Seed point detection with Livecyte system November 2016

- Automatically detect cell seed points within a label-free heterogeneous population
- Obtain an accurate cell count at every time point in a label-free population
- Higher success percentage compared to commonly used alternative algorithms
- Reveal dynamic phenotypes by enabling robust tracking of individual cells over a time course
- Confront the deficiencies associated with population averaged approach and alternative manual solutions to characterising cell behaviour

1. Introduction

Robust segmentation of individual cells allows more refined data to be extracted from a cell culture in comparison to the alternative population averaged approach. Morphological and dynamic phenotypic parameters can be investigated, as well as the ability to employ more accurate cell models such as label-free heterogeneous populations. Manual segmentation of cells comes at a great cost in terms of time, repeatability and accuracy and as such an automated approach is far superior. In this technical note we introduce an automated seed point detection algorithm, which outperforms alternative automated techniques. Seed point detection is a vital step to robust cell segmentation and achieving all the advantages associated with the procedure.

2. Phasefocus Water Balloon seed point algorithm

The Phasefocus Livecyte system allows morphological and dynamic phenotypic parameters to be extracted from a heterogeneous population in a quick and easy manner. This is achieved by an automated seed point detection algorithm which identifies and subsequently enables the robust segmentation of individual cells through an entire time lapse.
The Water Balloon algorithm is

- Easy to use
  - Has 3 simple sliders to control the seed point detection process
- More consistent than manual seed points
  - Gives the same seed points every time when the same thresholds are used
  - Remains consistent and does not fatigue as a human operator
- Novel technique developed by Phasefocus
  - Not available in any other software package

3. Results

Fig. 1 displays a number of cell shapes, types and confluency examples with the water balloon algorithm applied. The location of each seed point, as calculated by the Livecyte system, is represented by a red cross overlaid on each cell. The individual frames are extracts from a longer time lapse study.

The Water Balloon algorithm was compared with three alternative algorithms namely, Extended h-maxima [1], Threshold and Erode [2] and, Level Set Voting and Mean Shift [3]. Fig. 2 shows the percentage of manually identified cells that contain exactly one seed point after running the various algorithms on the same datasets. Each algorithm was tuned for optimal performance on each dataset. The Phasefocus algorithm consistently out-performs the other techniques within this study (see Fig. 2).

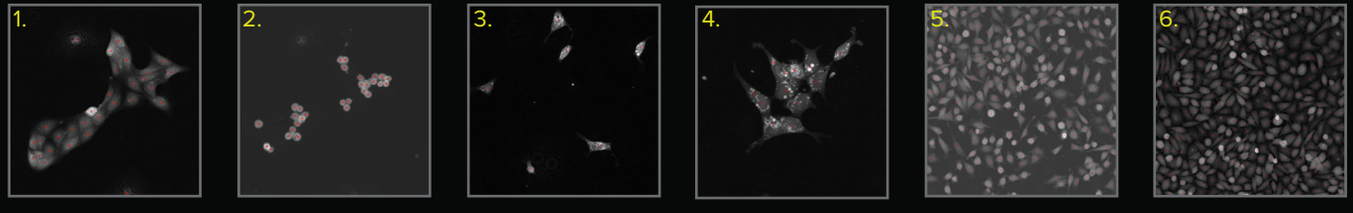

Fig. 1: Images of the data used for this study with Seed Points represented by a red cross.

4. Conclusion

As a result of non-invasive acquisition (label free with no risk of photo-toxicity) and robust seed point detection, the Livecyte system gives a far more accurate and in depth analysis of the cell behaviour compared to single time point and/or a population averaged approach. A manual approach to extracting the full suite of morphological and dynamic phenotypic parameters is time consuming and prone to subjective errors. The Phasefocus automated approach to analysis in combination with the non-invasive nature of its acquisition, allows more accurate cell models to be analysed quickly and robustly with the added confidence that the cells are behaving in a more native and natural manner.

References

1. Pinidiyaarachchi A. and Wählby C. 2005 Seeded Watersheds for Combined Segmentation and Tracking of Cells. Image Analysis and Processing – ICIAP 3617, pp.336-343

2. Barry D., Durkni C., Abella J. and Way M. 2015 Open source software for quantification of cell migration, protrusions, and fluorescence intensities. The Journal of Cell Biology 209(1) pp.163-180

3. Qi Z., Xing F., Foran D. and Yang L. 2011 Robust Segmentation of Overlapping Cells in Histopathology Specimens Using Parallel Seed Detection and Repulsive Level Set. IEEE Transactions on Biomedical Engineering 59(3), pp.754-765

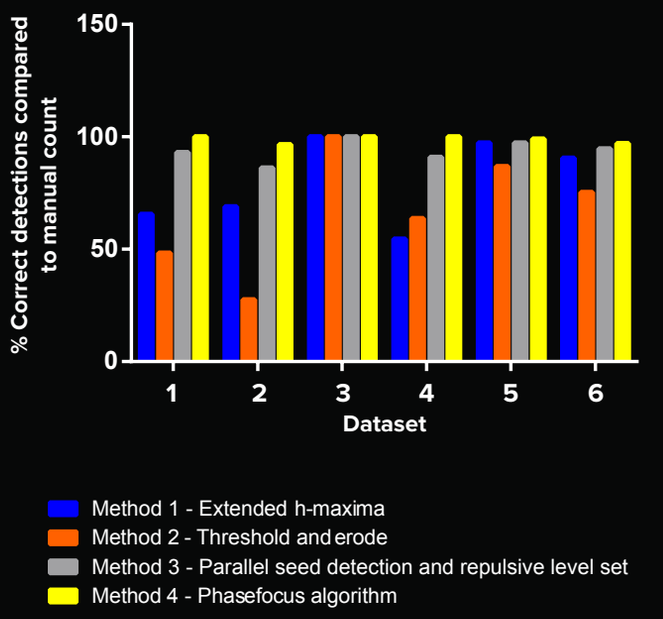

Fig. 2: Percentage of correct detections for each method on every dataset.

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For more information on the benefits of the Livecyte system, to access application notes and for additional product information, please visit: www.phasefocus.com/livecyte

A sample of time-lapse videos can be found at: www.youtube.com/phasefocuslimited

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