Chapter 3

Image Processing and Reconstruction of Cultured Neuron Skeletons

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Abstract. One approach to investigating neural death is through systematic studies of the changing morphology of cultured brain neurons in response to cellular challenges. Image segmentation and neuron skeleton reconstruction methods developed to date to analyze such changes have been limited by the low contrast of cells. In this paper we present new algorithms that successfully circumvent these problems. The binary method is based on logical analysis of grey and distance difference of images. The spurious regions are detected and removed through use of a hierarchical window filter. The skeletons of binary cell images are extracted. The extension direction and connection points of broken cell skeletons are automatically determined, and broken neural skeletons are reconstructed. The spurious strokes are deleted based on cell prior knowledge. The reconstructed skeletons are processed furthermore by filling holes, smoothing and extracting new skeletons. The final constructed neuron skeletons are analyzed and calculated to find the length and morphology of skeleton branches automatically. The efficacy of the developed algorithms is demonstrated here through a test of cultured brain neurons from newborn mice.

Keywords: Neuron cell image, image segmentation, grey and distance difference, filtering window, neuron skeleton, skeleton reconstruction, skeleton branch.
1 Introduction

The molecular pathogenesis of neurodegeneration is poorly understood. These challenges include high energy demand, the length of the neural axon and dendrites - and resulting susceptibility to cytoskeletal transport defects - and their high metabolic rate, which together with their relative paucity of antioxidant capacity, makes them highly susceptible to damage caused by reactive oxygen species (ROS) [1] [2] [3].

A number of methods have been developed for analysis of neuron extraction and imaging. These include an algorithm for fast, automatic extraction of neurite structures based on soma segmentation, seed point detection, recursive center line detection, and 2D-curve smoothing [4], an automated neurite analysis method for extracting single and connected centerlines along neurites [5], and an interactive technique for the tracing and quantification of elongated image structures [6].

In order to binary object images from poor quality images, it is essential to threshold the image reliably. Although many thresholding techniques, such as global and local thresholding algorithms, multi thresholding methods [7] [8] [9] [10] and unimodal thresholding [11] have been developed in the past, it is still difficult to deal with images with very low quality. Such a sample cell image is shown in Fig. 12(1), and its histogram is shown in Fig. 12(2). We can see that its histogram is unimodal because of poor background.

Information taken from images of neuron cells being grown in culture with oxidative agents allows life science researchers to compare changes in neurons. It is clear that image analysis and recognition are useful tool to help our study of the neuron degeneration in a human disorder called Zellweger syndrome. In morphological terms, we expect to see this initial deterioration as the contraction, and eventually loss, of processes of neurons grown in culture. Therefore, not only segment neuron cell images, but also it is important to reconstruct broken neuron cell skeletons caused by segmentation of neuron cell images with more poor background than that of some neuron images in the previous research methods [1] [2] [3].

In this paper, preprocessing procedure, segmentation of neuron images with poor background, is introduced in Section 2. In Section 3, the reconstruction of cell skeletons is developed. We then conclude our analyzer in the final section.

The contribution of the paper is that new segmentation method is firstly used to segment the cultured neuron cell images, novel reconstruction and analysis of neuron skeleton are firstly developed and the neuron skeleton length (neural axons) is automatically analyzed and calculated.

2 Segmentation of Cultured Neurons Using Logical Analysis of Grey and Distance Difference

To date, a number of methods have been developed for analysis of neurite extraction and imaging. These include an algorithm for fast, automatic extraction of neurite structures based on soma segmentation, seed point detection,