Using the CytoSMART™ Exact for accurate cell counting

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Introduction

Cell counting is one of the most frequently performed tests in life sciences laboratories for obtaining accurate and consistent experimental results. Many laboratories still manually count cells using a hemocytometer due to the expenses associated with automated cell counters (e.g. purchase of disposable slides). However, manual cell counting can be subjective and labor intensive.

To overcome this problem, the CytoSMART™ Exact combines the speed of automated cell counting with the cost-effectiveness of a reusable counting slide. Here, we compare manual cell counting with automated cell counting using the CytoSMART™ Exact.

Materials and methods

A stock solution of $2 \cdot 10^7$ cells/mL C6 (rat glial tumor) cells was used to prepare 11 sample concentrations ranging between $5 \cdot 10^4$ cells/mL and $1 \cdot 10^7$ cells/mL. Each solution was mixed with an equal volume of 0.4% trypan blue to distinguish the dead cells from the live cells. 10 μL cell suspension was loaded into a Neubauer Improved hemocytometer.

The counting chamber was placed on the CytoSMART™ Exact to count the cells using the cell counting software in the CytoSMART™ Connect Cloud. Subsequently, the cells within the four corner squares of the Neubauer Improved hemocytometer were manually counted and averaged to obtain the cell concentration. All counts were performed in triplicate.

Results

When counting manually, dead cells are distinguished from live cells via Trypan Blue staining. Using the same method, the CytoSMART™ Exact software is also able to detect dead cells (Figure 1).

The total amount of cells (live and dead) was used to determine the cell counting accuracy of the CytoSMART™ Exact. The cell counts were compared to those obtained manually with a Neubauer Improved hemocytometer (Figure 2). In all cases the standard deviation of the cell count performed with the CytoSMART™ Exact was smaller or equal to that of the manually count (Figure 2).

Due to the low amount of cells in the solution with the lowest cell concentration, the precision of the cell count was lower compared to the other concentrations. The samples with the highest cell concentrations (above $1 \cdot 10^6$) cannot be counted accurately by manual counting, while the CytoSMART™ Exact was able to accurately count the cells since individual cells could be detected within clusters of cells (Figure 3).
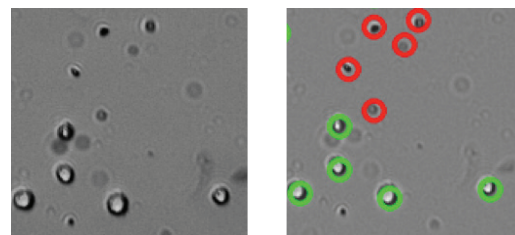

Figure 1. Dead cells stained with Trypan Blue detected by the CytoSMART™ software. Red circles represent dead cells, green circles represent live cells.
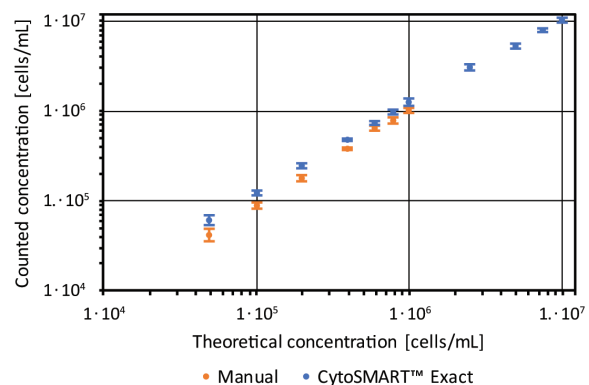

Figure 2. Different concentrations of C6 cells were counted manually and using the CytoSMART™ Exact (n = 3). In both cases, the count corresponds well with the theoretical concentration (error bars represent the standard deviation).

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Conclusion

The CytoSMART™ Exact was able to count cells with similar accuracy as manual cell counting. The advantage of the CytoSMART™ Exact cell counter compared to manual counting are the speed at which counts can be achieved (< 3 seconds*) and the wider range of cell concentrations that can be counted. Furthermore, manual counting can be operator dependent since each operator handles different criteria to include or exclude a cell in the count. Since the CytoSMART™ Exact software always handles the same criteria to count the cells, the operator dependency decreases significantly. The CytoSMART™ Exact combines the speed of automated cell counting with the accuracy of manual cell counting, making it an excellent option for counting cells.

* The counting speed depends on the upload and download speed of the internet connection as the cell count is performed in the CytoSMART™ Connect Cloud.
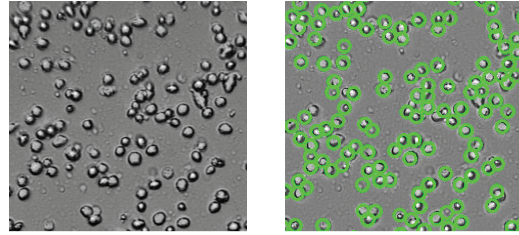

Figure 3. The CytoSMART™ software is able to detect individual cells within clusters of cells. Red circles represent dead cells, green circles represent live cells.