

# The SHARON Trial: Melphalan, BCNU, B12b, Asc, and Stem Cells for Pancreatic and Breast Cancer and an Inherited BRCA/PALB2 Mutation

Kenneth H. Yu, M.D.<sup>1</sup>; Parastoo B. Dahi, M.D.<sup>1</sup>; Colin D. Weekes, M.D., Ph.D.<sup>2</sup>; Yi-Bin Chen, M.D.<sup>2</sup>; Lauren Levy<sup>1</sup>; Katherine Nagel<sup>1</sup>; Mary Larsen<sup>1</sup>; Angela Chan<sup>1</sup>; Erin DiGugliemo<sup>1</sup>; Carly Schwartz<sup>1</sup>; Amin Yaqubie<sup>1</sup>; Jeffrey A. Glazier<sup>3</sup>; Arnold Glazier, M.D.<sup>3</sup>; Sergio A. Giral, M.D.<sup>1</sup>; Eileen M. O'Reilly, M.D.<sup>1</sup>



Memorial Sloan Kettering  
Cancer Center



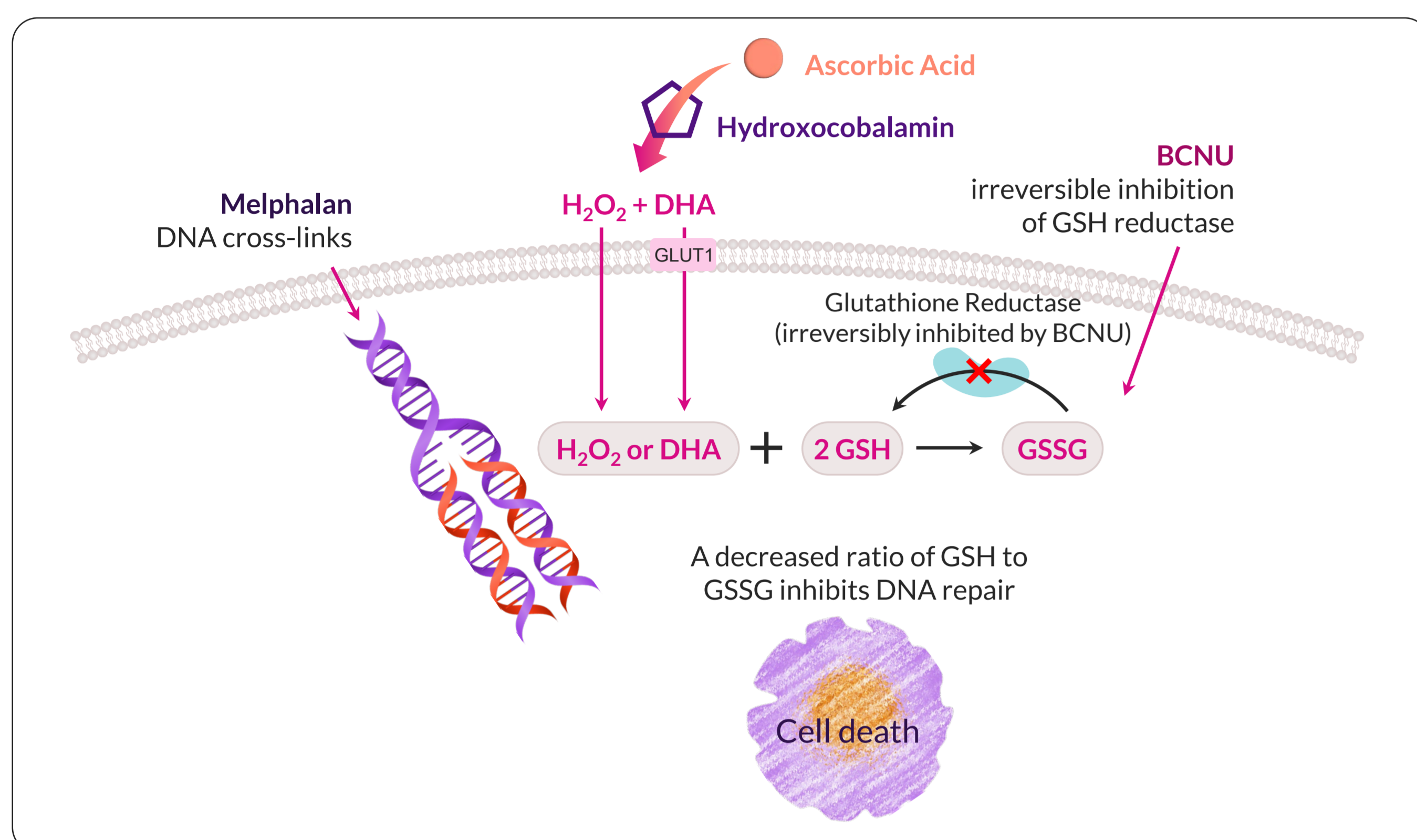
Massachusetts General Hospital  
Founding Member, Mass General Brigham

Contact: Kenneth Yu, M.D. (yuk1@mskcc.org)

<sup>1</sup> Memorial Sloan Kettering Cancer Center, New York, NY. <sup>2</sup> Massachusetts General Hospital, Boston, MA. <sup>3</sup> General Oncology, Inc.

## 1 Background

Pancreatic cancer arising in the setting of deleterious germline BRCA 1/2 and PALB2 mutations is usually accompanied by functional BRCA deficiency and hypersensitivity to DNA cross-linking agents; however, secondary genetic alterations can restore BRCA function and cause drug resistance. Oxidizing cellular GSH can sensitize cells to melphalan and inhibit DNA repair.<sup>1,2,3</sup> In the SHARON trial, BCNU, hydroxocobalamin (B12b), and ascorbic acid (Asc) are used to oxidize GSH. B12b catalyzes the oxidation of Asc to dehydroascorbic acid (DHA) and generates H<sub>2</sub>O<sub>2</sub>.<sup>4</sup> DHA is preferentially taken up by GLUT-1, which is over expressed in KRAS-mutated pancreatic cancer.<sup>5</sup> The proposed mechanism is shown below:



Low dose I.V. ethanol was used to protect red blood cell catalase for 23 hours after drug therapy.<sup>6</sup>

## 2 Study Design

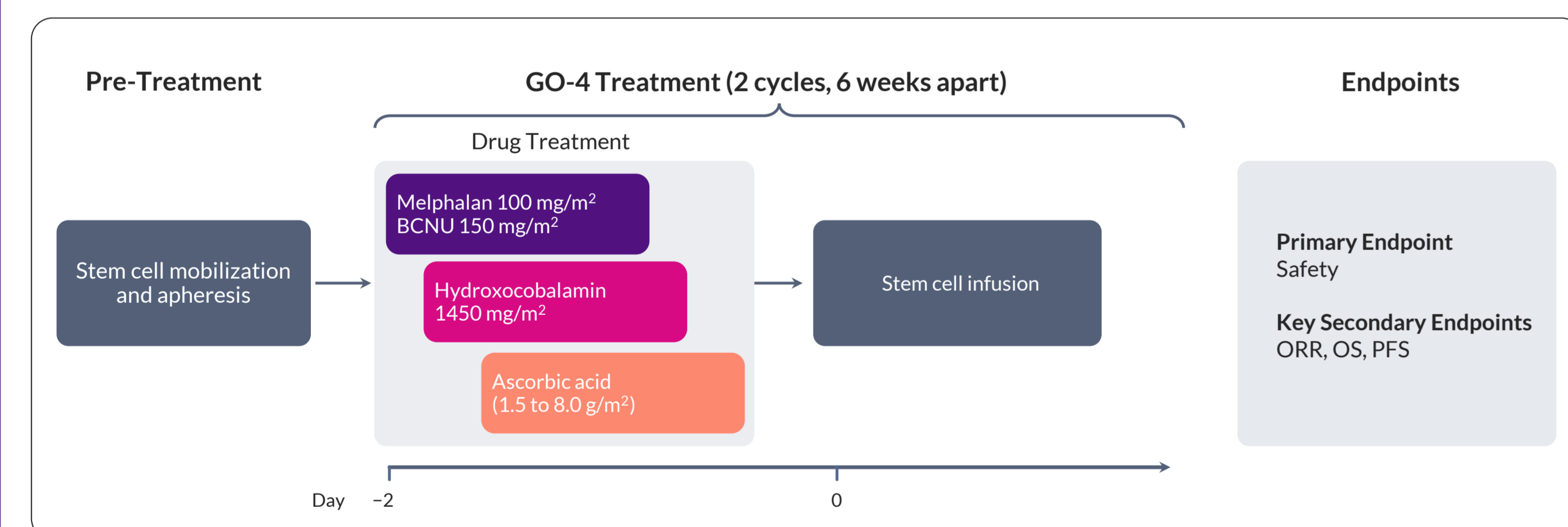
### Key Inclusion Criteria

- Stage 4 pancreatic ductal adenocarcinoma or breast adenocarcinoma and a deleterious germline BRCA1/2 or PALB2 mutation
- Expected survival time  $\geq$  6 months, as determined by the investigator
- Multiple lines of prior therapy were allowed

### Key Exclusion Criteria

- Conditions that would preclude BCNU, melphalan, and stem cell treatment
- G6PD deficiency, catalase deficiency, elevated plasma oxalate
- Metastatic disease to the CNS

### Investigational Treatment



An I.V. infusion of ethanol 0.5 g/h for 23 h was administered after completion of the BCNU infusion. Dose escalation for Asc from 3 to 6 to 8 g/m<sup>2</sup> with a 2+4 dose escalation pattern was planned but modified due to infusion reactions.

The following modifications were made to the protocol. Hydralazine was given prior to the hydroxocobalamin infusion and as needed for hypertension. Dexamethasone was started 24 hours prior to chemotherapy and continued for 2 days for prevention of cytokine release syndrome. Patients were given oral magnesium supplements beginning ~ 4 days prior to chemotherapy. Dantrolene 100 mg was given 90 minutes prior to chemotherapy to lessen muscle hyperactivity. The Asc dose was de-escalated.

### Primary Endpoint:

- Safety of the treatment protocol, evaluated according to CTCAE Version 5.0

### Secondary Endpoints:

- Objective response rate (ORR), progression-free survival (PFS), overall survival (OS)

Each of the efficacy endpoints were assessed 30 days ( $\pm$  5 days) after the first investigational treatment and 1, 3, 6, 9, and 12 months after the final investigational treatment. Longer term follow-up data was obtained on patients when available.

## 3 Results

### Patient Demographics

Demographics	N = 12
Sex	
Female	6 (50%)
Male	6 (50%)
Age	
Mean (SD)	62.0 (10.9)
Min, Max	40, 75
# Prior Lines of Treatment	
Mean (SD)	3.2 (1.9)
Median	3
Min, Max	1, 7
Cancer Type	
Pancreatic ductal adenocarcinoma	11 (91.7%)
Breast adenocarcinoma	1 (8.3%)
Germline Mutation	
BRCA1	4 (33.3%)
BRCA2	7 (58.3%)
PALB2	1 (8.3%)
Sites of Lesions	
Pancreas	5 (41.7%)
Liver	5 (41.7%)
Lung	3 (25.0%)
Lymph Node	2 (16.7%)
Peritoneum	2 (16.7%)
Kidney	1 (8.3%)
Bone	1 (8.3%)
Stomach	1 (8.3%)

### Safety and Tolerability\*

The investigational treatment was generally well tolerated with manageable and mostly expected side effects.

Treatment-Related Adverse Events Occurring in More than 10% of Patients (N=12)	Any Grade	Grade 3	Grade 4
Neutropenia and Thrombocytopenia	12 (100%)	(0%)	12 (100%)
Infusion-Related Reaction	12 (100%)	(0%)	(0%)
Anemia	6 (50%)	1 (8%)	(0%)
Fatigue	5 (42%)	(0%)	(0%)
Mucositis	5 (42%)	1 (8%)	(0%)
Diarrhea	3 (25%)	(0%)	(0%)
Engraftment Syndrome	3 (25%)	2 (17%)	(0%)
Hypertension	3 (25%)	2 (17%)	(0%)
Nausea	3 (25%)	(0%)	(0%)
Alopecia	2 (17%)	(0%)	(0%)
Anorexia	2 (17%)	(0%)	(0%)
Blood Bilirubin Increased	2 (17%)	(0%)	(0%)
Constipation	2 (17%)	(0%)	(0%)
Dysgeusia	2 (17%)	(0%)	(0%)
Febrile Neutropenia	2 (17%)	2 (17%)	(0%)
Fever	2 (17%)	(0%)	(0%)
Headache	2 (17%)	1 (8%)	(0%)
Hypotension	2 (17%)	(0%)	(0%)
Maculopapular rash	2 (17%)	(0%)	(0%)
Non-Cardiac Chest Pain	2 (17%)	(0%)	(0%)
Sinus Bradycardia	2 (17%)	(0%)	(0%)

There have been no reported long-term adverse events; Maximum tolerated dose not yet reached

\* Adverse reactions that are components of other more clinically relevant adverse reactions are not included above. For example, the % subjects with fever does not include subjects with febrile neutropenia.

No treatment-related mortality was observed.

One subject who received 3.5 g/m<sup>2</sup> of Asc developed grade 3 cytokine release syndrome ~13 hours after the start of the chemotherapy of cycle 2. The subject was treated with supportive care and dexamethasone and the problem resolved within 48 hours.

One subject with a history of a gastric ulcer presented on day 22 of cycle 2 with a complex fluid/gas abdominal collection extending from the margin of the pancreatojejunostomy suspicious for an anastomotic leak. The problem resolved with drain placement, antibiotics, and antifungal therapy.

### Infusion-Related Reactions

Infusion-related reactions variably characterized by increased blood pressure, muscle hyperactivity (e.g., fasciculations, migratory spasms, jaw pain, chest tightness), abdominal pain, burning sensations, and pain were seen in all subjects during or shortly after the Asc infusion. The infusion reactions resolved within ~ 1 hour without any lasting sequelae. The infusion reactions were generally managed with I.V. hydralazine, famotidine, narcotics, and lorazepam. Two subjects given oral magnesium supplementation and 100 mg dantrolene prior the chemotherapy did not display muscle hyperactivity; additional data is needed to evaluate the effectiveness of magnesium and dantrolene in this setting.

### Cytokine Release Syndrome (CRS)

After Asc dose reduction and the introduction of dexamethasone prior to chemotherapy and for 48 hours thereafter, CRS was not observed and plasma cytokine screens were not elevated in the 48 hours post-chemotherapy. Additional data at higher Asc doses is needed and planned.

### Maximum Tolerated Dose (MTD) of Ascorbic Acid

The maximum tolerated dose of Asc is  $\geq$  1.5 g/m<sup>2</sup>. The protocol-specified limit of two grade 3 dose-limiting toxicities attributable to Asc has not yet occurred.

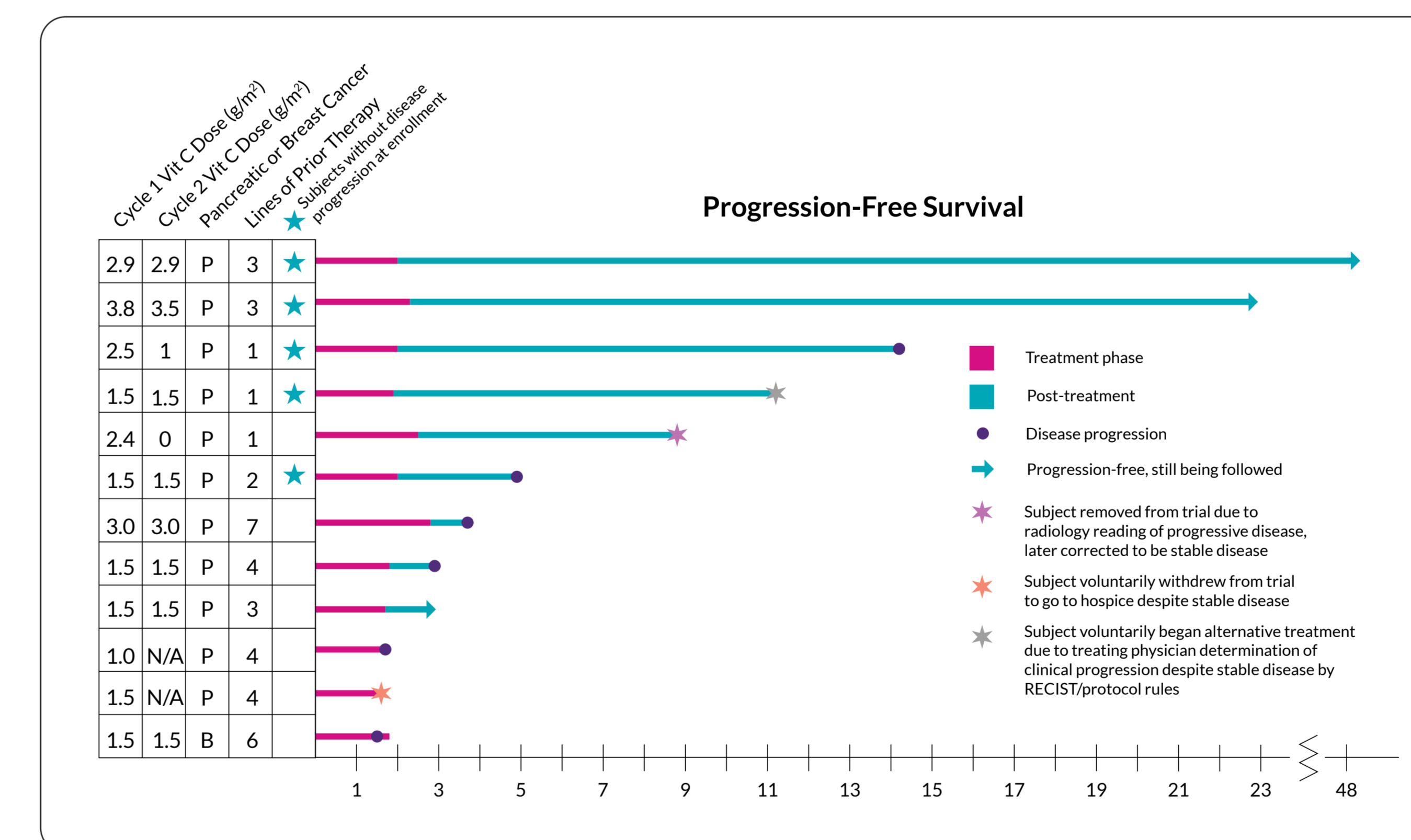
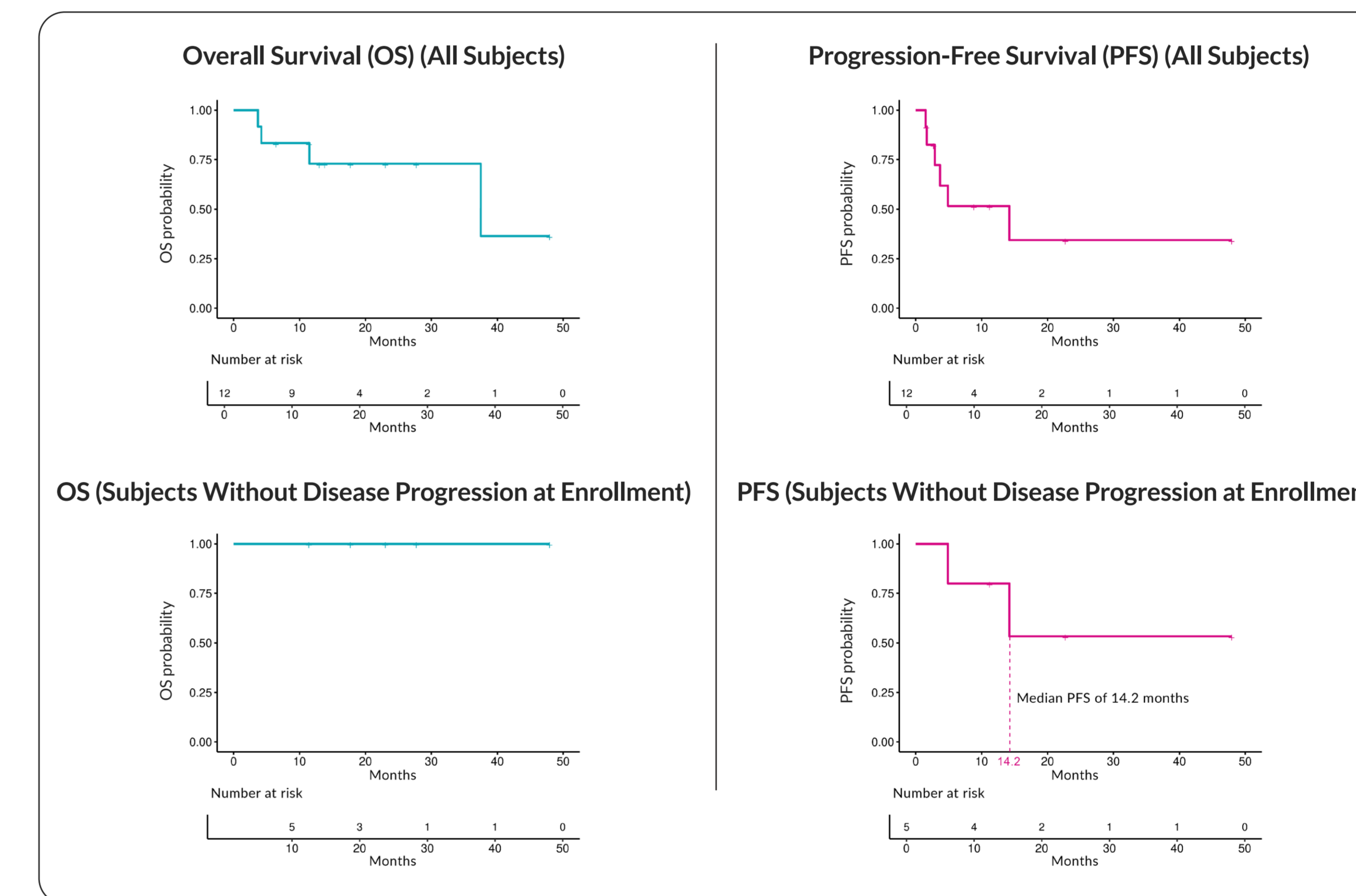
### Time to Neutrophil and Platelet Engraftment for both Cycles\*

- An absolute neutrophil count  $\geq$  500/ $\mu$ L for 3 days, with the date of engraftment being the first of those 3 days (mean 10 days, min 9 days, max 13 days)
- A platelet count  $\geq$  20,000/ $\mu$ L for 3 days, with the date of engraftment being the first of those 3 days (mean 14 days, min 10 days, max 21 days)

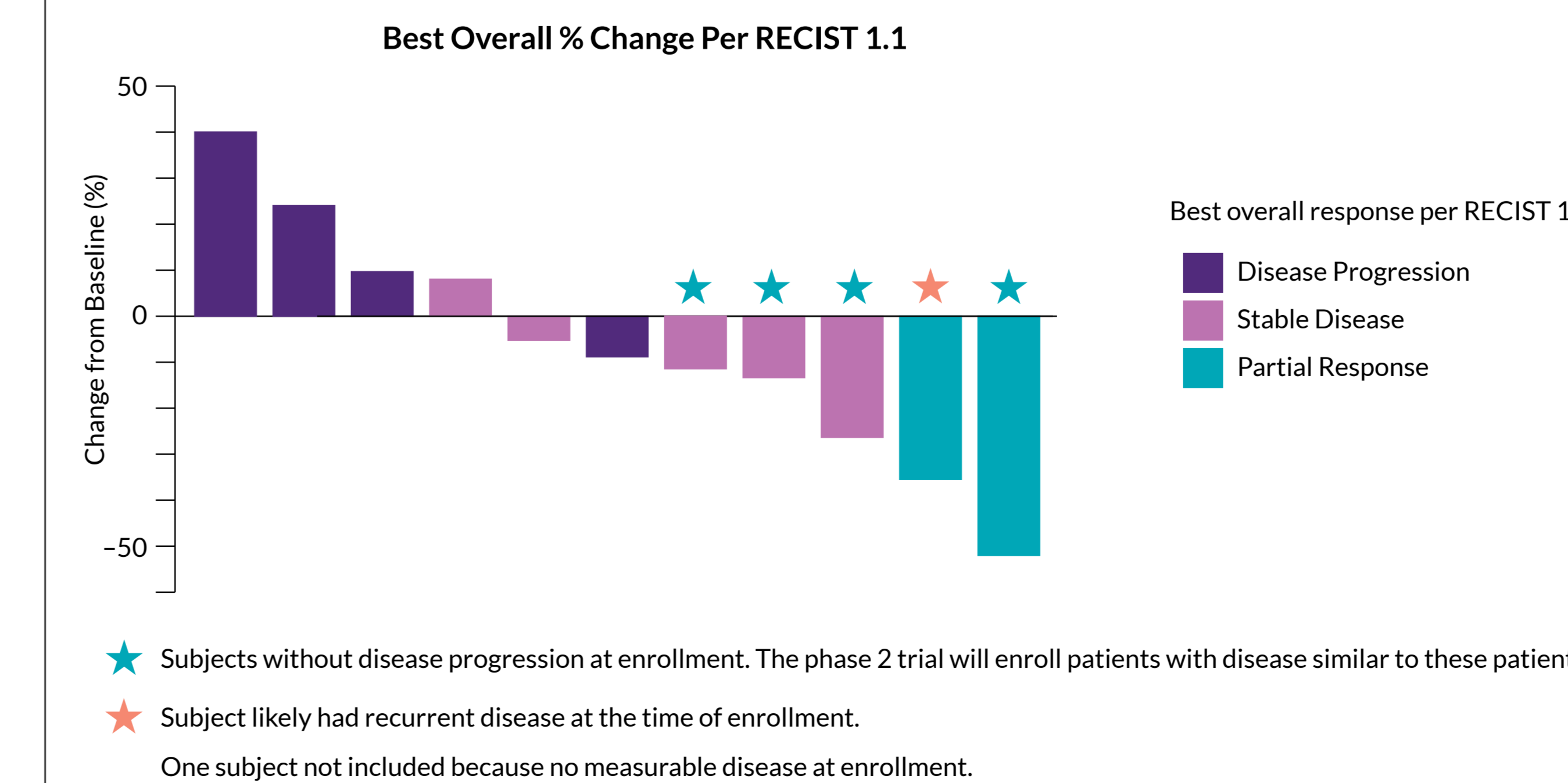
\* One subject had delayed platelet engraftment after cycle 1 and was removed from the trial due to disease progression. Another subject withdrew after cycle 1 to go to hospice despite stable disease.

### Efficacy

The investigational treatment displayed a strong signal of efficacy.



### Preliminary Analysis Best Overall Response



## 4 Discussion

The investigational treatment was generally well-tolerated with manageable and expected adverse effects typically seen with melphalan/BCNU and autologous stem cell infusions. A strong signal of efficacy was seen in subjects without disease progression at enrollment. For pts with PDAC with stable/responding disease at entry, median PFS was 14.2 months and N = 2 (18%) are free of disease at 23 and 48 months and off all therapy.

Infusion reactions characterized by increased BP and signs and symptoms of muscle hyperactivity (e.g., fasciculations, migratory muscle spasms, tremors, jaw pain, rigors) have been observed in subjects during or shortly after administration of Asc. The infusion reactions resolved without sequela with ~ 1 hour.

The hypertensive response may be due to interference with physiological nitric oxide-mediated vasodilation by inhibition of endothelial NOS by B12b and scavenging of nitric oxide by B12r, the one-electron reduction product of B12b.<sup>7,8</sup> The reduction of B12b by Asc generates B12r. DHA could also play a role in the hypertensive response. I.V. DHA causes a hypertensive response in rats.<sup>9</sup> In this trial, the administration of I.V. hydralazine prior to B12b and as needed after Asc provided good BP control.

The muscle hyperactivity could be due to perturbations in calcium homeostasis due to activation of the skeletal isoform of ryanodine receptors (RyR1). RyR1 is an ion channel, which upon activation releases Ca<sup>2+</sup> from the sarcoplasmic reticulum into the cytoplasm and thereby triggers actin-myosin cross-bridge formation and muscle contraction. RyR1 receptors are highly susceptible to redox modifications. Oxidation of thiol groups in RyR1 with agents such as hydrogen peroxide and GSSG activates RyR1 and triggers Ca<sup>2+</sup> release.<sup>10,11,12</sup> Mg<sup>2+</sup> and dantrolene inhibit RyR1 activation and decrease release Ca<sup>2+</sup> from the sarcoplasmic reticulum.<sup>13,14,15</sup> Two subjects given oral magnesium and dantrolene prior the chemotherapy did not display muscular hyperactivity; additional data is needed to evaluate the effectiveness of this intervention.

Since the addition of prophylactic dexamethasone, CRS and elevated plasma cytokines have not been observed. The MTD of Asc has not yet been determined. Asc dose escalation is planned using prophylactic dexamethasone and the discussed measures to ameliorate infusion reactions.

## 5 Conclusions

The regimen was safe and feasible, and a promising signal of efficacy was observed in PDAC patients with deleterious BRCA1/2 germline mutations and responding disease at enrollment. Further study is warranted.

The trial enrolled at Memorial Sloan Kettering Cancer Center and Massachusetts General Hospital. ClinicalTrials.gov Identifier: NCT04150042

### Disclosures

Kenneth H. Yu, M.D has the following conflicts: General Oncology, BioNTech, OncoC4, Ipsen, Episteme Genomics, AstraZeneca, SCG Cell Therapy

### Study Sponsor

The study was sponsored by General Oncology, Inc.

### Corresponding email addresses

Kenneth H. Yu, M.D., Yuk1@mskcc.org

### Acknowledgements

We are deeply grateful to the patients and their caregivers who made this study possible. Arny Glazier would like to thank Sharon; her courage and grace paved the way for this trial.

### References

- Jevtovic-Todorovic V, et al. J Cancer Res Clin Oncol. 1991;117(4):313-20.
- Solovieva ME, et al. Eur J Pharmacol. 2007 Jul 2;566(1-3):206-14.
- Akatov VS, et al. Biosci Rep. 2000 Oct;20(5):411-7.
- Nazhat NB, et al. J Inorg Biochem. 1989 Jun;36(2):75-81.
- Spiehlholz C, et al. Cancer Res. 1997 Jun 15;57(12):2529-37.
- Kirkman HN, et al. J Biol Chem. 1999 May 14;274(20):13908-14.
- Weinberg JB, et al. Free Radic Biol Med. 2009 Jun 15;46(12):1626-32.
- Wolak M, et al. J Am Chem Soc. 2001 Oct 10;123(40):9780-91.
- Patterson JW, et al. Am J Physiol. 1951 Oct;167(1):119-26.
- Oba T, et al. J Appl Physiol (1985). 2002 Dec;93(6):1999-2008.
- Jabe AC, et al. J Biol Chem. 1997 Mar 14;272(11):7069-77.
- Hidalgo C, et al. Biol Res. 2002;35(2):183-93.
- Zhao F, et al. J Biol Chem. 2001 Apr 27;276(17):13810-6.
- Oo YW, et al. Mol Pharmacol. 2015 Jul;88(1):57-63.
- Fruen BR, et al. J Biol Chem. 1997 Oct 24;272(43):26965-71.