A Comparative Evaluation of Texture Features for Semantic Segmentation of Breast Histopathological Images

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ABSTRACT
Breast histopathological image analysis helps in understanding the structure and distribution of the nucleus, thereby assisting in the detection of breast cancer. But analysis of histopathological image is challenging due to various reasons such as heterogeneity of nucleus structure, overlapping nuclei, clustered nuclei, variations in illumination, presence of noise etc. Limited availability of breast histopathological image dataset with fine annotations for detection of nucleus has restricted the analysis of histopathological images at the pixel-level. This paper presents fine annotations for nucleus segmentation of breast histopathological image datasets. Various textures such as Filter Banks, Gray Level Co-occurrence matrix and Local Binary Patterns are studied along with colour features for semantic segmentation of nuclei from histopathological images. Support Vector Machine and Multi Layer Perceptron algorithms are trained to perform pixelwise classification. The performance of the three texture features are evaluated on the two datasets and the results are presented in this paper.

INDEX TERMS
Computer-aided diagnostic, semantic segmentation, texture features, machine learning.

I. INTRODUCTION
Breast cancer is the most common malignancy found in women [1]. Early detection of cancer helps in providing better treatment and improves the survival rate. Microscopic analysis of tumour helps in the detection of malignancy. Histopathological imaging has been considered as a ‘gold standard’ in recognizing almost all sorts of cancers since it captures microscopic structures of the cells and tissues. For accurate identification of breast cancer, a biopsy accompanied by microscopic examination is an essential step. In the biopsy, a small section of tissue from the suspicious region of the body is removed, processed and stained for further evaluation. The nuclei of the tissue are expressed in dark purple and other structure in pink colour when stained with Hematoxylin and Eosin (H&E). Subsequently, pathologists perform microscopic examination of stained tissues. The manual evaluation of histopathological images is a tedious and highly time-consuming task. Hence, Computer-Aided Diagnostic (CAD) system plays an important role in evaluation of histopathological images. The primary step in analyzing histopathological image is to identify potential Region Of Interest (ROI) such as the nucleus, mitotic division, hyperchromatism etc. The identification of this potential ROI can aid in further analysis of image and detection of malignancy. However, analysing histopathological images using CAD system is a challenging task due to poor sample preparation such as overstaining, thick sections, poorly fixed tissue, complex appearance, inconsistent staining, variation in illumination, overlapping nuclei and cluster nuclei [2]–[4].

The most prominent ROI in breast histopathological images are nuclei since studying the structure of nuclei reveals the condition of the tumour. Hence, the extraction of nuclei is an important step towards classification of histopathological images as benign and malignant. Detection of nucleus from histopathological images by utilizing a CAD system involves the application of various image processing techniques such as feature extraction, segmentation, classification etc. Deep learning based CAD systems are trained in an end-to-end fashion and are dependent on availability of large datasets. Several datasets on breast histopathological images are available but annotations are provided for classification at

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the image level. There is a need for datasets with images annotated for nuclei regions. Hence, to study the histopathological images at a finer level, pixel-level annotations identifying the potential region of interest are necessary.

In the present study, fine annotations for the nucleus are created. Subsequently, pixel-level information such as texture and colour features are extracted to semantically segment the nucleus by using traditional machine learning algorithms and Artificial Neural Networks (ANN). Evaluation of the proposed methods are carried out on BreakHis [5] and KMC histopathological image dataset. The adopted methodology involves preprocessing of input images using colour normalization—this is followed by extraction of texture and colour features at the pixel-level. Subsequently, Support Vector Machines (SVM) and Multi-Layer Perceptron (MLP) classifiers are used to perform semantic segmentation of nuclei.

The contributions of the proposed work are as follows:

- Fine pixel level annotations for nuclei are created for semantic segmentation of BreakHis breast histopathological images.
- Identification of appropriate template image for colour normalization of breast histopathological images.
- Suitable pixel level texture and colour features are identified which are useful for semantic segmentation of the nucleus.
- A comparative analysis of texture and colour features showed that colour features alone is not sufficient for semantic segmentation.
- A comparison of MLP and SVM classifiers are performed in which MLP was found to give an accuracy of 83%.

This paper is organized as follows: In section II review of various studies on colour normalization and segmentation of the nucleus is explored. Section III provides the methodology of our study and section IV details the results and discussion, section V provides the comparison and performance evaluation. The paper ends with a conclusion in section VI.

II. RELATED METHODS
Segmentation of nucleus from histopathological images have received much attention in the past decade. Due to inconsistency in staining and image acquisition, histopathological images suffers from colour variation and illumination. Traditional image processing methods utilizes watershed based segmentation, thresholding, active contour based segmentation etc, for nuclei segmentation [6]–[9], [10]. Recently, deep learning methods are attracting widespread interest in nucleus detection from histopathological images as they can learn deep features [11]–[14], [15]. But these deep learning algorithms are dependent on the availability of large dataset with annotations. Gurcan et al. [16] summarized the recent state of the art CAD technology for histopathological image analysis. The authors have also emphasized on usage of standard dataset for evaluation of developed CAD systems since it helps in easier analysis and comparison [16].

In literature, many studies have reported use of publicly available histopathological image datasets [5], [17]–[21]. Some of these datasets [18], [19] etc. are histopathological images from various organs whereas BreakHis [5], MITOS-ATYP1A-14 [20] and BreCaHAD [21] are focussed on breast histopathological images. However, all these datasets provide annotations at the image level for image classification [22]. This limits the application of image processing methods and analysis of histopathological images at pixel-level.

Histopathological images take up stains inconsistently which results in colour variations in the images [23]. The presence of artifacts and errors in microscope during image acquisition results in illumination variations in histopathological images [23]. Handling these variations is difficult which leads to inappropriate results [23]. Many researchers have proposed various colour normalization techniques to handle colour variations of histopathological images [24]–[27]. In [24], the colour transformation was performed from RGB to LAB colour space. The authors of [26] utilized stain colour descriptors to identify stains and subsequently applied a relevance vector machine to locate the stains. Non-linear spline-based colour normalization method was applied to transfer the colour distribution from target image to the input image. Various deep learning methods are explored in literature to perform stain colour normalization [28], [29]. Generative Adversarial Networks (GAN) are also utilized to perform colour normalization of histopathological images [29] where the proposed network learns the stain properties and colour transformations.

From the literature, it is observed that several researchers have studied the effect of texture features for histopathological image classification [30], [31] and segmentation [32]–[34]. The texture and colour distribution of nuclei varies from one region to other regions in histopathological images [16], [33]. In [32], the authors proposed to segment the histopathological images by utilizing spatio-colour-texture graph segmentation method. Texture features such as GLCM, graph run-length matrix and Euler number were used along with linear discriminant analyzer to classify images. Doyle et al. [30] distinguished low and high grades of breast cancer by utilizing texture and nuclear structure-based features along with SVM classifier. In [33], the authors segmented histopathological images to identify and separate clustered cells. The authors extracted colour-texture features at each pixel neighborhood from the most discriminant colour space and used it to perform pixel classification. A novel colour-texture based analysis of histopathological images which captures pixel-level tissue characteristics was proposed by Sertel et al. [35].

Watershed-based segmentation algorithms are widely explored in literature for segmenting nuclei in histopathological images [7], [8], [10]. Veta et al. [7] presented a
marker-controlled watershed based technique to segment the cancerous nuclei at different scales and used different markers in H&E stained whole slide images of breast. These algorithms used handcrafted features to identify different regions using a rule-based classifier. Dundar et al. [8] reported an algorithm for automatic classification of breast microscopic images. The authors initially segmented the cell region by converting the RGB image to LAB colour space and used four component Gaussian mixture model and expectation maximization algorithm on 'A' and 'B' channels. Watershed-based segmentation algorithm was used to identify the individual cells from the segmented regions. Reported an overall accuracy of 87.9%. Kowal et al. [9] compared four different clustering algorithms for nuclei segmentation and developed two-stage segmentation approach using adaptive thresholding and clustering algorithms such as K-means, Fuzzy C-means, Competitive Neural Network and Gaussian mixture model. K-Nearest Neighbors (KNN) was used in the study and an accuracy of 86.6% were reported. Huang et al. [10] proposed a method to segment the nuclei from hepatocellular biopsy images. Nuclei were segmented by using marker-controlled watershed transform and shapes of the nuclei are extracted by using snake model. For classification, SVM based decision graph classifier was used. The classification accuracy of 94.54% were reported.

Deep learning algorithms have attracted the wide attention from scientific community as they can learn deep features and capture spatial information [11], [12], [14], [15]. In [15], CNN was used to detect touching nuclei by formulating segmentation task as a regression problem. Several works are available in the literature for detecting mitotic division from histopathological images using CNNs [14], [36]. Naylor et al. [13] developed a fully automated workflow for segmenting nuclei in histopathological images using known architectures in semantic segmentation such as PangNet, Fully Convolutional Net (FCN), DeconvNet and an Ensemble classifier. In [37], the authors proposed capsule network for image classification. But the usage of deep learning on nucleus detection and segmentation from histopathological images was limited due to the availability of large dataset with annotations.

It is observed from literature that, various colour normalization techniques are developed for preprocessing histopathological images. Both traditional and deep learning based algorithms are explored in literature for histopathological image segmentation and classification. However, to analyze the histopathological images at the pixel-level, annotations at pixel-level are necessary. It is observed that there is no publicly available breast histopathological image datasets with annotations at the pixel-level for segmentation of the potential region of interest such as the nuclei, mitosis, hyperchromatism etc. which is the first step towards the detection of malignancy. Hence, a novel dataset with annotations at the pixel-level for identifying the various potential region of interests is need of the hour.

III. SEMANTIC SEGMENTATION OF BREAST HISTOPATHOLOGICAL IMAGES

The present study is focused on detecting nucleus from histopathological images using semantic segmentation. The methodology adopted involves three steps namely preprocessing, feature extraction and segmentation of the nucleus using pixelwise classification. Initially, breast histopathological images are colour normalized. These colour normalized images are further processed to extract fine pixel level features such as texture and colour. Subsequently, machine learning algorithms are trained on these features to classify individual pixels as the nucleus and background. The details of these steps and the datasets used in the study are described in this section. The overview of the proposed system is shown in Figure 1.

A. DATA COLLECTION

In the present study two datasets are used namely 'BreakHis' and 'KMC dataset'. The details of the datasets are discussed in the below sections.
and image acquisition. Hence, a preprocessing step known as colour normalization which removes the variations in colour is necessary. The selection of template image is an important step in colour normalization. In the present study, a template image is selected such that, the nuclei are prominent and distinguishable from other tissues and regions. This template image is contrast enhanced by utilizing Contrast Limited Adaptive Histogram Equalization (CLAHE) [38]. The contrast enhancement of the template image results in darker nucleus thereby aids in detection of pale nucleus. The colour distribution of the template image is subsequently transferred to the input image by performing histogram matching.

1) CONTRAST LIMITED ADAPTIVE HISTOGRAM EQUALIZATION (CLAHE)

CLAHE [39] enhances the contrast of the image in the spatial domain. In this method, contrast enhancement is limited and hence noise amplification is reduced. The input image is divided into different non-overlapping regions called tiles. A transformation function is extracted by applying the contrast limiting step at each neighbourhood. Histogram of all these regions are calculated and a clip limit is selected based on the required contrast expansion. The histogram of all these regions is redistributed such that the peak remains within the clip limit. The transformed gray levels with uniform distribution are given by,

\[
l = (l_{\text{max}} - l_{\text{min}}) \times P(x) + l_{\text{min}}
\]

where, \( l \) represents calculated pixel value, \( l_{\text{max}} \) represents maximum pixel value, \( l_{\text{min}} \) represents minimum pixel value and \( P(x) \) represents Cumulative probability distribution.

Template image is first converted from RGB to LAB colour space. Subsequently, the template image is divided into \( 8 \times 8 \) grid. Bilinear interpolation is utilized to combine all the neighbouring tiles. CLAHE is applied to L channel of LAB colour space with a clip limit set to 3.

2) HISTOGRAM MATCHING

Histogram matching is a transformation applied to an image such that the histogram of the image matches the specified histogram. The histogram of the input image is transformed to match the histogram of the contrast enhanced template image thereby colour normalizing the input image. Cumulative distribution of the histogram of template and input image is calculated as \( C_1 \) and \( C_2 \). For every intensity level \( X_1 \), intensity level \( X_2 \) is found such that \( C_1(X_1) = C_2(X_2) \). This is applied to every pixel of the input image. The resultant images are contrast enhanced and colour normalized by this approach. The description of the preprocessing step is shown in Figure 4.

C. FEATURE EXTRACTION

The nucleus is characterised by distinct texture, colour and intensities. Hence, detection of nucleus is largely dependent on identification of these features. In the present study, texture features are extracted by utilizing Gray Level Co-occurrence...
Matrix (GLCM), Local Binary Patterns (LBP) and Filter banks. Along with these texture features, colour information is considered in RGB and LAB colour space. The details of these feature descriptors are presented in this section.

1) COLOUR FEATURES
Generally it is observed that, nucleus has different colour distribution as compared to other tissues and cells in histopathological images. This is due to the fact that, H&E stains the nucleus in dark purple colour and other tissues in light pink colour. To capture this information, colour features of each pixel in RGB and LAB colour space are extracted. From these colour features, RGB and LAB colour spaces are selected.

2) FILTER BANK
The proposed filter bank consists of Gaussian and Laplacian filters at different scales. The convolution of these filters with an image results in feature maps representing different texture features. The convolution operation is given by,

\[ F(x, y) = W * g(x, y) = \sum_{s=-a}^{a} \sum_{q=-b}^{b} W(s, q) * g(x-s, y-q) \]  

where \( W \) is the kernel, \( g \) is an image and \( F(x, y) \) is the filtered image. In the present study, a 17-dimensional filter bank is considered to extract texture features from the histopathological images. The filter bank consists of 17 filters, the details of which are provided in table 1. The histopathological images are convolved with the filter bank in order to obtain the feature maps. The same is shown in Figure 5.

\[ G(a, b) = \frac{1}{2\pi \sigma^2} e^{-\frac{a^2 + b^2}{2\sigma^2}} \]  

\[ \text{LoG}(a, b) = -\frac{1}{\pi \sigma^4} \left(1 - \frac{a^2 + b^2}{2\sigma^2}\right) \left(1 - e^{-\frac{a^2 + b^2}{2\sigma^2}}\right) \] 

where, \( a \) and \( b \) represents the spatial locations of the pixels and \( \sigma \) represents the standard deviation of the distribution.

3) GRAY LEVEL CO-OCCURRENCE MATRIX (GLCM)
GLCM is a statistical method of analysing texture features which captures the spatial relationship between pixel values. GLCM is created by counting the occurrence of pixel pairs with specific intensity values. Different statistical features are calculated based on GLCM. In the present study, statistical
features such as entropy, energy, homogeneity, contrast and correlation are calculated. Figure 6 shows the GLCM feature extraction.

The GLCM is calculated by initially converting the colour normalized image to grayscale image. Quantization is performed on this grayscale image to reduce the number of gray level intensities to 16 from 256. A symmetric GLCM is created for every pixel with a window size of $5 \times 5$ from the colour quantized image. Four co-occurrence matrices are created considering spatial relationships at 0, 45, 90 and 135 degrees. Features namely entropy, energy, homogeneity, contrast and correlation are calculated from the four co-occurrence matrices.

In Algorithm 1, $i$ and $j$ represents the index of the co-occurrence matrix, $P_{ij}$ represents an element of the co-occurrence matrices, $N$ represents the quantization level, $u$ represents the GLCM mean and $\sigma$ represents the variances.

### Algorithm 1 GLCM Texture Features

1. Convert image to grayscale
2. Perform image quantization from 256 to 16 levels
3. For every pixel select a neighborhood of $5 \times 5$ pixels do
   1. Calculate the co-occurrence matrices (0, 45, 90, 135 degrees) for the neighborhood
   2. For each co-occurrence matrix obtain features corresponding to each of GLCM using the following equation
      - **Contrast** $= \sum_{i,j=0}^{N-1} P_{ij}(i-j)^2$
      - **Energy** $= \sum_{i,j=0}^{N-1} P_{ij}^2$
      - **Entropy** $= \sum_{i,j=0}^{N-1} -\ln(P_{ij})P_{ij}$
      - **Homogeneity** $= \sum_{i,j=0}^{N-1} \frac{P_{ij}}{1+(i-j)^2}$
      - **Correlation** $= \sum_{i,j=0}^{N-1} \frac{(i-u)(j-u)}{(\sigma)^2}$
4. end for
5. end for

### D. PIXELWISE CLASSIFICATION: SVM AND MLP

Features extracted from colour normalized images are used to perform pixelwise classification. Two algorithms are utilized to perform pixelwise classification namely, Support Vector Machine and Multi-Layered Perceptron. The performance of these two classifiers is evaluated for segmentation of the nuclei. SVM is trained by using a radial basis function kernel. The parameters for SVM are determined by utilizing 10-fold cross-validation. MLP is fully connected and consists of two hidden layers with 32 neurons each. Rectified Linear Unit (ReLU) activation function is used in each layer and initial weights are assigned by using the normal distribution. Batch normalization and dropout layers are considered in second
and third layers, which help in fast convergence and avoiding over-fitting. Gradient descent is used to reduce the error and learn the model parameters. Softmax layer is utilized in the last layer to obtain the probability distribution of each pixel belonging to different classes. Binary cross-entropy is used as the loss function. The architecture of MLP is shown in Table 2.

### IV. RESULTS AND DISCUSSION

The present work is aimed at semantic segmentation of nucleus from histopathological images. The results of feature extraction using Filter bank, GLCM and LBP are shown in Figure 11, 12 and 13 respectively. SVM and MLP classifiers are used to perform the classification of pixels. The proposed method for nucleus segmentation is evaluated on BreakHis [5] and KMC dataset. SVM and MLP classifiers are trained on 70% of the data, 10% of the data is used for validation and 20% of the data is used for testing. Performance metrics such as precision, recall, F1-score, mean Intersection over Union (mIoU), Pixel Accuracy (PA) and mean Pixel Accuracy (mPA) are used to evaluate the performance of the classifier. These parameters are calculated as follows,

\[
mIoU = \frac{\sum_i x_{ii} - \sum_j x_{ij} \cdot \sum_j x_{ji}}{C \sum_i \sum_j x_{ij} + \sum_j x_{ji} - \sum_i x_{ii}} \tag{6}
\]

\[
PA = \frac{\sum_i x_{ii}}{\sum_i \sum_j x_{ij}} \tag{7}
\]

\[
mPA = \frac{1}{C} \sum_i x_{ii} \tag{8}
\]

| Layers | No. of Neurons | Type             |
|--------|----------------|------------------|
| 1      | 32             | Desnsly connected|
| 2      | -              | Batch normalization layer |
| 3      | -              | Dropout layer    |
| 4      | 32             | Desnsly connected|
| 5      | -              | Batch normalization layer |
| 6      | -              | Dropout layer    |
| 7      | 2              | Softmax layer    |
This section presents the various results obtained for colour normalization (Section IV-A), semantic segmentation of BreakHis dataset (Section IV-B) and semantic segmentation of KMC dataset (Section IV-C).

A. COLOUR NORMALIZATION

Generally, the selection of template image for colour normalization is dependent on dataset as colour distribution of images varies between datasets. There is no standard evaluation method for selection of template image, however, [41]

\[
\text{Precision} = \frac{TP}{TP + FP} \quad (9)
\]

\[
\text{Recall} = \frac{TP}{TP + FN} \quad (10)
\]

\[
F1 \text{ score} = 2 * \frac{\text{Precision} \cdot \text{Recall}}{\text{Precision} + \text{Recall}} \quad (11)
\]

where \(u_{ij}\) represents the number of pixels belonging to class \(i\) and predicted as class \(j\), \(C\) is two representing the number of classes in the present study, \(TP\), \(FP\) and \(FN\) are true positive, false positive and false negatives.

FIGURE 10. First row represents the template image and its corresponding contrast enhanced image along with its histogram. Column (a) represents the original histopathological images with variations in colour distribution and column (b) shows the corresponding histogram distribution. Column (c) shows the corresponding colour normalized image and column (d) represents the normalized histogram distribution.
suggests an approach considered for selection of the template image and the effect of good template image on colour normalization. In the present study, an experiment is carried out to study the effect of different images with varying colour distribution for colour normalization. However, the focus of the present paper is to perform semantic segmentation of the nucleus hence, the template image for colour normalization is selected such that the nuclei are distinct from background [26], [41]. A few sample template images are shown in Figure 8. It is observed that, a template image with pale colour distribution and low contrast reduces the contrast of the input image (Figure 8 (b) and (c)). This results in poor feature extraction and pixel classification. In Figure 8, (b) and (c) shows the results of colour normalization by utilizing low contrast template image. Hence the selection of template image has great influence on segmentation of nucleus as demonstrated in Figure 8. In the present study, a contrast enhanced breast histopathological image is selected as reference image in colour normalization.

The results of applying CLAHE on template image are shown in Figure 9. It is seen that the CLAHE algorithm produces a flat histogram distribution improving the contrast
FIGURE 14. (a), (b) and (c) shows few colour normalized images of BreakHis dataset. (d), (e) and (f) shows the corresponding segmentation output obtained by using filter bank and SVM classifier. Images (g), (h) and (i) shows the corresponding segmentation output obtained by using filter bank and MLP classifier.

of the image. From Figure 9 it is observed that the nuclei are distinct and have different colour distribution as compared to other tissues and cells. The colour distribution of the contrast enhanced template image is transferred to the colour distribution of the input images via histogram matching. In Figure 10, a few sample images with different colour distributions are shown along with its corresponding histograms in columns a and b. The results of applying colour normalization on these images and their corresponding histograms are shown in columns c and d of Figure 10. The resultant image of this procedure is a contrast enhanced and colour normalized image which has a flat histogram distribution as in the case of template image. These images are further processed to extract features and perform classification.

**B. EVALUATION ON BreakHis DATASET**

The training set of BreakHis dataset consists of 56 images while validation and test split consists of 8 and 16 images respectively. The weights of MLP classifier are determined by using gradient descent and binary cross-entropy loss. The various performance metrics of SVM and MLP classifier on BreakHis dataset are shown in Table 3, 4 and 7. The results obtained on BreakHis dataset are presented in this section.

1) **FILTER BANK AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION**

The semantic segmentation results of SVM and MLP classifiers by utilizing filter bank features are shown in Figure 14.

The SVM and MLP classifiers achieved an average F1-score of 0.82 and 0.83 respectively on BreakHis dataset. The SVM classifier has fewer false positives as compared to MLP classifier as shown in Figure 14. MLP classifier has higher false negatives as it fails to detect a pale nucleus. From Figure 11, it can be seen that, x and y derivatives of gaussian aids in easier detection of edges and boundaries of nucleus. It is also observed that, texture features within the regions of nucleus varies and MLP classifiers fails to model these variation as seen from Figure 14 (i).

2) **GLCM AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION**

The results of GLCM texture feature and colour features using SVM and MLP classifiers are shown in Figure 15. MLP and SVM classifier have similar F1-score of 0.82 for nucleus class (Figure 15). As compared to filter bank features, second-order GLCM features capture larger spatial information (window size of $5 \times 5$ pixels) there by capturing larger spatial relationships of pixels. The homogeneity feature of GLCM helps in detection of regions with similar texture patterns such as regions with in the nucleus. The same is shown in Figure 15.

3) **LBP AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION**

The results of LBP features for semantic segmentation of the nuclei by utilizing SVM and MLP classifiers are presented in Figure 16. It is observed that precision for nucleus class is less which is due to the presence of higher false positives. The edges of the clustered nuclei are not clearly distinguished which results in higher false positives. The LBP features fail to detect nuclei which is pale in colour since both colour and LBP features are local in nature. The SVM and MLP classifier achieved an average F1-score of 0.79 and 0.82 respectively.
FIGURE 15. (a), (b) and (c) shows few colour normalized images of BreakHis dataset. Images (d), (e) and (f) shows the corresponding segmentation output obtained by using GLCM and SVM classifier. (g), (h), (i) shows the corresponding segmentation output obtained by using GLCM and MLP classifier.

TABLE 6. Precision, Recall and F1-score of MLP classifier on KMC dataset.

| Features Utilized | Precision | Recall | F1-Score |
|-------------------|-----------|--------|----------|
| Filter Bank + Colour | 0.78 | 0.71 | 0.73 |
| GLCM + Colour | 0.79 | 0.82 | 0.80 |
| LBP + Colour | 0.78 | 0.76 | 0.77 |

TABLE 7. Performance metrics of algorithms on BreakHis [5] dataset.

| | Pixel Accuracy | Mean Pixel Accuracy | mIoU |
|-------------------|----------------|--------------------|------|
| Filter Bank + SVM | 0.85 | 0.82 | 0.70 |
| Filter Bank + MLP | 0.86 | 0.83 | 0.74 |
| GLCM + SVM | 0.85 | 0.83 | 0.70 |
| GLCM + MLP | 0.84 | 0.84 | 0.72 |
| LBP + SVM | 0.82 | 0.80 | 0.66 |
| LBP + MLP | 0.85 | 0.82 | 0.71 |
| Miško Veta et. al [7] | 0.82 | 0.80 | 0.73 |

C. EVALUATION ON KMC DATASET

The train, validation and test split of KMC dataset consists of 56, 8 and 16 images respectively. The various performance metrics of SVM and MLP classifier on KMC breast histopathological dataset are shown in Table 5, 6 and 8. The results of pixelwise classification using various texture features of KMC dataset are presented below.

1) FILTER BANK AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION

A few sample results of semantic segmentation of KMC histopathological images using filter bank and colour features are shown in Figure 17. It can be seen that prominent nuclei are detected accurately. However, the lightly stained nucleus is not detected by the SVM classifier due to variations in the colour of the pixels. The algorithm achieved an average F1-score of 0.72. The MLP algorithm along with filter bank and colour features achieved an average F1-score of 0.73. It is observed that MLP classifier can detect most prominent nuclei accurately as compared to SVM with filter bank features. SVM classifier produces higher false positives and false negatives as compared to MLP classifier on KMC histopathological image dataset.

2) GLCM AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION

The results of SVM and MLP classifier for semantic segmentation of histopathological images by utilizing GLCM and colour features are shown in Figure 18. From table 6...
it is observed that MLP algorithms along with GLCM features have an average precision and F1-score in the order of 0.79 and 0.80 respectively. From table 5 it can be observed that F1-score of SVM algorithm using GLCM features is higher than other texture features like filter bank and LBP. This is due to the reason that, GLCM is a second order feature and captures larger spatial relation ships of pixels as compared to other features. The GLCM feature which captures the correlation of each pixels with its neighbours helps in easier detection of pale nucleus. It is also observed that, contrast feature helps in easier detection of boundaries of nucleus and hence reduces false positives. The same is also observed by MLP classifier which has fewer false negatives for nucleus class as compared to SVM classifier. The average F1-score of SVM and MLP classifier for both the classes are in the order of 0.76 and 0.77 respectively.

3) LBP AND COLOUR FEATURES FOR NUCLEUS SEGMENTATION

LBP and colour features are extracted at the pixel-level to classify each pixel using SVM and MLP classifier. A few sample results of semantic segmentation using SVM and MLP classifiers using LBP and colour features are listed in Figure 19. Mean Pixel Accuracy and Pixel Accuracy and mIoU obtained for SVM and MLP using LBP features are shown in Table 8. The SVM and MLP classifier achieved an average F1-score of 0.76 and 0.77 respectively. The SVM classifier has higher recall values as compared to MLP classifier. Both the algorithms have similar F1-score for nucleus class by using LBP and colour features.

V. COMPARISON AND PERFORMANCE EVALUATION

In the present work, various texture features are analysed to perform semantic segmentation of nucleus from histopathological images. The performance of the algorithms are analysed on BreakHis [5] and KMC Breast histopathological image datasets.

The filter bank and GLCM features capture larger spatial information when compared to LBP features. Due to this reason, filter bank and GLCM features perform better in segmenting nucleus which has non-uniform texture and colour distribution on both the datasets. Since filter banks considered in the present study extracts texture features in L (derivative of gaussians and Laplacian of gaussians) space of the LAB colour channel, it is capable of handling illumination variations (Figure 17, (a) and (c)). It is also observed from Figure 18 (c), the GLCM texture features fails to handle border pixels of darkly stained and clustered nucleus due to poor contrast of these regions. However, the LBP feature is a local feature descriptor and hence models local texture patterns. Hence, SVM and MLP classifier trained on LBP and colour features are not able model the texture patterns with in
the nucleus and results in large false negatives. An experiment is carried out in order to analyze the effect of colour feature alone on semantic segmentation of the nucleus. Both SVM and MLP classifiers were trained to classify individual pixels based on R,G,B and L,A,B colour features alone. These colour features are dependent on pixel intensity values and are local to individual pixels. It is observed that these colour features fails to detect the nucleus from histopathological images as shown in Figure 20. Due to the variations in colour and texture features of the nucleus, colour features alone is insufficient for semantic segmentation of the nucleus.

The SVM is trained on predefined RBF kernel limits the SVM to model the texture and colour features. MLP classifier which can model nonlinear hyperplanes outperforms SVM classifier on both datasets for pixelwise classification of the nuclei. MLP classifier along with GLCM texture features has pixel accuracy of 83% on KMC histopathological dataset which is followed by MLP (LBP features) and SVM (GLCM

![Figure 19](image1.png)  
**FIGURE 19.** (a), (b) and (c) shows few original colour normalized images of KMC dataset. (d), (e) and (f) shows the corresponding segmentation output obtained by using LBP and SVM classifier. (g), (h) and (i) shows the corresponding segmentation output obtained by using LBP and MLP classifier.

![Figure 20](image2.png)  
**FIGURE 20.** Results of pixelwise classification of histopathological images by using colour features alone. Figure (a) shows the original images, Figure (b) and (c) shows the semantic segmentation results of SVM and MLP classifier respectively.

![Figure 21](image3.png)  
**FIGURE 21.** ROC curves of MLP and SVM algorithm along with various features.

![Figure 22](image4.png)  
**FIGURE 22.** Precision-recall curves of MLP and SVM algorithm along with various features.
features) with pixel accuracy of 82%. The Receiver Operating Characteristics (ROC) curve of these algorithms are plotted in Figure 21. It is observed that, area under the curve of MLP classifier with filter bank features is largest followed by MLP classifier with GLCM and LBP features. Precision-recall curve for SVM and MLP classifier using various texture features are shown in Figure 22. It is observed that, area under the curve for MLP classifiers is larger than the SVM classifiers. This shows that, the MLP classifiers performs better in segmenting the nucleus as compared to SVM classifier. Figure 23 shows the comparison of mIoU of MLP and SVM classifiers on BreakHis and KMC histopathological image dataset by utilizing various texture and colour features. From Figure 23 it can be seen that, MLP classifier achieves higher mIoU on both the datasets.

In [7], the authors proposed a watershed-based segmentation method to segment the histopathological images. The identified regions are subsequently classified as a nucleus by using a rule-based approach with out the usage of learning algorithm. This approach achieved a pixel accuracy of 0.74 and mPA of 0.71 on KMC dataset. However, in the present paper, the proposed methodologies decision to classify the pixels are learnt by the learning algorithm during the training procedure. It is seen that the learning algorithm can make reasonably good predictions based on texture and colour features.

VI. CONCLUSION

Malignancy of tumors in the breast is primarily decided by studying the structure and distribution of the nucleus in breast histopathological images. However, developing a semantic segmentation algorithm for nucleus segmentation in breast histopathological images is challenging due to lack of finely-annotated datasets. Hence, this paper presents fine annotations for nucleus segmentation from publicly available BreakHis image dataset. The texture features for breast histopathological images are studied at the region level which fails to capture variations at the pixel level. This paper analyzes various texture features of breast histopathological images at the pixel level to segment the nucleus. The input histopathological images are preprocessed by performing contrast enhancement and colour normalization to eliminate the variations in illumination and colour. GLCM, filter bank and LBP texture features are extracted along with various colour features in RGB and LAB colour space from breast histopathological images at pixel-level. SVM and MLP classifiers are trained on these features to classify each pixels as nucleus and background. These classifiers are selected since they are independent of the large annotated dataset for semantic segmentation. The performance of these two classifiers and texture features for segmentation of nucleus are evaluated on two datasets namely BreakHis and KMC breast histopathological image dataset.

From the study it is observed that, selection of template image for colour normalization helps in segmentation of nucleus from histopathological images since low contrast template image results in degradation of input image and loss of texture and colour features. It is also seen that contrast enhancement plays an important role in easier detection of the nucleus by highlighting the features of the nucleus. GLCM texture features along with RGB and LAB colour features out performs other texture features like filter bank and LBP in detection of nucleus from breast histopathological images. The average mIoU produced by MLP classifier is greater than SVM classifier on both the datasets since it can learn more complex hyperplanes. However, the performance of these algorithms can be improved by using edge and gradient features along with texture and colour features.

The fine pixel level annotations presented in this paper provides a platform for developing new nucleus semantic segmentation algorithms. This study also gives an insight into the effect of contrast enhancement of template image on colour normalization. The semantic segmentation of the nucleus achieved by the proposed algorithm can be further utilized to study the structure and distribution of the nucleus. This can aid in developing a decision support system for classifying histopathological images as benign and malignant. In the future, CNN based algorithms can also be explored for feature extraction and image classification provided that a reasonable number of images with annotations are available.

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