Image processing on animal cell cultures: a refined technique

Mariana Xavier, Daniela P. Mesquita, António L. Amaral, Eugénio C. Ferreira, Lígia R. Rodrigues and Leon D. Kluskens*

kluskens@deb.uminho.pt

Centre of Biological Engineering, Universidade do Minho
Campus de Gualtar, 4710-057 Braga, Portugal

Abstract

The process of microscopic animal cell counting can be a time-consuming process, resulting in a subjective analysis varying according to the researcher's perception. Regarding the ideal moment to divide the cells, the decision is performed in an empirical manner and is affected by the complexity of cell morphology and density. Searching for a way to overcome these problems, and considering the decreasing costs of computational data processing, a window was found for new methodologies to quickly characterize a given structure.

Advances in digital imaging allow the extraction of quantitative information, opposite to the qualitative and subjective evaluation of human analysis. Thus, microscopy image analysis techniques have gained, during the last years, an unquestionable role in several fields of research. The purpose of an image processing step resides in obtaining a final image holding significant information for a given application. These techniques should be automated as much as possible to avoid subjectivity. Thus, several segmentation techniques have been already proposed. For segmentation to take place, usually a threshold value(s) must be defined to allow the differentiation between the objects and background. Other methods, such as region growing, mathematical morphology and watershed are also used for this purpose. These are simple algorithms that when appropriately used can provide promising results and oftentimes with a low computation complexity. Nevertheless, the previous methods have some limitations, including non-uniform intensity variations, low-contrast images, irregular segmentation and over-segmentation. More sophisticated methods based on frame-

works of active contours (e.g. snakes, level-sets) or graph-cuts can also be applied to segment cells with positive results. Nonetheless, these algorithms present high computational complexity.

The main goal of this work was to develop an image processing tool using several algorithms in order to improve cell segmentation processing for different morphological cells and densities. For that purpose, different cells were used – MDA-MB-231 and -435, both cancer cell lines, and MCF-10-2A, a non-tumorigenic line. Cells were observed in a Leica DM IL inverted contrasting microscope, in phase-contrast at 100x total magnification, coupled with a Leica D-LUX 3 camera, ensuring the same acquisition conditions. Despite the variability in their morphology, preliminary results demonstrated that the segmentation process was fairly successfully. As a result, the previously described flaws were minimized, leading to more efficient animal cell culturing with less variability.

All authors contact details

mariana.nx91@gmail.com
{daniela, lpamaral, ecferreira, lrmr, kluskens}@deb.uminho.pt