Abstract 2646

Development and testing of an ex vivo, live tumor fragment platform for the prediction of immune checkpoint inhibitor response and relationship to approved biomarkers

David Braun,[1] Nicholas Dana,[2] Hilary R. Hernan,[2] Shalini Sahni,[2] Christina Scribano,[2] Christin Johnson,[2] Lindsey Vedder,[2] Ellen Wargowski,[2] Aishwarya Sunil,[2] Deepa Sharma,[2] Katherine Rexroad,[2] Payton McDonnell,[2] Victor Jergens,[2] Catarina Costa,[2] Richard Zuniga,[3] Giuseppe Toia,[4] Paras Patel,[5] Robert Martin,[6] Umair Majeed,[7] Debabrata Mukhopadhyay,[7] Yanyan Lou,[7] Nima Kokabi,[8] David Hays,[9] Andrew K. Godwin,[10] Victoria Giffi,[11] Alexander Gelbard,[12] Roxana S. Dronca,[7] Ruqin Chen,[7] Heather Chalfin,[13] Barbara Broome,[14] Hani M. Babiker,[7] Tarun Chandra,[15] Chetan Sood,[2] Laura C. F. Hrycyniak,[2] Janis Taube,[16] Sean Caenepeel,[2] Hilario Ramos,[2] Premal Patel,[2] Hinco J. Gierman,[2] Pooja Advani[7]

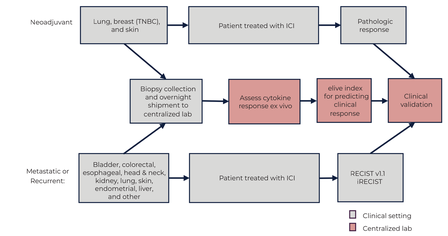
1. Yale School of Medicine, New Haven, CT; 2. Elephas Biosciences, Madison, WI; 3. New York Cancer & Blood Specialists, New York, NY; 4. University of Wisconsin, Madison, WI; 5. JPS Health Network, Fort Worth, TX; 6. University of Louisville School of Medicine, Louisville, KY; 7. Mayo Clinic Jacksonville, Jacksonville, FL; 8. University of North Carolina at Chapel Hill, Chapel Hill, NC; 9. CARTI, Little Rock, AR; 10. University of Kansas Medical Center, Kansas City, KS; 11. Meritus Health, Hagerstown, MD; 12. Vanderbilt University Medical Center, Vanderbilt University, Nashville, TN; 13. Frederick Health, Frederick, MD; 14. Orlando Health, Orlando, FL; 15. EmpiriQA LLC, Long Grove, IL; 16. Johns Hopkins University School of Medicine, Baltimore, MD

Introduction

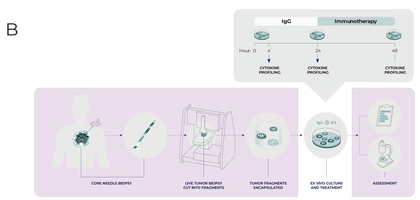
- Immune checkpoint inhibitors (ICIs) have transformed cancer care, but predicting which patients benefit remains a critical unmet need[1]
- Approved biomarkers (PD-L1, dMMR/MSI-H, TMB) capture only static features of the tumor microenvironment (TME) and inadequately predict response[2-4]
- Live tumor fragments (LTFs) preserve TME complexity and enable functional ex vivo ICI assessment; however, prior platforms required large, resected specimens from single academic centers[5]
- The elive platform has been developed to assess ICI response in standard-of-care biopsies shipped overnight from multiple clinical sites across the US[6]
- Here, we evaluated the predictive performance of the elive platform using core needle and forceps biopsies from multiple clinical sites and compared with approved biomarkers

Methods

A



B



Patients with eligible solid tumors (where ICI has an indication, including renal, non-small cell lung, bladder, colorectal, and triple-negative breast cancer, and others) were enrolled in ongoing, prospective observational trials (NCT05478538, NCT05520099, NCT06349642). Biomarker status was determined as indicated for PD-L1 or MMR protein expression, MSI, and/or TMB from patient records.

Biopsies were cut into LTFs, encapsulated in hydrogel, and treated sequentially with IgG antibody followed by ICI (α PD-[L]1 with or without αCTLA-4).

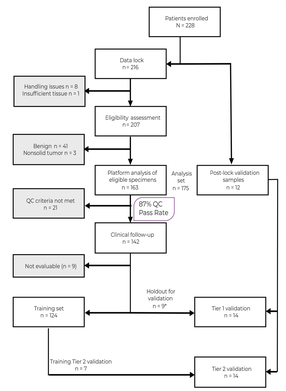
Cytokine production was assessed by a multiplexed bead-based immunoassay at multiple time points during ex vivo treatment.

Using receiver operating characteristic and precision recall analyses, we identified 8 predictive cytokines (including IFN-γ, granzyme B, and CXCL10), which were used to develop the elive index to assess cytokine response to ex vivo ICI treatment.

Results

1 Study Design and Patient Characteristics

A



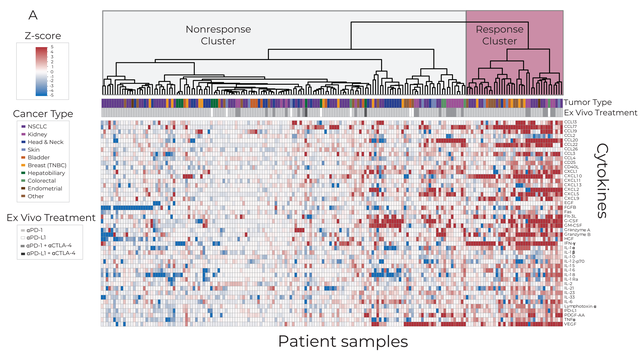
B

Patient Demographics		
Age at diagnosis (Years)		
Range	31-81+	
Median	69	
	N	%
Sex at Birth		
Female	80	46
Male	94	54
Unknown	1	1
Race		
White	151	86
Black	12	7
Asian	1	1
Other/unknown	11	6
Ethnicity		
Hispanic	14	8
Non-Hispanic	155	89
Not reported/ unknown	6	3
US region of treatment		
Midwest	39	22
South	134	77
Northeast	1	1
West	1	1
Tumor type		
NSCLC	70	40
Kidney	22	13
Head & Neck	20	11
Bladder	9	5
Skin	11	7
Breast (TNBC)	8	5
Hepatobiliary	6	3
Colorectal	3	2
Endometrial	3	2
Other/Unknown	23	13
Biopsy site		
Primary	99	57
Metastatic	54	31
Lymph node	22	13
Biopsy number (CNBs; N = 117)		
1	58	50
2+	59	50

A. Of the 163 patient specimens eligible for inclusion in the training set, 87% (n = 142) passed quality control metrics and 124 were used for model development with the remainder withheld for validation. Tier 1 Validation dataset = patients whose in vivo ICI treatment fully matched to ex vivo treatment and patients with a partial treatment match who experienced disease progression. Tier 2 validation = specimens with a predicted ex vivo response to ICI in patients who received ICI in combination with another therapy and had a clinical response. Notably, the development of the elive index did not use clinical response, and the Tier 1 and Tier 2 Validation datasets were combined into a Validation Cohort. **B**. Table showing patient demographics. Percentages may not sum to 100 due to rounding.

2 Development of elive Index

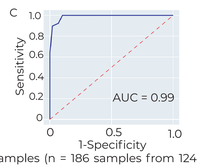
A



B

Analyte	Normalized Threshold	Weight	HM Sensitivity	HM Specificity
IFN-γ	4.7	2	0.74	0.97
CCL22	3.2	2	0.54	0.97
CXCL10	2.8	2	0.74	0.93
Lymphotoxin-α	1.5	2	0.51	0.99
TNFα	0.9	2	0.59	0.93
CCL17	8.4	1	0.46	0.97
Granzyme B	1.2	1	0.77	0.83
IL-10	0.4	1	0.72	0.83

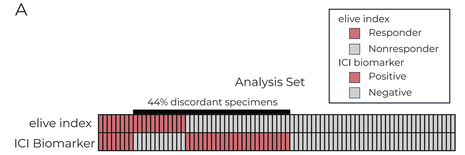
C



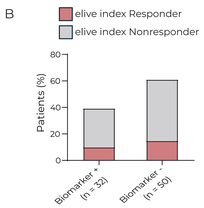
A. Heatmap shows z-scores of cytokine measurements across patient samples (n = 186 samples from 124 patient tumors) following ex vivo ICI treatment, with hierarchical clustering identifying a response cluster comprising 25% of patients (n = 38 samples from 31 patient tumors) and a nonresponse cluster (n = 148 samples from 93 patient tumors). Heatmap color reflects modified z-scores of the delta in cytokine production rates between the ICI and IgG treatment phases. **B**. Table of cytokines included in the elive index listing normalized threshold values and corresponding weights as well as sensitivity and specificity metrics describing each analyte's ability to distinguish between response and nonresponse clusters. **C**. AUROC demonstrating the performance of the elive index in predicting sample classification (response vs nonresponse clusters) within the heatmap.

3 Discordance Between elive Index Response Prediction and Standard-of-Care ICI Biomarkers

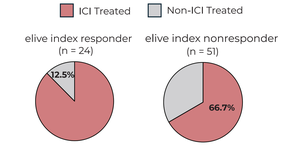
A



B



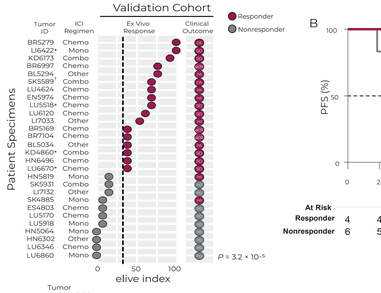
C



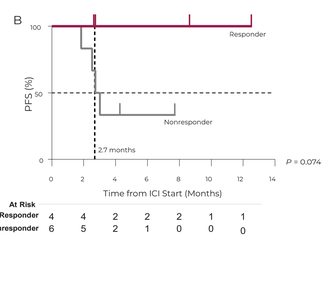
A. Box plot of elive index–predicted ICI response and ICI biomarker status for all patients in the analysis set with both data points. **B.** Stacked bar plot showing the percentage of patients classified as predicted responders or nonresponders by the elive index, stratified by clinical biomarker status (ICI biomarker–positive vs ICI biomarker–negative) for all evaluable data (n= 82). **C.** Distribution of ICI- vs non–ICI-treated patients among elive index–classified responders and nonresponders for all data where treatment is confirmed (n = 75).

4 elive Index Accurately Predicts Clinical Response to ICI Treatment in 93% of Patients

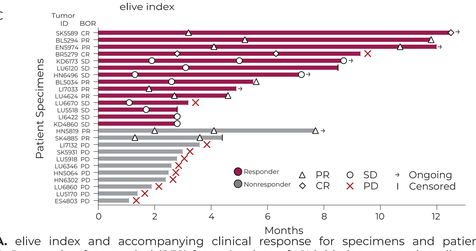
A



B



C



A. elive index and accompanying clinical response for specimens and patients in Validation Cohort. **B**. Progression-free survival (PFS) from the time of ICI initiation, comparing elive predicted responders with nonresponders. Median PFS for nonresponders was 2.7 months; median PFS for responders was not reached. **C**. Swimmer plot showing time to progression and clinical course for each patient, stratified by predicted responders and nonresponders. Symbols denote best overall response. CR = complete response, pCR = complete pathological response, PR = partial response, SD = stable disease, and PD = progressive disease. Tumor types: BL, bladder; BR, breast; EN, endometrial; ES, esophageal; HN, head and neck; KD, kidney; LI, liver; LU, lung; SK, skin. *<6 months SD; †Tier 2 Validation ICI combinations.

Scan to learn more

Copies of this poster obtained through Quick Response (QR) Code are for personal use only and may not be reproduced without permission from ASCO® or the author of this poster.

Conclusions

- The elive platform uses molecular data from a compilation of 8 cytokines collected during ex vivo ICI treatment to predict which patients are most likely to benefit from ICI
- In many patient samples, elive platform predictions revealed discordance with ICI biomarkers approved by the US Food and Drug Administration
- Using routine pretreatment tumor biopsy specimens, the elive platform shows high concordance (>90%) with clinical response to ICI therapy

Future Directions

Prospective clinical trials are underway and will further define the accuracy of the elive platform in specific ICI therapy indications.

Acknowledgments

We thank the patients, clinicians, research staff, and Elephas employees for their contributions to this work.

References

1. Vallejo, J. et al. (2024). *The Oncologist*. 29, 422–430.
2. Hu, X. et al. (2023). *eBioMedicine*. 97, 104816.
3. Yamaguchi, H. et al. (2024). *Cell Rep. Med.* 5, 101621.
4. Yao, J. et al. (2024). *Hum. Vaccines Immunother.* 20, 2406063.
5. Voabil, P. et al. (2021). *Nat. Med.* 27, 1250–1261.
6. Ramasubramanian, T.S., et al. (2026). *J. Transl. Med.* 24, 18.