

A New Approach for Detecting Eosinophils in the Gastrointestinal Tract and Diagnosing Eosinophilic Colitis

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Abstract: *Eosinophilic Gastrointestinal Diseases (EGIDs) represent a rare group of disorders that can have various clinical presentations dependent on the involved segment within the gastrointestinal tract. Eosinophilic Colitis is considered as an under-diagnosed disease, which requires more attention and correct diagnosis. Our research aims to develop an image processing and machine learning approach that can be utilized by pathologists to diagnose patients with Eosinophilic Colitis in an easy and fast manner. The approach tends to enable pathologists to detect eosinophils in the microscopic sections of the gastrointestinal tract including; the esophagus and colon. We proposed an approach that relies on applying advanced image processing techniques on the digitally acquired images of microscopic biopsies to extract the primary features of the eosinophils and to estimate the count of the eosinophils in the given patient's slide. These counts were used as inputs to machine learning algorithms including, Support Vector Machine (SVM) and Neural Networks in order to decide whether the patient has eosinophilic colitis disease or not. The accuracy of detecting Eosinophilic Colitis using SVM classifier is 85.71%, and in neural network is 93.8%.*

Keywords: *Eosinophilic colitis, eosinophils, eosinophilic gastroenteritis, image processing, digital images, neural network, SVM.*

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1. Introduction

Within the Eosinophilic Gastrointestinal Disorders (EGIDs), Eosinophilic Gastroenteritis (EGE) represents one member that includes Eosinophilic Esophagitis (EoE), gastritis, enteritis, and colitis [1]. Through a characteristic biopsy and/or Eosinophilic ascites fluid, and in the absence of other causes of gut Eosinophilia, the EG diagnosis can be confirmed. The disease can affect patients of any age [15].

The clinical manifestations of EGE are proteinic and associated with organs, tissue layers and the intensity of Eosinophilic inflammation [15]. Some patients present with dominant gastric or duodenal disease. Dominant gastric disease often presents with nausea, vomiting and early satiety [15]. In addition to the variation in the distribution of Eosinophils along the Gastro Intestinal (GI) tract, multiple reports have indicated subtypes of EGE based on the difference in the depth of eosinophil infiltration [15]. The diagnosis is based on typical symptoms coupled with increased gastric or intestinal eosinophils. Our work aims to help pathologists to detect eosinophils in the microscopic sections of the gastrointestinal tract. Our focus is on Eosinophilic Colitis.

2. Related Work

EGIDs represent a rare group of disorders that can have various clinical presentations dependent on the involved segment within the gastrointestinal tract. Eosinophilic gastritis presents with abdominal pain or vomiting; eosinophilic colitis can present with diarrhea or bloody stools [4]. Primary eosinophilic colitis may be related to altered hypersensitivity. The true frequency is not known, probably an under diagnosed entity, which is attributed to the lack of defined histological criteria. The clinical presentation varies between mild abdominal pain, diarrhea that might be bloody, and weight loss.

Digital pathology has emerged as an innovative field of study. Through the use of digital pathology, digitized tissue slides are produced to provide high-resolution images. Additionally, Advances in computational technology and storage have allowed fast processing of large-scale pathology images [14].

Several research projects and algorithms have been developed to analyze pathology images. Some algorithms have focused on segmentation and/or classification of micro-anatomic objects, while others have focused on generating automated image classifications. Most of these analysis algorithms produced collections of features

corresponding to micro-anatomic objects such as nuclei [4].

The traditional approach to pathology images characterization is to employ algorithms to extract imaging features [9]. Features can be classified as pixel-level, object-level, or semantic-level based.

Pathology imaging informatics refer to the analytical and computational methods for handling, analyzing, and exploring histopathological images and their associated clinical data in order to achieve a medical goal [9]. In the study of McCann *et al.* [11], researchers have presented a framework for the classification of colon biopsy images into two diagnostic categories: normal or colitis. Using image-processing techniques, they could extract different features and classify images into different diagnostic categories [11].

Liu and colleagues [10] have proposed a fully automated system for colitis detection in Computed Tomography (CT) images using Regional Convolutional Neural Network (R-CNN), which includes three major steps: in the first step, category-independent region proposals were generated for each input image. In the second step, a fixed-length feature vector was computed using a large Convolutional Neural Network (CNN) for each region proposal. In the third step, a linear Support Vector Machine (SVM) was used to classify colitis. In R-CNN, high-capacity CNN and bottom-up region proposals were combined for more accurate object detection [10]. Microscopic Colitis (MC) is microscopically characterized by normal or near normal colonic mucosa. It also characterized by an increased number of intraepithelial lymphocytes (IELs) and mononuclear cell infiltration in the underlying Lamina Propria (LP) [7]. Göransson *et al.*, [7] have characterized the inflammatory cells involved in mucosal inflammation using immunohistochemistry. Computerized image analysis of the immunohistochemical staining was used to calculate areas of stained lymphocytes in the surface, crypt epithelial, and the LP [7].

Recently, deep learning methods have been widely used to develop Artificial Intelligence (AI) in almost every field [8]. Machine learning is one of the most common branches of artificial intelligence, which is considered as one of the modern methods for prediction, determination and supporting of the decision-making process [2]. Early detection of colitis is necessary to early intervention that lessens the duration and severity of the ailment. In the current clinical workflow, each image is manually examined to detect instances of colitis. Manual methods of analysis require a lot of time and efforts. In addition to the possibility of human errors and the subjective evaluation from different pathologists. Our research aims to develop an approach that helps pathologists to diagnose patients with Eosinophilic Colitis in an easy and fast manner. The approach can detect eosinophils

in the gastrointestinal tract including; the esophagus and colon.

3. Problem Statement

Early detection of colitis is necessary to early intervention that lessens the duration and severity of the disease. In the current clinical workflow, each image is manually examined to detect instances of colitis. Manual methods of analyzing are time and resource consuming. Additionally, human error may occur due to lack of concentration. This research aims to develop an algorithm that will be utilized by pathologists to diagnose patients with Eosinophilic Colitis in an easy and fast way. We also aim to allow the machine to understand and classify images into cases and controls and thus give the correct diagnosis in an objective way without the need for the subjective evaluation by different pathologists.

4. Methods and Materials

Our approach relies on applying advanced image processing techniques on the digitally acquired images of microscopic biopsies to detect Eosinophilic colitis. Each image contains a large number of erythrocytes that are similar to Eosinophils; we segmented the image to find the region of interest (Eosinophils). We used the color of the cell nucleus and the size features to identify the Eosinophils. Figure 1 presents our methodology.

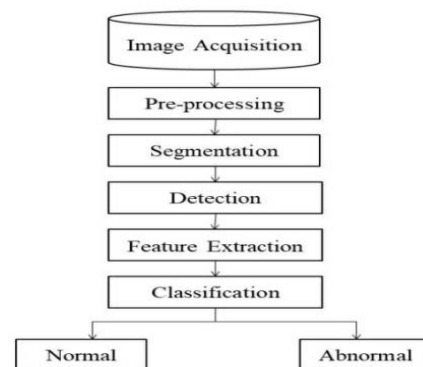


Figure 1. Proposed methodology.

4.1. Image Acquisition

The colored microscopic digital images were captured for 114 studied biopsies, which were taken in the department of pathology at King Abdullah University Hospital (KAUH), Irbid, Jordan. Images were captured using an Olympus soft imaging camera, with a resolution of 1600*1200 pixels. Images were captured at 40x magnification using an Olympus lens on Olympus microscope in order to capture the most accurate field from the tissue as possible. These images were divided into 38 groups. Each patient has three images. In our approach, the three patient images should be read. Each image is stored in a matrix so that each of them can be independently processed.

4.2. Pre-Processing

Pre-processing is used to enhance the image's accuracy and interpretation. In medical image processing, pre-processing is very important since the extracted image has impurities that make it hard for the images to be ready for the coming steps including, segmentation and feature extraction. After capturing the image, the digital image of the biopsy was preprocessed by cutting the image into four equal parts, in which each part was treated as a separate image for the purpose of enhancing the accuracy of diagnosing the disease. After that, we could adjust the image contrast using "imadjust" MATLAB function. We adjusted the contrast in the image using "stretchlim" function to set the limits.

4.3. Segmentation

Image segmentation is one of the most common and important operations of digital image processing. The goal of segmentation is to separate the image domain into dissimilar regions to extract distinctive objects in an image [3], in which each one has a consistent trait for further processing [18]. During this step, our goal was to separate the Eosinophils and distinguish them from other different components in the microscopic image. In our work, the segmentation was conducted using our proposed segmentation approach, which consists of several processes including, converting Red, Green and Blue (RGB) color space to $L^*a^*b^*$ color space, applying K-means clustering, and applying image indexing.

4.3.1. Converting RGB Color Space to $L^*a^*b^*$ Color Space

During image processing, we need to get more information about the colors inside the image. Thus, sometimes we need to convert the image from one color space to another color space. MATLAB provides a Toolbox that covers all color space transformation. In our work, we used "makecform" function to create the color transformation structure in order to perform RGB to $L^*a^*b^*$ transformation, which in turn facilitates easy quantifying of the visual differences between colors. The input for this function is an RGB image and the output is the LAB image that consists of L^* , a^* , and b^* channel L^* indicates lightness, a^* is the red/green coordinate, and b^* is the yellow/blue coordinate. $L^*a^*b^*$ is device-independent that is not sensitive to the existence of noise in the chrominance channels [12].

4.3.2. Applying K-Means Clustering

In our work, we used clustering to improve the detection efficiency [19]. Given low-quality images, the accuracy could be lower than the normal and high-quality images. Therefore, our images can be

supported using clustering that helps to increase the efficiency and accuracy of the images. K-Means is the most widely used mechanism in image processing [17]. It is considered a simple method that gives an effective result. We used K-Means algorithm since it represents a general variance based on clustering [6] and we basically rely on the color feature to detect the Eosinophils.

MATLAB toolbox contains the k-means function, which was used to cluster the images. This function gives a set of data and several k clusters. When we say $k=3$ clusters, this implies to partition the data into three clusters. For each pixel of an image, it calculates the distance between the center and each pixel of an image based on the distance function that is called "sqEuclidean". Our goal was to find a certain group of images based on the similarity in the data with the number of groups that is represented by k value. After that, three images were produced from this cluster, and then one of the most appropriate images was chosen. After that, we re-entered the resulting image from the first cluster into a second cluster with the same specifications as in the first cluster in order to obtain more accurate and clear results.

4.3.3. Image Indexing

The RGB image consists of a pixel array and a color map matrix. The color map matrix is an array of class double that contains values in the range [0, 1]. Each row of the map specifies the red, green, and blue channels of a single color. The indexed image is used to map each pixel in the array to its appropriate color map values [13]. The pixel values are direct indices into a color map. Color map also supports setting a transparent color. Coarsening transparent pixels are very helpful when it comes to images with irregular shapes. Therefore, this step is one of the most important steps in our work since it helps us to deal with and handle the colors for each pixel in the image in order to extract the required area and convert the image from 3D to 2D while maintaining the colors. In the index image, we identified the red layer in the range of [0.73-0.97] on the images of Eosinophilic Colitis, and the rest of the image was zero, which should be eliminated.

4.4. Detection

After we identified the required area by specifying the color range in the colormap, we could find that some of the parts in the image might have colors in common with eosinophils. Thus, more processes had to be performed to detect and extract the eosinophils. Our detection approach consists of the following steps; applying morphologic operations, converting RGB Images to HSV Images, multiplying images, and applying boundary detection.

4.4.1. Applying Morphological Operations

Morphological operations can be defined as simple operations that help to extract the most useful components in the image components [16], which support the representation and description of the regions based on the

shape of the image. We used the following morphological techniques:

- **Dilation:** Dilation is an operation that gradually expands the boundaries of the foreground pixels, in which holes within those regions become smaller [5, 16]. We used the MATLAB image processing toolbox function "imdilate" to perform dilation.
- **Closing:** It joins narrow breaks, fills long thin gulfs, and fills small holes from an image while maintaining the shape and size of objects in the image [16]. It tends to smooth the contours of objects. We used the MATLAB "imclose" function to perform closing.

4.4.2. Applying Boundary Detection

Medical imaging allows researchers to explore the contours and boundary that are derived from different body organs and tissues and use them in numerous applications [21].

After obtaining the image, we were interested in identifying eosinophils and drawing some boundaries around them. Using "bwboundaries" function that traces the exterior boundaries of objects, we inserted an image that contains the set of objects. After tracing the boundaries of the objects, each object was created with a matrix that contains the set of points surrounding it. Afterwards, we could measure properties of image regions using "regionprops" function that returns measurements for the set of properties specified by properties of the target component (object) in the binary image. After that, we determined the range of the required eosinophils and extracted them.

4.5. Feature Extraction

Feature extraction is considered as one of the processes that aims to reduce the amount of redundant data to perform a specific analysis and thus facilitate the learning generalization steps in the machine learning process [22]. In our approach, we selected color and texture features. Color is the first candidate feature since the human vision system is sensitive to color information. We also used the texture feature to describe the contents of the image.

We extracted all the features from the cluster image, we applied a set of 7 measures including, mean, standard deviation, entropy, Root Mean Square error (RMS), variance, kurtosis, and skewness. The computing formulas are given in Table 1. N is the number of pixels and $I_{i,n}$ is the quantized pixel value in the i th color channel. We used 114 test images to extract the features for our matrices. Our features matrix size is 21x38, in which each patient has three images. Each image has 7 features.

Table 1. Color histogram statistics.

Statistic	Formula
Mean	$\mu_i = \frac{1}{N} \sum_{n=1}^N I_{i,n}$
Standard deviation	$\sigma_i = \sqrt{\frac{1}{N} \sum_{n=1}^N (I_{i,n} - \mu_i)^2}$
Variance	$\sigma_i^2 = \frac{1}{N} \sum_{n=1}^N (I_{i,n} - \mu_i)^2$
Kurtosis	$k_i = \frac{1}{N} \sum_{n=1}^N \frac{(I_{i,n} - \mu_i)^4}{(\sigma_i)^4}$
Skewness	$s_i = \frac{1}{N} \sum_{n=1}^N \frac{(I_{i,n} - \mu_i)^3}{\sigma_i^3}$

4.6. Classification

During the classification step, we focused on the number of eosinophils in the microscopic image. As we explained earlier in this paper, each patient has three images and each image should be examined based on the threshold value that was determined by an expert pathologist (the threshold value is 20). Therefore, the image that contains more than 20 eosinophils would be classified as colitis case, while the image that contains less than 20 eosinophils would be classified as normal. We used two classification algorithms including, SVM and Neural Network.

- **SVM:** involves three phases: training, testing, and validation phase. The 114 images of our dataset were divided into 80% training examples, and 20% test examples in Eosinophilic Colitis. The training phase takes two inputs: a feature matrix that contains the values of each of the 7 features for each training example, and a label vector that contains an integer that represents the two classes, including normal and abnormal.
- **Neural Network:** ANN can learn more complex nonlinear (input-output) relations. So it is used in solving complex problems [5]. We used the Pattern Recognition tool in MATLAB R2017b (nprtool) to generate a neural network for our feature and target matrices. We used the MATLAB defaults for weights and biases and tested multiple numbers of hidden layers. 20 hidden layers gave us the best testing results. Figure 2 shows the neural network architecture where 21 inputs are introduced into 20 hidden layers, which perform processing to produce a single output that represents the class.

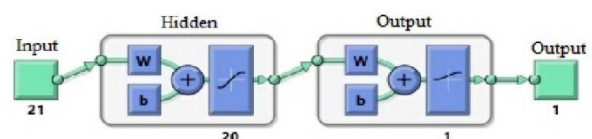


Figure 2. Our output neural network architecture.

The inputs to the neural network include the feature

matrix and the target matrix. We used nprtool to automatically separate the input matrix into the training (70%), validation (15%), and test (15%) matrices. After loading the data, a neural network with 20 hidden layers was generated using MATLAB.

5. Results and Discussion

5.1. Evaluation Metrics

The results of the classification step of machine learning is provided through a confusion matrix, where the relevant evaluation index is a performance evaluation metric that is often calculated from the confusion matrix. The matrix consists of True Positive Rate (Sensitivity, Recall), True Negative Rate (Specificity), Positive Predictive Value (PPV, Precision), and Accuracy [20].

$$\text{Accuracy} = (\text{TP} + \text{TN}) / (\text{TP} + \text{TN} + \text{FP} + \text{FN}) \quad (1)$$

$$\text{Sensitivity} = \text{TP} / \text{P} = \text{TP} / (\text{TP} + \text{FN}). \quad (2)$$

$$\text{Specificity} = \text{TN} / (\text{TN} + \text{FP}). \quad (3)$$

$$\text{Precision} = \text{TP} / (\text{TP} + \text{FP}). \quad (4)$$

Where:

- True Positive (TP): means correctly identified. Normal patient is correctly identified as Normal.
- False Positive (FP): means incorrectly identified. Normal patient is incorrectly identified as Abnormal.
- True Negative (TN): means correctly rejected. Abnormal patient is correctly identified as Abnormal.
- False Negative (FN): means incorrectly rejected. Abnormal patient is incorrectly identified as Normal.

5.2. Experimental Results

Eosinophilic Colitis (EC) is a rarely observed disease of unknown origin. The diagnosis of this disease is difficult and requires more attention, accurate diagnosis, and high index of clinical suspicion. Therefore, in our work, we studied computer-based eosinophilic colitis and produced an approach that can count eosinophils and diagnose eosinophilic colitis through microscopic images using image-processing and machine learning techniques.

The process of detecting and counting eosinophils is a stressful process that needs a lot of time. The pathologist may take around 10-15 minutes to count eosinophils under a microscope within one image. However, through our approach, we could perform the task in much lower time.

114 medical images (related to 38 patients) were tested by our approach. They were all blinded and uncategorized, which means that we don't know which images are related to Eosinophilic Colitis and which images are not related to Eosinophilic Colitis. Our approach could detect 72 images (related to 24 patients) with Eosinophilic Colitis and 42 images (related to 14 patients) without Eosinophilic Colitis.

These results were evaluated and accepted by our specialist pathologists.

Our approach is represented by several steps including, pre-processing, segmentation, detection, feature extraction, and classification. Image pre-processing techniques are important to prepare the images to be easily handled by machines. The aim of the first step was to divide the image into multiple parts to identify relevant information and features. The aim of the second step was to adjust the image intensity values to improve the image for effective diagnosis. Figure 3 shows the results of pre-processing.

Segmentation is a process that separates the images into sections with similar characteristics. We applied segmentation with K-Means method. This step depends on the color of the eosinophils that distinguishes them in the picture. Figure 4 shows the results of the two K-Means clusters. Figure 5 shows the identified eosinophils. This means that our approach was able to distinguish the eosinophils.

Once the eosinophils were detected, we could count the eosinophils in each of the four parts of the image. After that, the numbers were grouped into each part and the cumulative count of the eosinophils was identified in the image. Figure 6 shows the detection and counting of eosinophils in each part of image and finally, the cumulative counting of the eosinophils is shown.

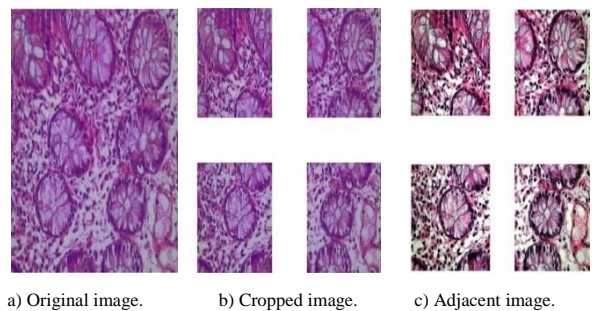


Figure 3. Pre-processing results.

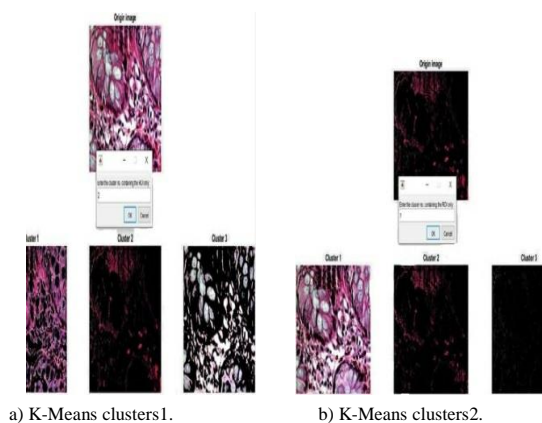


Figure 4. Segmentations results.

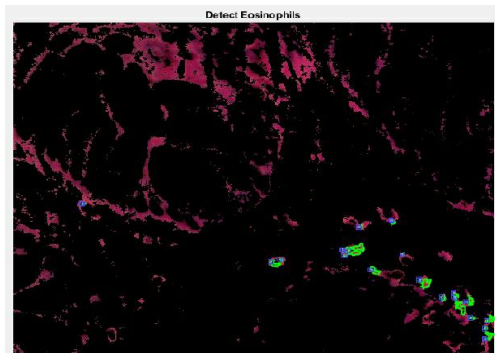


Figure 5. Eosinophils detection.

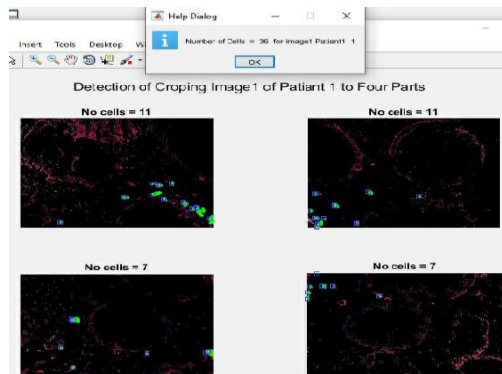


Figure 6. Eosinophils detection and counting.

Microscopic images from 38 patients with colitis were used to evaluate our approach for detecting eosinophils and diagnosing Eosinophilic Colitis. For every patient, 3 images were selected. Therefore, our dataset includes a total of 114 images. 80% of them (91 images) were randomly selected for the training set and the remaining 23 images were assigned to the testing set for the evaluation using the SVM classifier. Additionally, 70% of the total images (80 images) were randomly selected for the training set and the remaining 34 images were assigned to the testing set for the use of the neural network classifier.

The accuracy of detecting the Eosinophilic Colitis on the testing images is 85.71% using the SVM classifier. The accuracy of detecting the Eosinophilic Colitis on the testing images is 93.8% using the neural network classifier. The sensitivity/ Recall is 88.9%, which means that 88.9% of the time, our approach could predict positive classes that are positive in reality. The precision of detecting Eosinophilic Colitis on the testing images is 100%. Figure 7 shows the accuracy of our approach using four different confusion matrices.

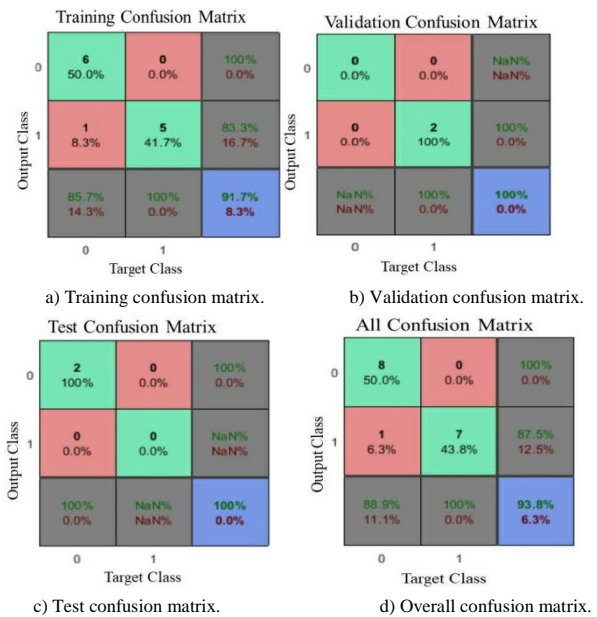


Figure 7. Confusion matrices.

The rows in Figure 7 correspond to the predicted class and the columns correspond to the true class. In our approach, class 0 denotes normal, and class 1 denotes abnormal. The red blocks show the wrong classifications, and the green blocks show the number of patients who were correctly classified. The blue block at the bottom rightmost corner shows the overall accuracy. In the training confusion matrices, the blue block shows that we achieved 91.7% accuracy in the training phase. The testing confusion matrix and the validation matrix provide accuracy of 100%. The overall confusion matrix provides 93.8% accuracy, and the sensitivity is 88.9%.

6. Conclusions

we described an approach for automatic detection of Eosinophilic Colitis disease. Our approach consists of image processing and machine learning algorithms. Both SVM and Neural Networks algorithms were applied. In the Eosinophilic Colitis detection, the SVM performance was 85.71%. Additionally, in neural network, the detection performance was 93.8%. Our approach was able to distinguish eosinophils from other parts of the image and count them. It was also able to diagnose patients with Eosinophilic Colitis disease in short time. The pathologist may take 10-15 minutes to count the eosinophils under a microscope for one image. However, through our approach, we could save a lot of time required by the pathologist. So, instead of spending 10-15 minutes to count the eosinophils in one image, our approach spends approximately 6-9 minutes to count the eosinophils in three images. Our approach can facilitate the work of the pathologists by saving their time and efforts.

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