#7595: Implementation of a Novel Immune Monitoring Strategy for Multicenter Clinical Trials to Enable Equity in Cancer Prognosis for Individuals From Remote Settings of Australia

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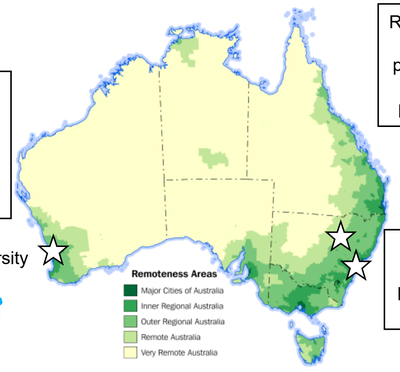
Introduction

Improved cancer prognosis begins with the collection of high-quality data from clinical trials. The implementation of **longitudinal immune phenotyping** studies is complex, requiring **specialised training and equipment**. The challenges of immune monitoring are exacerbated in the **remote and rural communities of Australia**, which are burdened by both an increased prevalence and worse prognosis of cancer[1].

Hub for Western Australia. Experienced in blood processing but not immune phenotyping.

Perth
Edith Cowen University



Regional hub that provides services to 100,000+ people. Limited laboratory facilities, no immune phenotyping experience.

Dubbo
Western Cancer Centre
Western NSW Local Health District

Central lab site. Experienced in blood processing and immune phenotyping.

Sydney
University of Sydney

Recent advances in CyTOF™ immune monitoring technology now enable us to expand clinical immune monitoring to remote settings, **compatible with current hospital facilities,** improving patient care[2].

Study aims

Design and workshop an easily implementable CyTOF immune phenotyping protocol.

Implement the optimised protocol to monitor the immune system of cancer patients treated with ICIs at 3 sites across Australia.

Summary and conclusions

Comprehensive and **robust** immune profiling from a single blood sample achieved with **simplified** CyTOF workflow.

This workflow enables access to underserved communities, facilitating equity in immune phenotyping studies.

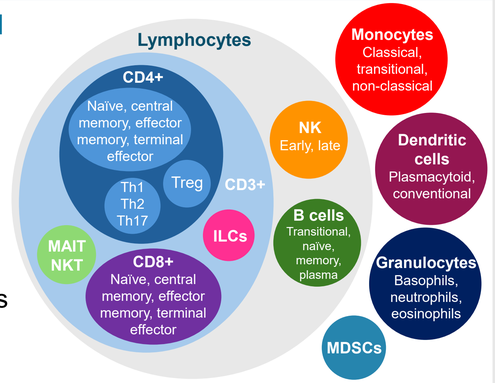
A simplified immune monitoring strategy

50+ markers included

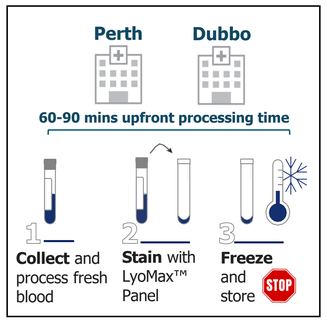
Custom LyoMax CyTOF Panel

View panel

Dried antibody format
Single-tube assay



Identify 30 cell populations
Further characterise with 25+ functional markers



Immune profiling in a rural hospital environment:

Laboratory facilities at Dubbo Hospital contain only a fridge, centrifuge and freezer.

Cutting-edge data made possible without cutting-edge equipment.

"The team at Dubbo Base are so grateful and excited about the research. [We] really feel valued and listened to." – Emma, Dubbo

Implementation across Australia

Adaption of approach for whole blood staining:

Preparation of samples can be simplified to be compatible with resource-limited environments.

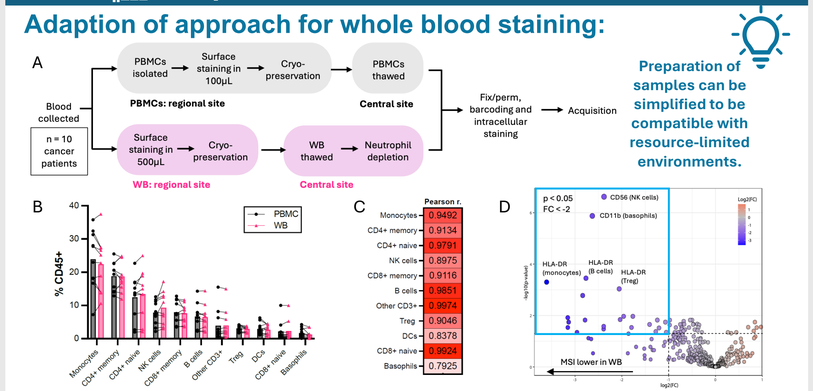


Figure 1: Population frequencies are highly correlated between PBMC and WB sample preparations, despite reduced resolution for some myeloid cell surface antigens. (A) Whole blood (WB) was collected from 10 cancer patients. 500µL of WB was stained for surface antigens with a custom LyoMax CyTOF Panel and cryopreserved. PBMCs were isolated from the remaining WB, stained for surface antigens and then cryopreserved. After thawing, neutrophils were depleted from WB then paired samples were barcoded, pooled, stained for intracellular targeted and acquired. **(B)** Multiple paired t-tests of frequency of each cell population in PBMCs (black) and neutrophil-depleted WB (pink). **(C)** Heatmap of Pearson's R values for the correlation between the abundance of each population in matched PBMCs and neutrophil-depleted WB. **(D)** Median signal intensity (MSI) of each antigen in relevant cell populations calculated. Volcano plot of fold-change vs. significance for differential MSI values between matched PBMC and WB samples. Top 5 MSI values lower in WB based on –value are annotated.

Implementation at 3 sites across Australia:

First application of high-parameter cytometry to study a regional population in Australia.

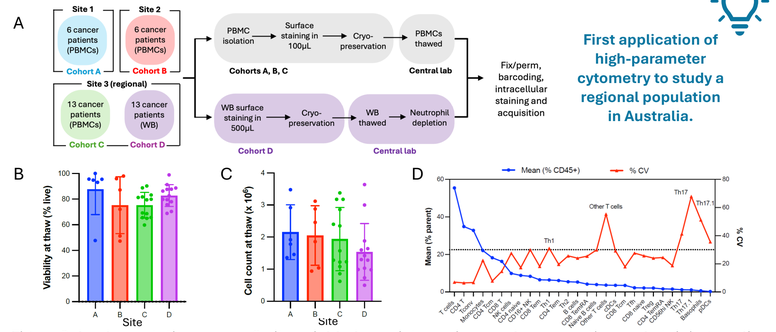


Figure 2: Implementation across 3 sites yielded consistency in sample preparation and staining quality control parameters. (A) Blood was collected from 38 cancer patients at 4 sites across Australia. PBMCs were processed, stained for surface antigens and then cryopreserved in cohort A (Sydney, n = 6), cohort B (Perth, n = 6) and cohort C (Dubbo, n = 13). WB was stained for surface antigens in cohort D (Dubbo, n = 13). Samples were shipped to the central site where staining was completed and samples were acquired. **(B-C)** Bar charts showing the **(B)** viability and **(C)** cell count at sample thaw. **(D)** The mean and CV of each cell population expressed as a percentage of live lymphocytes; data pooled for the 38 donors across the 4 cohorts. Dotted line at CV = 30%. Populations with CV greater than 30% annotated.

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Acknowledgements: Study made possible through a generous research collaboration with **Standard BioTools Inc.** and patient contributions. Funding support received from National Health and Medical Research Council, Cancer Council NSW, Sydney Cancer Partners, International Society for the Advancement of Cytometry Marylou Ingram Scholars Program.

References: 1) Olver, I., Marine, F., & Grogan, P. (2011). Disparities in cancer care in Australia and the Pacific. *The Oncologist*, *16*(7), 930–934. **2)** Schmidt, D., Webster, E., & Lyle, D. (2025). Mechanisms to build research capacity in the rural health workplace: a realist synthesis. Front Med, 12, 1584904.

Read more about this innovative immune monitoring method:
Smith, N., et al. (2026). Workflow to Achieve High-Dimensional Immunophenotyping in Resource-Limited or Decentralized Environments. *Current Protocols*, Vol 6, Issue 3, *e70335*.