cystic metastases can be considered as measurable lesions, if they meet the definition of measurability described above. However, if non-cystic lesions are present in the same patient, these are preferred for selection as target lesions.

Target lesions. All measurable lesions up to a maximum of 2 lesions per organ and 5 lesions in total, representative of all involved organs, should be identified as **target lesions** and recorded and measured at baseline. Target lesions should be selected on the basis of their size (lesions with the longest diameter), be representative of all involved organs, but in addition should be those that lend themselves to reproducible repeated measurements. It may be the case that, on occasion, the largest lesion does not lend itself to reproducible measurement in which circumstance the next largest lesion which can be measured reproducibly should be selected. A sum of the diameters (longest for non-nodal lesions, short axis for nodal lesions) for all target lesions will be calculated and reported as the baseline sum diameters. If lymph nodes are to be included in the sum, then only the short axis is added into the sum. The baseline sum diameters will be used as reference to further characterize any objective tumor regression in the measurable dimension of the disease.

Non-target lesions. All other lesions (or sites of disease) including any measurable lesions over and above the 5 target lesions should be identified as **non-target lesions** and should also be recorded at baseline. Measurements of these lesions are not required, but the presence, absence, or in rare cases unequivocal progression of each should be noted throughout follow-up.

6.3.2 Methods for Evaluation of Measurable Disease

All measurements should be taken and recorded in metric notation using a ruler or calipers. All baseline evaluations should be performed as closely as possible to the beginning of treatment and never more than 4 weeks before the beginning of the treatment.

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Imaging-based evaluation is preferred to evaluation by clinical examination unless the lesion(s) being followed cannot be imaged but are assessable by clinical exam.

Clinical lesions: Clinical lesions will only be considered measurable when they are superficial (e.g., skin nodules and palpable lymph nodes) and ≥10 mm diameter as assessed using calipers (e.g., skin nodules). In the case of skin lesions, documentation by color photography, including a ruler to estimate the size of the lesion, is recommended.

Chest x-ray: Lesions on chest x-ray are acceptable as measurable lesions when they are clearly defined and surrounded by aerated lung. However, CT is preferable.

Conventional CT and MRI: This guideline has defined measurability of lesions on CT scan based on the assumption that CT slice thickness is 5 mm or less. If CT scans have slice thickness greater than 5 mm, the minimum size for a measurable lesion should be twice the slice thickness. MRI is also acceptable in certain situations (e.g. for body scans).

Use of MRI remains a complex issue. MRI has excellent contrast, spatial, and temporal resolution; however, there are many image acquisition variables involved in MRI, which greatly impact image quality, lesion conspicuity, and measurement. Furthermore, the availability of MRI is variable globally. As with CT, if an MRI is performed, the technical specifications of the scanning sequences used should be optimized for the evaluation of the type and site of disease. Furthermore, as with CT, the modality used at follow-up should be the same as was used at baseline and the lesions should be measured/assessed on the same pulse sequence. It is beyond the scope of the RECIST guidelines to prescribe specific MRI pulse sequence parameters for all scanners, body parts, and diseases. Ideally, the same type of scanner should be used and the image acquisition protocol should be followed as closely as possible to prior scans. Body scans should be performed with breath-hold scanning techniques, if possible.

6.3.3 Response Criteria

All the scans performed at Baseline and other imaging performed as clinically required (other supportive imaging) need to be repeated at subsequent visits. In general, lesions detected at Baseline need to be followed using the same imaging methodology and preferably the same imaging equipment at subsequent tumor evaluation visits.

Brain CT / MRI scan should be performed, if clinically indicated by development of new specific symptoms or on the discretion of the Principal Investigator. For each subject, the Investigator will designate 1 or more of the following measures of tumor status to follow for determining response: CT or MRI images of primary and / or metastatic tumor masses, physical examination findings,

and the results of other assessments. All available images collected during the trial period will be considered. The most appropriate measures to evaluate the tumor status of a subject should be used. The measure(s) to be chosen for sequential evaluation during the trial have to correspond to the measures used to document the progressive tumor status that qualifies the subject for enrollment. The tumor response assessment will be assessed and listed according to the Study Calendar **3.7**.

The foreseen treatment duration is until disease progression verified by a scan subsequent to the initial documentation of PD, unacceptable toxicity, or any criterion for withdrawal from the trial occurs (see Section **3.8**). **Before stopping the treatment, progressive disease should be confirmed by imaging 4 to 6 weeks (preferably 6 weeks, but not later) after progression has been diagnosed according to RECIST 1.1.** If progression is based on the occurrence of a new lesion in an area not scanned at Baseline, a further on-study scan 6 weeks later should be considered before performing the 28-Day Safety Follow-up visit. Treatment may be continued despite progression according to RECIST 1.1 at any time if:

- There are no new symptoms or worsening of existing symptoms.
- There is no decrease in ECOG PS.
- The Investigator does not consider it necessary to administer a salvage therapy.

The treatment should be stopped immediately, if the subject does not tolerate M7824 in combination with gemcitabine anymore or if therapeutic failure occurs, which requires urgent treatment with an additional drug or results in clinically significant progression / deterioration.

Tumor responses to treatment will be assigned based on the evaluation of the response of target, non-target, and new lesions according to RECIST 1.1 (all measurements should be recorded in metric notation).

- To assess objective response, the tumor burden at baseline will be estimated and used for comparison with subsequent measurements. At baseline, tumor lesions will be categorized in target and non-target lesions according to RECIST 1.1.

Results for these evaluations will be recorded with as much specificity as possible so that pre- and post-treatment results will provide the best opportunity for evaluating tumor response.

Any CR or PR should be confirmed according to RECIST 1.1 (**Table 9**). **In the case of a PR or CR, a confirmatory CT or MRI scan should be done no sooner than 4 weeks (preferably at the scheduled 6-week interval)**.

The Investigator may perform scans in addition to a scheduled trial scan for medical reasons or if the Investigator suspects PD.

Table 9. Response Criteria for Patients with Measurable Disease (i.e., Target Disease)

Target Lesions	Non-Target Lesions	New Lesions	Overall Response	Best Overall Response when Confirmation is Required*
CR	CR	No	CR	≥4 wks. Confirmation**

CR	Non-CR/Non-PD	No	PR	≥4 wks. Confirmation**
CR	Not evaluated	No	PR	
PR	Non-CR/Non-PD/not evaluated	No	PR	
SD	Non-CR/Non-PD/not evaluated	No	SD	Documented at least once ≥4 wks. from baseline**
PD	Any	Yes or No	PD	no prior SD, PR or CR
Any	PD***	Yes or No	PD	
Any	Any	Yes	PD	

* See RECIST 1.1 manuscript for further details on what is evidence of a new lesion.

** Only for non-randomized trials with response as primary endpoint.

*** In exceptional circumstances, unequivocal progression in non-target lesions may be accepted as disease progression.

Note: Patients with a global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time should be reported as *"symptomatic deterioration."* Every effort should be made to document the objective progression even after discontinuation of treatment.

For Patients with Non-Measurable Disease (i.e., Non-Target Disease)

Non-Target Lesions	**New Lesions**	**Overall Response**
CR	No	CR
Non-CR/non-PD	No	Non-CR/non-PD*
Not all evaluated	No	not evaluated

Unequivocal PD	Yes or No	PD
Any	Yes	PD
* 'Non-CR/non-PD' is preferred over 'stable disease' for non-target disease since SD is increasingly used as an endpoint for assessment of efficacy in some trials so to assign this category when no lesions can be measured is not advised		

Subjects who have experienced SD, PR, or CR should continue treatment through the end of 24 months, although additional treatment is possible. If the Investigator believes that a subject may benefit from treatment beyond 24 months, it may be permissible.

6.3.4 Responses

6.3.4.1 Best overall response (BOR)

The duration of best overall response is measured from the time measurement criteria are met for CR, PR or SD (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started).

The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that progressive disease is objectively documented.

6.3.4.2 Duration of stable disease

Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started, including the baseline measurements. Stable Disease (SD) when sum of all target lesions does not qualify for CR/PR/PD for Target Lesion Response and Persistence of non-target lesions on Non-Target Lesion Response.

6.4 Immune-related Response Criteria (irRECIST)

Modified immune-related response criteria (irRECIST) will also be employed in this study. This new classification is based on the recent learning from clinical studies with cancer immunotherapies that even if some new lesions appear at the beginning of a treatment or if the total tumor burden does not increase substantially, tumor regressions or stabilizations might still occur later. For this trial, the concepts of the irRECIST are combined with RECIST 1.1. Please refer to **Appendix C** for further details.

6.4.1 Immune-related BOR (irBOR)

The duration of Immune-related best overall response is measured from the time measurement criteria are met for irCR or irPR (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented (taking as reference for progressive disease the smallest measurements recorded since the treatment started). irSD, not meeting criteria for irCR or irPR, in absence of irPD.

The duration of overall irCR is measured from the time measurement criteria are first met for irCR until the first date that progressive disease is objectively documented. Confirmation of assessment might be delayed up to 12 weeks to confirm PD to account for flare.

6.5 Toxicity Criteria

The following adverse event management guidelines are intended to ensure the safety of each patient while on the study. The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50).

The safety assessments will be performed according to the Study Calendar**,** Section **3.7**.

7 SAFETY REPORTING REQUIREMENTS / DATA AND SAFETY MONITORING PLAN

7.1 Definitions

7.1.1 Adverse Event

Any untoward medical occurrence in a human subject, including any abnormal sign (for example, abnormal physical exam or laboratory finding), symptom, or disease, temporally associated with the subject's participation in research, whether or not considered related to the subject's participation in the research.

7.1.2 Suspected adverse reaction

Suspected adverse reaction means any adverse event for which there is a reasonable possibility that the drug 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.

7.1.3 Unexpected adverse reaction

An adverse event or suspected adverse reaction is considered "unexpected" if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed; or, if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan or elsewhere in the current application. "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 from the pharmacological properties of the drug but are not specifically mentioned as occurring with the particular drug under investigation.

7.1.4 Serious

An Unanticipated Problem or Protocol Deviation is serious if it meets the definition of a Serious Adverse Event or if it compromises the safety, welfare or rights of subjects or others.

7.1.5 Serious Adverse Event

An adverse event or suspected adverse reaction is considered serious if in the view of the investigator or the sponsor, it results in any of the following:

- Death,
- A life-threatening adverse drug experience

- Inpatient hospitalization or prolongation of existing hospitalization
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

7.1.6 Disability

A substantial disruption of a person's ability to conduct normal life functions.

7.1.7 Life-threatening adverse drug experience

Any adverse event or suspected adverse reaction that places the patient or subject, in the view of the investigator or sponsor, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that had it occurred in a more severe form, might have caused death.

7.1.8 Protocol Deviation (NIH Definition)

Any change, divergence, or departure from the IRB-approved research protocol.

7.1.9 Non-Compliance (NIH Definition)

The failure to comply with applicable NIH Human Research Protections Program (HRPP) policies, IRB requirements, or regulatory requirements for the protection of human research subjects.

7.1.10 Unanticipated Problem

Any incident, experience, or outcome that:

- Is unexpected in terms of nature, severity, or frequency in relation to

  (a) the research risks that are described in the IRB-approved research protocol and informed consent document; Investigator's Brochure or other study documents, and

  (b) the characteristics of the subject population being studied; **AND**
- Is related or possibly related to participation in the research; **AND**
- Suggests that the research places subjects or others at a *greater risk of harm* (including physical, psychological, economic, or social harm) than was previously known or recognized

7.2 NIH Intramural IRB and Clinical Director Reporting

7.2.1 NIH Intramural IRB and NCI CD Expedited Reporting of Unanticipated Problems and Deaths

The Protocol PI will report in the NIH Problem Form to the NIH Intramural IRB and NCI Clinical Director:

- All deaths, except deaths due to progressive disease
- All Protocol Deviations

- All Unanticipated Problems
- All non-compliance

Reports must be received within 7 days of PI awareness via iRIS.

7.2.2 NIH Intramural IRB Requirements for PI Reporting at Continuing Review

The protocol PI will report to the NIH Intramural IRB:

1. A summary of all protocol deviations in a tabular format to include the date the deviation occurred, a brief description of the deviation and any corrective action.
2. A summary of any instances of non-compliance
3. A tabular summary of the following adverse events:
   - All Grade 2 **unexpected** events that are possibly, probably or definitely related to the research;
   - All Grade 3 and 4 events that are possibly, probably or definitely related to the research;
   - All Grade 5 events regardless of attribution.
   - All Serious Events regardless of attribution.

   NOTE: Grade 1 events are not required to be reported.

7.2.3 NIH Intramural IRB Reporting of IND Safety Reports

Only IND Safety Reports that meet the definition of an unanticipated problem will need to be reported to the NIH Intramural IRB.

7.3 IND Sponsor Reporting Criteria

From the time of the first study treatment through the first 28 days after the subject receives the last investigational agent/intervention, the investigator must **immediately** report to the sponsor, using the mandatory MedWatch form 3500a or equivalent, any serious adverse event, whether or not considered drug related, including those listed in the protocol or investigator brochure and must include an assessment of whether there is a reasonable possibility that the drug caused the event. For events that occur more than 28 days after the last administration of investigational agent/intervention, only report serious adverse events that have an attribution of at least possibly related to the agent/intervention.
Required timing for reporting per the above guideline:

- Deaths (except death due to progressive disease) must be reported via email within 24 hours. A complete report must be submitted within one business day.
- Other serious adverse events including deaths due to progressive disease must be reported within one business day

Events will be submitted to Center for Cancer Research (CCR) at:

CCRsafety@mail.nih.gov and to the CCR PI and study coordinator.

7.3.1 Reporting Pregnancy

7.3.1.1 Maternal exposure

If a patient becomes pregnant during the course of the study, the study treatment should be discontinued immediately, and the pregnancy reported to the Sponsor. The potential risk of exposure of the fetus to the investigational agent(s) or chemotherapy agents (s) should be documented in box B5 of the MedWatch form "Describe Event or Problem".

Pregnancy itself is not regarded as an SAE. However, as patients who become pregnant on study risk intrauterine exposure of the fetus to agents which may be teratogenic, the CCR is requesting that pregnancy should be reported in an expedited manner as **Grade 3** "***Pregnancy, puerperium and perinatal conditions - Other (pregnancy)***" under the ***Pregnancy, puerperium and perinatal conditions*** SOC.

Congenital abnormalities or birth defects and spontaneous miscarriages should be reported and handled as SAEs. Elective abortions without complications should not be handled as AEs. The outcome of all pregnancies (spontaneous miscarriage, elective termination, ectopic pregnancy, normal birth, or congenital abnormality) should be followed up and documented.

If any pregnancy occurs in the course of the study, then the investigator should inform the Sponsor within 1 day, i.e., immediately, but **no later than 24 hours** of when he or she becomes aware of it.

The designated Sponsor representative will work with the investigator to ensure that all relevant information is provided to the Sponsor within 1 to 5 calendar days for SAEs and within 30 days for all other pregnancies.

The same timelines apply when outcome information is available.

7.3.1.2 Paternal exposure

Male patients should refrain from fathering a child or donating sperm during the study and for 4 months after the last treatment with M7824 or gemcitabine whichever occurs later. Pregnancy of the patient's partner is not considered to be an AE. However, the outcome of all pregnancies (spontaneous miscarriage, elective termination, ectopic pregnancy, normal birth, or congenital abnormality) occurring from the date of the first dose until 4 months after the last treatment with M7824 or gemcitabine whichever occurs later should, if possible, be followed up and documented.

7.4 Reporting Criteria to the Pharmaceutical Collaborators

All events listed below must be reported in the defined timelines to CCRsafety@mail.nih.gov.

The CCR Office of Regulatory Affairs will send all reports to the manufacturer as described below, except as noted.

7.4.1 EMD-Serono

The following reportable events must be submitted to EMD Serono within 2 business days or 3 calendar days (whichever comes first) using the Medwatch form or equivalent. The Sponsor will assume responsibility for submitting the reportable event(s) to EMD Serono as well as ensuring that any local reporting requirements are completed in parallel.

- Serious Adverse Events
- Exposure during Pregnancy or Breastfeeding (even if not associated with an adverse event)
- Occupational exposure (even if not associated with an adverse event)

- Potential drug-induced liver injury (Hy's Law cases): These events are considered important medical events and should be reported as SAEs.
- In addition, all AEs will be collected in tabulated form and reported to EMD Serono as outlined in the Collaborative Agreement. **Note:** The PI and Research Team will be responsible for reporting the AEs that are not specifically listed above.

7.4.1.1 Reporting of Overdose of M7824

An overdose is defined as any dose 5% greater than the highest dose included in the clinical trial protocol. Any overdose must be recorded in the trial medication section of the eCRF.

For monitoring purposes, any case of overdose, whether or not associated with an AE (serious or non-serious), must be reported to the sponsor.

There are no known symptoms of M7824 overdose to date. The Investigator should monitor closely for AEs should an overdose occur and use his or her clinical judgment in providing symptomatic / supportive care as medically indicated. There is no known antidote for M7824.

Contact information for submission of reportable events to EMD Serono:

E-mail: ICSR_CT_GPS@merckgroup.com

Specifying:

PROTOCOL Number and/or Title

EMD Serono assigned Study Number (MS200647_0015)

SUBJECT Number

PI Name

SAE/ONSET DATE.

7.5 Data and Safety Monitoring Plan

7.5.1 Principal Investigator/Research Team

The clinical research team will meet on a regular basis (every two weeks by teleconference with the pharmaceutical collaborator) when patients are being actively treated on the trial to discuss each patient, enrollment and data management issues. Decisions about dose level enrollment and dose escalation will be made based on the toxicity data from prior patients.

All data will be collected in a timely manner and reviewed by the principal investigator or a lead associate investigator. Adverse events will be reported as required above. Any safety concerns, new information that might affect either the ethical and or scientific conduct of the trial, or protocol deviations will be immediately reported to the IRB using iRIS.

The principal investigator will review adverse event and response data on each patient to ensure safety and data accuracy. The principal investigator will personally conduct or supervise the investigation and provide appropriate delegation of responsibilities to other members of the research staff.

7.5.2 Sponsor Monitoring Plan

As a sponsor for clinical trials, FDA regulations require the CCR to maintain a monitoring program. The CCR's program allows for confirmation of: study data, specifically data that could

affect the interpretation of primary study endpoints; adherence to the protocol, regulations, and SOPs; and human subjects protection. This is done through independent verification of study data with source documentation focusing on:

- Informed consent process
- Eligibility confirmation
- Drug administration and accountability
- Adverse events monitoring
- Response assessment.

The monitoring program also extends to multi-site research when the CCR is the coordinating center.

This trial will be monitored by personnel employed by a CCR contractor. Monitors are qualified by training and experience to monitor the progress of clinical trials. Personnel monitoring this study will not be affiliated in any way with the trial conduct.

8 STATISTICAL CONSIDERATIONS

8.1 Statistical Considerations

8.1.1 Primary efficacy endpoints

The primary objectives of this trial are to determine the safety and tolerability, measured by number, severity and duration of treatment-related AEs for the combination treatment M7824 and gemcitabine according to CTCAE v5.0, and the occurrence, severity, and duration of Treatment-Emergent Adverse Events (TEAEs) for the combination treatment M7824 and gemcitabine according to the NCI-CTCAE v5.0. and the efficacy, as measured by Best Objective Response (BOR) rate according to RECIST 1.1

8.1.2 Secondary efficacy endpoints

The secondary endpoints are the following:

- Progression-free Survival (PFS)
- Overall survival (OS)
- Immune-related BOR (irBOR) using the immune-related RECIST (irRECIST)

8.2 Sample size determination

Prior to amendment B, the protocol enrolled 6 evaluable patients into safety run-in and according to DLT definition, DLTs were recorded. We want to change DLT definition and the results from these initial 6 patients will be reported as a separate group, and accrual will begin again using the same phase IB design as initially proposed, but with new DLT definition described in amendment B. Total amount of patient on trial will be the same as with this amendment we are removing 200 mg M7824 dose level.

Phase IB:

This combination will be first given to 6 patients as a safety run-in: the doses are 1,200 mg for M7824 and an assigned gemcitabine dose: 600 or 1,000 mg/m^2 weekly according to baseline

pharmacogenomic profiling. If there are 0-1 grade 3 and 4 DLTs, the study will proceed to the phase 2 cohorts. If there are ≥2 grade 3 and 4 DLTs, there will be a dose reduction to 500 mg M7824 and the safety run-in in 6 patients will be repeated. With these 6 patients who have 0-1 DLTs in 6 patients at the reduced level of M7824, the trial will then be carried out in the phase 2 cohorts. If 2 or more grade 3 or greater toxicities are seen in this second run-in of 6 patients, the study will be halted.

Thus, no more than 12 evaluable patients will be enrolled in the phase IB cohort with Amendment B.

Phase II:

The study will be conducted using a Simon two-stage phase 2 minimax design. The objective of the trial will be to determine whether this novel combination can be associated with a response rate (PR + CR; RECIST) that can rule out 5% (p0=0.05) in favor of an improved response rate of 25% (p1=0.25). Using alpha=0.10 (probability of accepting a poor agent) and beta=0.10 (probability of rejecting a good agent), initially 13 evaluable patients will be enrolled into the phase II study, including up to 6 patients from the completed safety cohort if they are completely eligible for the phase II portion of the trial. If 0 of 13 patients respond, then no further patients will be enrolled. If 1 or more of the first 13 evaluable patients enrolled have a response (PR or CR), then accrual will continue until a total of 20 evaluable patients have been enrolled. Since it may take several weeks for a response to be identified, a pause in the accrual of 6 weeks will be made before accruing to the second stage. If 1 to 2 of the 20 has a clinical response, then this will be considered inadequate for further investigation of this regimen. If 3 or more of 20 (15%) respond, then this will warrant further investigation in a subsequent trial. Under the null hypothesis (5% response rate), the probability of early termination is 51.3%.

Although participants who are in the phase II cohort will receive different levels of gemcitabine based on their genetic results, the participants will have their outcomes combined and will be evaluated using a single two-stage design with up to 20 participants. This will then assess the ability of the treatment strategy of gemcitabine (regardless of dose 600 or 1000 mg/m2) and M7824 at a consistent dose to act together and produce responses.

Thus, with 6 patients enrolled prior to Amendment B, up to 12 participants in the phase IB cohort starting with Amendment B and up to 20 evaluable participants in phase II, the trial may require up to 38 participants to be treated. This number may be reduced by up to 6 participants treated at the RP2D in the phase IB portion of the study. To allow for a small number of inevaluable patients, the accrual ceiling will be set at 41 participants. With an anticipated accrual rate of 10-12 patients per year, it is expected that the trial may complete accrual within 3 to 4 years.

8.2.1 Populations for analysis

Evaluable for objective response: all patients who receive at least one dose of M7824 in combination with gemcitabine at the dose level determined to be safe in the initial safety run-in in 6 patients will be included in the statistical analyses performed.)

Evaluable Non-Target Disease Response: all patients who receive at least one dose of M7824 in combination with gemcitabine at the dose level determined to be safe in the initial safety run-in in 6 patients will be included in the statistical analyses performed. The response assessment is based on the presence, absence, or unequivocal progression of the lesions.

Evaluable for toxicity: All patients will be evaluable for toxicity from the time of their first treatment with M7824.

Evaluable for RP2D: Subject enrolled to the safety run in portion of the study are evaluable. However, subjects who do not complete the DLT observation period for reasons other than a DLT will be replaced and not included in the evaluation.

8.3 Description of Statistical Analyses

8.3.1 General Considerations

Patients who are in the safety evaluation will have the number of patients with a DLT determined. Patients who are in the efficacy evaluation and patients from the safety run-in administered the M7824 dose used in the phase II expansion will have the fraction of clinical responses determined.

8.3.2 Analysis of the primary efficacy endpoints

Patients who are in the efficacy evaluation will have the fraction of clinical responses determined and reported along with two sided 80% and 95% confidence intervals.

8.3.3 Analysis of the secondary efficacy endpoints

Overall survival and PFS will be determined in all patients treated at the RP2D of the combination of M7824 and gemcitabine using Kaplan-Meier curves. Any comparisons with published results will be interpreted as being exploratory. The immune-related BOR (irBOR) rate will be determined by dividing the number of patients with an immune-related response by the number of evaluable patients who are treated at the RP2D. The fraction with irBOR will be reported along with two-sided 80% and 95% confidence intervals.

At the end of the trial, as a secondary analysis, a retrospective evaluation will be performed to determine the individual response rates based on the participants' genetic /treatment results. This evaluation will include 80% and 95% two-sided confidence intervals for each of the treatment arms within the phase II cohort.

8.3.4 Safety analyses

Patients in the phase I portion of the trial will be assessed for toxicity by reporting the grades of toxicity and the type of toxicity observed for all patients. Patients will be enrolled and evaluated in the phase IB cohort of the trial as described in the phase IB sample size determination section. Patients will continue to be enrolled onto the phase II cohort if the cumulative fraction of patients with greater or equal grade 3 DLT is less than 1/3.

8.3.5 Baseline descriptive statistics

Limited demographic and clinical characteristics of all patients will be reported, separately for the phase IB and the phase II cohorts.

8.3.6 Planned interim analyses:

Toxicity will be assessed within each group of 6 patients accrued in the phase IB cohort. An evaluation of the fraction of patients who respond in the phase IB part will be included in the phase II cohort, as indicated in the phase II sample size determination section.

8.3.7 Exploratory analyses

Exploratory objectives of the study are to compare each of following parameters between patients in the phase II cohorts of the trial who are clinical responders vs. non-responders, using the approach described below for each:

Tumor perfusion measured by Dynamic Contrast Enhanced MRI (DCE-MRI); this will primarily compare the Houndsfield units obtained between responders and non-responders using an exact Wilcoxon rank sum test.

Impact of baseline somatic genomic profile of advanced pancreatic cancers on clinical response to M7824 in combination with gemcitabine. The number of somatic mutations obtained for each patient will be compared between responders and non-responders. This will be done using either a Fisher's exact test, Cochran-Armitage trend test, or exact Wilcoxon rank sum test depending on the distributions of values.

Determination of circulating free tumor DNA (cftDNA) levels to correlate with clinical course of pancreatic cancer patients treated with M7824 in combination with gemcitabine; the levels will be compared between responders and non-responders using an exact Wilcoxon rank sum test.

Measures of intra-tumoral immune cell fractions to evaluate the intra-tumoral immunogenicity of M7824 in combination with gemcitabine; these fractions per patient will be compared between responders and non-responders using an appropriate non-parametric test.

Health-related Quality of Life (HRQoL) scores measuring patient-reported disease-specific symptoms and treatment-related concerns; these will be compared between responders and non-responders using an exact Wilcoxon rank sum test.

9 COLLABORATIVE AGREEMENTS

9.1 Cooperative Research and Development Agreement (CRADA)

A CRADA (02666) is in place with EMD Serono for the supply of M7824.

10 HUMAN SUBJECTS PROTECTIONS

10.1 Rationale For Subject Selection

Subjects from all racial and ethnic groups are eligible for this trial if they meet the eligibility criteria. Efforts will be made to extend the accrual to a representative population.

10.2 Participation of Children

Individuals under the age of 18 will not be eligible to participate in this study because they are unlikely to have pancreatic cancer, and because of unknown toxicities in pediatric patients.

10.3 Participation of subjects unable to give consent

Adults unable to give consent are excluded from enrolling in the protocol. However, re-consent may be necessary and there is a possibility, though unlikely, that subjects could become decisionally impaired. For this reason and because there is a prospect of direct benefit from research participation (Section **10.4.2**), all subjects will be offered the opportunity to direct their wishes for research and care to a surrogate, and assign a substitute decision maker on the "NIH Advance Directive for Health Care and Medical Research Participation" form so that another person can make decisions about their medical care in the event that they become incapacitated or cognitively impaired during the course of the study. Note: The PI or AI will contact the NIH Ability to Consent Assessment Team (ACAT) for evaluation as needed for the following: an independent assessment of whether an

individual has the capacity to provide consent; assistance in identifying and assessing an appropriate surrogate when indicated; and/or an assessment of the capacity to appoint a surrogate. For those subjects that become incapacitated and do not have pre-determined substitute decision maker, the procedures described in MEC Policy 87-4 and NIH HRPP SOP 14E for appointing a surrogate decision maker for adult subjects who are (a) decisional impaired, and (b) who do not have a legal guardian or durable power of attorney, will be followed.

10.4 Evaluation of Benefits and Risks/Discomforts

The primary risk to patients participating in this research study is from the toxicity of M7824 and gemcitabine, or both drugs. M7824 is an investigational agent designed to enhance antitumor efficacy of standard treatment. Gemcitabine is an approved agent for treatment of pancreatic cancer. The protocol provides for detailed and careful monitoring of all patients to assess for toxicity. Toxicity data from the phases I and II dose levels will be collected and reviewed to ensure that there were no severe toxicities that would preclude further patient enrollment. Patients will be treated with therapeutic intent and response to the therapy will be closely monitored.

10.4.1 Risks

10.4.1.1 Risk of Biopsies

All care will be taken to minimize risks that may be incurred by tumor sampling. Biopsies will be taken by Interventional Radiology sampling the lesion which is most accessible and can be sampled with the least morbidity. Up to 6 passages are allowed if deemed safe by the Interventional Radiologist. All procedure-related risks (such as bleeding, infection and visceral injury) that will be explained fully during informed consent.

10.4.1.2 Risks of exposure to ionizing radiation

This research study involves two optional CT guided biopsies collected for research purposes. Subjects undergoing two biopsies collections will be exposed to 1.6 rem. This amount of radiation is below the guideline of 5 rem per year allowed for adult research subjects by the NIH Radiation Safety Committee.

10.4.1.3 Risks of Dynamic Contrast Enhanced MRIs (DCE-MRI)

This research study involves up to 5 Dynamic Contrast Enhanced MRIs (DCE-MRI) performed for research purposes. The main risk is allergic reaction to IV administered contrast agent. This risk is in the range of 1 to 1.5 %.

10.4.1.4 Research Blood Collection Risks

Risks of blood draws include pain and bruising in the area where the needle is placed, lightheadedness, and rarely, fainting. When large amounts of blood are collected, low red blood cell count (anemia) can develop.

10.4.1.5 Other Risks

Risks include the possible occurrence of any of a range of side effects which are listed in the Consent Document or this protocol document. Frequent monitoring for adverse effects will help to minimize the risks associated with administration of the study agents.

10.4.1.6 Non-Physical Risks of Genetic Research

Risk of receiving unwanted information

Anxiety and stress may arise as a result of the anticipation that unwanted information regarding disease related DNA sequencing or disease tendencies, or misattributed paternity. Patients will be clearly informed that the data related to DNA sequencing and genetic analysis is coded, investigational and will not be shared with patients, family members or health care providers.

Risk related to possibility that information may be released

This includes the risk that data related to genotype, DNA sequencing or risk for disease tendency or trait can be released to members of the public, insurers, employers, or law enforcement agencies. Although there are no plans to release results to the patients, family members or health care providers, this risk will be included in the informed consent document.

10.4.2 Benefits

The potential benefit to a patient that goes onto study is a reduction in the bulk of their tumor which may or may not have favorable impact on symptoms and/or survival.

10.5 Risks/Benefits Analysis for All Participants

For patients with advanced pancreas cancer, median survival is in the range of 6 months. It is possible that treatment on this protocol may reduce tumor burden or lessen symptoms caused by the cancer. While treatment on this protocol may not individually benefit subjects, the knowledge gained from this study may help others in the future who have pancreatic cancer. Potential risks include the possible occurrence of any of a range of side effects listed. If patients suffer any physical injury as a result of the biopsies, immediate medical treatment is available at the NIH's Clinical Center in Bethesda, Maryland. Although no compensation is available, any injury will be fully evaluated and treated in keeping with the benefits or care to which patients are entitled under applicable regulations.

10.6 Consent Process and Documentation

The investigational nature and research objectives of this trial, the procedures and treatments involved and their attendant risks and discomforts and benefits, and potential alternative therapies will be carefully explained to the patient, and a signed informed consent document will be obtained by a study investigator prior to entry onto the study.

The PI or associate investigator will meet with the patient to discuss the protocol treatment and alternative options in detail. It will be stated clearly that participation in the research study is voluntary and that participants can withdraw from the study without losing benefits they would otherwise be entitled to. The patient will be encouraged to ask questions, and additional meetings to discuss the treatment options will be arranged as necessary.

10.6.1 Re-Consent via Telephone

The informed consent document will be sent to the subject. An explanation of the study will be provided over the telephone after the subject has had the opportunity to read the consent form. The subject will sign and date the informed consent.

The original informed consent document will be sent back to the consenting investigator who will sign and date the consent form with the date the consent was obtained via telephone.

A fully executed copy will be returned via mail for the subject's records.

11 PHARMACEUTICAL INFORMATION

11.1 M7824

Other names: Anti PDL-1 TGF Beta-Trap, "TRAP", MSB0011359C

The finished injectable dosage form is manufactured by EMD Serono Research & Development Institute, Inc.

11.1.1 Status:

For Investigational Use Only.

11.1.2 Brief Mechanism of Action/Drug Classification:

M7824 is a fully human antibody of the IgG1 isotype designed as a single molecule that blocks the following two major immune escape mechanisms (negative immunoregulatory pathways) simultaneously: PD-L1/PD1 signaling and TGF-beta signaling. PDL1 is highly expressed in tumor cells, which strongly correlates with poor prognosis in a variety of human cancers. TGF-beta is associated with malignant progression, evasion of immune surveillance, invasion and metastasis. Elevated levels of TGF-beta correlate with poor outcome in many different human cancers.

11.1.3 How Supplied:

M7824 is supplied in two different formulations: a sterile lyophilized powder and a sterile liquid.

Lyophilized (freeze-dried) formulation:

Supplied as a Type 1 glass vial containing a white lyophilized powder. Each vial contains 45 mg of M7824 and 6% (w/v) trehalose dihydrate, 40 mM sodium chloride, 5 mM L-methionine and 0.05% (w/v) polysorbate 20 (Tween 20) and 10 mM L-histidine at pH 5.5.

Liquid formulation:

Supplied as a Type 1 glass vial containing an extractable volume of 60 mL of a sterile, clear, colorless solution. Each mL contains 10 mg of M7824 and 6% (w/v) trehalose dihydrate, 40 mM sodium chloride, 5 mM L-methionine and 0.05% (w/v) polysorbate 20 (Tween 20) and 10 mM L-histidine at pH 5.5.

11.1.4 Storage:

Store intact vials, both lyophilized and liquid formulations, in the refrigerator (2°C – 8°C). Do NOT freeze the vials. The vials must be stored in the original packaging and protected from light until use.

Store reconstituted lyophilized vials and diluted infusion solutions at room temperature.

11.1.5 Reconstitution:

For lyophilized formulation only:

Allow each vial to equilibrate to room temperature preferably for 10 to 20 minutes.

Reconstitute each 45 mg vial with 4.5 mL of Sterile Water for Injection, USP (SWI) using a sterile syringe and needle. An 18 gauge needle is suggested. Avoid forceful impact of the SWI on the powder and avoid foaming. Slowly and gently add the SWI to the vial, aiming the flow of water towards the vial wall and NOT directly onto the lyophilized powder cake. Start the clock at this point. Gently invert each vial several times to mix. Do not shake or agitate vigorously. Stop the

clock after the product is completely dissolved. The reconstitution time MUST be no more than 120 seconds. When reconstituted as recommended, each mL will contain 10 mg of M7824 as a clear, colorless solution.

NOTE: If the reconstitution time is greater than 120 seconds, put the vial aside to return to IDMRS and get a new vial to prepare the dose.

11.1.6 Stability:

Reconstituted lyophilized vials, if not used immediately, are stable at room temperature for up to 24 hours.

When diluted with 0.9% Sodium Chloride Injection, USP to a concentration of 0.16 mg/mL to 9.6 mg/mL, chemical and physical in-use stability has been demonstrated for 72 hours when stored at room temperature.

11.1.7 Administration:

M7824 is administered as an intravenous (IV) infusion via a peripheral OR central vascular access device (VAD). Confirm patient has a titanium port before accessing the Central VAD. A 0.2 micron polyethersulfone (PES) in-line filter is mandatory for administration.

11.1.8 Compatibility:

M7824 is compatible with 0.9% Sodium Chloride Injection, USP.

M7824 is compatible with DEHP-free infusion bags composed of polyethylene and/or polypropylene. Acceptable bags include B. Braun EXCEL® containers prefilled with 0.9% Sodium Chloride Injection, USP. Baxter INTRAVIA™ empty containers and AVIVA pre-filled bags may also be used.

M7824 is compatible with DEHP-free polyvinylchloride (PVC) and polyurethane (PUR) infusion lines. A polyethersulfone (PES) 0.2 micron in-line filter is required during administration.

11.1.9 Preparation of Infusion:

1. If using the liquid formulation, allow each vial to equilibrate to room temperature preferably for 10 to 20 minutes. If using the lyophilized formulation, first follow reconstitution instructions above.

2. Gently invert the vial(s) several times before use. Do NOT shake or agitate vigorously.

3. Calculate the volume of M7824 (10 mg/mL) required to prepare a patient's dose. Note: Pharmacy will add 25 mL overfill (fluid plus drug).

4. Prepare a bag containing the amount of 0.9% Sodium Chloride Injection, USP required to dilute the dose to a total volume of 250 mL. Note: Pharmacy will add 25 mL overfill (fluid plus drug).

5. With a syringe and sterile 18 gauge needle, withdraw from the vials containing the M7824 the volume calculated in Step #2 and inject the drug into the bag containing the 0.9% Sodium Chloride Injection, USP.

6. Gently mix the solution by slowly inverting the container 10 times to ensure a homogeneous clear solution free of visible particles. Do NOT shake the admixture to mix.

7. Purge all air from the container.

8. Attach to the container an administration set for ambulatory pump with an integral 0.2 micron filter. Prime the tubing close to its distal end and cap the administration set with a Luer-locking cap.

9. The prescription label will identify the total volume to infuse: "Volume to Infuse is 250 mL" and will state the product contains overfill (25 mL fluid + drug).

11.1.10Hazards and Precautions:

M7824 should be handled and labeled as a hazardous drug

11.1.11Labeled:

M7824 AAPC.

11.2 Gemcitabine

Gemcitabine HCl is a white to off-white solid. It is soluble in water, slightly soluble in methanol, and practically insoluble in ethanol and polar organic solvents. Gemcitabine will be provided and prepared by the Clinical Center Pharmacy Department. The clinical formulation is supplied in a sterile form for intravenous use only. Vials of gemcitabine contain either 200 mg or 1 g of gemcitabine HCl (expressed as free base) formulated with mannitol (200 mg or 1 g, respectively) and sodium acetate (12.5 mg or 62.5 mg, respectively) as a sterile lyophilized powder. Hydrochloric acid and/or sodium hydroxide may have been added for pH adjustment

11.2.1 Toxicity

Data in **Table 10** are based on 979 patients receiving gemcitabine as a single-agent administered weekly as a 30-minute infusion for treatment of a wide variety of malignancies. The gemcitabine starting doses ranged from 800 to 1250 mg/m^2. The frequency of all grades and severe (WHO Grade 3 or 4) adverse events were generally similar in the single-agent safety database of 979 patients and the subset of patients with pancreatic cancer. Adverse reactions reported in the single-agent safety database resulted in discontinuation of gemcitabine therapy in about 10% of patients. In the comparative trial in pancreatic cancer, the discontinuation rate for adverse reactions was 14.3% for the gemcitabine arm and 4.8% for the 5-FU arm. All WHO-graded laboratory events are listed in **Table 10**, regardless of causality. Non-laboratory adverse events were reported, regardless of causality, for at least 10% of all patients, except the categories of Extravasation, Allergic, and Cardiovascular and certain specific events under the Renal, Pulmonary, and Infection categories (from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf).

Table 10. Selected WHO-graded AEs in patients receiving single-agent gemcitabine (from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf)

	All Patients[a]			Pancreatic Cancer Patients[b]			Discontinuations (%)[c]
	All Grades	Grade 3	Grade 4	All Grades	Grade 3	Grade 4	All Patients
Laboratory[d]							
Hematologic							
Anemia	68	7	1	73	8	2	<1
Leukopenia	62	9	<1	64	8	1	<1
Neutropenia	63	19	6	61	17	7	-
Thrombocytopenia	24	4	1	36	7	<1	<1
Hepatic							<1
ALT	68	8	2	72	10	1	
AST	67	6	2	78	12	5	
Alkaline Phosphatase	55	7	2	77	16	4	
Bilirubin	13	2	<1	26	6	2	
Renal							<1
Proteinuria	45	<1	0	32	<1	0	
Hematuria	35	<1	0	23	0	0	
BUN	16	0	0	15	0	0	
Creatinine	8	<1	0	6	0	0	
Non-laboratory[e]							
Nausea and Vomiting	69	13	1	71	10	2	<1
Pain	48	9	<1	42	6	<1	<1
Fever	41	2	0	38	2	0	<1
Rash	30	<1	0	28	<1	0	<1
Dyspnea	23	3	<1	10	0	<1	<1
Constipation	23	1	<1	31	3	<1	0
Diarrhea	19	1	0	30	3	0	0
Hemorrhage	17	<1	<1	4	2	<1	<1
Infection	16	1	<1	10	2	<1	<1
Alopecia	15	<1	0	16	0	0	0
Stomatitis	11	<1	0	10	<1	0	<1
Somnolence	11	<1	<1	11	2	<1	<1
Paresthesias	10	<1	0	10	<1	0	0

Grade based on criteria from the World Health Organization (WHO).
[a] N=699-974; all patients with laboratory or non-laboratory data.
[b] N=161-241; all pancreatic cancer patients with laboratory or non-laboratory data.
[c] N=979.
[d] Regardless of causality.
[e] Table includes non-laboratory data with incidence for all patients ≥10%. For approximately 60% of the patients, non-laboratory events were graded only if assessed to be possibly drug-related.

Most adverse events are reversible and do not need to result in discontinuation, although doses may need to be withheld or reduced. There was a greater tendency in women, especially older women, not to proceed to the next cycle. Gemcitabine clearance is affected by age. There is no evidence, however, that unusual dose adjustments are necessary in patients over 65 years of age, and in general, adverse reaction rates in the single-agent safety database of 979 patients were similar in patients above and below 65 years. Grade 3/4 thrombocytopenia was more common in the elderly. Gemcitabine clearance is affected by gender: in the single-agent safety database (N=979 patients), however, there is no evidence that unusual dose adjustments are necessary in women. In general, in single-agent studies of gemcitabine, adverse reaction rates were similar in men and women, but women, especially older women, were more likely not to proceed to a

subsequent cycle and to experience Grade 3/4 neutropenia and thrombocytopenia. Gemcitabine should be used with caution in patients with preexisting renal impairment or hepatic insufficiency. Gemcitabine has not been studied in patients with significant renal or hepatic impairment.

11.2.2 Formulation

Gemcitabine 200 mg white, lyophilized powder in a 10-mL size sterile single use vial (No. 7501) NDC 0002-7501-01 1 gram white, lyophilized powder in a 50-mL size sterile single use vial (No. 7502) NDC 0002-7502-01 Store at controlled room temperature (20° to 25°C) (68° to 77°F).

(from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf).

11.2.3 Preparation

The recommended diluent for reconstitution of Gemzar is 0.9% Sodium Chloride Injection without preservatives. Due to solubility considerations, the maximum concentration for Gemzar upon reconstitution is 40 mg/mL. Reconstitution at concentrations greater than 40 mg/mL may result in incomplete dissolution, and should be avoided. To reconstitute, add 5 mL of 0.9% Sodium Chloride Injection to the 200-mg vial or 25 mL of 0.9% Sodium Chloride Injection to the 1-g vial. Shake to dissolve. These dilutions each yield a gemcitabine concentration of 38 mg/mL which includes accounting for the displacement volume of the lyophilized powder (0.26 mL for the 200-mg vial or 1.3 mL for the 1-g vial). The total volume upon reconstitution will be 5.26 mL or 26.3 mL, respectively. Complete withdrawal of the vial contents will provide 200 mg or 1 g of gemcitabine, respectively. The appropriate amount of drug may be administered as prepared or further diluted with 0.9% Sodium Chloride Injection to concentrations as low as 0.1 mg/mL. Reconstituted Gemzar is a clear, colorless to light straw-colored solution. After reconstitution with 0.9% Sodium Chloride Injection, the pH of the resulting solution lies in the range of 2.7 to 3.3. The solution should be inspected visually for particulate matter and discoloration, prior to administration, whenever solution or container permit. If particulate matter or discoloration is found, do not administer.

(from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf).

Caution should be exercised in handling and preparing gemcitabine solutions. The use of gloves is recommended. If Gemcitabine solution contacts the skin or mucosa, immediately wash the skin thoroughly with soap and water or rinse the mucosa with copious amounts of water. Although acute dermal irritation has not been observed in animal studies, 2 of 3 rabbits exhibited drug-related systemic toxicities (death, hypoactivity, nasal discharge, shallow breathing) due to dermal absorption.

(from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf).

11.2.4 Stability and Storage

When prepared as directed, gemcitabine solutions are stable for 24 hours at controlled room temperature 20° to 25°C (68° to 77°F). Discard unused portion. Solutions of reconstituted gemcitabine should not be refrigerated, as crystallization may occur. The compatibility of gemcitabine with other drugs has not been studied. No incompatibilities have been observed with infusion bottles or polyvinyl chloride bags and administration sets. Unopened vials of gemcitabine are stable until the expiration date indicated on the package when stored at controlled room temperature 20° to 25°C (68° to 77°F).

(from https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/020509s033lbl.pdf).

11.2.5 Administration Procedures

See Section **3.3.2**.

11.2.6 Incompatibilities and Overdose

The compatibility of gemcitabine with other drugs has not been studied. No incompatibilities have been observed with infusion bottles or polyvinyl chloride bags and administration sets. There is no known antidote for overdoses of gemcitabine. Myelosuppression, paresthesias, and severe rash were the principal toxicities seen when a single dose as high as 5,700 mg/m^2 was administered by I.V. infusion over 30 minutes every 2 weeks to several patients in a Phase 1 study. In the event of suspected overdose, the patient should be monitored with appropriate blood counts and should receive supportive therapy, as necessary.

12 REFERENCES

1. Rahib L, Smith BD, Aizenberg R, Rosenzweig AB, Fleshman JM, Matrisian LM: **Projecting cancer incidence and deaths to 2030: the unexpected burden of thyroid, liver, and pancreas cancers in the United States**. *Cancer Res* 2014, **74**(11):2913-2921.

2. Siegel RL, Miller KD, Jemal A: **Cancer statistics, 2016**. *CA Cancer J Clin* 2016, **66**(1):7-30.

3. Conroy T, Desseigne F, Ychou M, Bouche O, Guimbaud R, Becouarn Y, Adenis A, Raoul JL, Gourgou-Bourgade S, de la Fouchardiere C *et al*: **FOLFIRINOX versus gemcitabine for metastatic pancreatic cancer**. *N Engl J Med*, **364**(19):1817-1825.

4. Von Hoff DD, Ervin T, Arena FP, Chiorean EG, Infante J, Moore M, Seay T, Tjulandin SA, Ma WW, Saleh MN *et al*: **Increased survival in pancreatic cancer with nab-paclitaxel plus gemcitabine**. *N Engl J Med*, **369**(18):1691-1703.

5. Hingorani SR, Harris WP, Beck JT, Berdov BA, Wagner SA, Pshevlotsky EM, Tjulandin SA, Gladkov OA, Holcombe RF, Korn R *et al*: **Phase Ib Study of PEGylated Recombinant Human Hyaluronidase and Gemcitabine in Patients with Advanced Pancreatic Cancer**. *Clin Cancer Res* 2016, **22**(12):2848-2854.

6. Thind K, Padrnos LJ, Ramanathan RK, Borad MJ: **Immunotherapy in pancreatic cancer treatment: a new frontier**. *Therap Adv Gastroenterol* 2017, **10**(1):168-194.

7. Hanks BA, Holtzhausen A, Evans KS, Jamieson R, Gimpel P, Campbell OM, Hector-Greene M, Sun L, Tewari A, George A *et al*: **Type III TGF-beta receptor downregulation generates an immunotolerant tumor microenvironment**. *J Clin Invest* 2013, **123**(9):3925-3940.

8. Lebrun JJ: **The Dual Role of TGFbeta in Human Cancer: From Tumor Suppression to Cancer Metastasis**. *ISRN Mol Biol* 2012, **2012**:381428.

9. Wrzesinski SH, Wan YY, Flavell RA: **Transforming growth factor-beta and the immune response: implications for anticancer therapy**. *Clin Cancer Res* 2007, **13**(18 Pt 1):5262-5270.

10. Hao NB, Lu MH, Fan YH, Cao YL, Zhang ZR, Yang SM: **Macrophages in tumor microenvironments and the progression of tumors**. *Clin Dev Immunol* 2012, **2012**:948098.

11. Smirne C, Camandona M, Alabiso O, Bellone G, Emanuelli G: **[High serum levels of Transforming Growth Factor-beta1, Interleukin-10 and Vascular Endothelial Growth Factor in pancreatic adenocarcinoma patients]**. *Minerva Gastroenterol Dietol* 1999, **45**(1):21-27.

12. Hotchkiss RS, Moldawer LL: **Parallels between cancer and infectious disease**. *N Engl J Med* 2014, **371**(4):380-383.

13. Burris HA, 3rd, Moore MJ, Andersen J, Green MR, Rothenberg ML, Modiano MR, Cripps MC, Portenoy RK, Storniolo AM, Tarassoff P *et al*: **Improvements in survival and clinical benefit with gemcitabine as first-line therapy for patients with advanced pancreas cancer: a randomized trial**. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 1997, **15**(6):2403-2413.

14. Donkor MK, Sarkar A, Li MO: **Tgf-beta1 produced by activated CD4(+) T Cells Antagonizes T Cell Surveillance of Tumor Development**. *Oncoimmunology* 2012, **1**(2):162-171.

15. Topalian SL, Drake CG, Pardoll DM: **Targeting the PD-1/B7-H1(PD-L1) pathway to activate anti-tumor immunity**. *Curr Opin Immunol* 2012, **24**(2):207-212.

16. Song S, Yuan P, Wu H, Chen J, Fu J, Li P, Lu J, Wei W: **Dendritic cells with an increased PD-L1 by TGF-beta induce T cell anergy for the cytotoxicity of hepatocellular carcinoma cells**. *Int Immunopharmacol* 2014, **20**(1):117-123.

17. Morris JC, Tan AR, Olencki TE, Shapiro GI, Dezube BJ, Reiss M, Hsu FJ, Berzofsky JA, Lawrence DP: **Phase I study of GC1008 (fresolimumab): a human anti-transforming growth factor-beta (TGFbeta) monoclonal antibody in patients with advanced malignant melanoma or renal cell carcinoma**. *Plos One* 2014, **9**(3):e90353.

18. Goudie DR, D'Alessandro M, Merriman B, Lee H, Szeverenyi I, Avery S, O'Connor BD, Nelson SF, Coats SE, Stewart A *et al*: **Multiple self-healing squamous epithelioma is caused by a disease-specific spectrum of mutations in TGFBR1**. *Nat Genet* 2011, **43**(4):365-369.

19. Akhurst RJ, Hata A: **Targeting the TGFbeta signalling pathway in disease**. *Nat Rev Drug Discov* 2012, **11**(10):790-811.

20. Khan SA, Joyce J, Tsuda T: **Quantification of active and total transforming growth factor-beta levels in serum and solid organ tissues by bioassay**. *BMC Res Notes* 2012, **5**:636.

21. Farrell JJ, Bae K, Wong J, Guha C, Dicker AP, Elsaleh H: **Cytidine deaminase single-nucleotide polymorphism is predictive of toxicity from gemcitabine in patients with pancreatic cancer: RTOG 9704**. *Pharmacogenomics J* 2012, **12**(5):395-403.

22. Okazaki T, Javle M, Tanaka M, Abbruzzese JL, Li DH: **Single Nucleotide Polymorphisms of Gemcitabine Metabolic Genes and Pancreatic Cancer Survival and Drug Toxicity**. *Clin Cancer Res* 2010, **16**(1):320-329.

23. Joerger M, Burgers SA, Baas P, Smit EF, Haitjema TJ, Bard MPL, Doodeman VD, Smits PHM, Vincent A, Huitema ADR *et al*: **Germline polymorphisms in patients with advanced nonsmall cell lung cancer receiving first-line platinum-gemcitabine chemotherapy A Prospective Clinical Study**. *Cancer* 2012, **118**(9):2466-2475.

24. Carpi FM, Vincenzetti S, Ubaldi J, Pucciarelli S, Polzonetti V, Micozzi D, Micinini F, Napolioni V: **CDA gene polymorphisms and enzyme activity: genotype-phenotype relationship in an Italian-Caucasian population**. *Pharmacogenomics* 2013, **14**(7):769-781.

25. Sugiyama E, Kaniwa N, Kim SR, Kikura-Hanajiri R, Hasegawa R, Maekawa K, Saito Y, Ozawa S, Sawada J, Kamatani N *et al*: **Pharmacokinetics of gemcitabine in Japanese cancer patients: The impact of a cytidine deaminase polymorphism**. *Journal of Clinical Oncology* 2007, **25**(1):32-42.

26. Serdjebi C, Gagniere J, Desrame J, Fein F, Guimbaud R, Francois E, Andre T, Seitz JF, Monterymard C, Arsene D *et al*: **FFCD-1004 Clinical Trial: Impact of Cytidine**

Deaminase Activity on Clinical Outcome in Gemcitabine-Monotherapy Treated Patients. *Plos One* 2015, **10**(8).

27. Li H, Wang XL, Wang XW: **The impact of CDA A79C gene polymorphisms on the response and hematologic toxicity in gemcitabine-treated patients: A meta-analysis**. *Int J Biol Marker* 2014, **29**(3):E224-E232.

28. Ding XX, Chen WW, Fan HJ, Zhu B: **Cytidine deaminase polymorphism predicts toxicity of gemcitabine-based chemotherapy**. *Gene* 2015, **559**(1):31-37.

29. Rothenberg ML, Moore MJ, Cripps MC, Andersen JS, Portenoy RK, Burris HA, 3rd, Green MR, Tarassoff PG, Brown TD, Casper ES *et al*: **A phase II trial of gemcitabine in patients with 5-FU-refractory pancreas cancer**. *Ann Oncol* 1996, **7**(4):347-353.

30. Herbertz S, Sawyer JS, Stauber AJ, Gueorguieva I, Driscoll KE, Estrem ST, Cleverly AL, Desaiah D, Guba SC, Benhadji KA *et al*: **Clinical development of galunisertib (LY2157299 monohydrate), a small molecule inhibitor of transforming growth factor-beta signaling pathway**. *Drug Des Devel Ther* 2015, **9**:4479-4499.

31. Zitvogel L, Kepp O, Kroemer G: **Immune parameters affecting the efficacy of chemotherapeutic regimens**. *Nat Rev Clin Oncol* 2011, **8**(3):151-160.

32. Zitvogel L, Galluzzi L, Smyth MJ, Kroemer G: **Mechanism of action of conventional and targeted anticancer therapies: reinstating immunosurveillance**. *Immunity* 2013, **39**(1):74-88.

33. Weiss GJ, Waypa J, Blaydorn L, Coats J, McGahey K, Sangal A, Niu J, Lynch CA, Farley JH, Khemka V: **A phase Ib study of pembrolizumab plus chemotherapy in patients with advanced cancer (PembroPlus)**. *Br J Cancer* 2017, **117**(1):33-40.

34. Langer CJ, Gadgeel SM, Borghaei H, Papadimitrakopoulou VA, Patnaik A, Powell SF, Gentzler RD, Martins RG, Stevenson JP, Jalal SI *et al*: **Carboplatin and pemetrexed with or without pembrolizumab for advanced, non-squamous non-small-cell lung cancer: a randomised, phase 2 cohort of the open-label KEYNOTE-021 study**. *The lancet oncology* 2016, **17**(11):1497-1508.

35. Webster K, Cella D, Yost K: **The Functional Assessment of Chronic Illness Therapy (FACIT) Measurement System: properties, applications, and interpretation**. *Health Qual Life Outcomes* 2003, **1**:79.

36. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M *et al*: **New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1)**. *Eur J Cancer* 2009, **45**(2):228-247.

37. Wolchok JD, Hoos A, O'Day S, Weber JS, Hamid O, Lebbe C, Maio M, Binder M, Bohnsack O, Nichol G *et al*: **Guidelines for the evaluation of immune therapy activity in solid tumors: immune-related response criteria**. *Clin Cancer Res* 2009, **15**(23):7412-7420.

13 APPENDICES

13.1 Appendix A Performance Status Criteria

ECOG Performance Status Scale		**Karnofsky Performance Scale**	
Grade	Descriptions	Percent	Description
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.	100	Normal, no complaints, no evidence of disease.
		90	Able to carry on normal activity; minor signs or symptoms of disease.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (e.g., light housework, office work).	80	Normal activity with effort; some signs or symptoms of disease.
		70	Cares for self, unable to carry on normal activity or to do active work.
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	60	Requires occasional assistance, but is able to care for most of his/her needs.
		50	Requires considerable assistance and frequent medical care.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	40	Disabled, requires special care and assistance.
		30	Severely disabled, hospitalization indicated. Death not imminent.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	20	Very sick, hospitalization indicated. Death not imminent.
		10	Moribund, fatal processes progressing rapidly.
5	Dead.	0	Dead.

13.2 Appendix B Fact-Hep

FACT-Hep (Version 4)

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

FACT-Hep (Version 4)

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

	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

FACT-Hep (Version 4)

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

	ADDITIONAL CONCERNS	Not at all	A little bit	Some-what	Quite a bit	Very much
C1	I have swelling or cramps 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
C4	I can digest my food well	0	1	2	3	4
C5	I have diarrhea (diarrhoea)	0	1	2	3	4
C6	I have a good appetite	0	1	2	3	4
Hep 1	I am unhappy about a change in my appearance	0	1	2	3	4
CNS 7	I have pain in my back	0	1	2	3	4
Cx6	I am bothered by constipation	0	1	2	3	4
H17	I feel fatigued	0	1	2	3	4
An7	I am able to do my usual activities	0	1	2	3	4
Hep 2	I am bothered by jaundice or yellow color to my skin	0	1	2	3	4
Hep 3	I have had fevers (episodes of high body temperature)	0	1	2	3	4
Hep 4	I have had itching	0	1	2	3	4
Hep 5	I have had a change in the way food tastes	0	1	2	3	4
Hep 6	I have had chills	0	1	2	3	4
HN 2	My mouth is dry	0	1	2	3	4
Hep 8	I have discomfort or pain in my stomach area	0	1	2	3	4

13.3 Appendix C: Modified Immune-related Response Criteria (irRECIST)

This new classification is based on the recent learning from clinical studies with cancer immunotherapies that even if some new lesions appear at the beginning of a treatment or if the total tumor burden does not increase substantially, tumor regressions or stabilizations might still occur later. The irRC were created using bi-dimensional measurements (as previously widely used in the World Health Organization criteria). For this trial, the concepts of the irRC are combined with RECIST 1.1 to come up with the modified irRC.

For modified irRC, only target and measurable lesions are taken into account. In contrast to the RECIST 1.1 criteria, the modified irRC criteria (a) require confirmation of both progression and response by imaging at 6 weeks after initial imaging and (b) do not necessarily score the appearance of new lesions as progressive disease if the sum of lesion diameters of target lesions (minimum of 10 mm per lesion, maximum of 5 target lesions, maximum of 2 per organ) and measurable new lesions does not increase by ≥ 20%.

The same method of assessment and the same technique should be used to characterize each identified and reported target lesion(s) at baseline, during the trial, and at the end of trial visit. All measurements should be recorded in metric notation. The modified irRC based on RECIST 1.1 are displayed below.

Modified immune-related response criteria are defined as follows:

New measurable lesions: Incorporated into tumor burden.

New non-measurable lesions: Do not define progression but precludes (irCR).

Overall irCR:	Complete disappearance of all lesions (whether measurable or not) and no new lesions. All measurable lymph nodes also must have a reduction in short axis to 10 mm.
Overall irPR:	Sum of the longest diameters of target and new measurable lesions decreases ≥ 30%.
Overall irSD:	Sum of the longest diameters of target and new measurable lesions neither irCR, irPR, (compared to baseline) or irPD (compared to nadir).
Overall irPD:	Sum of the longest diameters of target and new measurable lesions increases ≥ 20% (compared to nadir), confirmed by a repeat, consecutive observations at least 4 weeks (normally it should be done at 6 weeks) from the date first documented.

Overall Responses Derived from Changes in Index, Non-Index, and New Lesions

Measurable Response	Non-Measurable Response		Overall Response Using Modified irRC
Index and New, Measurable Lesions (Tumor Burden)[1]	**Non-Index Lesions**	**New, Non-Measurable Lesions**	
Decrease 100%	Absent	Absent	irCR[2]
Decrease 100%	Stable	Any	irPR[2]
Decrease 100%	Unequivocal progression	Any	irPR[2]
Decrease ≥ 30%	Absent / Stable	Any	irPR[2]
Decrease ≥ 30%	Unequivocal progression	Any	irPR[2]
Decrease < 30% to increase < 20%	Absent / Stable	Any	irSD
Decrease < 30% to increase < 20%	Unequivocal progression	Any	irSD
Increase ≥ 20%	Any	Any	irPD

[1] Decreases assessed relative to baseline
[2] Assuming that the response (irCR and irPR) and progression (irPD) are confirmed by a second, consecutive assessment at least 4 weeks apart (normally it should be done 6 weeks apart).