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912 A COMPARISON OF 37+ IMMUNE POPULATIONS IN WHOLE BLOOD BY CYTOF® FLOW CYTOMETRY COLLECTED WITH 4 DIFFERENT PRESERVATION REAGENTS

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Background Immune profiling studies using whole blood (WB) specimens are increasingly common in research and clinical research settings. It is crucial to standardize sample collection, shipping, and downstream processing of specimens for large studies. WB preservation reagents have been developed to address these logistics challenges.

Using a lyophilized CyTOF® 30-marker immune profiling panel, this study aims to assess the effect of Streck Cyto-Chex®, BD PAXgene® Blood DNA Tube, Smart Tube Proteomic Stabilizer PROT1, and Cytodelics preservatives on data quality.

CyTOF® flow cytometry allows for simultaneous detection of 50-plus parameters in a single tube without the need for single-stain controls. CyTOF uses typical antibody staining protocols and overcomes spillover and autofluorescence by using antibodies conjugated to heavy metal isotopes.

Methods We compared 37+ immune populations in WB drawn from three healthy donors into BD heparin, Cyto-Chex, and PAXgene blood collection tubes. As a baseline, blood from the BD heparin donors was stained with the Maxpar® Direct® Immune Profiling Assay kit and T Cell Panel 3 with <24h post-blood draw. For Cytodelics stabilizer and Prot1, aliquots from BD heparin donor tubes was combined with stabilizer and frozen <24h post-blood draw. Remaining blood from the BD heparin, Cyto-Chex, and PAXgene tubes was stored at room temperature (RT). At 48h post-draw, blood stabilized in Cytodelics stabilizer and PROT1 was thawed and RBC lysed. All samples were stained following the Maxpar Direct Immune Profiling Assay protocol.

Samples were acquired on the CyTOF XT™. Data was analyzed manually and by automated analysis using Maxpar Pathsetter™ software.

Results Compared to the baseline at 24h, the frequencies of most populations remained similar across conditions however, the median staining intensity of most markers was reduced for all conditions. Major immune cell populations were identified when WB was stored at RT in Cyto-Chex and PAXgene tubes. Major populations were also preserved in PROT1 and Cytodelics, however, the expression of chemokine receptors was altered.

Conclusions This study provides a comparison of WB preservation methods and their compatibility with a lyophilized CyTOF immune profiling panel. All stabilizers allowed major immune populations to be identified at 48h post thaw, although signal intensities of markers were reduced. An appropriate preservation method combined with lyophilized CyTOF antibody panels enables deep immune profiling when access to instruments, expertise and sample volume is limited in large/multi-site studies.

Ethics Approval The samples obtained for this study were sourced from an accredited commercial provider.