BINARIZATION OF NOISY MICROSCOPY IMAGES THROUGH SIGNAL RECONSTRUCTION USING ITERATIVE DETECTION NETWORK

Tomáš Lukeš, Daniel Kekrt, Karel Fliegel, Miloš Klíma

Department of Radioelectronics, Faculty of Electrical Engineering
Czech Technical University in Prague, Czech Republic

ABSTRACT

We propose a novel binarization method based on a signal reconstruction using an iterative detection network. The algorithm simulates the whole image acquisition process taking into account a point spread function of the imaging system and its noise characteristics. The negative influence of image blur and noise is effectively suppressed by iterative detection network based on the criterion of maximum a posteriori probability. The proposed method was successfully applied to noisy microscopy images. Experiments show that the proposed method due to the noise suppression and deconvolution properties provides for noisy images significantly better results compared to common thresholding techniques. Binarized images obtained by the proposed method can be particularly useful for particle detection and analysis of cell samples.

Index Terms—Binarization, Microscopy Images, Image Processing, Image Reconstruction, Iterative Detection Network

1. INTRODUCTION

In many applications, thresholding that leads to a binary image is an important step of image analysis. Examples of such applications are document analysis, cellular imaging and various segmentation techniques for nondestructive testing. Successful thresholding should separate objects in the foreground from the background. Thresholding methods can be divided into two main groups, local and global methods. The global thresholding methods try to find one optimal intensity threshold value. A typical representative of such an approach is the often used Otsu’s method [1]. Local adaptive methods such as Niblack’s [2] and Sauveola’s methods [3] use different thresholds for distinct image regions. An extensive survey of image thresholding methods can be found in [4]. A large number of algorithms have been proposed for successful document image binarization [5, 6]. They were designed to handle images of documents acquired under difficult illumination conditions, some of them even with low levels of noise [7]. In the case of microscopy imaging with short acquisition times (in our experiments 53µs - 200µs) the biggest issue is not the uneven illumination but the high level of noise. With a certain level of noise, the image histogram exhibits a nearly unimodal character and the signal to noise ratio (SNR) is so low (approx. 10 – 20 dB) that common thresholding techniques fail. Additionally, thresholding techniques often rely on an operator who has to adjust the thresholding parameters. The results are then dependent on a subjective decision of a human operator. That is impractical in the case of an analytical tool which should provide the result automatically and objectively in an easily repeatable manner.

We propose a different approach that does not search for any threshold values. Our method simulates the whole image acquisition chain and evaluates probabilities of all possible values of each pixel. The iterative detection network decides for each pixel whether its value will be black or white by maximizing the corresponding a posteriori probability that the pixel contains the useful signal (object in the foreground) or not (background noise etc.). The problem can be seen as a decoding task similar to the decoding of one-dimensional communication signals where the iterative detection is broadly used [8]. Kekrt et al. applied the principle of iterative detection on two-dimensional signals [9]. The efficiency of the iterative detection has been shown on simulations with black and white motion blurred images [10]. In this work, we present an algorithm applicable to grayscale microscopy images with high level of noise.

2. THEORETICAL BACKGROUND OF THE PROPOSED METHOD

Let us suppose that \( d(i,j) \) denotes pixel values of an ideal image \( D \) of a microscopic sample. The transmission of the signal through the optical system (2D linear and space invariant system) can be described by the convolution:

\[
q(i,j) = h(i,j) * d(i,j)
\]

\[
= \sum_{0\leq m, n \leq M, N} h(m,n) d(i+m,j+n)
\]

where \( h(m,n) \) denotes the point spread function (PSF) of the optical system and \( M, N \) are the height and width of the convolution kernel, respectively. The two-dimensional PSF
of a microscope can be well approximated by Gaussian model [11]. The signal \( q(i,j) \) is corrupted by noise \( w(i,j) \) at the camera sensor, such that: \( x(i,j) = q(i,j) + w(i,j) \). It can be rewritten in a matrix form as: \( X = Q + W \).

Digital image \( x_d(i,j) \) is obtained by a quantization of the captured signal \( x(i,j) \) with a quantization step \( \Delta_d \) in the N-bit A/D converter.

**Fig. 1.** Image acquisition model used by the iterative detection network

If we assume that noise sources in the cells of the sensor are mutually independent and each value \( x(i,j) \) depends only on the corresponding \( q(i,j) \), then the whole imaging path can be considered as a memory less signal channel with independent eliminated channel states (IECS-ML channel [8]). Under these assumptions, the joint probability density function (PDF) of noise is given as a product of marginal densities:

\[
p_W(\Xi) = \prod_{l,j} p_w(\xi)
\]  

where \( \xi \) is a possible value of noise and \( \Xi \) is a matrix of all possible noise values for the whole image. The likelihood function of the acquired image \( X \) is given by:

\[
p_X(\Xi|Q) = \prod_{l,j} p_X(\xi|q(l,j))
\]  

The optimal MAP detector is based on the maximum a posteriori probability criterion:

\[
\hat{d}(i,j) = \arg\max_{\hat{d}(i,j)} \left[ \sum_{[D:\hat{d}(i,j)]} P[X,D] \right]
\]

where \( \hat{d}(i,j) \) is the estimated value of an ideal image \( D \), \( \hat{d}(i,j) \) is a hypothetical pixel value, \( \hat{D} \) is one hypothetical realization of the ideal image \( D \), \( \{D: \hat{d}(i,j)\} \) marks a set of all hypothetical image realizations which contain the particular pixel value at the \( i,j \) position. \( P[X,\hat{D}] \) denotes a joint probability that is further expressed as:

\[
P[X,\hat{D}] = \prod_{l,j} p(x(l,j)|\Omega(i,j) \in \hat{D}) \times \prod_{l,j} p(\hat{d}(l,j))
\]

where \( \Omega(i,j) \) is the convolution region. Let \( \hat{D} \) be the estimated image composed from the \( \hat{d}(i,j) \) pixels. Direct estimation of \( \hat{D} \) using the MAP criterion (4) is not possible, but (based on the IECS-ML channel assumption) the estimation problem can be decomposed from the entire image level to the single convolution region level. Therefore, the single-stage MAP detector can be substituted by an iterative detection network [8]. The iterative detection network (IDN) is formed from functional blocks which perform the soft inversion. It is a process that can be divided into two steps. Firstly, the input probabilities \( P[I(S(\ell))] \) are combined to the joint probabilities:

\[
P[N] = \prod_{S(\ell) \in N} P[I(S(\ell))]
\]

where \( \ell \) is a number of all inputs and outputs of one functional block and \( N \) is a set of all these inputs and outputs \( N = \bigcup S(\ell) \). Secondly, all joint probabilities \( P[N] \) are marginalized into the outputs of the functional block:

\[
PO[S(\ell)] = \left( \max_{N:S(\ell)} P[N] \right) / P[I(S(\ell))]
\]

\( S(\ell) \) represents the subset of the convolution region. There are many ways to divide the convolution region. This results in different types of possible topologies of the network. In our case, the network is composed from blocks with horizontal, vertical, and diagonal connections (Fig. 2).

The outputs of one functional block are further connected to the inputs of another functional block of the IDN. One iteration of the whole IDN means the activation of soft inversion in all functional blocks of the network. More about marginalization strategies within the functional block can be found in [12]. The algorithm performs a defined number of iterations of IDN and then a hard decision is made according to the following formula:

\[
\hat{d}(i,j) = \arg\max_{d(i,j)} \left[ PI[\hat{d}(i,j)] \times PO[\hat{d}(i,j)] \right]
\]

The IDN works with probability densities instead of brightness values. Therefore, there is a gateway in front of the IDN that calculates signal and noise probability densities from the input image and known characteristics of noise. The gateway then provides the input probabilities \( PI[q(i,j)] \) for the IDN.

**Fig. 2:** a) IDN using marginalization on the pixel block level with horizontal, vertical and diagonal connections.

b) Soft inversion (IDN cell at the \( i,j \) position)
3. EXPERIMENTAL RESULTS

The proposed method was examined using test images with simulated additive white Gaussian noise (AWGN) and blur. SNR of the tested images ranged from 14.3 dB to 29.4 dB. PSF is modeled by a Gaussian function with a 3x3 kernel, standard deviation $\sigma = 0.7$, and mean value $\mu = 0$. Experiments show that good results can be obtained by a small number of iterations (Fig. 3). The root-mean-square error (RMSE) falls rapidly with an increasing iteration number.

The performance of the proposed method (7th iteration) was compared to Otsu’s and Niblack’s binarization methods (Fig. 4). The proposed method reached the best results with the lowest RMSE calculated between the ground truth image and the estimated image. Fig. 5 shows the blurred and noisy test image (SNR = 14.3 dB) together with binarized images obtained by the tested methods. The advantage of the proposed method is visible on the small details (1-5 px in diameter). Common binarization methods are not able to distinguish these details from noise.

4. APPLICATIONS IN LIGHT MICROSCOPY

We used an Olympus CX41 microscope equipped with a 40x/0.65 NA Olympus objective and a conventional CCD camera (Infinity 2, Lumenera). The iterative detection network takes into account image distortions caused by the optical system. Several parameters of the acquisition model have to be set. It includes the camera parameters, the 2D
microscopy PSF, and the mean value of camera noise. The advantage is that the camera parameters are known, the PSF and noise can be measured, so there are no thresholding parameters that rely on a subjective judgment of a human operator.

The algorithm uses the following camera parameters: full well capacity (7950 electrons), read out noise (10 electrons), and bit depth (8 bits). The 2D PSF was estimated directly from the acquired image by measuring the edge spread function and fitting a Gaussian function at its first derivative. For the estimation of thermal noise, we have used five images taken under the same conditions. As a sample, we used a stage micrometer (Pyser-SGI, pattern S12, 0.1mm in 0.002mm division). Fig. 6 shows the image acquired with short exposure time (53μs) which depicts a central part of the stage micrometer. The line spacing is 2μm. The proposed method provided good results despite the high level of noise. On the contrary, common binarization methods fail under these conditions. The histogram of the acquired image in Fig. 5 has an unimodal character and there is not a single threshold value that would be suitable for successful background subtraction.

5. CONCLUSION AND FUTURE WORK

A novel binarization method has been proposed. Unlike traditional approaches, this method uses an iterative detection network and works with probabilities of possible pixel states instead of searching for a threshold value. The performance of the proposed method was evaluated and compared with Otsu's and Niblack binarization by measuring the RMSE. The proposed method offers better results in exchange for higher computational demand. However, there is a great potential to significantly reduce the complexity of the proposed algorithm. A large number of marginalizations can be omitted because their values are irrelevant due to the input probability distribution which is equal to zero over a large interval. We intend to focus our future work on the development of an adaptive table that would redefine which marginalizations do not have to be evaluated.

The advantage of the algorithm is clearly visible when it is applied to optical microscopy images with low SNR. Under these conditions, Otsu’s and Niblack’s methods fail but the proposed method provides good results by the 3rd iteration of the IDN. The drawback of the current implementation is the necessity to set up parameters of the acquisition model. Our future research effort is going to address this issue. We are developing a concept of a network synchronizer that would automatically optimize defocus parameters and simplify the calibration process. The results have proven that our proposed method is able to effectively suppress the negative influences of noise and blur and provides well binarized images even when the SNR is very low. These properties make the method suitable for applications in the field of microscopy image analysis.

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