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The system for histopathology images analysis of spinal cord slices

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Abstract

In this paper, a specialized system for NeuN-stained neurons detection in two dimensional images of cat spinal cord slices is proposed. Considered approach of using interactive algorithm based on threshold and morphological transformations, in combination with hand correction of detection errors, provides some interesting results. The system accurately processing large images and collects all data of provided experiments of neurons detection in the centralized data base, which provides approximation of large collection of resulting data.

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Keywords: neurons detection; spinal cord slices; image processing; software; interactive algorithm; NeuN.

1. Introduction

An important part of the spinal cord experimental research is a visualization of neuronal activity, based on two-dimensional images of the spinal cord slices derived from neurophysiological experiments on animals. A three-dimensional model of neuronal activity constructed from two-dimensional images analysis results of various patterns of experimental animal movement. This improves the efficiency of studying the structure and functions of motion control neural networks. To solve this problem, must be solved the range of subtasks, in this paper supervised subtask of NeuN-stained neurons detection on two-dimensional images of the cat spinal cord slices, also described applied methods and some system features.

The task is complicated by the several factors, such as some neurons covered by thin layer of tissue, therefore part of neurons have variously transparence, also axon can be marked with the neurons body, it leads to errors in

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neurons size calculation or axon can be incorrectly recognized as individual neuron. Furthermore images of slice may contain physical tissue defects, such as gaps and indurations. Important to note that every image contain neurons with different size and the smallest neuron may be 6-8 times smaller than the largest. In addition, different imaging conditions, such as the light source intensity and transparency of the specimen, can cause variations of the image intensity in each of the three RGB channels resulting in neurons appearing with different colors in different image parts. Image example is shown in Fig. 1.

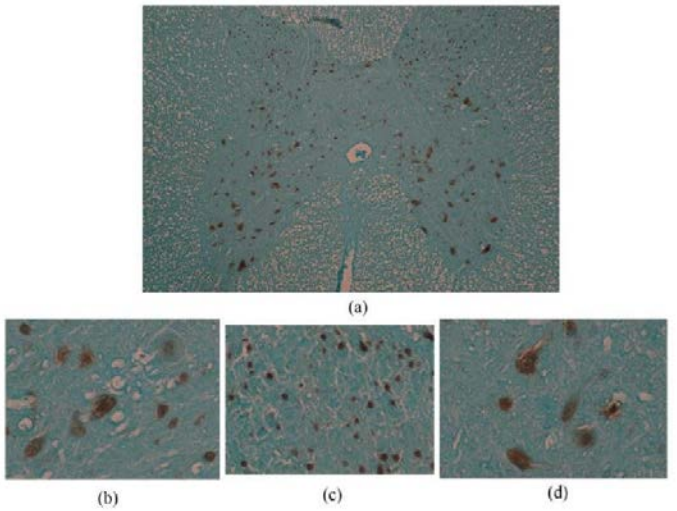

Fig. 1. (a) image of spinal cord slice; (b), (d) image fragments with large neurons; (c): image fragments with small neurons. Fragments (b) - (d) have same scale relative to the original.

2. Related works

Cell detection on histopathological images is quite widely research field of bioinformatics, because each new combination of the neuronal marker, the slice making approach, the resulting image dimension and the type of supervised tissue may generate a new type of histopathological images. We consider two-dimensional images of paraffin NeuN-stained cat spinal cord slices. For this type of images we didn't find any suitable articles. Otherwise, we should base on the experience of neurons detection in other types of two-dimensional histopathological images[1,2], but these methods don't work in case of processing images with physical defects. Furthermore, most of currently studying approaches work on 3D confocal images[3,4,5,6], but sometimes automated cell detection methods are not suitable replacement for manual cell detection[7]. It is useful to note some articles[8,9] considering processing images with inhomogeneous background and analysis object, part of the information from these articles may serve as a help for further development. From existing software should to note ImageJ[10] – public domain, Java-based image processing program, however, in order to improve performance decided to develop new application.

3. Algorithm

Process of neurons detection is presented on Fig. 2. Module "AlgoritmProcess" in this diagram is a combination of methods, configured manually or automatically according to precedents. The general scheme on Fig. 3 shows the operation of the algorithm for binary image producing.

Module "NeuronsMarker" allocates connected regions in the binary image; connectivity value is 8 that mean that the eight nearest neighbor pixels will be considered. Next, rectangles describing the connected regions that have passed the binary limits, drawn on the vector graphics scene "CorrectionScene" over the original slice images, in such format the user has the ability to manually add or remove the neurons outlining rectangles from the scene.