STEM CELL (L) AUTOCLASSIFICATION

ASSAY COMPATIBILITY

CATALOG NUMBER: AI004-IL

ASSAY NAME: Stem Cell RTF Assay (L)

ASSAY DESCRIPTION: CD34 Stem Cell panel with lysis buffer for mobilized leukapheresis samples.

PANEL DESCRIPTION

MARKER	FLUOROPHORE	CLONE
CD45	PE/Cy7	HI30
CD34	PE	581
Viability	DiYO-3	N/A
Count beads	N/A	N/A

The Accellix Platform automates your entire GMP cell phenotyping process from sample preparation to data acquisition and analysis, in order to generate rapid results directly in the manufacturing suite. Central to our enabling technology are standard and custom assays. These assays allow for sample preparation in a single-use microfluidic cartridge using unitized and dried reagents that are stable at ambient temperatures. The dried reagent also has control beads, enabling cell counting and in-run QC for every assay. Each assay incorporates an Accellix cartridge with a unique QR code, simplifying the workflow and reducing the chances for human error. The autoclassification software (algorithm-based analysis) automatically analyzes the compensated flow cytometry data using multi-parametric analysis and provides real-time results on the Accellix instrument screen at the conclusion of each run.

DESCRIPTION AND INTENDED USE

The Accellix Stem Cell (L) autoclassification data analysis software complements the Stem Cell RTF Assay (L) to automatically classify and enumerate CD34+ stem cells using multi-parametric analysis, dynamically setting gates for each marker in each run. The assay and software are intended for both fresh and frozen mobilized leukapheresis samples. The Accellix instrument tabulates the autoclassification results on-screen immediately after completion of the assay run and reports the results in a digital PDF file along with traceability information such as operator, Accellix assay lot, and software version. In addition, graphical results are generated for rapid and clear visual review of the data as well as providing an indication of how the software performed. To allow flexibility for oversight by subject matter experts, Flow Cytometry Standard (FCS) files are generated and exported along with the automated results.

KEY BENEFITS

+ **Autoclassification improves reproducibility by removing subjectivity from manual gate placement.**

+ **Automating analysis greatly shortens time to results in comparison to manual gating by applying objective logic for gate placement and classification of cells over manual analysis, reducing potential errors in data interpretation.**

+ **Automated results are generated immediately after each run, enabling users to quickly make informed decisions.**

+ **Assay reportables are configurable and include warning flags for flow disturbances, low viability, and low event counts.**

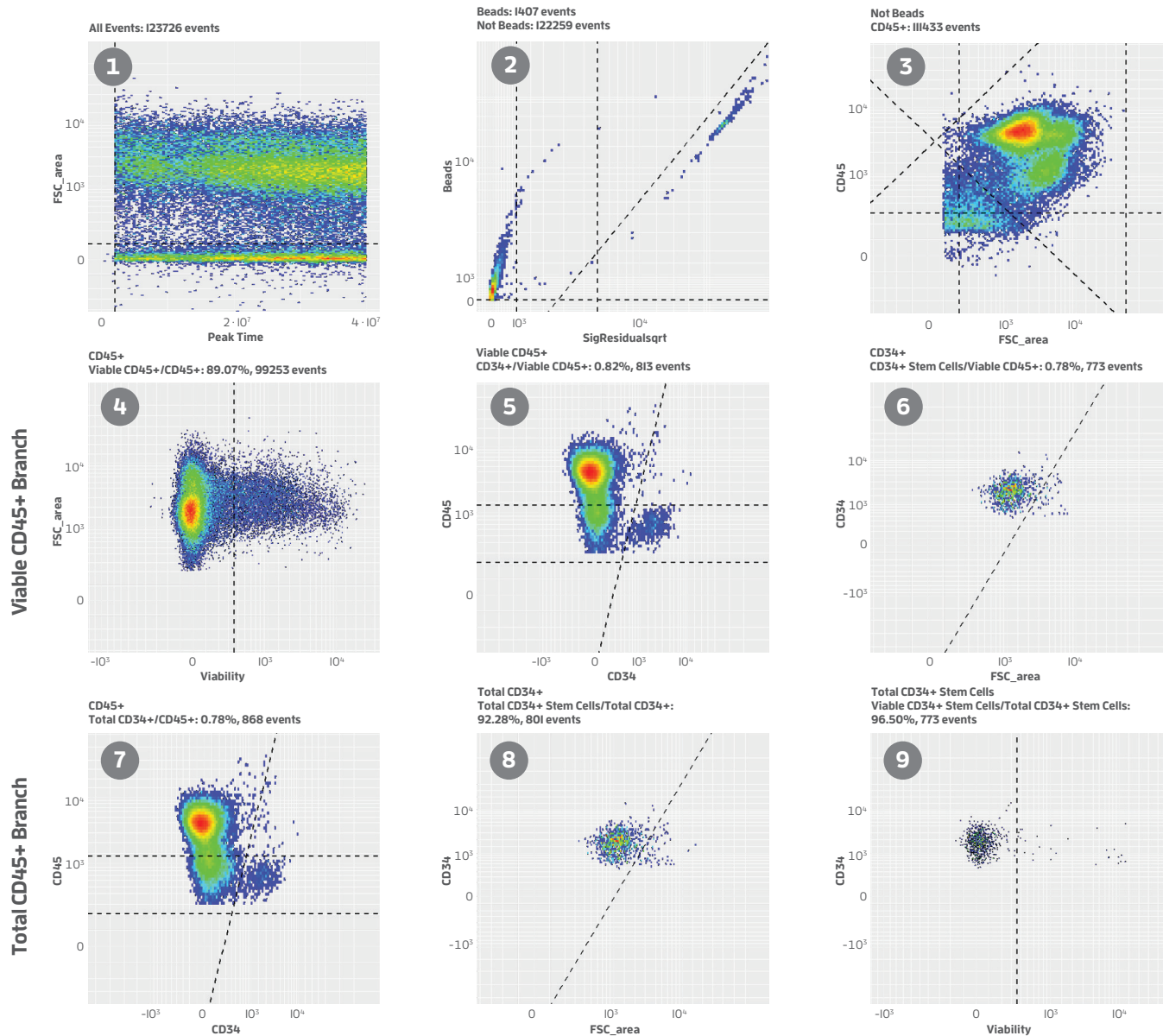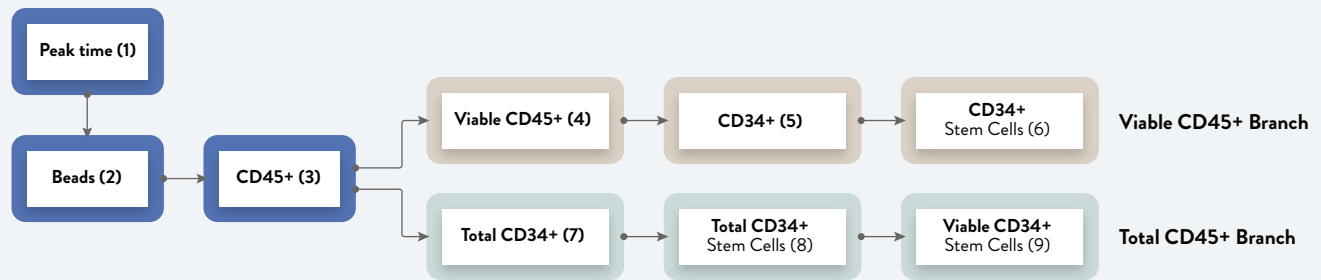# AUTOMATED ANALYSIS: MOBILIZED LEUKAPHERESIS

Figure 1 - Gating hierarchy and plots of representative Stem Cell RTF Assay (L) (Cat. No. AI004-IL) run with mobilized leukapheresis sample on the Accellix Platform. Assay-specific autoclassification software classified and enumerated CD34+ Stem Cells. From left to right, top to bottom: time gate (plot I) to exclude flow disturbances, identification and exclusion of assay beads used for ratiometric cell counting (plot 2), and CD45+ leukocytes (plot 3) followed by either the viable CD45+ branch consisting of viable cells of CD45+ (plot 4), CD34+ of viable CD45+ cells (plot 5) and CD34+ Stem Cells of viable CD45+ cells (plot 6*), or the total CD45+ branch consisting of total CD34+ cells (plot 7), followed by total CD34+ Stem Cells of total CD34+ cells (plot 8*), and viable CD34+ Stem Cells of total CD34+ Stem Cells (plot 9).

*Clean up gates have been added to remove debris and false positives.