Semantic Segmentation of Anaemic RBCs Using Multilevel Deep Convolutional Encoder-Decoder Network

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ABSTRACT Pixel-level analysis of blood images plays a pivotal role in diagnosing blood-related diseases, especially Anaemia. These analyses mainly rely on an accurate diagnosis of morphological deformities like shape, size, and precise pixel counting. In traditional segmentation approaches, instance or object-based approaches have been adopted that are not feasible for pixel-level analysis. The convolutional neural network (CNN) model required a large dataset with detailed pixel-level information for the semantic segmentation of red blood cells in the deep learning domain. In current research work, we address these problems by proposing a multi-level deep convolutional encoder-decoder network along with two state-of-the-art healthy and Anaemic-RBC datasets. The proposed multi-level CNN model preserved pixel-level semantic information extracted in one layer and then passed to the next layer to choose relevant features. This phenomenon helps to precise pixel-level counting of healthy and anaemic-RBC elements along with morphological analysis. For experimental purposes, we proposed two state-of-the-art RBC datasets, i.e., Healthy-RBCs and Anaemic-RBCs dataset. Each dataset contains 1000 images, ground truth masks, relevant, complete blood count (CBC), and morphology reports for performance evaluation. The proposed model results were evaluated using crossmatch analysis with ground truth mask by finding IoU, individual training, validation, testing accuracies, and global accuracies using a 05-fold training procedure. This model got training, validation, and testing accuracies as 0.9856, 0.9760, and 0.9720 on the Healthy-RBC dataset and 0.9736, 0.9696, and 0.9591 on an Anaemic-RBC dataset. The IoU and BFScore of the proposed model were 0.9311, 0.9138, and 0.9032, 0.8978 on healthy and anaemic datasets, respectively.

INDEX TERMS CNN, deep learning, blood cells, segmentation analysis, semantic segmentation, multi-level deep convolutional encoder-decoder network.

I. INTRODUCTION

With the quick expansion of CNN architecture, deep learning has attained a great milestone in computer vision. The development of AlexNet [1] architecture brings the convolutional neural network (CNN) to a dominant position within the domain of computer vision. After that, several neural networks based on CNNs were developed one after another. Out of them, the most important ones are ResNet [2], DenseNet [3], GoogLeNet [4], CliqueNet [5], VGG-Net [6], ResNeXt [7], Xception [8] Inception-V4 [9] and NasNet [10] etc. all these outperform several times during ImageNet

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competition. Due to the deep internal framework of these models, CNNs have the upper hand concerning accuracy, performance and are sometimes more efficient than human observation in some areas. Recently, CNNs have been widely used in the three core domains of digital image processing, i.e., object detection, image segmentation (Semantic Segmentation), and classification [11]–[13]. Image segmentation refers to identifying the region of interest (ROI) within an image. With promising results, various traditional segmentation methods [14]–[20] have been used in the medical image domain, especially for blood cell segmentation. But they failed to analyze the blood image at pixel level and ignored the semantics of ROI, especially in Anaemic red blood cells (RBCs.)

Anaemia is a condition in which a significant change in shape, size, and count of RBCs mass occurs [21]. Several techniques have been published to identify Anaemia in RBCs but ignore the pixel-level deep machine learning analysis. However, diagnosing blood-related disease greatly assisted with cytogenetic, molecular, and immunological tests. But, physical and quantitative descriptors like morphological, texture, and color play a crucial role in the diagnostic purpose of several morphological blood-related diseases like anaemia [22]. Recent research in semantic segmentation ignores the blood cell analysis, especially segmentation and pixel-level classification of abnormal RBC morphology due to Anaemia [39]–[45]. Red blood cells mostly show relatively the same geometry for shape and size along with overlapped structure. Most of the prior approaches do not consider the overlapped structure of RBCs and perform segmentation only on a single cell at the region level, not pixel-level. Segmentation of blood cells considering overlapping structures is still challenging due to the ambiguous, complex, and homologous assembly of cells. Current research also ignores the ground truth for crossmatching analysis and evaluation of their architecture. Precise identification of pixel-level information regarding the shape and size of infected red blood cells to normal is still challenging. Detection and treatment of blood-related disease at an earlier stage may overcome the worst results of the condition. Several recent research works [42], [46]–[48] used U-Net, Seg-Net, and FCN-AlexNet for blood cell segmentation. However, the issues regarding the prediction of change in morphological features of RBCs, like changes in shape and size of infected cells, remain to be solved.

For pixel-level segmentation, semantic segmentation gives outstanding results along with preserving semantic information. The fundamental concept of semantic segmentation is assigning distinct labels to each pixel of an image which is a challenging task in computer vision [14]. Due to the provision of pixel-level segmentation and classification, several real-world application got benefits from this technique i.e. video object segmentation [15]–[17], pedestrian detection [18]–[20], [23], [24], defect detection[25], therapy planning [26], [27], medical image diagnosis [28]–[30], computer-aided diagnosis [31], [32] and other computer vision fields [33]–[36]. The pixel-level semantic information assists the intelligent systems in grasping spatial positions for making important decisions. With the rapid advent of deep learning approaches, semantic segmentation primarily depended on deep learning. Concerning the characteristics and different processing granularity, deep learning-based semantic segmentation is divided into two categories, i.e., 1) Semantic segmentation based on pixel classification and 2) Regional classification. Semantic segmentation based on pixel classification extracts semantic information and features from many labeled images. Using this information, it labeled and classified each pixel during end-to-end training to achieve semantic segmentation.

In contrast, semantic segmentation based on regional classification clubs the classifier with traditional image processing approaches for segmentation and classification purpose. Traditional methods segment the ROI within an image, and then semantic classification is carried out using a classifier. Several semantic segmentation approaches use CNNs for the assignment of distinct labels to each pixel [5], [9], [37], [38].

This research proposed a multi-level deep convolutional encoder-decoder (DCED-Net) network for semantic segmentation of blood cells to overcome the above problems. We also developed two state-of-the-art blood cell datasets that will help the research community for the testing of semantic segmentation algorithms along with ground truth for crossmatch analysis. For experimental purposes, we have collected a total of 2000 annotated blood images from Shaukat Khanum Memorial Cancer Hospital and Research Centre Lahore (SKMCH&RC) under the supervision of an expert pathologist. This dataset comprises 1000 normal and 1000 anaemic RBCs images along with CBC and morphology reports. Each image is further divided into manually generated ground truth under the supervision of a pathologist for the authentication of segmented results.

The key contribution of this research work is as follows:

1. First, we developed a multi-level deep convolutional encoder-decoder (DCED-Net) network for semantic segmentation with forward and reversed connections between two consecutive levels to exchange semantic information.
2. We find the semantic segmentation of normal and anaemic RBCs with accurate global and class-wise pixel accuracy that helps to diagnose disease.
3. Development of two state-of-the-art Anaemic RBCs datasets comprises 2000 images, including CBC and morphology reports of each image.
4. This dataset is also equipped with 2000 manually generated ground truth for each image that authenticates the performance of the proposed architecture using crossmatch analysis.

The rest of the paper is organized as follows: Section II comprises related work. Section III describes the proposed methodology and data collection/preparation of the current work. Experimental work, results and discussion, conclusion and future work are described in sections IV-VII.

II. RELATED WORK

Automated blood cell segmentation plays a vital role in quantitative analysis in biomedical image processing [40]. Cell segmentation approaches are divided into supervised and unsupervised categories. Un-supervised segmentation includes classical approaches like watershed transformation [49], region growing, gradient flow [50], semi-automated methods [51], etc. While fully convolutional networks (FCN) and CNN have been used in supervised segmentation methods. In [40], authors developed a deep semantic segmentation-based architecture for the classification of RBCs. They

enhanced the classic U-net architecture by adding deformable convolutional layers to correct sickle cell disease's red blood cells classification. But this model needs more work for the correct identification of cell counts in overlapped areas. Another work has been done by [26] using a sliding window approach for aggregate labelling of each pixel in the blood cell. But this work reduces the resolution of segmentation. The authors [52] used an online region-based segmentation method to detect cancerous red blood cells in the BCCD dataset. This study helps the radiologist for the detection of cancerous cells but is not able to analyze them at the pixel level. For the maximization of semantic information, [38] introduces a loop-based c with an encoder-decoder structure. They created a loop between two layers by adding reverse and forward connections. Each layer works in both directions, i.e., forward and reverse direction. Due to the clique block structure, the calculation cost increases as the number of layers increases. The total depth of the network was 194 layers with encoder (82 layers) and decoder (112 layers). This network outperforms at CamVid and Cityscapes. Dynamic attention networks have been developed by [48] for semantic segmentation of general-purpose images using PASCAL VOC 2012 [52] and Cityscapes [53] datasets. In [48] author developed a network with two modules: Deformable attention pyramid (DAP) and Fusing Attention Interim (FAI) modules to perform self-adjustable descriptions of high-level output and back-propagation guidance of information. Semantic features of images were extracted by using Res-Net 101 [2] and DAP. Fusing Attention Interim used global average pooling and feature fusion into the block to build the smooth connection between decoder and encoder.

Another semantic segmentation network has been designed by [54] to recognize the complex Chinese character. They combined three modules, i.e., FCN-ResNet50, Wubi-like label coding, and CRF, to develop LCSegNet. For pixel-level classification, FCN-ResNet50 [55] was used, Wubi-like label coding was used to convert Chinese characters into 140-bits labels. In contrast, the CRF module overcame invalid code labeling for the transformation matrix among different groups. ResUNet [56] framework comprises Dice loss function along with hybrid UNet encoder-decoder architecture. The author used the dice loss function and pyramid scene parsing pooling, residual connection, multi-tasking inference, and atrous convolution to account for the precise semantics of remotely sensed data. The experiment was performed on the ISPRS 2D Postsdam dataset. Selection loss and attention loss were used by [57] with noisy annotation for semantic segmentation of the PASCAL VOC 2012 dataset. This architecture works in three steps: First, it generates initial proxy annotation. In the second step, the mask coring strategy was used for the selection of high-quality annotation. The Selection and Attention (SAL) network was applied to retrain the segmentation network in the last step. For the quantitative measurement of edge segmentation, [58] proposed a region-based evaluation index architecture. This work aimed to enhance the segmentation at the region of interest (ROI) edges

within an image by using two losses, i.e., los Lce and loss Lhp. Task decomposition technique has been used by [59] for semantic segmentation of Robotics scene, Brain tumour, and Retinal fundus images. To reduce the distance between the results of pixel-level semantic segmentation and the instance class prediction tasks, they used a novel sync-regularization multi-step approach. The substructure in the satellite image dataset was addressed by [62] using a cross-channel structure and large kernel size. To boosts the image features, they used the GAN network with a feature attention mechanism. They got mIOU with 88.15% at PASCAL VOC 2012 dataset. Semantic segmentation was also used as transfer learning [60] between various deep learning classifiers to identify different types of crops. The author used a shortened version of the SegNet framework, known as SegNet-Basic. A hybrid version of SegNet known as Random-Walk-SegNet (RWSNet) was developed by [61] for enhancing blurry object boundaries in remote sensing images. Undirected graph, gradient, probability map of SegNet, and random walk algorithm were used with basic SegNet to develop the RWSNet framework. Several recent research works [42], [46]–[48] used U-Net, Seg-Net, and FCN-AlexNet for blood cell segmentation. However, the prediction of percentage change in morphological blood cell features like change in shape and size of infected cells remains to be solved.

Previously developed blood cell datasets like ALL-IDB-I, ALL-IDB-II [65], extended ALL-IDB [66], BCCD, IUMS-IDB [67], SMC-IDB [68], BS_DB3 [69], Ash bank, BBBC [70] dataset equipped with small no of images with little RBCs elements as shown in Table 2. ALL-ID-I contains only 108 images with just 39000 blood elements. All 108 images have no masks of authentication of proposed results. Other datasets also lack detailed information regarding manual ground truth for authentication purposes and blood cells elements. SMC-IDB contains only 367 WBC-type images without any ground truth. IUMS-IDB and malarial datasets comprise 196 (WBCs) and 848 (RBCs) images, respectively. Approximately all datasets have no ground truth for the authentication of proposed techniques except extended ALL-IDB. No dataset targets the morphological changes in RBCs due to anaemia disease.

III. METHODOLOGY

The workflow chart of the proposed semantic segmentation architecture is shown in Figure 1. This architecture is mainly divided into the following steps:

- Image data collection and Pre-processing
- Network Architecture
- Segmentation

A. IMAGE DATA COLLECTION AND PRE-PROCESSING

The proposed work randomly selected a total of 100 participants for blood image collection. Out of which, 50 were healthy, and 50 were anaemic patients. Data collection was carried out under the supervision of an expert pathologist from Shaukat Khanum Memorial Cancer Hospital and
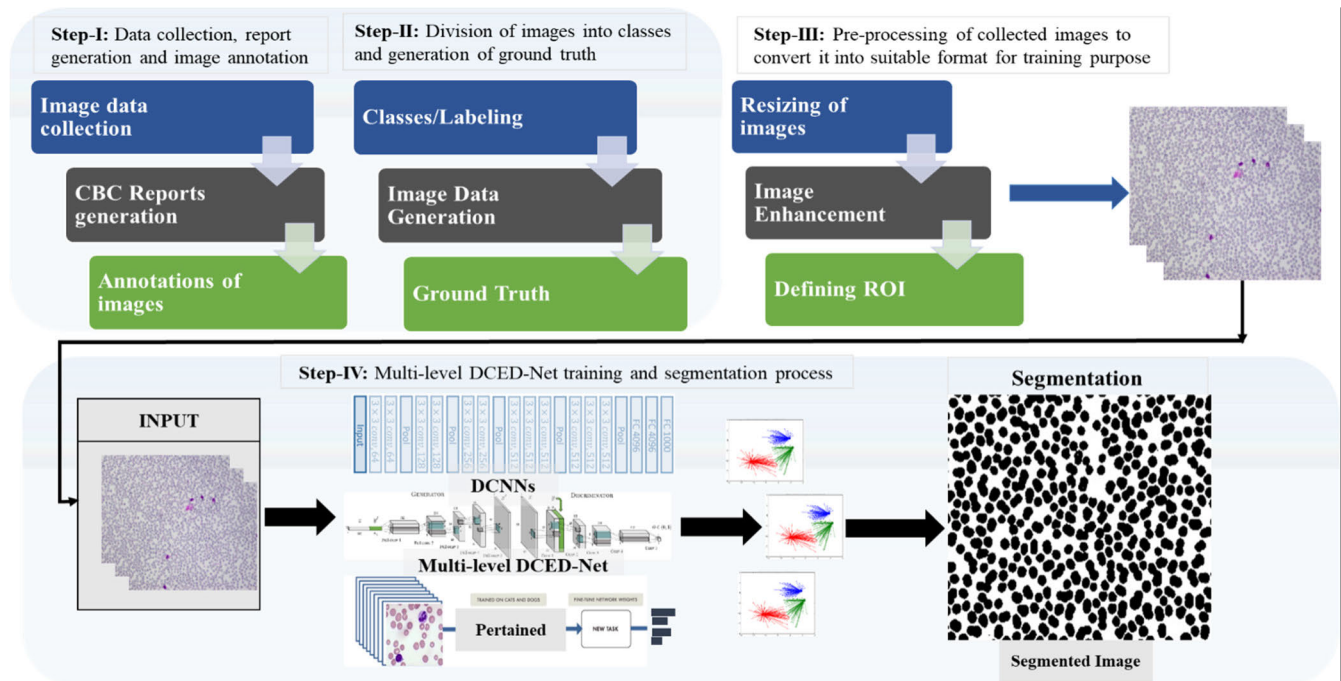
FIGURE 1. Flow chart of multilevel deep convolutional encoder-decoder network (DCED-Net). Step-I: The first step included blood image collection from Shaukat khanum hospital, Lahore, CBC and morphology reports generation of each image and image annotation abut pathologist decision. Step-II: This step involves dividing these images into classes, i.e., healthy or anaemic image and ground truth generation of each image for evaluation purposes. Step-III: In a pre-processing step, image resizing, enhancement and definition of region of interest (ROI) are carried out. Step-IV: In this step, training of DCED-Net is carried out that produces a segmented image as output.

Research Centre (SKMCH&RC) Pakistan. Olympus Dp27 8.9-megapixel CMOS sensor captured RBC images with a 4K resolution of microscopic digital camera. Twenty blood smear samples were obtained from each of the 100 participants. Using these samples, we have prepared 2,000 blood smear slides from the routine workload of the hospital pathology lab. A total of 2,000 digital images (490 × 480 pixels resolution) were collected along with relevant CBC and morphology reports of each image. The ground truth of each image was also prepared under the supervision of an expert pathologist to authenticate generated output. After collecting the RBC dataset, all the images were categorized into two main groups based on their cell type: Group I: Healthy RBCs image and Group II: Anaemic RBCs.

The quality of blood images and cell morphology regarding healthy and infected cells was critically examined by a hematologist before the image processing. This critical examination helps us for the selection of anaemic and healthy images. Each image contains approximately 83 RBCs elements. Therefore, our dataset comprises 4000 images (2000 original, 2000 ground truth) and about 1,992,000 RBC elements. These 1,992,000 RBC elements include normal microcytes, macrocytes, elliptocytes, and target RBCs. Samples images from healthy and anaemic RBCs are shown in Figure 2. The pre-processing step involves three primary activities, i.e., first: grayscale conversion of RGB images, second: pixel fusing, and then unity mask generation. Contrast normalization was used to reduce noisy, blurry patterns to produce sharp

edges with high contrast images. Resizing of each image is also done according to the underlying model, i.e., 320 × 320 × 3. Pre-processing phase comprises the following steps, also shown in Figure 3:

- Algorithm read both type of images (original and fine-tuned manual generated masks) from memory.
- All the images were checked for three channels of color, and pixel labelling fusing was also initiated in this step.
- Wiener and Laplace's filters were applied in spatial domains to separate the region of interest (ROI) from the background.
- A pixel ID, PID = 0, was assigned to all pixels of ROI to generate the unity mask, and PID = 1 was given to the image background.
- All the pixels of the RBC element and background are pointed out individually.
- After that, segmented ROI (RBC element) was generated with pixel value zero.
- After that, each image was resized into 320 × 320 × 3 resolution and passed to the model for training.

B. NETWORK ARCHITECTURE
Clique Net inspires a Multi-level deep convolutional encoder-decoder network's design. DCED-Network is arranged to maximize the transmission of semantic information and best optimum features selection from one level to another. Each

level of DCED-Net comprises the encoding and decoding phase.

1) ENCODING

During pre-processing step, all the images are resized into $320 \times 320 \times 3$ resolution. These $320 \times 320 \times 3$ images are given as input to the encoding block of the DCED-Net for extracting features to encode them into optimum features. The encoding block comprises five pools of layers. Each pool comprised convolution layers with stride size 3, batch normalization, and average pooling layers. The internal operation of each layer's pool is shown in Figure 4. After convolution operation, the output is concatenated with the original input and given to the next pool during encoding and decoding phases. The dropout was added only on the last two pools to reduce and prevent the selection of irrelevant features. The first pool receives input images with size $320 \times 320 \times 3$ and reduces them into $160 \times 160 \times 32$ by applying 3×3 convolutions and 2×2 average pooling, as shown in Figure 5. On the 2nd and 3rd pools, the input image reduces upto $40 \times 40 \times 128$. On the 4th and 5th pools, additional features and attributes are extracted from the image to reduce them into $10 \times 10 \times 512$. At the end of the encoding block, the input image obtained with $10 \times 10 \times 512$ size contains the best optimum feature for decoding purposes. The encoder performs a downsampling operation, as shown in Figure 5.

2) DECODING

The decoding block performs transpose convolution (TC) on the output of the encoding block to generate a segmented image from $10 \times 10 \times 512$ to $320 \times 320 \times 3$. It consists of 5 pools of layers. Each pool consists of the same layer as the encoding block. Each pool in the decoding block is connected with the same pool in the encoding block using skip connection to preserve the semantics of the image, as shown in figure 4. A dropout of 0.2 was applied on the initial two pools to maintain the original texture and features of an image and reduce overfitting. Every decoding pool received two inputs: 1) One from the previous pool and 2) The second is from the corresponding same-sized encoding pool to improve the feature selection and texture preservation. The main operation of the decoding block is the upsampling of a low-resolution image to a high-resolution segmented image. The decoding block retrieves the high-resolution image by applying 3×3 transpose convolution with max pooling. Each level in DCED-Net receives input in image format and produces output in the segmented image. So the input from one level to the other is the image and also ground truth.

C. SEGMENTATION

Segmentation is the most challenging and critical task for digital blood image analysis. This is because of the complex and crowded structure of the blood cell elements. Accurate segmentation greatly assists in subsequent levels of classification. For accurate and efficient semantic segmentation of anaemic RBCs, we have used a DCED-Net along with

pre-processing steps. Here is the pseudo-code for our DCED-Net model and evaluation matrices used for performance measures.

```
BEGIN
# Two types of data will be loaded i.e. Anaemic Red Blood
Cells, ii. Healthy Red Blood Cells
def Load_Data:
    For each Image in AnemicImageSet:
        AnemicRBC[Image] = List of AnemicRBCImages
        AnemicLabelCount = AnemicLabelCount + 1
    Next
    For each Image in HealthyImageSet:
        HealthyRBC[Image] = List of HealthyRBCImages
        HealthyLabelCount = HealthyLabelCount + 1
    Next
    If data_loaded_successful
        Return True
    Else
        Return False
Return data_loading_status
# Pre-Processing Data Sets
While ImageCount <= AnemicLabelCount
    If(Typeof(AnemicRBC[Image])="RGB" than
        AnemicRBC[Image]) = ConvtoGrayScaleImg
        (AnemicRBC[Image])
    Else If(PixelLabelingMaskFusion == True)than
        PixelValue = 0
        Generate UnityMask
    End If
End While
While ImageCount <= HealthyLabelCount
    If (Typeof (AnemicRBC[Image]) = "RGB" than
        AnemicRBC[Image])= ConvtoGreyScaleImg
        (AnemicRBC[Image]))
    Else If (PixelLabelingMaskFusion == True) than
        PixelValue = 0
        Generate UnityMask
    End If
End While
# Generate Image with Segmented ROI having Zero-Pixel
Status
While ImagePixel(AnemicRBC[Image]) = 0 OR
ImagePixel(HealthyRBC[Image]) = 0
    If Background = 1 and RBCElemnet = 0 than
        Set MaskPixelID = UNIQUEPIXEL(RBC, Background)
        Resize MaskedImageWidth = 320
        Resize MaskedImageHeight = 320
        Set OriginalPixelID = UNIQUEPIXEL(RBC, Background)
        Resize OriginalImageWidth = 320
        Resize OriginalImageHeight = 320
    End If
    SaveFile "MaskedDataSet"
    SaveFile "OriginalDataSet"
End While
# FEATURES EXTRACTION
While Not All Data Loaded
```
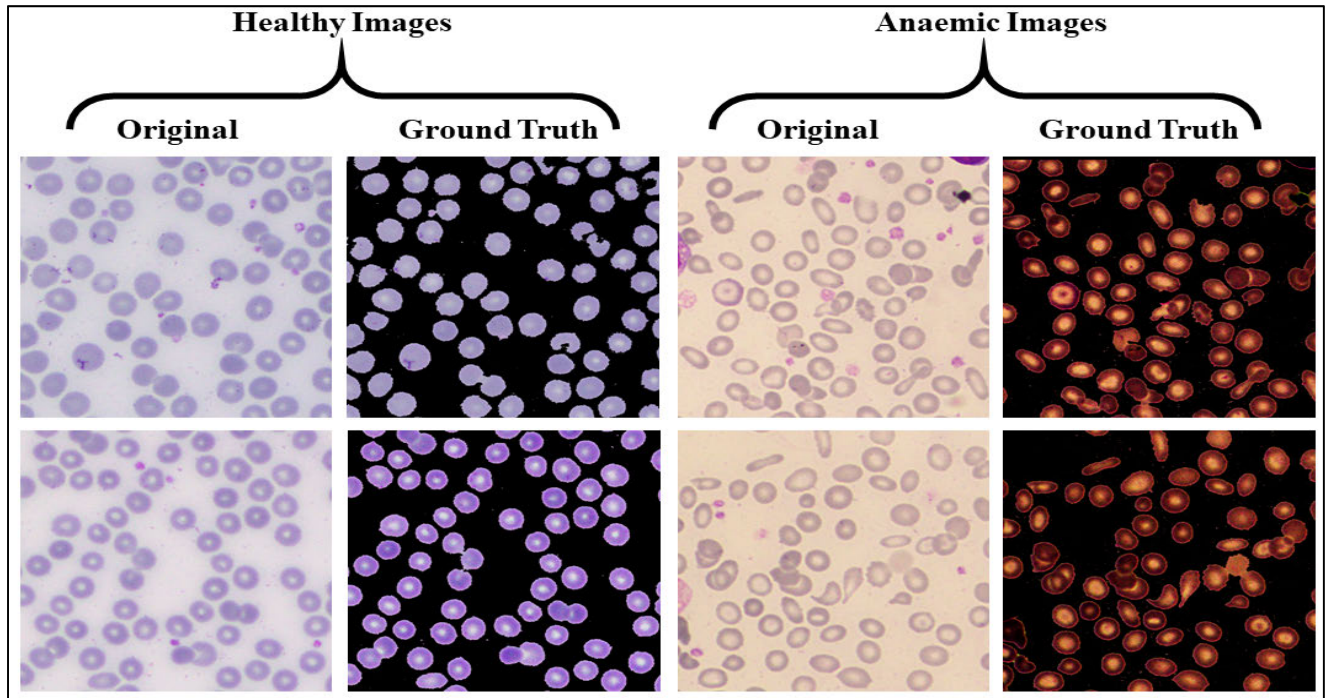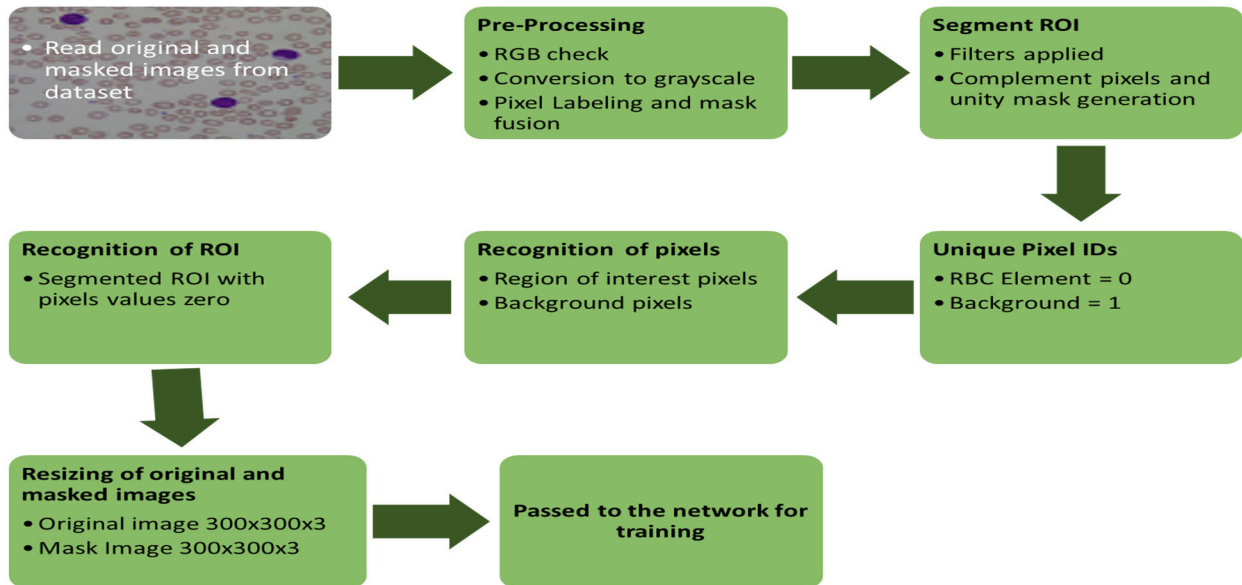
FIGURE 2. This diagram shows the samples from healthy and anaemic images from the proposed dataset. The images of this dataset are captured under various illumination as shown in the diagram.

FIGURE 3. All pre-processing steps of DCED-Net.

For each Image in MaskedDataSet:
 Extract(ListofFeaturesMaskedData)
Next
For each Image in OriginalDataSet:
 Extract(ListofFeaturesOrginalDataSet)
Next
TrainingDataSet = 80% of Extracted Features
TestDataSet = 20% of Extracted Features

End While
#LOADING DCED-NET and Configuring Training Parameters
While DCED-NET Loaded
 Configure Batch_Size_minimum ← 2
 Configure Total Epoch ← 300-500
 Configure Iterations per epoch ← 150
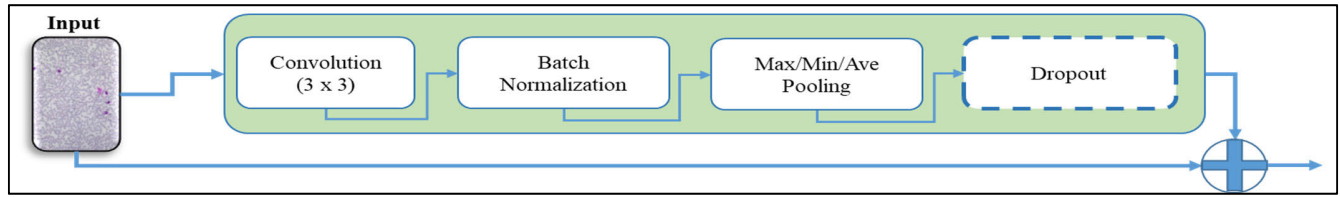 **Configure** Learning rate ← 1e-4

Input

FIGURE 4. The internal operation of each layer's pool in DCED-Network.

Procedure TrainModel
  For I = 1 to TotalEpoch
    Randomly sample the training data with
    replacement
    Count Healthy and Anaemic Pixels of RBC
    Create separate Segments for each Healthy and
    Anemic Pixels
  End For
  End Procedure
End While
 # Compute Evaluation Metrics
  Calculate Accuracy $\leftarrow \frac{TP+TN}{TP+TN+FP+FN}$

  Calculate IoU $\leftarrow \frac{Target \cap Prediction}{Target \cup Prediction}$

  Calculate BF Score $\leftarrow \frac{2(Precision*Recall)}{(Precision+Recall)}$
END

Above pseudo-code is the detailed description of the segmentation technique. For understanding, we have divided the segmentation process into 05 primary steps: 1) In the first step, the system uploads original and manually generated images for pre-processing. 2) During pre-processing, unity mask generation, pixel fusing, and implementation of morphological operation were performed. Resizing and removing noisy and blurry patterns of an image were also handled during this phase. 3) In the 3rd step, all the input data is divided into test and train images. 4) Proposed DCED-Net was loaded for training purposes. Network architecture diagram of DCED-Net are shown in Figure 5. 5) In the last step, segmented healthy and Anaemic RBCs were generated along with evaluation matrices measurement. A brief description of semantic segmentation is given in Figure 6.

Inspired by LSTM and recurrent network structure, we developed a new multi-level deep convolutional encoder-decoder network. Unlike prior CNN structures, we divided the encoder-decoder network into different levels with intra-level, inter-level forward, and reverse connections between any two consecutive layers and levels/groups, respectively, as shown in Figure 6. We address DCED-Net as $\mathbb{N}$. The network $\mathbb{N}$ is a composition of multiple levels of the encoder-decoder networks. For simplicity, each level is represented as $\acute{L}$. The DCED-Net is defined in equation 1.

$$\mathbb{N} = \acute{L}_1 + \acute{L}_2 + \acute{L}_3 + \acute{L}_4 \qquad (1)$$

The last level of architecture is also connected with all previous tiers. The final layer of each pool is equipped with two

variables, i.e., 1) Threshold value (T_o), and 2) Comparison function (C_o). The value of T_o is set as 50%, 80%, and 95% for levels 1,2, and 3 of DCED-Net, respectively. These values are set as a condition along with training parameters at the start of the training. Now, each level received two inputs: one actual ground truth (G) and the second is the previous level's output. Every encoder pool extracts a feature map from the input image and generates a segmented image during decoding phases. At the last layer of the decoding phase, comparison function C_o compares the extracted features map (Fn) and generated image (Φ_n) of level n with the T_o value of nth level (Fn-1) and ground truth (G) respectively. Here n represents the current level of the network. If the C_o function got a value greater than or equal to the current level's threshold, the Φ_n and Fn passed as input to the $n + 1$ convolutional level for further processing.

On the other hand, if the value of C_o is less than T_o, the output of the nth convolutional level is passed as input again to the same level for more optimal feature selection. In this way, the best optimum feature selection is achieved, preserving pixel-level semantic information at each layer and architecture level. The whole comparison function is formulated in the following three equations.

$$C_o = \text{Fn} \equiv T_o \ \& \ \Phi_n \equiv G \qquad (2)$$
$$C_o \geq T_o \ \ then \ output \ \rightarrow \ n + 1 \ level \qquad (3)$$
$$C_o! \geq T_o \ \ then \ output \ \rightarrow \ n^{th} \ level \qquad (4)$$

Here $\equiv$ operator act as a comparison operator. The output of the last level compares with ground truth data. If the system achieves the required accuracy, it evaluates statistical matrices for performance evaluation. Otherwise, the last level output is sent back to the appropriate level for the re-selection of features. Each layer and level within the architecture act as input and output, which means they are more densely connected than prior networks [3], [5]. This strategy enhances the pixel-level feature selection and preservation of semantic information of ROI. The proposed Multi-level DCED-Net has some unique properties. A deep look at the architecture shown in Figure 6 explains that the model is more densely feature-demanding. With given block of n layers, CliqueNet [5] need A_n^2 (Permutation operator) group of feature for single clique block while DensNet [3] required C_n^2 (Combination operator), but we employed $\left(A_n^2 + A_{n+1}^2 + A_{n+2}^2 + A_{n+3}^2 + \ldots A_{n+n}^2\right)$ for each level as well as for every layer within each level. This
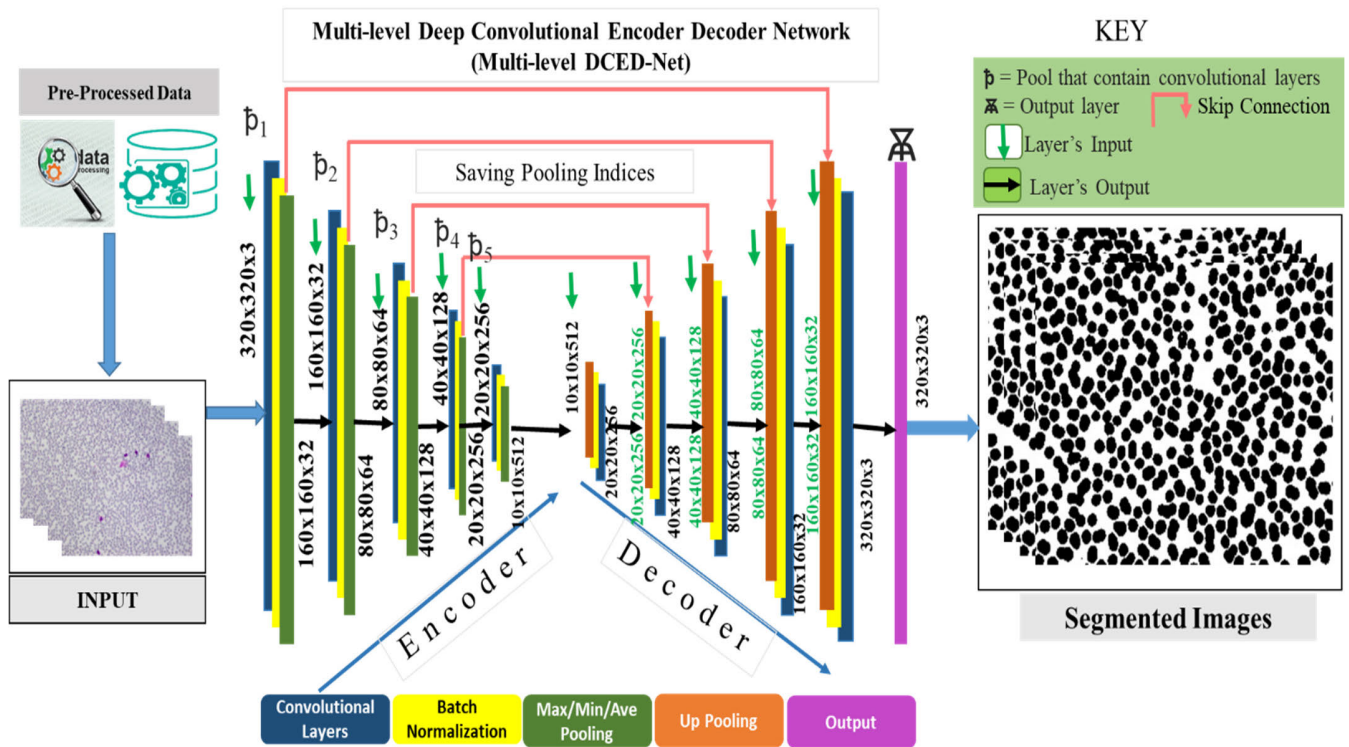
FIGURE 5. Details of layer's input and output of single network used in multilevel deep convolutional encoder-decoder network (DCED-Net) for semantic segmentation.
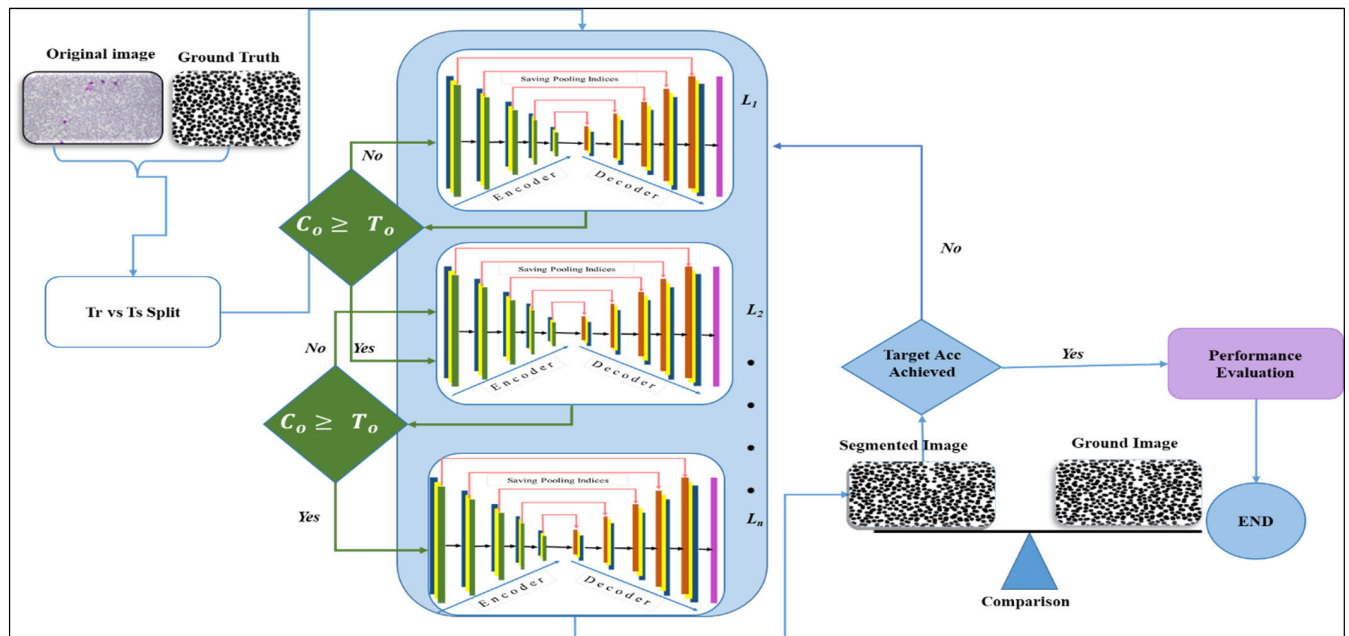
FIGURE 6. Synaptic view of semantic segmentation process in densely connected DCED-Net. C_O represents the comparison function, T_O represent the predefined threshold value., T_r means train dataset, T_S mean test dataset.

means that DCED-Net needs product of permutation features. The training process formulated in equation 5, as shown at the bottom of the next page. Here, $*$ denotes convolutional operations with feature parameters W. while Y is the non-linear activation function. Parameters weights W_{ij}, keeps the pixel features for comparison operations and future use. In DensNet, filters increase linearly as the depth of the net increases [62], which causes rapid growth of parameters,

that may feed irrelevant features. While in cliqueNet, every block provided their features to the next block beyond stage-II irrespective of block output accuracy. But, in the proposed model, the output of each level compares with the previous level's using the C_o function, and only those features are fed to the next level, which is optimal. It turns out that this is a more optimized feature selection way. Densely connected DCED-Net has a solid ability to preserve pixel-level semantic information with the help of recurrent structure at each layer and level with a feedback mechanism. Forward and feedback connections are densely connected at each level to maximize the best feature map and information flow. Due to this structure, our model outperforms for reducing background noises, paying more concentration only on ROI, and achieving competitive performance over existing techniques. For the evaluation and authentication of proposed model results, we used accuracy, IoU, and BF score. Crossmatch analysis was also performed with manually segmented ground truth under the supervision of expert pathologists.

1) LOSS FUNCTION

The loss function of the network is adjusted using the mean square error (MSE) technique. MSE is the average of the squares of the errors between predicted image (p) and ground truth (g). It is calculated by equation 6.

$$MSE = \frac{1}{n} \sum_{i=1}^{n} (g_i - p_i)^2 \quad (6)$$

MSE of the proposed model is measured at each network level, and feedback is given to the network for further training. MSE is measured continuously until or unless the network got a value greater than the predefined threshold value T_o. As the network got desired MSE value, the training of the current level was stopped, and output was given to the next level for further processing with an updated value of T_o.

IV. RESULT AND EVALUATION MATRICES

A. TRAINING AND TESTING

The proposed semantic segmentation architecture is trained and tested on two datasets, i.e., Healthy-RBC and Anaemic-RBC. A total of 2,000 (1000 healthy and 1000 anaemic) peripheral blood smear images were collected from 100 individuals, i.e., 20 images from each with approximately 1,992,000 (Original and Ground truth) RBCs elements. We have also generated 2,000 manually segmented ground truth for authentication purposes.

Figure 2 demonstrates the category distribution and dataset organization. Out of 2,000 images, the system randomly chooses 1800 images for training and 200 images for testing.

The proposed model was trained on 400 epochs, 150 iterations per epoch on both datasets with an initial learning rate of $1e^{-4}$ and a minibatch size of 2. Model execution carried out on GEFORCE RTX-3060 GPU with 24 GB of RAM.

B. EVALUATION

All the results of the underlying model were evaluated based on two evaluation projections. 1) Iteration vs Accuracy and Iteration vs Loss Projection, 2) Epochs vs Accuracy and Epochs vs Loss Projection.
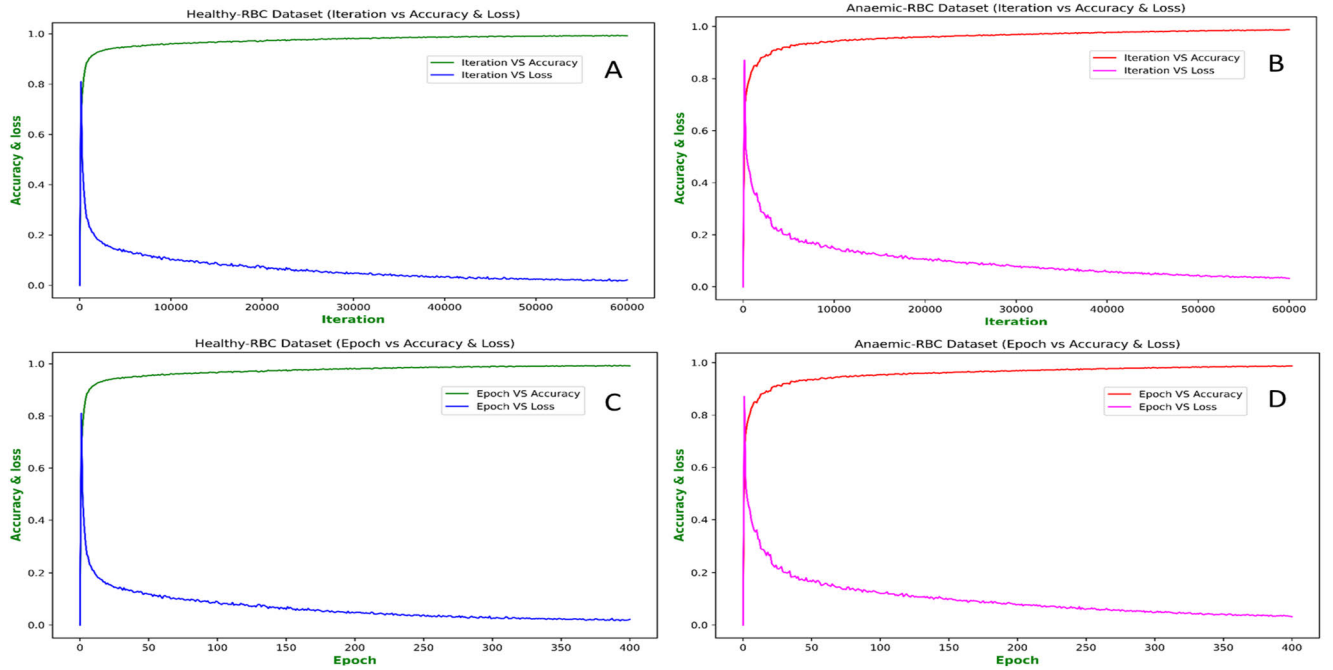
1) ITERATION VS ACCURACY AND ITERATION VS LOSS PROJECTION

The DCED-Net was trained on 400 epochs. The training procedure comprises 60,000 iterations with 150 iterations per epoch. In Figure 7 part (A, B) shows the loss and accuracy projection against the iterations on Healthy-RBC and Anaemic-RBC datasets, respectively. The green line demonstrates the ratio between min-batch accuracy and iteration, while the blue line explains the relation between min-batch loss with each iteration. On the Healthy-RBC dataset, with the start of the training procedure, the accuracy graph starts from 0 and directly jumped into 0.6689 with 0.8096 loss on completion of 150 epochs. But, the Anaemic-RBC dataset shows relatively less accuracy, i.e., 0.6577 with 0.8707 loss. After completing 162 epochs with 24,300 iterations, the accuracy graph goes to 0.9731 with downfall of the loss graph to 0.0634 from 0.8096 on Healthy-Dataset. While in Anaemic-dataset, after 24,300 iterations, the accuracy graph goes up to 0.9556 with 0.1266 loss. As the iteration goes to 45,000, the spike of the accuracy graph touches the highest point, i.e., 0.9856 and 0.9736 on healthy and anaemic datasets, respectively. While after 52,000 iterations, the model loss goes down to the lowest value, i.e., 0.0252 on the healthy dataset and 0.0351 on the anaemic dataset.

2) EPOCHS VS ACCURACY AND EPOCHS VS LOSS PROJECTION

In Figure 7 part (C) the green line explains the relationship between accuracy achieved against each epoch. The blue line explains loss during training on the healthy-RBC dataset. While part (D) shows the ratio between epoch vs loss and accuracy on the anaemic-RBC dataset. On the Healthy-RBC dataset, at the first epoch, the training accuracy was 0.6689 with 0.8096 loss. As the epochs increase to 100, the accuracy goes upward and touches 0.9707, decreasing the corresponding loss down to 0.0789. After that point, the accuracy and loss fluctuate between approximately 0.9754 to 0.9856 and 0.0317 to 0.0215 on both datasets, respectively. The highest accuracy is achieved at epoch no

$$Z_o^n = \begin{pmatrix} f_i \left(\sum_{i<j} W_{ij} * Y_i^n + \sum_{x>j} W_{xj} * Y_x^{n-1} \right) + f_{i+1} \left(\sum_{i+1<j+1} W_{ij+1} * Y_{i+1}^n + \sum_{x+1>j+1} W_{xj+1} * Y_{x+1}^{n-1} \right) \cdots \\ f_{i+n} \left(\sum_{i+n<j+n} W_{ij+n} * Y_{i+n}^n + \sum_{x+n>j+n} W_{xj+n} * Y_{x+n}^{n-1} \right) \end{pmatrix} \quad (5)$$

FIGURE 7. Part (A) Iteration vs accuracy and loss projection on the healthy-RBC dataset, Part (B) Iteration vs accuracy and loss projection on Anaemic-RBC dataset, Part (C) Epoch vs accuracy and loss projection on Healthy-RBC dataset, Part (D) Epoch vs accuracy and loss projection on Anaemic-RBC dataset.

345, i.e., 0.9856 and 0.9736 on healthy and anaemic datasets, respectively. At the same time, minimum validation loss against epoch is achieved at epoch no 324 with 0.0171 on the healthy dataset and 0.0724 on the anaemic dataset.

C. EVALUATION MATRICES
We have further evaluated our model on three fundamental statistical matrices, i.e., 1) Accuracy (mean and global); 2) MeanBFScor; and 3) Intersection over union (IoU) (weighted and mean)

1) ACCURACY
In semantic segmentation, accuracy is used to determine correctly classified pixels of an image. Accuracy is the ratio between the product of correctly identified negative pixels (TN) and correctly identified positive pixels (TP) over the product of falsely identified positive (FP), falsely identified negative pixels (FN) along with TN and TP as shown in equation 6:

$$Accuracy = \frac{TN + TP}{TN + TP + FN + FP} \qquad (7)$$

To find out the mean value of each accuracy, we have carried out a 5-fold training procedure. Table 1 shows the different accuracies of both RBCs datasets, i.e., healthy and anaemic, with global accuracy. Individual dataset training, validation, and testing accuracies with relevant losses are also shown in Table 1. Our model got 0.9856 and 0.9738 training accuracy for healthy and anaemic RBCs with 0.0253 and 0.0351 losses, respectively. While validation

and test accuracies were 0.9760 and 0.9720 for healthy and anaemic datasets. Global training, test, and validation accuracies on both datasets simultaneously were 0.9796, 0.9728, and 0.9655, respectively, shown in Table 1

2) MeanBFScore
The match between the predicted boundary of an image and the ground truth boundary is measured by BFScore matrix [63]. BFScore is calculated by taking the ratio of twice of recall and precision product over the sum of precision and recall as described in equation 7:

$$BFScore = \frac{2 * (Recall * Precision)}{Precision + Recall} \qquad (8)$$

The BFScore of both datasets was calculated as 0.9138 and 0.8978 for the healthy and anaemic datasets, respectively shown in Table 1. At the same time, the overall MeanBFScore on both datasets was 0.9058, shown in Table 1.

3) INTERSECTION OVER UNION IoU
Intersection over Union (IoU) [64] is another class of statistical matrix used to evaluate semantic segmentation results. The primary function of IoU is quantifying the ratio between overlapping of predicted output and targeted ground truth. It is calculated by finding the balance between the intersection of target and predicted pixels overall masks and original image pixels. The mathematical form of IoU is shown in equation 8:

$$IoU = \frac{Target \cap Prediction}{Target \cup Prediction} \qquad (9)$$

Our model attained IoU of 0.9311 with healthy-RBC while 0.9032 with Anaemic-RBC dataset, as shown in Table 1. Overall, MeanIoU was calculated as 0.9058 collectively on both datasets.

V. RESULT AND DISCUSSION

The proposed model was trained, validated, and tested on two RBCs datasets, i.e., Healthy and Anaemic-RBCs. Both datasets were collected from SKMCH&RC Lahore. Healthy-RBC datasets were collected from 50 patients with 20 samples from each. So, a total of 1000 healthy images were obtained. Likewise, 50 patients were selected randomly from the daily workload to collect anaemic-RBCs with 20 samples from each. So, a total of 1000 anaemic images were obtained. After the collection of images, ground truth was generated for both datasets to authenticate the predicted results. The proposed model is applied on both datasets individually and collectively to find the performance and accuracy. The model was implemented in two phases: 1) Pre-Processing phase, manually generated masks, and original images were pre-processed and modified according to the model requirement. 2) In the second phase, a deep convolutional encoder-decoder network (DCED-Net) was initiated. DCED-Net comprises an encoder-decoder network and pre-trained VGG-16. The densely connected deep convolutional encoder-decoder framework is used to train and test both RBC-datasets, while VGG-16 was used for pixel-level feature extraction. Evaluation matrices like Accuracy, IoU, and BFScore were calculated to prove the performance of the model.

A. DATASET

Previously developed blood cell datasets like ALL-IDB-I, ALL-IDB-II [65], extended ALL-ID [66], BCCD, IUMS-IDB [67], SMC-IDB [68], BS_DB3 [69], Ash bank, BBBC [70] dataset equipped with small no of images with little RBCs elements as shown in Table 2. ALL-ID-I contains only 108 images with just 39000 blood elements. All 108 images have no masks of authentication of proposed results. Other datasets also lack detailed information regarding manual ground truth for authentication purposes and blood cells elements. While in this research work, first, we have developed two state-of-the-art blood cell datasets: 1) Healthy-RBCs dataset and 2) Anaemic-RBCs dataset with their relevant CBC and morphology report. These reports will help the research community to authenticate the proposed model results in respect of anaemia disease. Both datasets are equipped with 2000 images (1000 healthy, 1000 anaemic) with the ground truth of each image. So, our dataset repository comprises a total of 4000 images (2000 original and 2000 ground truth) with approximately 1,992,000 RBC elements. RBCs elements are categorized into four classes, i.e., normal, microcytes, macrocytes, elliptocytes, and target RBCs, as shown in Figure 8.

B. ACCURACY

Table 1 shows the training, testing, and validation accuracy of both datasets. Our model outperforms on the healthy-RBC dataset with training, validation, and test accuracies of 0.9856, 0.9760, and 0.9720, respectively, compared to the anaemic-RBC dataset. At the same time, BFScore and IoU of the proposed model on anaemic-RBCs images were 0.8978 and 0.9032, respectively. The BFScore and IoU on the healthy-RBC dataset were calculated as 0.9138 and 0.9311, as shown in Table 1. All these figures reveal that the proposed model outperformed the healthy-RBC dataset compared to the anaemic-RBC images. This is because anaemic-RBCs images are more complicated than healthy RBCs. We have carried out 05-fold training, validation, and testing of the underlying model and got global training, validation, and testing accuracies 0.9796, 0.9728, and 0.9655, respectively, as shown in Table 1.

C. GLOBAL AND CLASS-WISE PIXEL COUNTING

Accurate blood element examination is beneficial for predicting blood-related diseases like Anaemia. In the medical image processing domain, precise pixel counting plays a significant role in diagnosing blood-related diseases. The proposed model is also outperformed in this regard. Table 3 shows the pixel count of healthy-RBCs and anaemic-RBCs images individually, along with global pixel counting in a single blood image.

D. COMPARATIVE ANALYSIS

A model proposed in [39] used 1,120 manually collected images for WBC detection without ground truth for authentication and pixel counting, which are key features for disease prediction using medical image processing. Authors in [44], Automated Semantic Segmentation of Red Blood Cells for Sickle Cell Disease target malaria and SCD without providing ground truth for crossmatching analysis. Research work carried out by [39], [42], [43], [71], [72] ignores the pixel counting, generation of ground truth, and targeting any disease. Most of the previous work ignores the training and validation accuracy with the highest accuracy of 98.87%, as shown in Table 4. This work first developed state-of-the-art 2 RBC blood image datasets equipped with 1000 Healthy RBCs and 1000 anaemic-RBC images with 2000 ground truth for crossmatching analysis and performance evaluation. We have also categorized our dataset into four different RBC classes to classify the abnormal shape of the RBCs due to anaemia disease. To authenticate the proposed results, we have calculated three different types of accuracies along with relevant losses, i.e., training, validation, and testing accuracies and losses individually for both datasets as well as globally. The proposed model achieved individual training, validation, and testing accuracies of 98.56%, 97.60%, and 97.20%, respectively. While global accuracies were 97.96%, 97.28%, and 96.55% for training, validation, and testing. In addition to this, our model can also precisely count the pixels for diagnosing anaemia disease.

TABLE 1. Individual and global training, validation, test accuracies, BFScore, and IoU with relevant losses of healthy and anaemic-RBCs dataset.

S #	Evaluation Matric	Healthy-RBC Dataset	Anaemic-RBC Dataset	Global
1	Training Accuracy	0.9856	0.9736	0.9796
2	Training Loss	0.0252	0.0351	0.0204
3	Validation Accuracy	0.9760	0.9696	0.9728
4	Validation Loss	0.0613	0.0776	0.0272
5	Test Accuracy	0.9720	0.9591	0.9655
6	Test Loss	0.0717	0.1085	0.0345
7	BFScore	0.9138	0.8978	0.9058
8	IoU	0.9311	0.9032	0.9171
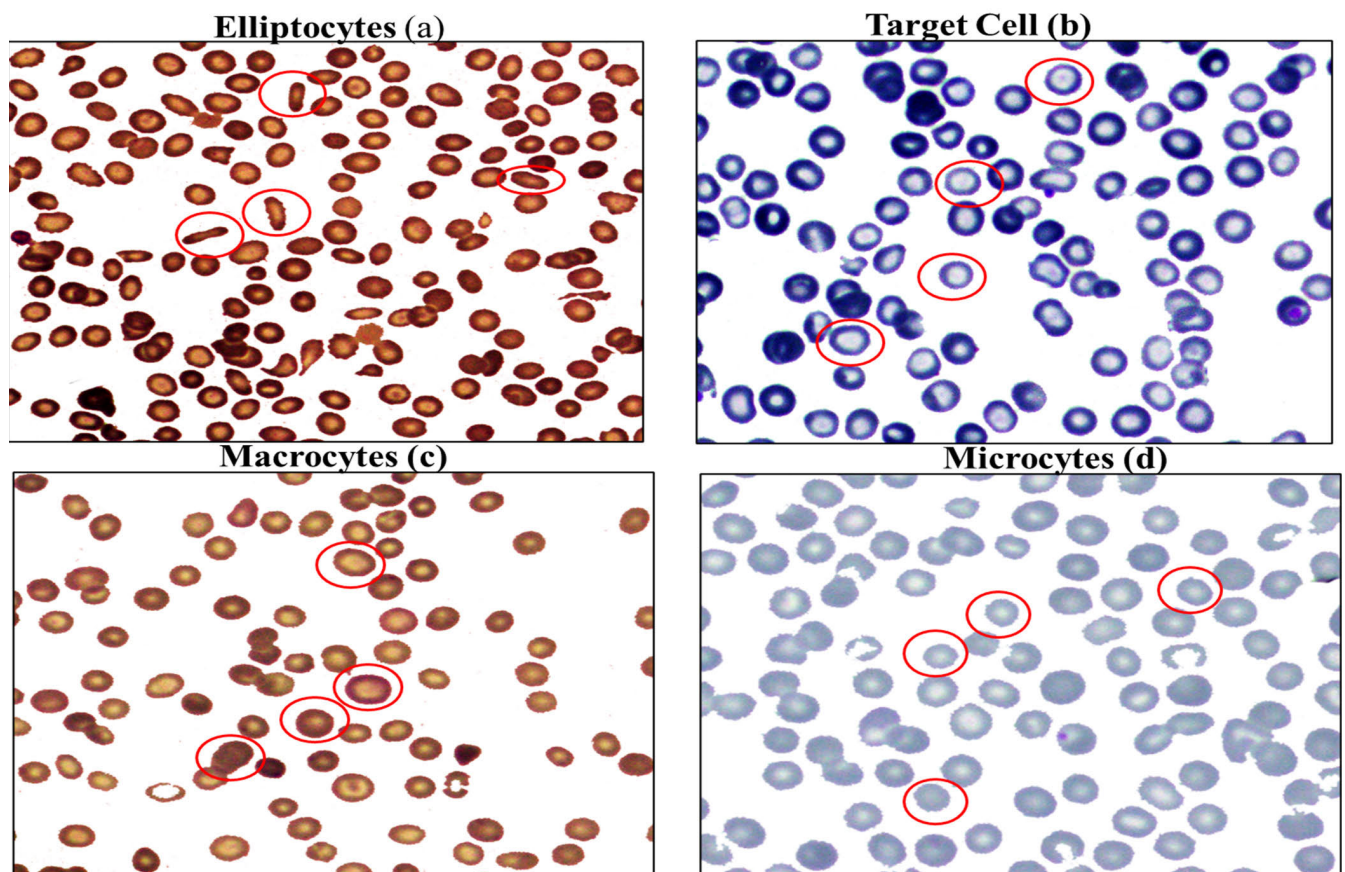
FIGURE 8. Shows the four categories of RBC elements, i.e., (a) Elliptocytes are elongated oval-shaped RBCs due to iron deficiency, (b) Target cells increase with excess cell membrane relative to cell volume, (c) Microcytes are those RBCs that have a size smaller than normal, and (d) Macrocytes RBCs are those whose size becomes larger than usual in healthy-RBC and Anaemic-RBC datasets.

E. ADVANTAGES OVER EXISTING TECHNIQUES

This research work has upper hand over current research work relevant to segmentation of RBC image in respect of the following:

1) We used a multi-level deep convolutional encoder-decoder network that preserved the semantic information of the blood element.
2) Each level of the network performs a segmentation training process upto a specific threshold value that helps to reduce unnecessary training.
3) We have developed two state-of-the-art RBCs datasets with 2000 original images (1000 healthy and 1000 anaemic) and 2000 manually generated ground truth with approximately 1,992,000 RBC elements for the research community.
4) Each level has its specific learning weights that help to extract feature maps upto a specific level. So the network is prevented from the extraction of unnecessary features.
5) The multi-level approach also reduces the gradient descent phenomenon during the training.
6) The chances of over-fitting and under-fitting of the network become negligible using T_o value at each level.

TABLE 2. Dataset comparison with proposed novel RBCs datasets. "X" represents non-availability and "✓" availability.

Dataset	No of Images	Blood elements	Ground truth	CBC reports	Morphology reports
ALL-IDB-I and II [65]	108 and 260	WBC and RBC	×	×	×
SMC-IDB[68]	367	WBC	×	×	×
IUMS-IDB [67]	196	WBC	×	×	×
Malarial dataset	848	RBC	×	×	×
Proposed Healthy-RBC Dataset	**2000**	**RBC**	✓	✓	✓
Proposed Anaemic-RBC Dataset	**2000**	**RBC**	✓	✓	✓

TABLE 3. Pixel count of healthy and anaemic-RBCs images along with global pixel count.

S #	Blood cell image Type	Pixel count
1	Healthy-RBCs Image	$3.2657e + 09$
2	Anaemic-RBC Image	$2.1728e + 08$
3	Global pixel count	$1.7414e + 09$

TABLE 4. Brief comparative analysis of proposed dataset and technique with current research work.

	Dataset Comparisons						Technique evaluation comparisons		
Work	Dataset	No of images	Ground Truth	Blood element	Pixel counting	Targeted Disease	Accuracy type	Evaluation matrices	Accuracy achieved
[39]	Manually collected	1,120	×	WBC	×	×	Mean accuracy	Accuracy	90.09%
[44]	Manually collected	848 and Malarial Dataset	×	RBC	×	Malaria	Global accuracy	Accuracy	93.72%
[66]	Extended ALL-IDB-I, II	Original 108,	Ground truth 108	RBC, WBC, plt	✓	×	Mean, global Accuracy	Accuracy, IoU and BFScore	97.18%
[71]	Manually collected	564	×	WBC	×	×	Accuracy	Precision, recall and specificity	100%
[72]	SMC-IDB, IUMS-IDB, ALL-IDB	367, 195 and 108	×	WBC	×	×	Accuracy	Accuracy	94.1%
[40]	Manually collected	314	×	RBC	✓	SCD	Not mentioned	Dice Coefficient, Jaccard Index, and Hausdorff Distance	Not mentioned
[42]	ALL-IDB-I and II	42 and 101	42	WBC and RBC	×	×	Accuracy	Global accuracy	93.3%
[43]	Manually collected	257	×	WBC	×	×	Accuracy	Accuracy	98.87
Proposed	**Healthy-RBCs and Anaemic-RBCs**	**2000 and 2000**	✓	**RBC**	✓	Anaemia	**Training Accuracy, Validation accuracy, Test Accuracy and Global accuracy**	**Accuracy, IoU and BFScore**	**Global Training = 97.96% Validation = 97.28%, Test = 96.55%**

7) We have performed 5-fold training, validation, and testing of blood segmentation to authenticate the proposed results.

8) We also compare the proposed results with manually generated ground truth and evaluate it by finding IoU. Our model got 0.9311and 0.9032 for the healthy and anaemic-RBC dataset with a MeanIoU of 0.9171 on both datasets.

VI. CONCLUSION

To handle the precise segmentation of blood elements in digital images, we proposed a novel model known as multi-level

DCED-Net. The multi-level concept enhances the preservation of semantic information at the pixel level, which helps for accurate, effective segmentation. For experimental purposes, we have developed two state-of-the-art RBC datasets for the research community. These datasets will play a pivotal role in determining an accurate model in the semantic segmentation domain and diagnosing anaemia deformities in RBC cells. During the experiment, we carried out 5-fold training, validation, and testing to obtain precise results. Our model got 0.9856, 0.9760, and 0.9720 training, validation, and testing accuracies on the healthy-RBC dataset, respectively. The anaemic-RBC dataset, training, validation, and testing accuracy were 0.9736, 0.9696, and 0.9591, respectively. We have generated a total of 2000 ground truth mask images for cross-matching analysis and got the IoU 0.9311 and 0.9032 with BFScore 0.9138, 0.8978 on healthy and anaemic datasets.

FUTURE WORK

In the future, we will enhance our model for pixel-level classification regarding RBCs classes, i.e., normal, microcytes, macrocytes, elliptocytes, and target RBCs. This work is the first phase of the Anaemia Disease Prediction Support System. The next paper will extend this dataset and classify each RBC type, i.e., normal, microcytes, macrocytes, elliptocytes, and target RBCs. This classification will help to detect anaemia disease automatically in blood cells.

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AUTHORSHIP CONTRIBUTION

Muhammad Shahzad: Conceptualization; Methodology; Formal analysis; Data curation; Writing - Original Draft; Writing - review & editing, Dataset Preparation. **Arif Iqbal Umar**: Supervision and reviewing. **Syed Hamad Shirazi:** Reviewing, Editing, Data Analysis. **Israr Ahmed Shaikh**: Image Collection, Image Preparation.

COMPETING INTEREST

All authors declare no conflict of interest.

AUTHORS APPROVAL

All authors have read and approved the final manuscript.

REFERENCES

[1] A. Krizhevsky, S. Ilya, and E. H. Geoffrey, "ImageNet classification with deep convolutional neural networks," *Commun ACM*, vol. 12, May 2017, Art. no. 04015009, doi: 10.1061/(ASCE)GT.1943-5606.0001284.

[2] K. He, X. Zhang, S. Ren, and J. Sun, "Deep residual learning for image recognition," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2016, pp. 770–778, doi: 10.1109/CVPR.2016.90.

[3] G. Huang, Z. Liu, L. van der Maaten, and K. Q. Weinberger, "Densely connected convolutional networks," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit.*, Jul. 2017, pp. 2261–2269, doi: 10.1109/CVPR.2017.243.

[4] C. Szegedy, W. Liu, Y. Jia, P. Sermanet, S. Reed, D. Anguelov, D. Erhan, V. Vanhoucke, and A. Rabinovich, "Going deeper with convolutions," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2015, pp. 1–9, doi: 10.1109/CVPR.2015.7298594.

[5] Y. Yang, Z. Zhong, T. Shen, and Z. Lin, "Convolutional neural networks with alternately updated clique," in *Proc. IEEE/CVF Conf. Comput. Vis. Pattern Recognit.*, Jun. 2018, pp. 2413–2422, doi: 10.1109/CVPR.2018.00256.

[6] K. Simonyan and A. Zisserman, "Very deep convolutional networks for large-scale image recognition," in *Proc. Int. Conf. Learn. Represent. (ICLR)*, 2015, pp. 1–14, doi: 10.2146/ajhp170251.

[7] S. Xie, R. Girshick, P. Dollar, Z. Tu, and K. He, "Aggregated residual transformations for deep neural networks," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jul. 2017, pp. 5987–5995, doi: 10.1109/CVPR.2017.634.

[8] M. Firdaus-Nawi, O. Noraini, M. Y. Sabri, A. Siti-Zahrah, M. Zamri-Saad, and H. Latifah, "DeepLabv3+_encoder-decoder with Atrous separable convolution for semantic image segmentation," *Pertanika J. Trop. Agric. Sci.*, vol. 34, no. 1, pp. 137–143, 2011.

[9] C. Szegedy, S. Ioffe, V. Vanhoucke, and A. A. Alemi, "Inception-v4, Inception-ResNet and the impact of residual connections on learning," in *Proc. AAAI Conf. Artif. Intell.*, 2017, pp. 4278–4284.

[10] B. Zoph, V. Vasudevan, J. Shlens, and Q. V. Le, "Learning transferable architectures for scalable image recognition," in *Proc. IEEE/CVF Conf. Comput. Vis. Pattern Recognit.*, Jun. 2018, pp. 8697–8710, doi: 10.1109/CVPR.2018.00907.

[11] P. Liu, H. Yu, and S. Cang, "Adaptive neural network tracking control for underactuated systems with matched and mismatched disturbances," *Nonlinear Dyn.*, vol. 98, no. 2, pp. 1447–1464, Oct. 2019, doi: 10.1007/s11071-019-05170-8.

[12] Z.-C. Tang, C. Li, J.-F. Wu, P.-C. Liu, and S.-W. Cheng, "Classification of EEG-based single-trial motor imagery tasks using a B-CSP method for BCI," *Frontiers Inf. Technol. Electron. Eng.*, vol. 20, no. 8, pp. 1087–1098, Aug. 2019, doi: 10.1631/FITEE.1800083.

[13] Z. Tang, H. Yu, C. Lu, P. Liu, and X. Jin, "Single-trial classification of different movements on one arm based on ERD/ERS and corticomuscular coherence," *IEEE Access*, vol. 7, pp. 128185–128197, 2019, doi: 10.1109/ACCESS.2019.2940034.

[14] S. Hao, Y. Zhou, and Y. Guo, "A brief survey on semantic segmentation with deep learning," *Neurocomputing*, vol. 406, pp. 302–321, Sep. 2020, doi: 10.1016/j.neucom.2019.11.118.

[15] D.-P. Fan, W. Wang, M.-M. Cheng, and J. Shen, "Shifting more attention to video salient object detection," in *Proc. IEEE/CVF Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2019, pp. 8546–8556, doi: 10.1109/CVPR.2019.00875.

[16] X. Lu, W. Wang, C. Ma, J. Shen, L. Shao, and F. Porikli, "See more, know more: Unsupervised video object segmentation with co-attention Siamese networks," in *Proc. IEEE/CVF Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2019, pp. 3618–3627, doi: 10.1109/CVPR.2019.00374.

[17] W. Wang, X. Lu, J. Shen, D. Crandall, and L. Shao, "Zero-shot video object segmentation via attentive graph neural networks," in *Proc. IEEE/CVF Int. Conf. Comput. Vis. (ICCV)*, Oct. 2019, pp. 9235–9244, doi: 10.1109/ICCV.2019.00933.

[18] A. Nurhadiyatna and S. Loncaric, "Semantic image segmentation for pedestrian detection," in *Proc. 10th Int. Symp. Image Signal Process. Anal.*, Sep. 2017, pp. 153–158, doi: 10.1109/ISPA.2017.8073587.

[19] G. Papandreou, L.-C. Chen, K. P. Murphy, and A. L. Yuille, "Weakly-and semi-supervised learning of a deep convolutional network for semantic image segmentation," in *Proc. IEEE Int. Conf. Comput. Vis. (ICCV)*, Dec. 2015, pp. 1742–1750, doi: 10.1109/ICCV.2015.203.

[20] X. Xie and Z. Wang, "Multi-scale semantic segmentation enriched features for pedestrian detection," in *Proc. 24th Int. Conf. Pattern Recognit. (ICPR)*, Aug. 2018, pp. 2196–2201, doi: 10.1109/ICPR.2018.8545414.

[21] N. Ivanova, V. Gugleva, M. Dobreva, I. Pehlivanov, S. Stefanov, and V. Andonova, *Laboratory Approach to Anemia*, vol. 1. Rijeka, Croatia: InTech, 2016, p. 13.

[22] A. Merino, L. Puigví, L. Boldú, S. Alférez, and J. Rodellar, "Optimizing morphology through blood cell image analysis," *Int. J. Lab. Hematol.*, vol. 40, pp. 54–61, May 2018, doi: 10.1111/ijlh.12832.

[23] F. Flohr and D. Gavrila, "PedCut: An iterative framework for pedestrian segmentation combining shape models and multiple data cues," in *Proc. Brit. Mach. Vis. Conf.*, 2013, pp. 1–11, doi: 10.5244/C.27.66.

[24] G. Brazil, X. Yin, and X. Liu, "Illuminating pedestrians via simultaneous detection and segmentation," in *Proc. IEEE Int. Conf. Comput. Vis. (ICCV)*, Oct. 2017, pp. 4960–4969, doi: 10.1109/ICCV.2017.530.

[25] X. Tao, D. Zhang, W. Ma, X. Liu, and D. Xu, "Automatic metallic surface defect detection and recognition with convolutional neural networks," *Appl. Sci.-Basel*, vol. 8, no. 9, pp. 1–15, Sep. 2018, doi: 10.3390/app8091575.

[26] Z. Wang, L. Wei, L. Wang, Y. Gao, W. Chen, and D. Shen, "Hierarchical vertex regression-based segmentation of head and neck CT images for radiotherapy planning," *IEEE Trans. Image Process.*, vol. 27, no. 2, pp. 923–937, Feb. 2018, doi: 10.1109/TIP.2017.2768621.

[27] Y. Guo, Y. Gao, and D. Shen, "Deformable MR prostate segmentation via deep feature learning and sparse patch matching," *IEEE Trans. Med. Imag.*, vol. 35, no. 4, pp. 1077–1089, Apr. 2016, doi: 10.1109/TMI.2015.2508280.

[28] M. Drozdzal, E. Vorontsov, G. Chartrand, S. Kadoury, and C. Pal, "The importance of skip connections in biomedical image segmentation," in *Deep Learning and Data Labeling for Medical Applications* (Lecture Notes in Computer Science), vol. 10008, G. Carneiro, D. Mateus, L. Peter, A. Bradley, J. M. R. S. Tavares, V. Belagiannis, J. P. Papa, J. C. Nascimento, M. Loog, Z. Lu, J. S. Cardoso, and J. Cornebise, Eds. Cham, Switzerland: Springer, 2016, pp. 179–187, doi: 10.1007/978-3-319-46976-8_19.

[29] H. Noh, S. Hong, and B. Han, "Learning deconvolution network for semantic segmentation," in *Proc. IEEE Int. Conf. Comput. Vis.*, May 2015, pp. 1520–1528.

[30] Z. Zhou, M. M. R. Siddiquee, N. Tajbakhsh, and J. Liang, "UNet++: Redesigning skip connections to exploit multiscale features in image segmentation," *IEEE Trans. Med. Imag.*, vol. 39, no. 6, pp. 1856–1867, Dec. 2019, doi: 10.1109/TMI.2019.2959609.

[31] X. Zhu, H. Suk, S. Lee, and D. Shen, "Subspace regularized sparse multitask learning for multiclass neurodegenerative disease identification," *IEEE Trans. Biomed. Eng.*, vol. 63, no. 3, pp. 607–618, Mar. 2015, doi: 10.1109/TBME.2015.2466616.

[32] X. Zhu, H.-I. Suk, and D. Shen, "A novel matrix-similarity based loss function for joint regression and classification in AD diagnosis," *NeuroImage*, vol. 100, pp. 91–105, Oct. 2014, doi: 10.1016/j.neuroimage.2014.05.078.

[33] C. Chen, J. Wei, C. Peng, W. Zhang, and H. Qin, "Improved saliency detection in RGB-D images using two-phase depth estimation and selective deep fusion," *IEEE Trans. Image Process.*, vol. 29, pp. 4296–4307, 2020, doi: 10.1109/TIP.2020.2968250.

[34] H. Ding, X. Jiang, B. Shuai, A. Q. Liu, and G. Wang, "Semantic segmentation with context encoding and multi-path decoding," *IEEE Trans. Image Process.*, vol. 29, pp. 3520–3533, 2020, doi: 10.1109/TIP.2019.2962685.

[35] M. Hassaballah and A. I. Awad, *Deep Learning in Computer Vision*. Boca Raton, FL, USA: CRC Press, Mar. 2020.

[36] L. Sun, C. Zhao, Z. Yan, P. Liu, T. Duckett, and R. Stolkin, "A novel weakly-supervised approach for RGB-D-based nuclear waste object detection," *IEEE Sensors J.*, vol. 19, no. 9, pp. 3487–3500, May 2018, doi: 10.1109/JSEN.2018.2888815.

[37] Y. Guo, Y. Liu, T. Georgiou, and M. S. Lew, "A review of semantic segmentation using deep neural networks," *Int. J. Multimedia Inf. Retr.*, vol. 7, no. 2, pp. 87–93, Jun. 2018, doi: 10.1007/s13735-017-0141-z.

[38] G. Wu and Y. Li, "CyclicNet: An alternately updated network for semantic segmentation," *Multimedia Tools Appl.*, vol. 80, no. 2, pp. 3213–3227, Jan. 2021, doi: 10.1007/s11042-020-09791-9.

[39] Q. Wang, S. Bi, M. Sun, Y. Wang, D. Wang, and S. Yang, "Deep learning approach to peripheral leukocyte recognition," *PloS One*, vol. 14, no. 6, 2019, Art. no. e0218808.

[40] M. Zhang, X. Li, M. Xu, and Q. Li, "Automated semantic segmentation of red blood cells for sickle cell disease," *IEEE J. Biomed. Health Informat.*, vol. 24, no. 11, pp. 3095–3102, Nov. 2020, doi: 10.1109/JBHI.2020.3000484.

[41] M. Zhang, X. Li, M. Xu, and Q. Li, "RBC semantic segmentation for sickle cell disease based on deformable U-Net," in *Medical Image Computing and Computer Assisted Intervention—MICCAI 2018*, vol. 5. Springer, 2018.

[42] T. Tran, T. Phuong, L. B. Minh, S. Lee, and K. Kwon, "Blood cell count using deep learning semantic segmentation," Tech. Rep., Sep. 2019, doi: 10.20944/preprints201909.0075.v1.

[43] M. R. Reena and P. M. Ameer, "Localization and recognition of leukocytes in peripheral blood: A deep learning approach," *Comput. Biol. Med.*, vol. 126, Nov. 2020, Art. no. 104034, doi: 10.1016/j.compbiomed.2020.104034.

[44] A. Merino, "A deep learning approach for segmentation of red blood cell images and malaria detection," *Entropy*, vol. 22, no. 6, p. 657, 2020.

[45] L. Chan, M. S. Hosseini, and K. N. Plataniotis, "A comprehensive analysis of weakly-supervised semantic segmentation in different image domains," *Int. J. Comput. Vis.*, vol. 129, no. 2, pp. 361–384, Feb. 2021, doi: 10.1007/s11263-020-01373-4.

[46] K. de Haan, H. C. Koydemir, Y. Rivenson, D. Tseng, E. Van Dyne, L. Bakic, D. Karinca, K. Liang, M. Ilango, E. Gumustekin, and A. Ozcan, "Automated screening of sickle cells using a smartphone-based microscope and deep learning," *NPJ Digit. Med.*, vol. 3, no. 1, pp. 1–9, Dec. 2020, doi: 10.1038/s41746-020-0282-y.

[47] J. S. Shemona and A. K. Chellappan, "Segmentation techniques for early cancer detection in red blood cells with deep learning-based classifier—A comparative approach," *Inst. Eng. Technol., Image Process.*, vol. 14, no. 9, pp. 1726–1732, 2020, doi: 10.1049/iet-ipr.2019.1067.

[48] F. Wu, F. Chen, X.-Y. Jing, C.-H. Hu, Q. Ge, and Y. Ji, "Dynamic attention network for semantic segmentation," *Neurocomputing*, vol. 384, pp. 182–191, Apr. 2020, doi: 10.1016/j.neucom.2019.12.042.

[49] K. Parvati, B. S. Prakasa Rao, and M. Mariya Das, "Image segmentation using gray-scale morphology and marker-controlled watershed transformation," *Discrete Dyn. Nature Soc.*, vol. 2008, pp. 1–8, Nov. 2008, doi: 10.1155/2008/384346.

[50] G. Li, T. Liu, A. Tarokh, J. Nie, L. Guo, A. Mara, S. Holley, and S. T. Wong, "3D cell nuclei segmentation based on gradient flow tracking," *BMC Cell Biol.*, vol. 8, no. 1, pp. 1–10, Dec. 2007, doi: 10.1186/1471-2121-8-40.

[51] A. Baâzaoui, W. Barhoumi, A. Ahmed, and E. Zagrouba, "Semi-automated segmentation of single and multiple tumors in liver CT images using entropy-based fuzzy region growing," *IRBM*, vol. 38, no. 2, pp. 98–108, Apr. 2017, doi: 10.1016/j.irbm.2017.02.003.

[52] M. Everingham, S. M. A. Eslami, L. Van Gool, C. K. I. Williams, J. Winn, and A. Zisserman, "The PASCAL visual object classes challenge: A retrospective," *Int. J. Comput. Vis.*, vol. 111, no. 1, pp. 98–136, Jan. 2014, doi: 10.1007/s11263-014-0733-5.

[53] M. Cordts, M. Omran, S. Ramos, T. Rehfeld, M. Enzweiler, R. Benenson, U. Franke, S. Roth, and B. Schiele, "The cityscapes dataset for semantic urban scene understanding," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2016, pp. 3213–3223, doi: 10.1109/CVPR.2016.350.

[54] X. Wu, Q. Chen, Y. Xiao, W. Li, X. Liu, and B. Hu, "LCSegNet: An efficient semantic segmentation network for large-scale complex Chinese character recognition," *IEEE Trans. Multimedia*, vol. 23, pp. 3427–3440, Sep. 2021, doi: 10.1109/tmm.2020.3025696.

[55] J. Long, E. Shelhamer, and T. Darrell, "Fully convolutional networks for semantic segmentation," in *Proc. IEEE Conf. Comput. Vis. Pattern Recognit. (CVPR)*, Jun. 2015, pp. 3431–3440, doi: 10.1109/CVPR.2015.7298965.

[56] F. I. Diakogiannis, F. Waldner, P. Caccetta, and C. Wu, "ResUNet—A: A deep learning framework for semantic segmentation of remotely sensed data," *ISPRS J. Photogramm. Remote Sens.*, vol. 162, pp. 94–114, Apr. 2020, doi: 10.1016/j.isprsjprs.2020.01.013.

[57] L. Zhou, C. Gong, Z. Liu, and K. Fu, "SAL:Selection and attention losses for weakly supervised semantic segmentation," *IEEE Trans. Multimedia*, vol. 23, pp. 1035–1048, Apr. 2021, doi: 10.1109/tmm.2020.2991592.

[58] D. He and C. Xie, "Semantic image segmentation algorithm in a deep learning computer network," *Multimedia Syst.*, Aug. 2020. [Online]. Available: https://link.springer.com/article/10.1007%2Fs00530-020-00678-1, doi: 10.1007/s00530-020-00678-1.

[59] X. Ren, L. Zhang, S. Ahmad, D. Nie, F. Yang, L. Xiang, Q. Wang, and D. Shen, "Task decomposition and synchronization for semantic biomedical image segmentation," 2019, *arXiv:1905.08720*.

[60] P. Bosilj, E. Aptoula, T. Duckett, and G. Cielniak, "Transfer learning between crop types for semantic segmentation of crops versus weeds in precision agriculture," *J. Field Robot.*, vol. 37, no. 1, pp. 7–19, Jan. 2020, doi: 10.1002/rob.21869.

[61] J. Jiang, C. Lyu, S. Liu, Y. He, and X. Hao, "RWSNet: A semantic segmentation network based on SegNet combined with random walk for remote sensing," *Int. J. Remote Sens.*, vol. 41, no. 2, pp. 487–505, Jan. 2020, doi: 10.1080/01431161.2019.1643937.

[62] Y. Chen, J. Li, H. Xiao, X. Jin, S. Yan, and J. Feng, "Dual path networks," 2017, *arXiv:1707.01629*.

[63] G. Csurka, D. Larlus, and F. Perronnin, "What is a good evaluation measure for semantic segmentation?" in *Proc. Brit. Mach. Vis. Conf.*, 2013, p. 5244, doi: 10.5244/C.27.32.

[64] A. Rahman and Y. Wang, "Optimizing intersection-over-union in deep neural networks for image segmentation," in *Proc. Int. Symp. Visual Comput.*, 2016, pp. 234–244.

[65] R. D. Labati, V. Piuri, and F. Scotti, "ALL-IDB: The acute lymphoblastic leukemia image database for image processing," in *Proc. IEEE Int. Conf. Image Process.*, Sep. 2011, pp. 2089–2092.

[66] M. Shahzad, A. I. Umar, M. A. Khan, S. H. Shirazi, Z. Khan, and W. Yousaf, "Robust method for semantic segmentation of whole-slide blood cell microscopic images," *Comput. Math. Methods Med.*, vol. 2020, pp. 1–13, Jan. 2020, doi: 10.1155/2020/4015323.

[67] O. Sarrafzadeh, H. Rabbani, A. Talebi, and H. U. Banaem, "Selection of the best features for leukocytes classification in blood smear microscopic images," *Proc. SPIE*, vol. 9041, Mar. 2014, Art. no. 90410P, doi: 10.1117/12.2043605.

[68] M. Mohamed, B. Far, and A. Guaily, "An efficient technique for white blood cells nuclei automatic segmentation," in *Proc. Conf. IEEE Int. Conf. Syst. Man Cybern.*, Oct. 2015, pp. 220–225, doi: 10.1109/ICSMC.2012.6377703.

[69] M. M. A. Mohamed and B. Far, "A fast technique for white blood cells nuclei automatic segmentation based on Gram–Schmidt orthogonalization," in *Proc. IEEE 24th Int. Conf. Tools With Artif. Intell. (ICTAI)*, vol. 1, Nov. 2012, pp. 947–952, doi: 10.1109/ICTAI.2012.133.

[70] V. Ljosa, K. L. Sokolnicki, and A. E. Carpenter, "Annotated high-throughput microscopy image sets for validation," *Nat. Methods*, vol. 9, no. 7, p. 637, 2012, doi: 10.1038/nmeth.2083.

[71] L. Alzubaidi, M. A. Fadhel, O. Al-Shamma, J. Zhang, and Y. Duan, "Deep learning models for classification of red blood cells in microscopy images to aid in sickle cell anemia diagnosis," *Electronics*, vol. 9, no. 3, p. 427, Mar. 2020, doi: 10.3390/electronics9030427.

[72] C. D. Ruberto, A. Loddo, and G. Puglisi, "Blob detection and deep learning for leukemic blood image analysis," *Appl. Sci.*, vol. 10, no. 3, p. 1176, Feb. 2020, doi: 10.3390/app10031176.

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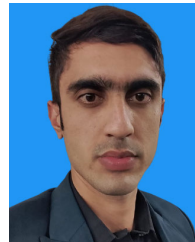
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