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Development of an Image Processing MATLAB Algorithm for Cell Growth Analysis

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Abstract
An image processing algorithm was developed using a MATLAB Graphical User Interface (GUI), allowing the automatic counting and analysis of fluorescent cells in microscope images. This software improves the reliability of results obtained by low-experienced researchers and can be used as feedback for maintaining the correct environmental conditions for fermentation.

Key words:
Image processing algorithm, MATLAB Graphical User Interface, Cell counting.

Introduction

In a biochemical process where the cells act as the catalyzer, the cells properties and their growth rate are of major importance for the data analysis, and one of the most used laboratory techniques for the kinetic evaluation is the optical microscopy.

The counting of fluorescent-marked cells is one of the most used procedures, but rely on the use of unfriendly interfaces and computer programs which demand the manipulation of different image processing parameters in order to obtain a reliable counting[1].

In this scenario, the development of an appropriate image processing algorithm with a user-friendly interface and a computer language common to the scientific community allow a low-experienced researcher to perform those analyses in a standardized and efficient way. The digital data obtained can then be used as feedback for improving the mathematical model used for the fermentation analysis and also allow the maintenance of the correct environmental conditions for fermentation[2].

Results and Discussion

A pre-processing of the images was performed for the cell counting, consisting of 4 major steps:

1. Conversion of image from RGB to gray scale;
2. Enhancement of image contrast through histogram equalization;
3. Removal of cells on the image borders;
4. Use of low-pass filter for noise removal.

After the pre-processing step, three different algorithms were applied to identify the cells: **Blob Analysis, Fill and Segmentation, and Watershed**.

To aid the tests a GUI, shown in **Figure 1**, was developed using the MATLAB (MathWorks) assistant.

For the counting on a chamber used for the culture of fluorescent *Saccharomyces cerevisiae* cells[3], the Blob Analysis showed the best results. The Watershed captured a great amount of noise and was very sensitive to the modification of the parameters, while the Fill and Segmentation counted the walls as cells. The results are shown in **Figure 2**.
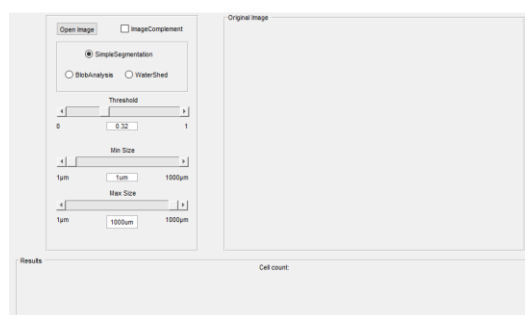
Image 1. GUI interface developed on MATLAB.
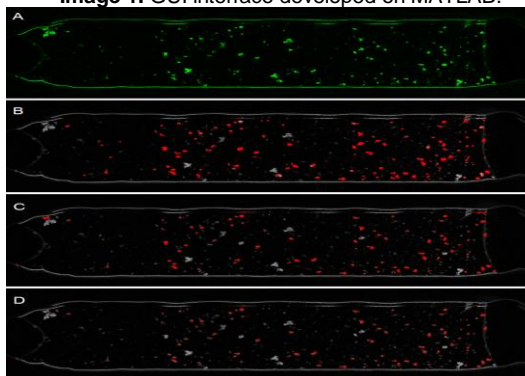
Image 2. (A) Chamber used for the culture of fluorescent-marked cells[3]; Segmentation and counting using 3 different algorithms: (B) Blob Analysis; (C) Fill and Segmentation; (D) Watershed.

Conclusions

All the algorithms showed results consistent with the visual counting, but still present problems in the isolation of single cells, mostly when they are grouped.

The Fill and Segmentation algorithm can give information on cell structures, so additional filters could solve the problems of walls identification. The Watershed lead to a counting higher than the visual procedure, counting the same cell multiple times. The Blob Analysis, despite having the best efficiency, can be improved by adjusting the contrast during the pre-processing step.

Acknowledgement

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[2] DORAN, P. *Bioprocess Engineering Principles*. Second Edition. Elsevier, **2013**.
[3] Oliveira *et al. Biochem. Eng. J.* 105 (**2016**) 288-295.