

# Multispectral Texture characterization: Application to Mitotic Count in Breast Cancer Histopathology

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## Abstract

*Multispectral Imaging (MSI) ...*

## 1. Introduction

In medical image analysis, computer aided detection/diagnosis (CAD) systems are popular for clinical purposes (medical procedures seeking to divulge, diagnose or examine disease) and medical science (including the study of body anatomy and physiology). CAD systems are extensively used in the detection and differential diagnosis of many different types of abnormalities in medical images obtained using different imaging modalities. Medical image analysis in radiology (x-rays, ultrasound, MRI, CT and mammogram) and cytology domains have been major research fields for several decades and numerous systems [32, 10, 33, 14, 2, 16, 29] and software platforms<sup>1, 2, 3, 4, 5, 6</sup> have been developed for these domains. The application of these systems to histopathology is complicated due to radically different imaging modalities and image characteristics. In general, histology images are considerably different from radiology images, having a large amount of objects of interest (like nuclei) widely distributed in the images and surrounded by different neighbouring tissues like blood vessels, muscle, stroma, and fat. In addition, histopathology tissues are generally stained with different colors while ra-

diology images usually contain only gray intensities. Although, cytology images have some similarities to histology images but both images are taken at different magnification level. In case of histology images, cellular structure and function is studied while in histology, whole tissue having cells and architecture like gland formation are studied.

According to World Health Organization, the reference process for breast cancer prognosis is histologic grading that combine tubule formation, nuclei atypia and mitotic counts [3, 9]. This assessment of tissue sample is synthesized into a diagnosis that would help the clinician determine the best course of therapy. We found several CAD systems for tubule formation [28, 27] and nuclei atypia [6, 7, 5, 8], but only few mitotic counts [17, 18]. One of the most difficult fields in histopathology imagery is spatial analysis, more specifically automated nuclei detection and classification [12]. The objective of nuclei classification is assign label to different type of nuclei as normal, cancer, mitotic, apoptosis, lymphocytes etc that in particular, a challenging problem to address in histopathology. In addition, quantitative characterization is important not only for clinical applications (e.g., to reduce/eliminate inter- and intra-observer variation in diagnosis) but also for research applications (e.g., to understand the biological mechanisms of the disease process [15]).

In histopathology, H&E is a well established staining technique that exploits intensity of stains in the tissue images to quantify the nuclei and other structures related to cancer developments. Image processing techniques in this context are devoted to the accurate and objective quantification and localization of such activity in specific regions of the tissue such as cytoplasm, membranes and nuclei. From the chromatic viewpoint, nuclear regions are characterized

<sup>1</sup>3D Slicer, <http://slicer.org/>

<sup>2</sup>ImageJ, <http://rsb.info.nih.gov/ij/>

<sup>3</sup>Mirada Medical, <http://www.mirada-medical.com/>

<sup>4</sup>Infinite Radiology, <http://infiniteradiology.com/>

<sup>5</sup>PATHOS-WEB, <http://pathos-web.sourceforge.net/>

<sup>6</sup>Xebra (medical imaging software), <http://en.vionto.com/show/me/Xebra>

by non-uniform stain intensity and color, thus preventing a trivial classification based on color separation. In addition, the superposition of tissue layers as well as the diffusion of the dyes on the tissue surface may bring the stains to contaminate the background or other cellular regions which are different from their specific target. Recent work [19, 17, 18] show great potential for CAD of histopathological datasets for breast cancer diagnosis.

Multispectral imaging (MSI) retrieves spectrally resolved information of an image scene at specific frequencies across the electromagnetic spectrum. MSI captures images with accurate spectral content correlated with spatial information and reveals the chemical and anatomic features of histopathology [21, 23]. This is because it provides option to biologists and pathologists to see beyond the RGB image planes that they are accustomed to. Recent publications [11, 24, 34, 20] have begun to explore the use of extra information contained in such spectral data. Specifically, there have been comparisons of spectral unmixing algorithms (to separate constituent dyes) which demonstrate the advantage of multispectral data [22, 13]. The added benefit of multispectral imaging for analysis of routine H&E histopathology imagery, however, is still largely unknown, although some promising results are presented in [30, 11, 20, 34].

While nuclei segmentation and classification using MSI in histopathology is new area of research, many researchers used single band of multispectral imagery for segmentation of tissue and nuclear region [4, 35, 26] and few used all or selected bands [11, 20]. As far as we know, there is no existing study of the advantage of multispectral imaging for automation of mitotic counts in breast cancer histopathology. We proposed here to extend the already successful work of [18] to automation of mitotic counts in multispectral imaging.

The reminder of the paper is organized as follows. Section 2 reviews the state-of-the-art multispectral methods, particularly in object or region detection in histopathology, related to this research work. Section 3 describes the proposed framework for mitotic figure detection. Experiment and results are presented in section 4. Finally, the concluding remarks with future work are given in section 5.

## 2. Literature Review

The main idea of extracting texture from multispectral images is the use of combined spectral and spatial information for discrimination of region or objects. We found few methods in the MSI literature for texture characterization of histopathological images. Some of them employed single band of MSI and other used multiple selected bands of MSI. Fernandez et al. [11] coupled high-throughput Fourier transform infra-red (FTIR) spectroscopic imaging of tissue microarrays with statistical pattern recognition of spectra indicative of endogenous molecular composition

and demonstrate histopathologic characterization of prostatic tissue. They explicitly defined metrics to consist of spectral features that have a physical significance related to tissue biochemistry and facilitated the measurement of cell types. The approach in [?] used hyperspectral images of colon biopsy slides whereby the classification algorithm was based on spectral analysis to discriminate between normal and cancerous biopsies of the colon tissue. In this study of hyper-spectral cancer analysis, they used Laplacian eigenmaps to take into account the non-linear geometry in the design of learning algorithms and evaluated two approaches for spectral feature selection: Haar wavelet packet best bases (active sensing) and random projections.

Masood et al. [26] proposed a colon biopsy classification method based on spatial analysis of hyperspectral image data from colon biopsy samples. Initially, using circular local binary pattern algorithm, spatial analysis of patterns are represented by a feature vector in selected spectral band. Later, classification is achieved using subspace projection methods like principal component analysis, linear component analysis and support vector machine. Boucheron et al. [4] presented an analysis of the utility of multispectral versus standard RGB imagery for pixel level classification of nuclei in H&E stained histopathological images and found that performance differences between single multispectral image bands and single RGB image bands are not statistically significant.

Khelifi et al. [20] proposed a spatial and spectral gray level dependence method in order to extend the concept of gray level co-occurrence matrix by assuming the presence of texture joint information between spectral bands. Malon et al. [25] demonstrated a segmentation based features with convolutional neural networks using additional focal planes and spectral bands for identification of mitotic nuclei in breast cancer histopathology and achieved best classification accuracy (F-measure = 59%) on multispectral dataset during ICPR context 2013 [31].

Recently, Wu et al. [34] proposed a multilayer conditional random field model using a combination of low-level cues and high-level contextual information for nuclei separation in high dimensional data set obtained through spectral microscopy. In this approach, the multilayer contextual information was extracted by an unsupervised topic discovery process from spectral images of microscopic specimen, which efficiently helps to suppress segmentation errors caused by intensity inhomogeneity and variable chromatin texture.

In the proposed methodology, we address limitation of the shortcomings in previous works, including (1) comprehensive analysis of multispectral spatial feature vector in all bands rather than single band [26, 35, 34] and (2) combining spatial feature with multispectral spatial feature vector in order to discriminate mitotic figures from other nuclei

and microscopic objects. The main novel contributions of proposed work are: (1) a multispectral spatial and morphology feature vector computation which inherit discriminant information from other nuclei and (2) an projection features from multispectral features vector for classification of mitotic figures in breast cancer histopathological images.

### 3. Proposed Method

#### 3.1. Dataset

We evaluated the proposed methodology on multispectral MITOS dataset [1], a freely available mitosis dataset. The data set is made up of 50 high power fields (HPF) coming from five different slides scanned at 40X magnification using a 10 bands multispectral microscope. There are 10 HPFs per slide and each HPF has a size of  $512 \times 512 \mu m^2$  (that is an area of  $0.262 mm^2$ ). The spectral bands are all in the visible spectrum. In addition, for each spectral band, the digitization has been performed at 17 different focus planes (17 layers Z-stack), each plane being separated from the other by 500 nm. For one HPF, there are 170 gray scale images (10 spectral bands and 17 layers Z-stack for each spectral band). These 50 HPFs contain a total 322 mitotic cells. The training data set consists of 35 HPFs containing 226 mitotic cells and evaluation data set consists of 15 HPFs containing 98 mitotic cells [31]. Figure 1 shows the spectral coverage of each of the 10 spectral bands of the multispectral microscope.

#### 3.2. Proposed Method

In this paper, we propose a mitotic figures detection algorithm based on spatial analysis of multispectral images in breast cancer histopathology. Initially, a z-stack plane is selected based on gradient and band is selected based on histogram analysis. Then, candidates for mitotic figures are computed on selected band and z-stack plane. A multispectral feature vector is computed for all detected candidates including intensity and texture features in all bands of multispectral images. In addition, using segmented regions of detected candidates, morphological features are also computed. A feature selection algorithm is employed on this multispectral features vector to save the computation cost and to discard any redundancy in the data in order to improve classification accuracy. Classification is achieved using support vector machine (SVM), Bayesian network (BN) as well as decision tree (DT). A side advantage of performing the spatial analysis on a multiple band is to investigate whether improvement in accuracy can be achieved with carefully selected multispectral features to those methods [26, 35, 34] which use single band data.

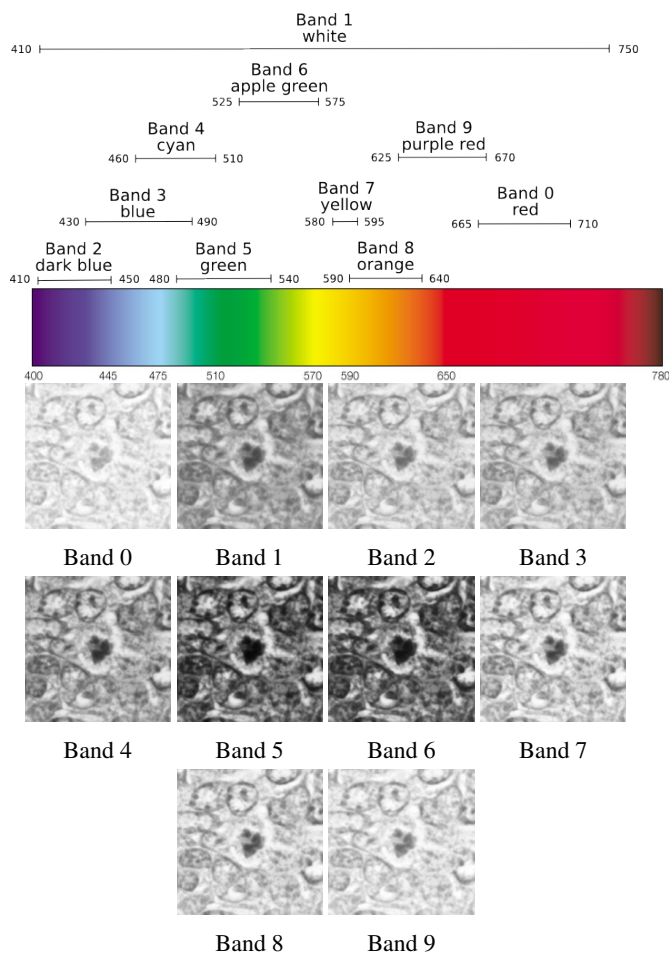


Figure 1: Spectral bands of the multispectral microscope and examples for each band.

### 4. Experiments and Results

#### 4.1. Experiments

#### 4.2. Results

### 5. Discussion and Conclusion

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