Segmentation and sub-cellular feature-based analysis of microscopy images

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***Abstract*— An image processing framework for the automated analysis of multichannel cell images is presented. It segments cells into subcellular regions, extracts a large number of image features and identifies image features that characterize cells grown under different conditions. The framework is applied to two sets of fibroblast cells, grown on stiff and soft matrixes. Textural features from the actin channel best differentiate cells grown on stiff and soft matrixes, suggesting that the arrangement of protein actin plays a fundamental role in cell response to the mechanical properties of the extracellular environment, according to the analyzed images.**

***Index Terms*—Extracellular matrix, fibroblast cells, actin, segmentation, feature extraction, feature-based analysis, subcellular analysis.**

I. INTRODUCTION

The mechanical properties of the extracellular environment are known to determine specific cell responses that play a fundamental role in pathologies such as cancer and cardiovascular diseases. In particular, the extracellular environment seems to affect cell shape, motility, and reproduction. To gain insight into the role of the extracellular environment, microscopy tools such as oil immersion and confocal microscopy, combined with simultaneous staining by different antibodies, have enabled the acquisition of high resolution multichannel images. This has enabled the visualization at subcellular level of proteins such as actin, myosin, focal adhesions, and vinculin, whose shape and spatial arrangement can be used to determine cell response [1, 2, 3, 4]. However, fully automated image processing tools with broad applicability to multichannel data are needed for effective image analysis.

II. TECHNICAL APPROACH

In this work, an image processing framework for the automated analysis of multichannel cell images was implemented. It enables the identification of subcellular regions, image features and channels that differentiate cells that were imaged under different conditions.

The framework consists of three modules: (1) a segmentation module to identify, for each channel, subcellular regions based on region geometry, intensity distribution and texture; (2) a feature extraction module to compute a large set of textural, geometric and intensity features; (3) a feature

analysis module to identify the image features that characterize cells grown under different conditions. The framework developed is flexible, user-friendly, and it has broad applicability to biology studies focusing on subcellular analysis.

III. SUBCELLULAR ANALYSIS

Two sets of multichannel images of fibroblast cells, grown respectively on stiff and soft matrixes, were analyzed using the image processing framework. The segmentation module consistently led to the identification of four well-known subcellular regions on the actin channel across all the cell images: lamellipodia, lamella, perinuclear region and nuclear region (Fig. 1). The obtained subcellular regions were validated by domain experts. Subsequently, image features were extracted from the subcellular regions and the four protein channels of interest: actin, myosin, focal adhesions and vinculin (module 2). Finally, the feature analysis (module 3) led to the identification of the image features that differentiate fibroblast cells grown on stiff ad soft matrixes.

Textural features in the lamella and lamellipodia regions from the actin channel are the most effective in cell differentiation. This suggests that actin, and in particular its textural characteristics, play a fundamental role in cell response to the extracellular environment. Future work will focus on increasing the set of features that are used for the analysis.

IV. DISCLAIMER

Commercial products are identified in this document in order to specify the experimental procedure adequately. Such identification is not intended to imply recommendation or endorsement by the National Institute of Standards and Technology, nor is it intended to imply that the products identified are necessarily the best available for the purpose.

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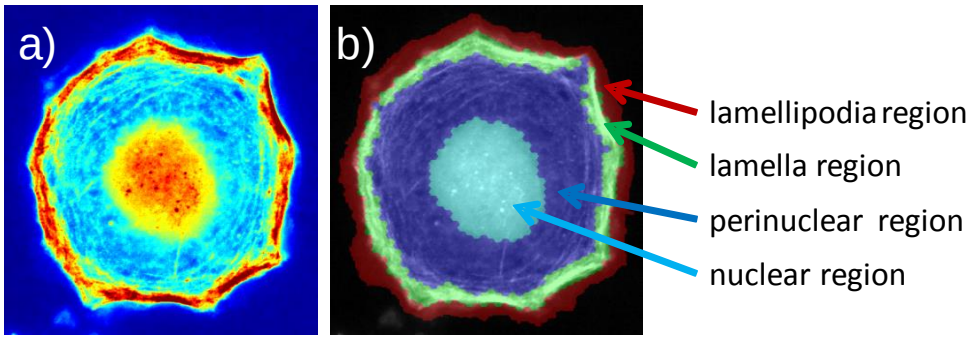

Figure 1: Actin-stained fibroblast cell, showing the actin fibers within the cytoskeleton (left). Four subcellular regions, which were detected using the segmentation module (right).