New robust method to normalize results of cell based assays with InCellis® Smart Cell Imaging System

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/ ABSTRACT

Single cell based assays are powerful tool for studying drug effects, protein interactions and signalling pathways. However, the assay analysis depends on several factors that can impact the results. An obtaining an accurate number of cells within the cell culture and/or calculating cell culture confluency are some of the important factors that need to be taken into consideration prior the optimization of the results.

Previously, we demonstrated the benefits of cell confluency application to define the perfect time to start the assay. In this study, we introduce a new cell culture InCellis App (InCellis® Smart Cell Imaging System (Bertin Technologies)), which is optimized for adherent cell counting as a new, rapid and accurate method to normalize results obtained from cell based assays in an automated fashion. An accuracy of cell counting between standard referent method and InCellis application is compared and presented here. Total cell counting application is a reliable alternative to a visual assessment of cell number, and is an efficient approach for any cell type analysis.

/ RESULTS

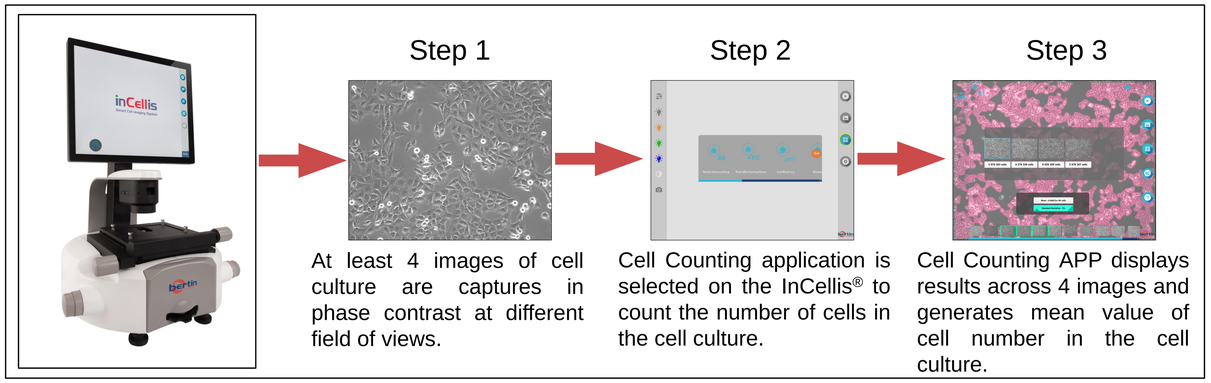

Figure 1. Automated total cell counting workflow using InCellis® application

/ CONCLUSION

The Total Cell counting application is a novel, robust, non-invasive (stain and trypsinisation free) method for a total cell counting.

An application provides accurate results when compared to the standard visual method.

Cell counting in an automated manner provides not only consistent results, but also allows the normalisation of cell-based assays by significantly reducing a hands-on time

Acknowledgement:

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/ METHODS

- MCF7 (breast cancer) cell lines were plated on 85 mm petri dishes in two cell densities : 1/10 and 1/5
- Cell lines were incubated at 37°C in appropriate cell culture media (DMEM) for 2 days
- A set of images of each petri dish with cell culture were captured using InCellis in phase contrast mode with 10x and 20x objectives
- Captured images were used to determine the total number of cells per petri dish using the Total Cell Counting application
- 8 different images of each dilution were captured in order to obtain statistically significant results
- Cell cultures were tripsinized, collected in cell culture media and an aliquoted cell suspensions were loaded on a Malassez cell. The number of cells was counted within 100 small squares following manufactures recommendations

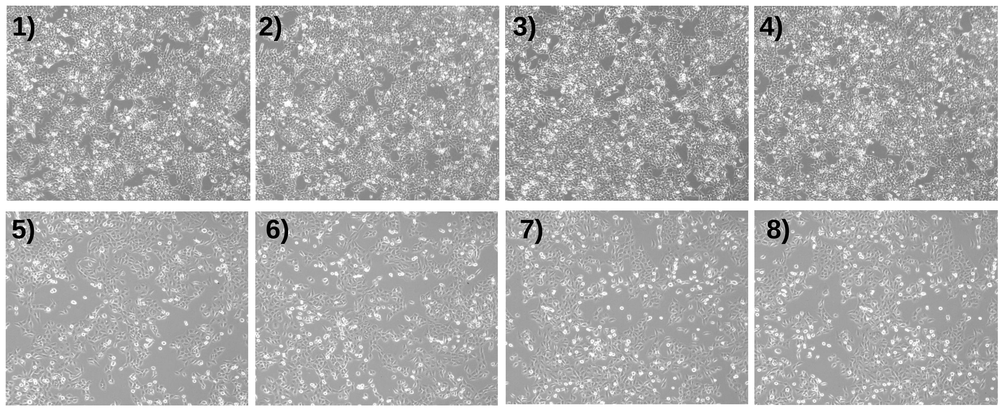

Figure 2. Four images of 1/5 (images 1-4)) and 1/10 (images 5-8) cell culture dilutions captured using 20x objective with InCellis. 6,3 M cells (mean values were obtained with a standard deviation of 3 % for 1/5 cell dilution and 3.5 % for 1/10 cell dilution.

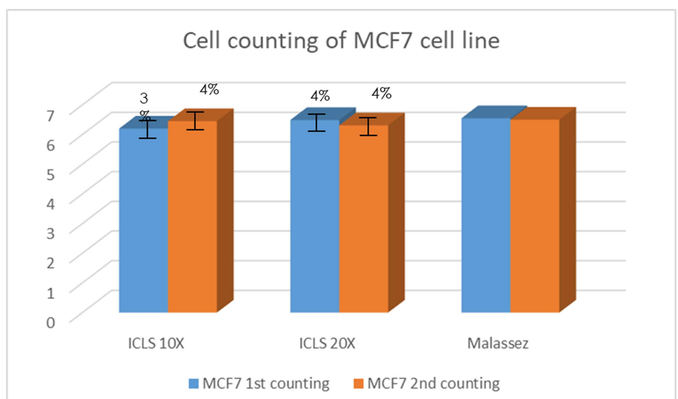

Figure 3. comparaison of total cell number of 1/10 MCF7 cell culture calculated with Total Cell counting automated application (InCellis) and manual cell counting on Malassez cell. ICLS 10X : images with 10X magnification. ICLS 20X : images with 20X magnification.

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