Automatic Detection of Filopodia from Fluorescence Microscopy Images

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Filopodia are thin, finger-like protrusions comprised of tight parallel bundles of filamentous actin. These protrusions are found at the leading edge of motile cells. Filopodia help cells to move through and sense their environment. Filopodia play crucial roles in processes such as development and wound-healing. Also, increases in filopodia number or size are characteristic of many invasive cancers and are correlated with increased rates of metastasis in mouse experiments. Thus, understanding the genetic and chemical factors that regulate filopodia is an important problem. There are presently no algorithms that automatically detect and accurately quantify filopodia in the range of sizes and numbers that are relevant to cancer cell motility. Instead, filopodia are detected by eye, and length or other spatial information are extracted by manually tracing filopodia using image manipulation software. Manual approach is tedious and slow, limiting the potential size of studies and their statistical power.

Here, we present FiloDetect, an automated tool for detecting, counting and measuring the length of filopodia in fluorescence microscopy images. The method first segments the cell from the background, using a modified triangle threshold method, and then extracts the filopodia using a series of morphological operations. Figure 1 shows the flowchart for the automatic detection system. Figure 2 shows the step by step results of FiloDetect system. We verified the accuracy of FiloDetect on Rat2 cell images, showing that per-cell filopodia counts and length estimates are highly correlated with the manual annotations. We then used FiloDetect to assess the role of a lipid kinase on filopodia production in breast cancer cells. Experimental results show that PI4KIIIβ expression leads to an increase in filopodia number and length, suggesting that PI4KIIIβ is involved in driving filopodia production.

FiloDetect will allow large scale comparative studies to be performed, assessing the effect of different genetic and chemical perturbations on filopodia production, in different cell types. In the context of cancer research, this tool can help to determine the mechanisms involved in cancer cell invasion, ultimately facilitating the development of cancer therapies that block metastasis by influencing filopodia production. Algorithms of FiloDetect were implemented in Matlab2009 and are available at `http://www.perkinslab.ca/Software.html`.

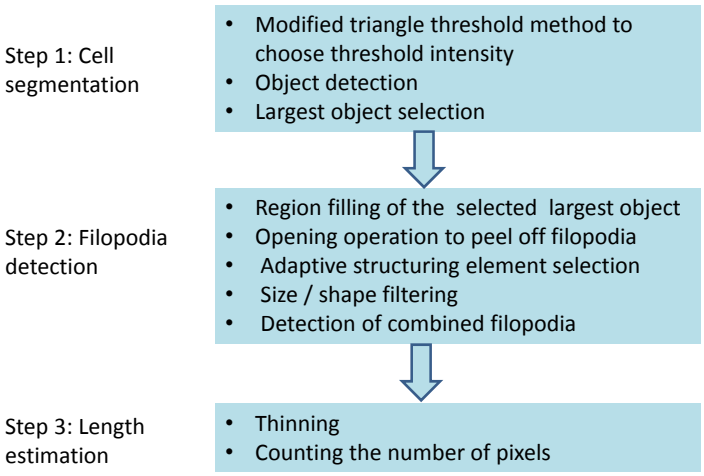

Fig. 1. Flowchart showing the steps of FiloDetect system.

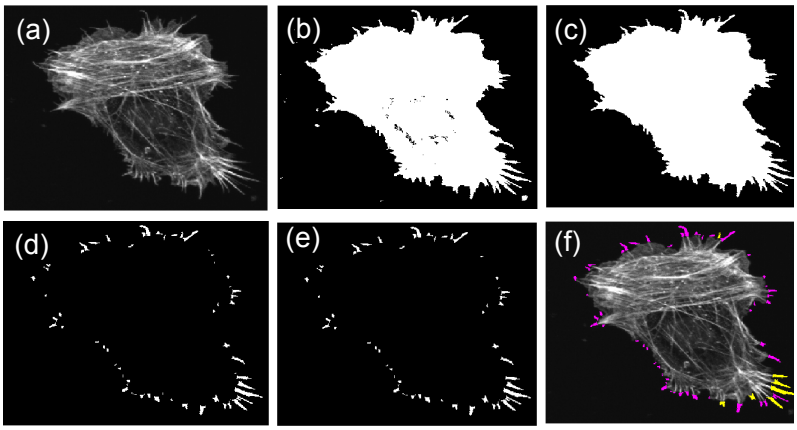

Fig. 2. Step-by-step results of FiloDetect system (a) input image, (b) thresholded image, (c) segmented cell after region filling, (d) candidate filopodia, (e) detected filopodia (pink represent single and yellow represent combined filopodia).