## A phase II study of post-remission therapy with pembrolizumab in older patients with acute myeloid leukemia

## Authors

Kevin Quann,<sup>1,2</sup> Konstantinos Lontos,<sup>2</sup> Alison Sehgal,<sup>2</sup> Anastasios Raptis,<sup>2</sup> Annie Im,<sup>1,2</sup> Robert L. Redner,<sup>1,2</sup> Kathleen A. Dorritie,<sup>1,2</sup> Mounzer Agha,<sup>2</sup> Jing-Zhou Hou,<sup>2</sup> Rafic Farah,<sup>2</sup> James Rossetti,<sup>2</sup> Daniel P. Normolle,<sup>3</sup> Theresa L. Whiteside,<sup>2,4</sup> Yen-Michael Sheng Hsu<sup>5</sup> and Michael Boyiadzis<sup>1</sup>

<sup>1</sup>Department of Medicine, Division of Hematology/Oncology, University of Pittsburgh School of Medicine, <sup>2</sup>UPMC Hillman Cancer Center, <sup>3</sup>Department of Biostatistics, University of Pittsburgh School of Public Health, <sup>4</sup>Department of Pathology, University of Pittsburgh School of Medicine and <sup>5</sup>Immunologic Monitoring and Cellular Products Laboratory, UPMC Hillman Cancer Center, Pittsburgh, PA, USA Correspondence: K. QUANN - quannka@pitt.edu

https://doi.org/10.3324/haematol.2024.285313

Received: February 18, 2024. Accepted: July 31, 2024. Early view: August 8, 2024. ©2024 Ferrata Storti Foundation Published under a CC BY-NC license





**Figure S1:** Kaplan Meier curves demonstrating relapse-free survival (**A**) and overall survival (**B**) according to subgroup by performance status, age, ELN risk stratification, and receipt of consolidation therapy. P-values between groups were computed by log-rank test.

## Supplemental Table 1: Adverse events

	Grade	Grade	Grade	Grade		Grade
	I _		III -	IV	Total	III-IV _
Platelet count decreased	5	0	2	3	10	5
Neutrophil count decreased	0	4	1	3	8	4
Anemia	3	2	1	0	6	1
Lymphocyte count decreased	0	2	3	0	5	3
Hyponatremia	4	0	1	0	5	1
Fatigue	1	4	0	0	5	0
Fever	2	2	0	0	4	0
Diarrhea	2	0	1	0	3	1
Hypocalcemia	1	1	0	0	2	0
Hypokalemia	1	1	0	0	2	0
Hypomagnesemia	2	0	0	0	2	0
Hypophosphatemia	2	0	0	0	2	0
Hypothyroidism	1	1	0	0	2	0
Creatinine increased	2	0	0	0	2	0
Pruritus	1	1	0	0	2	0
Aspartate aminotransferase increased	0	0	1	0	1	1
Alkaline phosphatase increased	1	0	0	0	1	0
Hyperglycemia	1	0	0	0	1	0
Hyperkalemia	1	0	0	0	1	0
Hypoalbuminemia	1	0	0	0	1	0
Hypoglycemia	1	0	0	0	1	0
Anorexia	1	0	0	0	1	0
Blurred vision	1	0	0	0	1	0
Chills	1	0	0	0	1	0
Cough	1	0	0	0	1	0
Dizziness	1	0	0	0	1	0
Edema limbs	1	0	0	0	1	0
Headache	0	1	0	0	1	0
Hematuria	0	0	1	0	1	1
Hypertension	0	0	1	0	1	1
Hypotension	0	1	0	0	1	0
Lung infection	0	1	0	0	1	0
Pneumonitis	0	1	0	0	1	0
Sore throat	1	0	0	0	1	0
Urinary tract infection	0	1	0	0	1	0

## Figure S2



**Figure S2:** PBMCs were immediately isolated from peripheral blood using Ficoll-Hypaque (Sigma Aldrich, St. Louis, MO) gradient centrifugation and cryopreserved. At time of flow cytometric analysis, PBMCs were thawed and resuspended in RPMI-1640 with 10% fetal bovine serum, 1% non-essential amino acids, 1 mM sodium pyruvate, 1% Glutamax and 10 mM HEPES (all from Thermo-Fisher, Waltham, MA) for culture and staining. Cells were stained for surface CD3 (clone: OKT3), CD4 (clone OKT4), CD8 (clone HIT8a), PD1 (clone EH12.2H7) and TIM-3 (clone F38-2E2, all from Biolegend, San Diego, CA). Intracellular staining for TCF-1 (clone C63D9, Cell Signaling, Danvers, MA) was performed using a FOXP3/transcription factor staining kit (Thermo-Fisher). A live/dead fixable dye (Biolegend) was added to exclude dead cells. Samples were acquired on an BD LSRFortessa flow cytometer (BD Biosciences, San Jose, CA) and analyzed using FlowJo software. The analysis gates were restricted to the lymphocytes based on forward and side scatter, followed by live T cells excluding viability dye that were CD3<sup>+</sup>, then CD4<sup>+</sup> and CD8<sup>+</sup> cells, on which PD-1<sup>+</sup>, TIM-3<sup>+</sup>, and TCF-1<sup>+</sup> subpopulations were gated. Shown are frequencies of PD-1<sup>+</sup>, TIM-3<sup>+</sup>, and TCF-1<sup>+</sup> populations among CD4+ (left column) and CD8<sup>+</sup> T cells (right column) at the end of pre-specified maintenance pembrolizumab cycles (1, 3, 7, 16, and 35), or at end of treatment or disease relapse. \*p<0.05, \*\*p<0.01, and \*\*\*p<0.001 by two-sided paired t-test compared to pre-treatment baseline control.