Classification of Red Blood Cells in Sickle Cell Anemia Using Deep Convolutional Neural Network

Laith Alzubaidi[1], Omran Al-Shamma[2], Mohammed A. Fadhel[3], Laith Farhan[4],

Jinglan Zhang[5]

[1,5]Faculty of Science & Engineering, Queensland University of Technology, Brisbane, Australia
[1, 2, 3] University of Information Technology and Communications, Baghdad, IRAQ
[4]School of Engineering, Manchester Metropolitan University, UK,
& University of Diyala, IRAQ

[1]laith.alzubaidi@hdr.qut.edu.au
[2]o.al_shamma@uoitc.edu.iq
[3]Mohammed.a.fadhel@uoitc.edu.iq
[4]l.al-bayati@mmu.ac.uk
[5]jinglan.zhang@qut.edu.au

Abstract. Sickle cell anemia is an abnormal red blood cell which leads to blood vessel obstruction joined by painful episodes and even death. It is also called abnormal hemoglobin. Hemoglobin is responsible for passing oxygen through the blood vessel for all over the body. Normal red blood cells are in a circular shape and they are compact and flexible, enabling them to move freely through small capillaries. On the other hand, abnormal red blood cells are in sickle shape and they are stiff and angular causing them to become stuck in small capillaries. Due to that, it will be a reason for pain to patients and lead to low oxygen and dehydration. The manual assessment, classification, and counting of biological cells require for an immense spending of time and it may lead to wrong classification and counting since red blood cells are millions in one smear. Also, cells classification is challenging due to heterogeneous and complex shapes, overlapped cells and a variety of colors. We overcome these drawbacks by introducing a new robust and effective deep Convolutional Neural Network to classify Red Blood Cells (RBCs) in three classes namely: normal ('N') abnormal (sickle cells anemia type ('S')) and miscellaneous('M'). In order to improve the results further, we have used our model as features extractor then we applied an error-correcting output codes (ECOC) classifier for the classification task. Our model with ECOC showed outstanding performance and high accuracy of 92.06%.

Keywords: Classification, Convolutional Neural Network, Cells counting, Sickle Cell Anemia (SCA), Red Blood Cells, ECOC classifier**.**

1 Introduction

Sickle Cell Anemia (SCA), is one of the inherited blood illnesses that described by irregular hemoglobin S (HbS) [1]. It is known as Sickle cell disease (SCD). Due to the oxygen need, HbS molecules are polymerized within RBCs, which influence mainly the RBCs adhesion properties, elasticity, and shape. In addition, the RBCs turn out to be extra fragile and stiff plus extremely heterogeneous shapes in the cell population [2]. This case can be a perfect applicant for morphological heterogeneity examination. These sickle RBCs tend to cluster together, and cannot easily move through the blood vessels and advance vaso-occlusion phenomena. Therefore, SCD patients are suffered from the danger of organ damage, stroke, and serious complications eventually, resulting to reduce average life. There are three main sorts of sickle cell disease. The first one is called Hb SS, when patients inherit sickle cell genes from both parents [20]. The second is a disease is called Hb SC, the patient inherits the sickle cell gene (S) and the second gene (C), produced from an abnormal kind of haemoglobin [21]. lastly, in S-beta thalassemia, the patient inherits one gene of sickle cell and beta thalassemia can be inherited from anemia.

Depending on a recent research [3], the number of people having SCD is about 3.2 million whereas an extra 43 million having sickle-cell symptoms, in 2013. As a result, 176,000 deaths, while in 1990, the number of deaths was 113,000 people, generally of African origin. The main sign of SCD is that the unexpected changing in its clinical severity. The current techniques are mostly helpful and objected at syndrome control, while their drawbacks are the absence of active health status monitoring, and the disease evolutional predicting in various clinical steps [4]. The recent techniques in medical image processing could support an efficient tool in controlling the case of SCD patients. A positive relationship between the number of protrusion and the volume of the cell utilizing X-ray tomography is recently demonstrated by Darrow, et al., [5].

Sickle imaging stream cytometry test, which is a useful technique in evaluating drug effectiveness in SCD, is utilized by Van beers, et al., [6] for obtaining a very precise and sensitive erythrocyte normality classification. Applying automatic, high-output cell classification technique may turn out to be an assisting technology to improve therapy planning, prediction of treatment outcome, and future clinical diagnosis. Conversely, numerous technical challenges are available for automated cell classification, such as; 1- overlapping between RBCs or seem like clusters in the image could lead a difficulty in detecting the cell hidden edges, 2- having low contrast in the RBC area and its background, 3- imaging procedure may also affect the RBCs boundaries and seems blurry, 4- the presence of heterogeneous and complex shapes in SCD, 5- the presence of artifacts like shading, various halos, and dirt on the path of image light, 6- finally, due to the lack of nuclei in RBCs, techniques employing nucleus position as a visible indicator for cell detecting and counting are not valid. In the recent years. Deep learning is a solution for many medical problems [18] and it could handle these challenges of cells classification. Deep learning shows a rapid progress in the field of computer-vision system development, and to solve the difficulties related to different areas in image processing. These difficulties focus on understanding

the images, which involve; label and multi-class classification, medical, pedestrian detection, and spectral [7] [8] [9] [10]. Unfortunately, machine learning algorithms and computer vision have many limitations for example; the need of countless manual tuning for each image, the difficulties in data representation with multi-level abstraction, and the processing ability of a large image data. The solution is a newly developed machine learning algorithm, termed as Deep Convolutional Networks [11]. This solution has the ability to obtain multi-level representation methods using non-linear simple modules first. Then, the simple feature representations are transformed into extra advanced abstract representations by these modules. In other words, the deep convolutional networks employ images as input for learning features, like locations from the pixel array values and edges at certain directions. Then, a combination of these edges has performed at a higher level to learn additional significant abstract features like desirable object components. The final stage is the connection of these components together to form the final objects [12]. Convolutional Neural Networks showed huge success in medical field [18]. In this paper, we propose a deep Convolutional Neural Network approach to classify red blood cells in three classes normal, abnormal(sickle cells anemia)and miscellaneous which includes any other red blood cells diseases such as Discocytes , Elongated , Oval and Echinocytes in blood smear.

2 Methodology

This part listed as following (i) dataset (ii) Data augmentation of Training patches (iii) Structure of our new model.

2.1 Dataset:

The initial stage was to collect a dataset of color images of RBCs. We collected 340 images of RBCs from different standardized websites such as [16, 17].We also collected blood samples from Wadsworth center data. These images are in different shape, color and size. Some images are noisy which make them challenging to classify. Fig.1 shows sample of input image. We cropped cells from all images with the size of 28*28. Total number of patches is 15600 patches of normal, abnormal (sickle type) and miscellaneous. The medical specialist labeled the ground truth of images in three groups Normal, Abnormal (sickle cells anemia) and miscellaneous (any other types of red blood diseases) with help of Labeler application in MATLAB R2018a. This app helps to label data so easily and save the ground truth in '.mat' (MATLAB). It also saves coordinates of cells with description for each cell. 2612 patches are sickle cells anemia type Hemoglobin SS, SC and SB+ (beta) thalassemia, 8016 patches are normal cells and 4972 patches are miscellaneous. At the end, we splitted the dataset into 80% of patches for training, 20% of patches testing set.

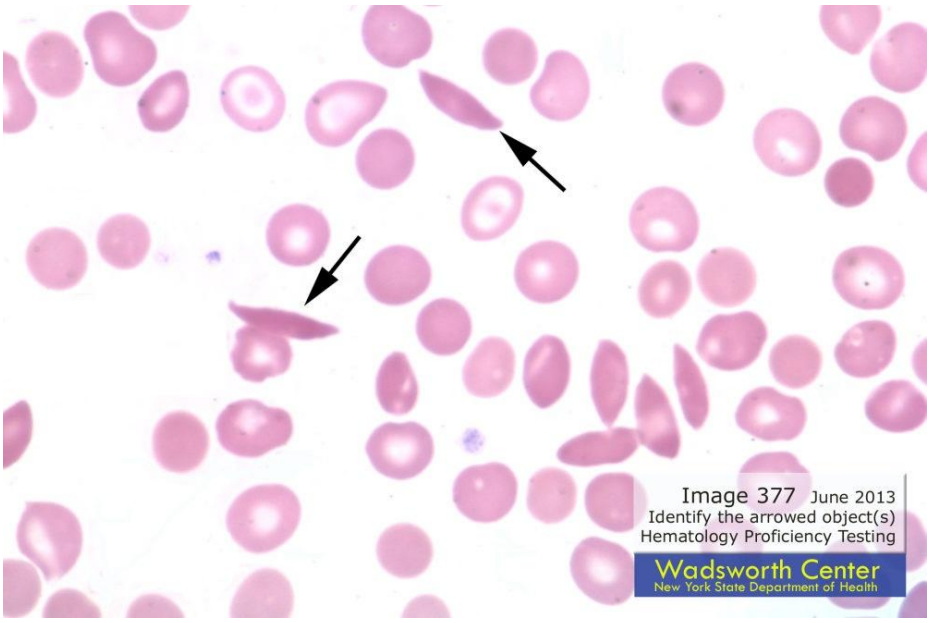

Fig. 1 sample of input image, the arrow points to sickle cells anemia.

2.2 Data augmentation of Training patches

Deep learning networks need a lot of training image data-set due to the big number of parameters, principally weights connected with convolutional layers demanded to be tuned by learning algorithms. Furthermore, collecting a big amount of medical data is costly and difficult task so, we applied data augmentation to enhance the performance of our model. We utilized the set of different image processing methods like flipping, rotation, contrast improvement, using various color models, and random scaling to complete data increment. The rotation is achieved by rotating the image by an angle of 90, 180, and 270. Then, three kinds of flipping (horizontal flip, vertical flip, and horizontal vertical flip) worked on the original patches. With data augmentation techniques, these patches are duplicated 10 times for training.

2.3 Fundamentals of Convolutional Neural Networks (CNNs)

Convolutional Neural Networks (CNNs) have effectively been applied for image classification. It can be classified as a feed-forward, deep Artificial Neural Networks (ANNs). CNNs consist of three main types of layers [11], which are described as follows:

The Convolutional (CONV) layer. Convolutional layer convolves the result of the previous layer with a set of learnable filters [19], where the weights specify the convolution filter. Each filter slides across the width and height of the input volume, producing a 2-dimensional activation map of that filter. The filters have the same depth as in

the input. The size of the output can be controlled by three hyper-parameters which are the depth, stride and zero-padding.
Depth: it is basically the number of filters that are applied to the input image. These filters detect structure such as edges, corners, blobs etc.
Stride: number of pixels the filter jumps while sliding over the image.
Zero-padding: padding zeros around the borders of the input to preserve its size.

Pooling layer. reduces the size of their input and allows multi-scale analysis. Max pooling and average-pooling are the most popular pooling operators .These operators compute the maximum or the average value within a small spatial block. Pooling with filters size of 2 × 2 with a stride of 2 is considered ideal.

The fully connected layers. Neurons are utilized to connect all activations in the previous layer, as obtained in regular Neural Networks (NN). Fully connected layers are typically used as last layer of the network and perform the classification.

2.4 The proposed approach architecture

Our model built based on the idea of Directed acyclic graph (DAG) network. A DAG network type is a neural network for deep learning with layers presented as a directed acyclic graph. A DAG network can have a more complex architecture in which layers have inputs from multiple layers and outputs to multiple layers. This type of networks has a single input layer and a single output layer. It has the great advantage of boosting its depth without drastically boosting its computational cost by making it more "wide", helping not only to boost the details possible to learn but also its accuracy[13,14].In our work, we adopted a deep CNN architecture with 18 layers, including 4 convolutional Layers(CONV1, CONV2, CONV3, CONV4). Convolutional Layers extract features such as shape , color and edges .Every convolutional Layer is followed by Batch Normalization and ReLU which is defined as f(x) =max (0, x). CONV3, CONV4 are concatenated in add layer then fed to average pooling layer to narrow down the features. Fully connected is then to pass one vector features to softmax function as shown in Fig. 2 .A and Table 2. In order to reduce a multiclass problem to a binary classification problem we have applied ECOC classifier. ECOC classifier has shown good results in different tasks [15] .Thus, we have used our model as features extractor by extract features from fully connected layer then we applied ECOC classifier for classification task as shown in Fig.2 B. Classification ECOC is an error-correcting output codes (ECOC) classifier for multiclass learning by reduction to multiple binary classifiers such as support vector machines (SVMs). Train a Classification ECOC classifier using Matlab function (fitcecoc) and the training data. In this work, Classification ECOC used SVM binary learners and a one-versus-one coding design. A one-versus-one coding design technique on our three classes of red blood cells introduces three binary learners of SVMs.

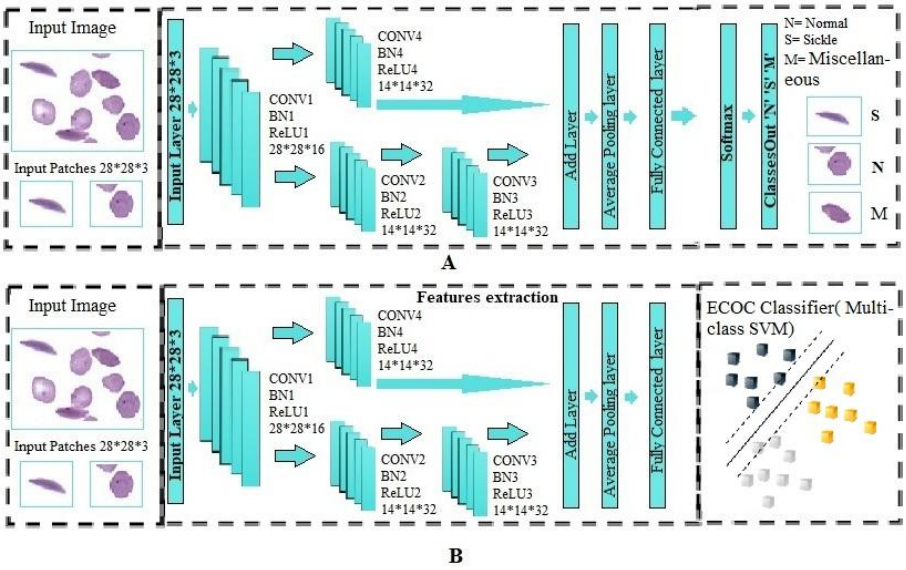

Fig. 2. Proposed Model architecture A) Our Model B) our model with ECOC classifier.

For each SVM classifier has one class is positive and another is negative. Trained Classification ECOC classifiers save the training data, parameter values, prior probabilities, and coding matrices. For ECOC classification, we required to construct a coding matrix as shown in Table 1.

Table 1: coding matrix

	SVM1	**SVM2**	**SVM3**
Class1	1	1	0
Class2	-1	0	1
Class3	0	-1	-1

The outputs of all the SVM classifiers are integrated to predict the final class.Our model has been trained for 100 epochs until the learning stopped. Fig. 3 shows sample of convolution kernels from first convolution layer. Lastly, the processor specifications used in this experiment are Intel (R) Core TM i7-5829K CPU @ 3.30 GHz, the RAM was 16 GB and the GPU was 8 GB. MATLAB script is utilized to diagnose sickle cell anemia in the bloodstream.

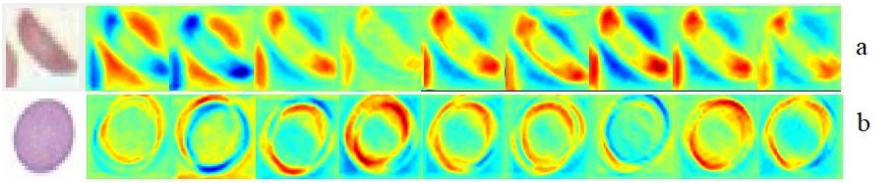

Fig. 3. Sample of convolution kernels from first convolution layer. a) Abnormal cell b) Sickle cell anemia.

Table 2. Model architecture

	Name	Activation
1	Input layer	28*28*3
2	Block 1 CONV1.BN1,ReLU1	28*28*16
3	Block 2 CONV1.BN2,ReLU2	14*14*32
4	Block 3 CONV3.BN3,ReLU3	14*14*32
5	Block 4 CONV4.BN4,ReLU4	14*14*32
6	Addition layer	14*14*32
7	Average Pooling layer	7*7*32
8	Fully connected layer	1*1*3
9	Softmax	1*1*3
10	Class out ‘S’ ‘N’ ‘M’	‘N’ =Normal ‘S’= Sickle ‘M’= Miscellaneous

3 Experimental Results

First, the accuracy of the network was evaluated as the ratio between the number of images classified correctly and the total number images evaluated. We test our model with different images which are different in color, shape and even overlapped cells.

Our model overcame majority of these challenges. Shape factor is the main feature to classify between three classes.

Table 3**.** Classification Results

	Models	Patch-wise%	Image-wise %
1	Our model	83.96	88.11
2	Our model+ECOC.	86.34	92.06

Table 3 reports Patch-wise and Image-wise classification accuracies of our model and our model with ECOC classifier. Patch-wise classification accuracies of our model and our model with ECOC classifier were 83.96% and 86.34, respectively.

In case of Image-wise, our model performance has been measured by classifying multiple patches cropped from each test image first. Then add the results of the classification of all patches throughout a majority voting process, to acquire the final class label for each image. Our model achieved whole image classification accuracy of 88.11%.

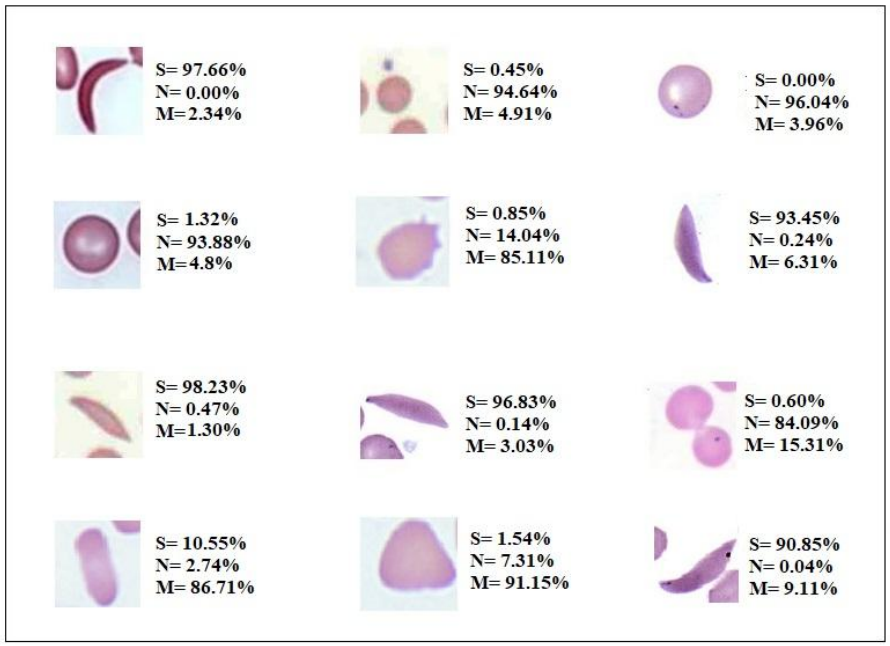

Fig. 4**.** Samples of test patches of our model with ECOC classifier.

Our model with ECOC classifier, on the other hand, achieved accuracy of 92.06%. Fig. 4 shows samples of test patches of our model with ECOC classifier. Overall, our model with ECOC classifier performed well on the classification of red blood patches. After classification process, it will be easy to count number of normal, abnormal red

blood cells (sickle cells anemia type) and other cells individually to evaluate the level of anemia danger.

4 Conclusion

In this paper, we proposed a deep Convolutional Neural Network framework to classify red blood cells into three classes Normal ,Abnormal (sickle cells anemia type) and miscellaneous. We applied ECOC classifier on top of our model in order to enhance the classification accuracy further. The results have shown that our proposed approach in task of classification is effective and robust. It accomplished an accurate prediction of normal red blood cells, sickle shaped cells anemia and miscellaneous. In addition, our approach showed the state-of-the-art performance in term of classification and it is more robust to the different variations in cell size, color, and shape. It also has the ability in discriminating between the noisy blood cells. Lastly our model is currently fine-tuned for only three classes; it will be further tested in the future to include many more classes.

References

1. Anglin C. Sickle Cell Disease. Journal of Consumer Health on the Internet. 2015; 19(2):122±131.
2. Fasano RM, Booth GS, Miles M, Du L, Koyama T, Meier ER, et al. Red blood cell alloimmunization is influenced by recipient inflammatory state at time of transfusion in patients with sickle cell disease. British journal of haematology. 2015; 168(2):291±300.
3. Abubakar I, Tillmann T, Banerjee A. Global, regional, and national age-sex specific all-cause and cause-specific mortality for 240 causes of death, 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet. 2015; 385(9963):117±171.
4. Milton JN, Gordeuk VR, Taylor JG, Gladwin MT, Steinberg MH, Sebastiani P. Prediction of fetal hemoglobin in sickle cell anemia using an ensemble of genetic risk prediction models. Circulation: Cardiovascular Genetics. 2014; p. CIRCGENETICS±113.
5. Darrow MC, Zhang Y, Cinquin BP, Smith EA, Boudreau R, Rochat RH, et al. Visualizing red blood cell sickling and the effects of inhibition of sphingosine kinase 1 using soft X-ray tomography. J Cell Sci. 2016; 129(18):3511±3517.
6. Van Beers EJ, Samsel L, Mendelsohn L, Saiyed R, Fertrin KY, Brantner CA, et al. Imaging flow cytometry for automated detection of hypoxia-induced erythrocyte shape change in sickle cell disease. American journal of hematology. 2014; 89(6):598±603.
7. Araújo, Teresa, et al. "Classification of breast cancer histology images using convolutional neural networks." PloS one 12.6 (2017): e0177544.
8. Alzubaidi, L., et al. "Nucleus detection in h&e images with fully convolutional regression networks." Proc. First International Workshop on Deep Learning for Pattern Recognition. 2016.
9. Albehadili, Hayder, et al. "Fast and Accurate Real Time Pedestrian Detection Using Convolutional Neural Network." The 1 st International Conference on Information Technology (ICoIT'17). 2017.
10. Zeiler, Matthew D., and Rob Fergus. "Visualizing and understanding convolutional networks." European conference on computer vision. Springer, Cham, 2014.

11. Y. LeCun, Y. Bengio, and G. Hinton, “Deep learning,” Nature, vol. 521,no. 7553, pp. 436–444, 2015.
12. C. Szegedy, W. Liu, Y. Jia, P. Sermanet, S. Reed, D. Anguelov, D. Erhan, V. Vanhoucke, and A. Rabinovich, “Going deeper with convolutions,” in Proceedings of the IEEE Conference on Computer Vision and Pattern Recognition, 2015, pp. 1–9.
13. Arenas, Javier O. Pinzón, Robinson Jiménez Moreno, and Ruben Darío Hernández Beleño. "Convolutional Neural Network with a DAG Architecture for Control of a Robotic Arm by Means of Hand Gestures." (2018).
14. Zhou, Jin, et al. "On applicability of auxiliary system approach to detect generalized synchronization in complex network." IEEE Transactions on Automatic Control 62.7 (2017): 3468-3473.
15. Ye, Qixiang, Jixiang Liang, and Jianbin Jiao. "Pedestrian detection in video images via error correcting output code classification of manifold subclasses." IEEE Transactions on Intelligent Transportation Systems 13.1 (2012): 193-202.
16. Homepage, https://www.nhlbi.nih.gov/health-topics/sickle-cell-disease, last accessed 2018/09/01.
17. Homepage, http://sicklecellanaemia.org/, last accessed 2018/09/01.
18. Litjens, Geert, et al. "A survey on deep learning in medical image analysis." Medical image analysis 42 (2017): 60-88.
19. Andrea Vedaldi and Karel Lenc. Matconvnet: Convolutional neural networks for matlab. In ACM International Conference on Multimedia, pages 689–692. ACM,2015.
20. D.1. Weatherall, "The importance of micro mapping the gene frequencies for the common inherited disorders of haemoglobin," British journal of haematology, vol. 149, pp. 635-637, 2010.
21. V. Marsh, F. Kombe, R. Fitzpatrick, T. N. Williams, M. Parker, and S. Molyneux, "Consulting communities on feedback of genetic findings in international health research: sharing sickle cell disease and carrier information in coastal Kenya," BMC medical ethics, vol. 14, p. 41, 2013.