Evaluation of the SpectraSlide® AP-1 for Accurate Counting of HL60 and Jurkat Cells

INTRODUCTION

Accurate quantification of suspension cell lines such as HL60 and Jurkat Cells are essential for a variety of applications in immunology, hematology, and other cell-based assays. These cell types are commonly used in high-throughput workflows and are typically processed in large volumes. Accordingly, achieving precision and consistency in cell counting is essential for ensuring the reliability and reproducibility of experimental outcomes.

The LUNA-FX7™ Automated Cell Counter, using acridine orange (AO) and propidium iodide (PI) dual fluorescence staining, provides a robust and efficient platform for assessing both cell concentration and viability. The PhotonSlide™ has served as a reliable standard for sample loading, and the SpectraSlide® AP-1 further enhances this workflow with pre-coated AO/PI staining and an intuitive "click–dip–release" design. This design eliminates the need for manual dye preparation and minimizes user-to-user variability during slide handling.
In this application note, we compare the performance of the SpectraSlide® AP-1 against the PhotonSlide™ for viability and concentration analysis of HL60 and Jurkat Cells using the LUNA-FX7™.

Figure 1. *Illustration of the SpectraSlide® AP-1 workflow. Press the sampling button, dip the sampling hole in the sample, release to draw in the liquid, and insert the slide into the LUNA-FX7™ for automated cell counting.*

MATERIAL AND METHODS

Cell Lines and Reagents

HL60 and Jurkat Cells were cultured under standard conditions in RPMI-1640 medium supplemented with 10 % fetal bovine serum and 1 % penicillin-streptomycin. Actively growing cells were harvested.

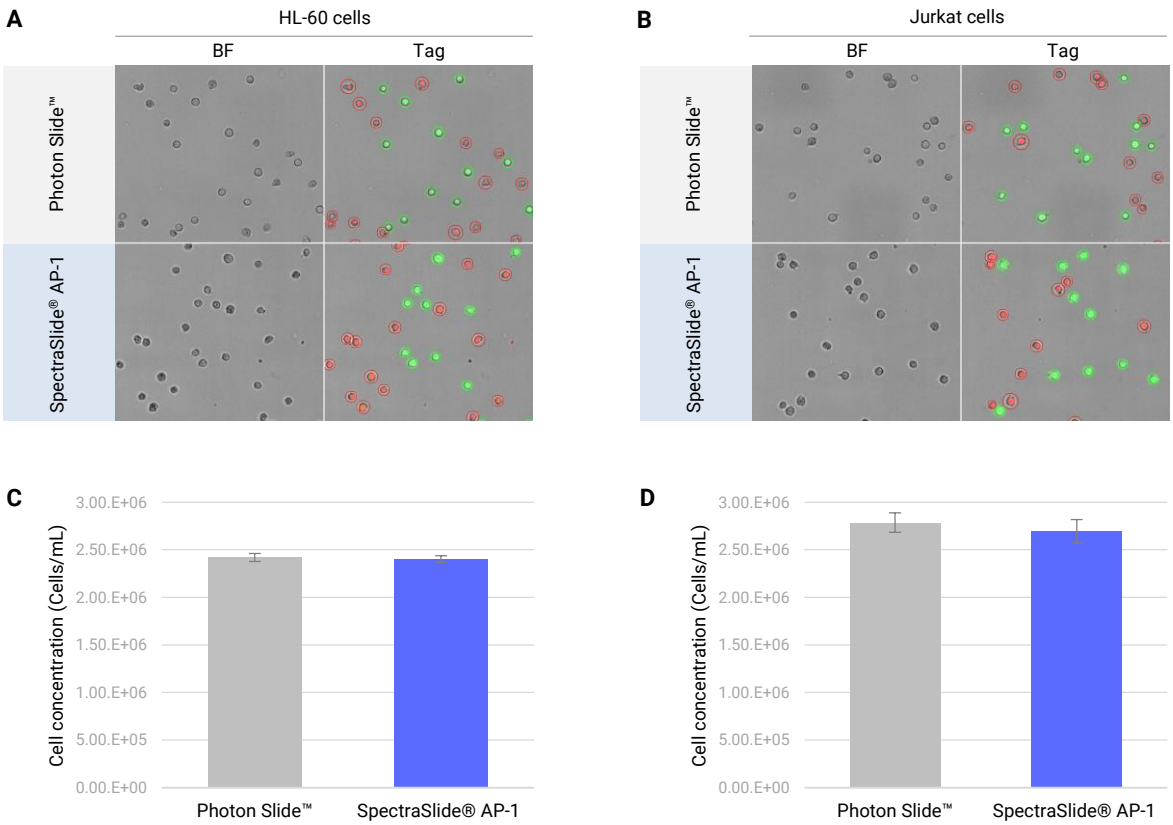
Comparison Between the SpectraSlide® AP-1 and the PhotonSlide™

Cells were prepared for concentration and viability measurements with a target viability of 50 %. Cell counting was performed on the LUNA-FX7™ using either the PhotonSlide™ (Cat# L12005) or SpectraSlide® AP-1 (Cat# L72061) under the default Fluorescence Cell Counting protocol. For PhotonSlide™ analysis, cells were stained with AO/PI reagent (Cat# F23001) at a 9:1 cell-to-dye ratio, and 10 µL of stained sample was loaded into the slide chamber. For SpectraSlide® AP-1, 500 µL of cell suspension was prepared in 1.5 mL tubes and loaded directly into the slide, eliminating the need for additional staining steps.

RESULTS

To assess the overall performance of the SpectraSlide® AP-1, both HL60 and Jurkat Cells were evaluated using the LUNA-FX7™ and compared with results obtained using the PhotonSlide™. Representative montage images of each cell type illustrate consistent detection of viable (green) and non-viable (red) cells across both slide types (Figure 2 A and B). The images confirm that the SpectraSlide® AP-1 enables high-quality fluorescence segmentation, comparable to the PhotonSlide™, for both cell types.

Quantitative analysis showed that cell concentrations measured with the SpectraSlide® AP-1 closely matched those obtained with the PhotonSlide™ for both HL60 (~2.4 × 10^6 cells/mL) and Jurkat Cells (~2.8 × 10^6 cells/mL) (Figure 2 C and D). Viability percentages were comparable between the two slide types for both HL60 and Jurkat Cells, with only minimal variation observed (Figure 2E and F).



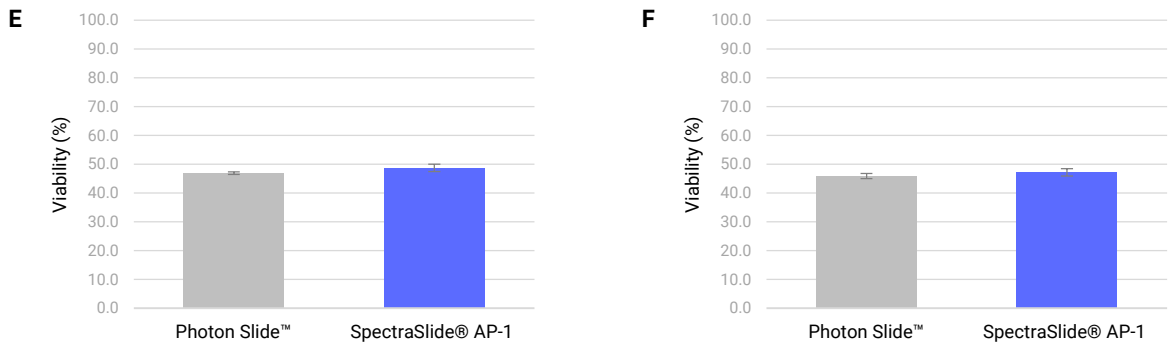


Figure 2. *(A, B) Representative montage images of HL60 cells (A) and Jurkat Cells (B) captured on the LUNA-FX7™. The left panel shows unstained brightfield images, while the right panel displays fluorescence overlays with viable cells stained green and non-viable cells stained red.*
(C, D) Bar graphs comparing total cell concentrations obtained using the PhotonSlide™ and SpectraSlide® AP-1 for HL60 (C) and Jurkat (D) cells.
(E, F) Viability percentages for HL60 (E) and Jurkat (F) cells, showing close agreement between the two slide types.

BF: Images captured in the brightfield channel.
Tag: Composite images of all channels, fluorescent and brightfield, with identified objects marked using red and green circles. Red circles indicate dead cells, while green circles represent live cells.

CONCLUSION

This study shows that using SpectraSlide® AP-1 with the LUNA-FX7™ offers a simple and reliable way to analyze small cells like splenocytes and nuclei. The slide comes pre-coated with AO/PI, allowing users to skip manual staining and reduce hands-on steps. Across both viability and concentration measurements, SpectraSlide® AP-1 showed excellent linearity and performance comparable to the PhotonSlide™, even in low-viability or debris-rich conditions. Additionally, the importance of allowing a brief settling period was confirmed to improve imaging focus and measurement accuracy. Together, these results highlight the SpectraSlide® AP-1 as a reliable, time-saving alternative for high-throughput and sensitive cell analysis workflows.