

Attributed Relational Graphs for Cell Nucleus Segmentation in Fluorescence Microscopy Images: Supplementary Material

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Abstract—More rapid and accurate high-throughput screening becomes possible with the development of automated microscopy imaging, for which cell nucleus segmentation commonly constitutes the core step. This technical report contains the supplementary material for the model-based algorithm that we developed for segmenting cell nuclei in fluorescence microscopy images [1].

Index Terms—Nucleus segmentation, model-based segmentation, fluorescence microscopy imaging, graph, attributed relational graph.

I. INTRODUCTION

WE recently developed a model-based segmentation algorithm, which we called *ARGraphs* [1]. In that work, we compared our algorithm against three different methods: adaptive h-minima [2], conditional erosion [3], and iterative voting [4]. We selected the external parameters of each comparison method as follows: we first listed a set of values for each of the method’s parameters, considered all possible combinations of the values of different parameters, and selected the combination that gave the best F-score for the training nuclei. In this technical report, we briefly explain the external parameters of the comparison methods along with the analysis of their effects on the segmentation performance. This analysis for our proposed algorithm can be found in [1].

II. ADAPTIVE H-MINIMA

The adaptive h-minima method [2] obtains a binary segmentation via active contours without edges and calculates an inner distance map that represents the distance from every foreground pixel to the background. Then, it identifies regional minima as the markers, found after applying the h-minima transform to the inverse of the map, and calculates an outer distance map representing the distance from the foreground pixels to their nearest marker. Finally, it grows the markers using the combination of the outer distance map and the grayscale image as a marking function.

This method has a single external parameter Δ , which is the gap value between shape markers in the adaptive h-minima

transform. In the experiments, it was selected as 1, considering the following set $\Delta = \{1, 2, 3, 4, 5\}$. However, when we analyze the effects of this parameter to the segmentation performance, we observe that it only slightly changes the F-scores (Fig. 1).

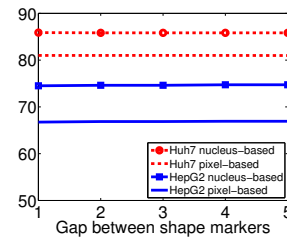


Fig. 1. For the Huh7 and HepG2 test sets, cell-based and pixel-based F-score measures as a function of the gap parameter Δ .

III. CONDITIONAL EROSION

The conditional erosion method [3] obtains a binary image by histogram thresholding and iteratively erodes its connected components by a series of two cell-like structuring elements of different sizes. It first uses the larger element until the sizes of the eroded components fall below a threshold. The component shapes are coarsely preserved due to the size of the structuring element and its round shape. It next uses the smaller element on the remaining components and stops the iterations just before the component sizes become smaller than a second threshold. Considering the eroded components as the markers, it then applies a watershed algorithm on the binary image.

This method has four external parameters: the size s_{large} of the larger structuring element, the threshold t_{large} used in the first erosion step, the size s_{small} of the smaller structuring element, and the threshold t_{small} used in the second erosion step. In the experiments, we considered the following sets for these parameters: $s_{large} = \{3, 5, 7, 9\}$, $t_{large} = \{1000, 1250, 1500, 1750\}$, $s_{small} = \{1, 2, 3, 4\}$, and $t_{small} = \{50, 100, 150, 200\}$. The selected values, which yielded the highest F-score for the training nuclei, were $s_{large} = 3$, $t_{large} = 1500$, $s_{small} = 2$ and $t_{small} = 150$. Figure 2 gives the effects of each parameter to the segmentation performance. As observed in this figure, the sizes of the structuring elements (s_{large} and s_{small}) affect the performance more than the thresholds. Moreover, the second threshold has a more significant effect than the first threshold.

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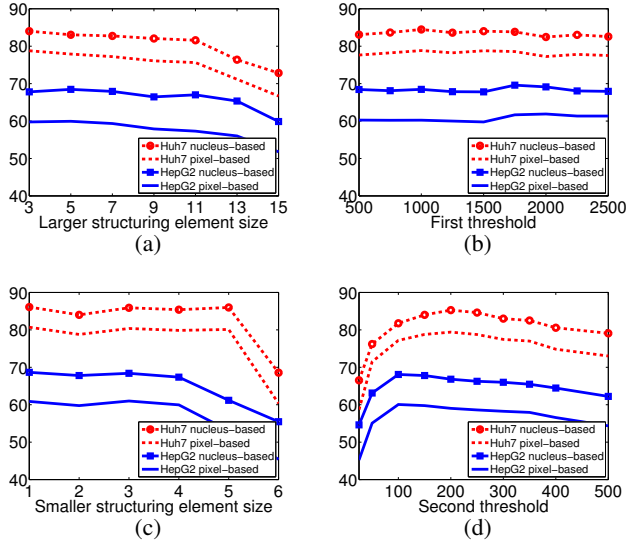


Fig. 2. For the Huh7 and HepG2 test sets, cell-based and pixel-based F-score measures as a function of the (a) larger structuring element size s_{large} , (b) threshold t_{large} used in the first erosion step, (c) smaller structuring element size s_{small} , and (d) threshold t_{small} used in the second erosion step.

IV. ITERATIVE VOTING

The iterative voting method [4] defines and uses a series of oriented kernels for localizing saliency, which corresponds to nucleus centers in a microscopic image. This study localizes the centers from incomplete boundary information by iteratively voting kernels along the radial direction. It continues iterations, in which the shape of the kernel and its orientation are refined, until convergence. It then identifies the centers by thresholding the vote image computed throughout the iterations and outputs a set of centers that can be used as the markers in a watershed algorithm. In our comparisons, we used the software provided by [4]’s authors, available at http://vision.lbl.gov/Publications/ieee_trans_ip07, to find the nucleus centers and apply a marker-controlled watershed algorithm on the binary image obtained by histogram thresholding.

This method has three external parameters: the diameter r of an object to be detected, the vote strength threshold Γ_v for selection or elimination of the detected points, and the scale σ for computing image gradients used in the voting process. In the experiments, we considered the following parameter sets: $r = \{25, 30, 35, 40\}$, $\Gamma_v = \{400, 500, 600\}$, and $\sigma = \{5, 6, 7, 8\}$. The parameters that gave the highest F-score are $r = 35$, $\Gamma_v = 500$, and $\sigma = 7$. The effects of these parameters to the segmentation performance are demonstrated in Fig. 3. As observed in this figure, the vote strength threshold Γ_v is less effective on the performance compared with the other two parameters.

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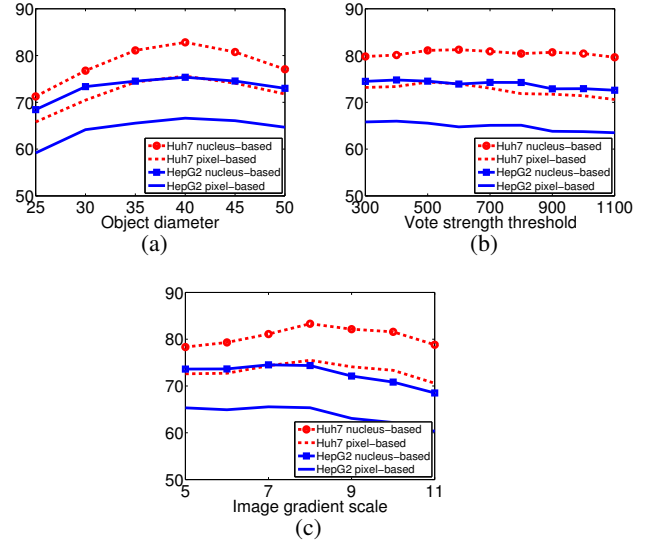


Fig. 3. For the Huh7 and HepG2 test sets, cell-based and pixel-based F-score measures as a function of the (a) object diameter r , (b) vote strength threshold Γ_v , and (c) image gradient scale σ .

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