主题: SPIE Paper Number 10137-19 Acceptance and Manuscript Information

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Dear Hui Hui,

Congratulations! The chairs of the upcoming "Biomedical Applications in Molecular, Structural, and Functional Imaging" conference have accepted your paper, "Brain vessels segmentation for light-sheet microscopy image using convolutional neural networks," for Oral presentation to be presented 13 February 2017.

Symposium: SPIE Medical Imaging

Symposium Dates: 11 - 16 February 2017

Symposium Location: Orlando, Florida United States.

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PAPER TITLE: Brain vessels segmentation for light-sheet microscopy image using convolutional neural

networks

**PAPER NUMBER: 10137-19** 

PRESENTATION DATE: 13 February 2017

**PRESENTATION TYPE:** Oral (determined by Conference Chairs)

PRESENTATION DURATION: 20 minutes (includes Q&A for oral presentations)

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This year's event includes an outstanding selection of presenters and we look forward to your participation!

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# Brain vessels segmentation for light-sheet microscopy image using convolutional neural networks

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**Abstract:** Blood vessel segmentation is an important step in image analysis for tumor angiogenesis study etc. With the purpose of extracting line structures of blood vessels, some filter-based methods are used to segment vessels. However, the design of accurate and automatic vessel segmentation algorithms is still challenging, due to the variety and complexity of images, especially in brain blood vessel segmentation. In this work, we address a problem of automatic segmentation of brain micro-vessels structures in light-sheet image stacks. To segment micro-vessels in large-scale image data, we propose a method using convolutional neural networks (CNN) as a pixel binary classifier. Three convolutional layers and one fully connected layer are used in the CNN model. We extract a patch of size 32x32 pixels in each acquired brain vessel image stack as training data set to feed into CNN for classification. This network is trained to output the probability that the center pixel of input patch belong to vessel structures. To build the CNN architecture, a series of mouse brain vascular images acquired from a commercial light sheet fluorescence microscopy (LSFM) system were used for training the model. The preliminary experimental results demonstrated that our approach is a promising method for effectively extracted micro-vessels structures in brain images.

**Keywords:** Brain vessel image segmentation, Convolutional neural networks, Light-sheet microscopic imaging

**Purpose:** Light sheet fluorescence microscopy (LSFM) is a single-cell resolution imaging technique which illuminates the fluorescence-labeled mouse brain vascular network with a thin light sheet. However, micro-vessel structures are high density with inhomogeneous gray level in LSFM images, which results in challenging for extracting vessel structures with traditional segmentation methods. In this work, we propose a vascular image segmentation method by classifying each pixel in brain vessel image using trained convolutional neural networks (CNN) model to automatic segment brain vessels.

## **Method:**

## 1. Image acquisition

The whole mouse brain was cleared with solvent-based clearing method [1]. The brain vessels were labeled by antibodies anti-CD31 and anti-PDGFR  $\beta$  to specifically target vascular endothelial and hemangiopericyte respectively. The intact mouse brain

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was imaged by a commercial light-sheet microscope equipped with a sCMOS camera and 2x/NA 0.5, 6mm working distance dipping cap.

## 2. CNN architecture

Our CNN model consists of three convolutional layers and one fully connected layer, as shown in Figure 1. The output of the model is binary label of the pixel. A patch of size 32x32 pixels in each acquired brain vessel image stack are taken as input pixels that are processed by series of convolutional and max-pooling layers in CNN [2]. Then the map between target output and input signal are achieved via fully connected layers. In each convolutional layer, each feature map is composed of one or more vessels and is extracted from the input patch of pixels by a convolution filter. The first convolutional layer in Figure 1 is composed of six feature maps, and each feature map contains a 28\*28 pixel array, and each pixel acquired by a convolution filter, which extract feature from a 5\*5 region. A softmax activation function is used for the last layer to get the probability of a pixel of the input image belonging to that class. Note that each group of pixels uses the same connection weights, which could reduce the train parameters.

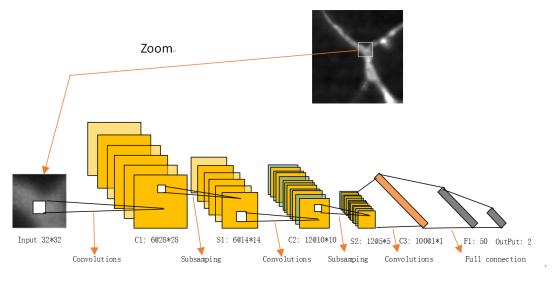


Figure 1. Convolutional neural networks for brain vessel segmentation

## 3. Data training

We acquire a series of mouse brain vascular images (2160\*2560\*700) from a commercial LSFM system in our lab. The method of training image data set contains four steps:

1) Random acquire a series of sub-images (500\*500) in a full-size image as shown in Figure 2 (b). 2) Then, the vessels in each sub-image were segmented manually to get the labels of pixels. 3) We random select some pixels from background, and then set the max intensity of selected pixel as threshold. 4) A patch of pixel and its label are fed into CNN if the intensity of pixel exceeds threshold, see Figure 2 (e).

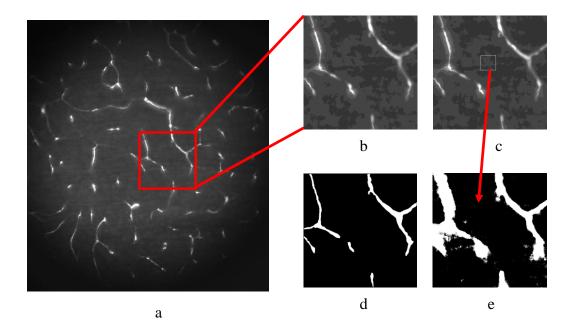


Figure 2. (a) The 121<sup>th</sup> slice from a series of mouse brain vascular images (2160\*2560\*700). (b) Random zoom in the rectangular frame. (c) The rectangular frame is random background pixels. (d) Manual segmentation results. (e) To balance the amount of pixels both in positive and negative, part of the negative pixels that greater than threshold value are set to positive. The positive pixels are used for training the classifier.

# New or breakthrough work to be presented:

We present a CNN model for brain vessel image segmentation in light-sheet microcopy images. This approach can be used for automatic segmentation of brain vessels.

### **Results:**

A CNN architecture has been built for automatic large-scale brain vessel image segmentation in light-sheet microcopy images. In preliminary study, 100 manually segment vessel image were used for training the classifier. The results illustrated that approach presented in this work is promising. However, to increase classification accuracy, more training data set have to be added into CNN model. The experiments are performed on a computer with a CPU Intel Xeon CPU E5-1620 4 cores @ 3.50GHz processor, 24GB of RAM, and a GPU NVIDIA Quadro K4200 graphics cards.

## **Conclusions:**

In this work, we have presented a method using convolutional neural networks (CNN) as a pixel binary classifier to address a problem of automatic segmentation of brain micro-vessels structures in light-sheet image stacks. The CNN architecture has been built by training the CNN model with three convolutional layers and one fully connected layer. The extracted feature by our approach can distinguish vessel and non-vessel pixels. This leads to a better performance than manual feature extraction. In future work, we will add more training data fed into our CNN model to increase

classification accuracy. And we will also optimize the GPU implementation for classifying each pixel, which will accelerate the computing process.

# **References:**

- [1] Renier, N., et al., iDISCO: a simple, rapid method to immunolabel large tissue samples for volume imaging. Cell 159, 896–910. 2014.
- [2] Dan, C Cirean, et al. Deep Neural Networks Segment Neuronal Membranes in Electron Microscopy Images. Advances in Neural Information Processing Systems 25(2012):2852-2860.