

Assessment of T cell activity by quantification of phosphorylated STAT5 using multicolour flow cytometry

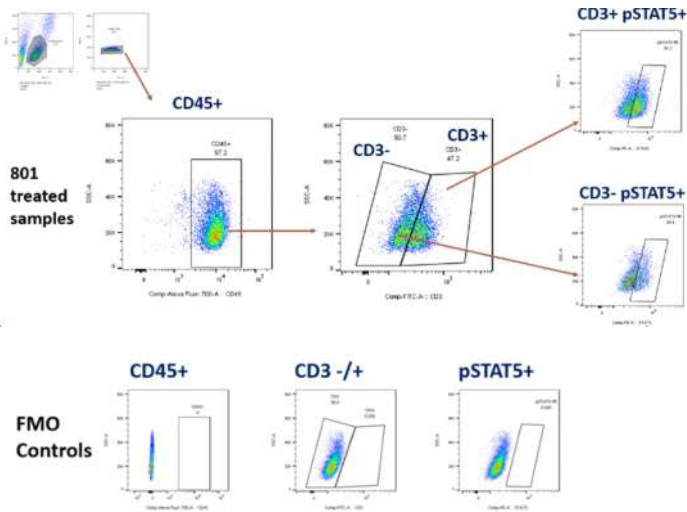
Authors: Narasimhulu Simma, Shalini Kolarkar, Tej Madineni, Murali Addepalli

INTRODUCTION

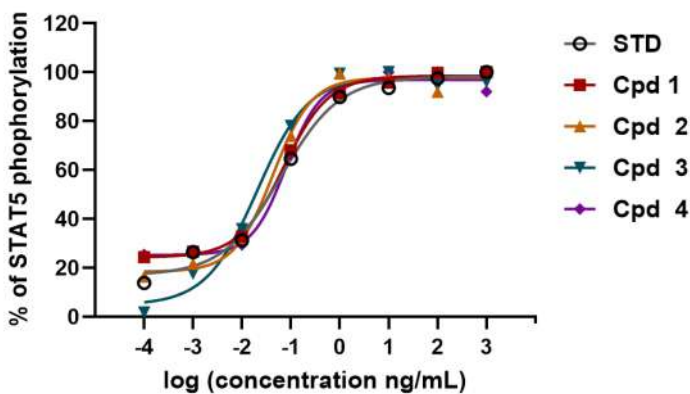
Multicolour flow cytometry offers rapid, multiparametric analysis of various cell populations of interest. Its applications cater to the needs of both researchers as well as clinicians. In drug discovery & development, multicolour flow cytometry is applied in immunology, immunoncology, biomarker discovery, virology, immune monitoring, etc.

Bifunctional engineered proteins, Compounds 1-4 (Cpd) are expected to stimulate the T cells by activation of the JAK1/ JAK3 tyrosine kinases and phosphorylate STAT5 (signal transducer and activator of transcription). Quantitative analysis of phosphorylation of STAT5 in a dose-dependent manner aids to determine the potency of T cell stimulators.

REPRESENTATIVE DOT PLOTS



Gating strategy for quantification of phosphorylated STAT5 in the human lymphocyte population (Lower panel indicates FMO controls)



Dose-response curves for compounds 1-4 in h PBMCs incubated for 20 min at 37°C

Test Compound	STD	Cpd 1	Cpd 2	Cpd 3	Cpd 4
EC ₅₀	0.06	0.07	0.04	0.02	0.08

*Determined by GraphPad prism ver. 9.5



CONCLUSION

All test compounds showed similar potency phosphorylating STAT5 and are found to be similar to that of the wild protein.

WHY LEXTRO

Scientists at Lextro Bio Solutions have extensive experience in working with multicolour flow cytometric analysis and cell sorting. The contributions of Lextro Scientists led to the advancement of various therapeutic molecules from Research to Clinics. Lextro offer services in immune therapeutics encompassing

- Hit to lead identification
- Immune cell phenotyping in Blood, Tumour or Tissues
- Biomarker analysis
- Receptor Occupancy Assays
- High-yielding clone isolations for recombinant proteins

Lextro Bio Solutions Pvt. Ltd.

#189, Synergy Square 1 Building, 2nd Floor, Genome Valley, Hyderabad-500 078, India
www.lextrobio.com | contact@lextrobio.com