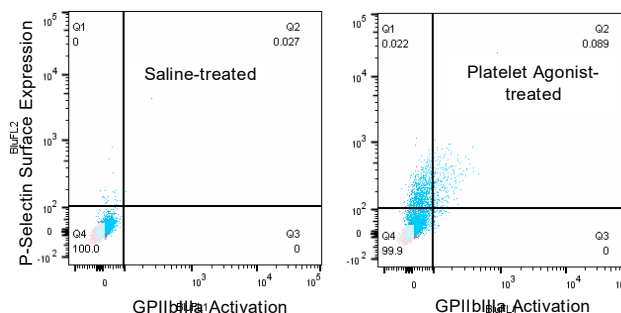


Platelet Function Analyzed by Flow Cytometry

Flow cytometry is a sensitive technique for assessing specific components of platelet function, including degranulation, receptor signaling and surface protein activation.

Platelet activation results in changes in platelet shape, degranulation, and signaling, including calcium flux, and protein phosphorylation. Agonists such as thrombin or ADP bind protease-activated receptors (in human platelets, PAR-1 and PAR-4) or purinergic receptors (P2Y₁₂), respectively, to induce platelet activation. In unlabeled whole blood or platelet-rich plasma samples, changes in platelet shape or granularity are measured as a shift in forward scatter and side scatter profile, respectively, on a flow cytometry instrument. Fluorescent antibodies are used to label surface P-Selectin (CD62P), whose expression is increased following alpha granule secretion, as well as conformationally-active platelet glycoprotein, GPIIb-IIIa. Flow cytometry is performed to measure the shift in fluorescence in agonist-treated samples relative to control-treated samples, indicative of platelet activation. Receptor-mediated signaling, such as calcium flux, cAMP levels, or protein phosphorylation (e.g., VASP), can also be measured.



Flow cytometry is utilized to evaluate markers of platelet activation following *in vitro* agonist stimulation in healthy donor platelets, or to assess patient platelet response to a panel of agonists.

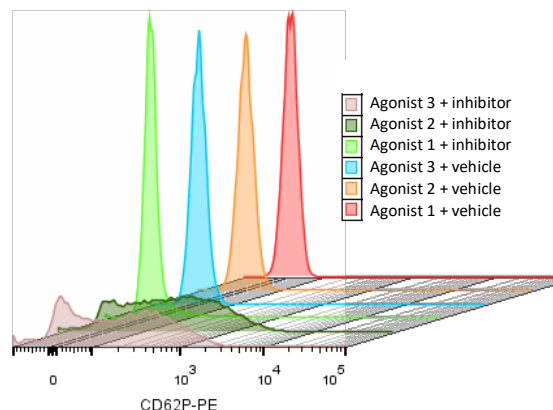
CirQuest routinely utilizes multi-color flow cytometry to evaluate platelet activation in whole blood, platelet-rich plasma, or washed platelets following agonist or drug treatment.

Routine Platelet Testing by Flow Cytometry

- ✓ Activation and shape change
- ✓ Degranulation and surface protein expression
- ✓ Receptor signaling and antagonism
- ✓ Platelet-Leukocyte Aggregates
- ✓ Microparticles

Clinical Trial Support

- ✓ Method customization and assay development
- ✓ Central lab capabilities for in-house testing
- ✓ Site training for study sample processing
- ✓ Provide reagent QC and kits



P-Selectin Surface Expression

Histograms are generated to highlight platelet responses to a panel of agonists following inhibitor treatment.