FLOW CONTRACT SITE LABORATORY

The Marker

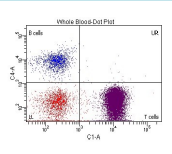
FCS Laboratory Quarterly Newsletter

2014 First & Second Quarter | Issue 14

This Issue:

Upcoming Events:

- ICCS Conference here in Seattle, October 10-14
- WBBA 25th Anniversary Summer Social, August 21st



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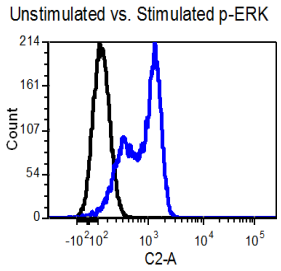
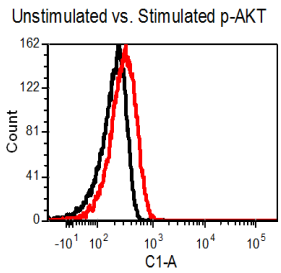
Development of a Novel Positive Signal Control for p-AKT and p-ERK

By: Esther Dubrovsky, MS, Lynette Brown, MS, CHT and Jennifer J Stewart, PhD

Over the past decade, the use of flow cytometry in clinical studies has grown substantially and has led to new and innovative flow cytometric methods for biomarker evaluation. Unfortunately there are few commercially available controls to evaluate assay reproducibility and reliability for many biomarkers of interest, especially intracellular proteins. Measurement of phospho (p)-AKT/PKB and p-ERK protein expression has been of increasing interest due to their key roles in cell growth, survival, and proliferation. However, p-AKT/PKB and p-ERK proteins are not readily detected in PBMC samples from normal donors due to the lack of the phosphorylated form under these conditions. In order to validate a method for evaluating these biomarkers by flow cytometry, a control was developed to show a positive signal over background in normal human PBMC. ERK was stimulated with PMA and AKT was stimulated with IGF-1 to produce phosphorylated forms, which can be detected by monoclonal antibodies directed against specific phosphorylated epitopes. The stimulated samples were fixed and frozen and then evaluated at weeks 1 and 2, and months 2, 3, and 4 post-stimulation to determine if the signal would be stable and reproducible within the specified storage conditions. Stability testing results showed that subject specimens can be tested up to a 3 month timeframe with precision of less than 10% CV consistently attained.





For more information on these controls, please contact us directly or visit our poster session at the upcoming ICCS meeting in Seattle on October 10-14th, 2014.

FCS Laboratory First & Second Quarter 2014 Updates

Jennifer presented at the AAPS National Biotech Conference in San Diego, CA this past May. The Round Table discussion was filled with excellent comments and conversations.

Stay tuned for a possible resurrection of the NW Regional Flow Cytometry Group!

Summer Events in Washington State

Bumbershoot August 30th– September 1st
www.bumbershoot.org
The nation's largest music and arts festival, attracting over 100,000 people

Seattle Seafair's 65th Year Anniversary
www.seafair.com
Events include a marathon, triathlon, parades, and the return of the U.S. Navy Blue Angels