

Survey of Changes in Complete Blood Count and Red Cell Indices of Whole Blood Incubated Different Temperatures up to 48 Hours

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ABSTRACT

The Coulter method accurately counts and sizes cells by detecting and measuring changes in electrical resistance when a particle (such as a cell) in a conductive liquid passes through a small aperture. Each cell suspended in a conductive liquid (diluent) acts as an insulator. As each cell goes through the aperture, it momentarily increases the resistance of the electrical path between the submerged electrodes on either side of the aperture. This causes a measurable electronic pulse. For counting, the vacuum used to pull the diluted suspension of cells through the aperture must be at a regulated volume.

KEYWORDS: *cbc, lymphocytes, neutrophils, monocytes*

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INTRODUCTION

The complete blood count report comprises of many blood report analysis which comes under a speculation test and it is the overall view of the human body that how it is working in the normal and abnormal favorable and unfavorable circumstances. Adaptation of these test and mindset behind doing this project is to analyze the test in the modes of non-temperament situation where a individual can analyze the situation of the body can go under process that is reality based functioned.

HEMOGLOBIN (hbg)

Hemoglobin molecules fill up the RBCs and carries oxygen giving RBCs a red color.

□The RBC indices are:

□MCV □ Shows the Size of RBC

□MCH □ mass of hemoglobin per RBC

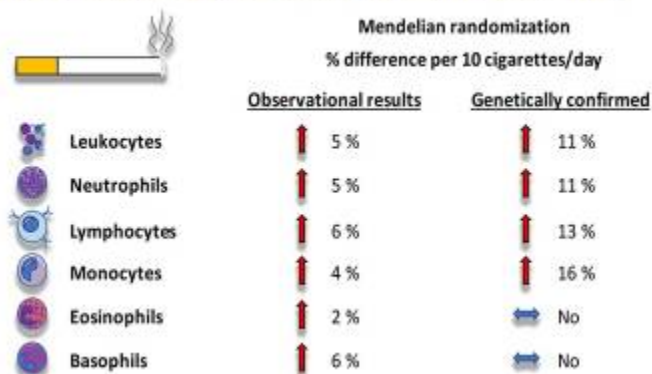
□MCHC □ Concentration of hemoglobin in RBC.

white blood cells

Blood and its components play a vital role in the proper functioning of the human body. The components of the blood can be broadly classified into erythrocytes (red blood cells), platelets and leukocytes (white blood cells). Of the three, WBCs are the primary component which is involved in the body's immune response and account for about 1 percent of the blood.

White blood cells are produced in the bone marrow and are present in blood and lymph tissues. They are further categorized into five groups: basophil, eosinophil, lymphocyte, monocyte, and neutrophil. Each type of WBC has its own significance. Monocytes, breaks down any bacteria that enters the body and tend live longer than the rest of WBCs. Lymphocytes, creates antibodies to fight against viruses like COVID-19

104 607 individuals from the Copenhagen General Population Study



Neutrophils are in larger number and are generally the first line of defense in the human body. They kill and digest bacteria and fungi. Eosinophils fights allergies, carcinogens and parasites. The last type, Basophils are minute and less in number.

clinical significance of wbc analysis

The normal total count of WBC in a person is $4500 - 11000$ per micro litre of blood. Variations in the count of WBCs and the concentration of different WBCs are analysed to determine the presence of an infection. There are many factors which are responsible for variations in the WBC count. For example, the study conducted by Pedersen et.al [4] shows that there is an increase in white blood cells among smoker as shown in figure 1.

The HemoCue^R WBC DIFF System is primarily used to detect these variations in WBC count and differential concentration. Conditions like 'leukopenia' where there is a decrease in total count of WBC can be diagnosed by observing the variation in total WBC count.

In recent times, the outbreak of COVID-19 has created shock waves all over the world. Immunity plays an important role in fighting

hemocue ab and hemocue^R wbc diff system

HemoCue AB. HemoCue AB is a global leader in the field of point-of-care testing. 'It must be possible'-conviction of HemoCue has been their driving force [1], and this has been a great motivational statement for us.

HemoCue^R WBC DIFF System, shown in figure 2 is a device used to measure the total white blood count with a 5-part differential in minutes. The system works in the following way: A droplet of blood is drawn into a microcuvette which is inserted into the device. The HemoCue^R WBC DIFF System gives the total count of WBC and the differential count in minutes

convolutional neural network

Convolutional neural networks are inspired by the neural networks in the human body and have been one of the most influential innovations in the field of

computer science. When we see a dog and a cat, we interpret different features like paws, face and so on to differentiate between the two animals. This interpretation is done with the help of our biological neural networks. In a similar way, the computer is able to perform this classification with the help of a convolutional neural network.

When the input is given to a convolutional neural network, it identifies low level features like edges and curves and when the input passes through its series of layers, and builds an abstract knowledge of unique features in the input. This is just an overview of the convolutional neural networks. More specifics about the convolutional neural networks are discussed in the coming sections.

approaches

In this thesis, we intend to use deep learning techniques for the classification of four different types of WBC (Lymphocytes, Monocytes, Neutrophils, and Eosinophils) and the total count of WBC in a sample.

There are different approaches that can be followed in field deep learning, depending on the data and requirements. We are presenting two different WBC classificational approaches namely, a multi-class classification and one-vs-all classification. Also, we will present two approaches at the architectural level of CNNs namely, multi-channel CNN classification and multi-input CNN classification.

Our intent is to build a model using deep learning techniques and to avoid in the manual computation of features. Also, use the depth of the image stack by using different image planes and make the algorithm more robust.

thesis disposition



Chapter 1 gives a short motivation to the research area. It also gives an introduction stating the aim of the thesis and the different techniques used.

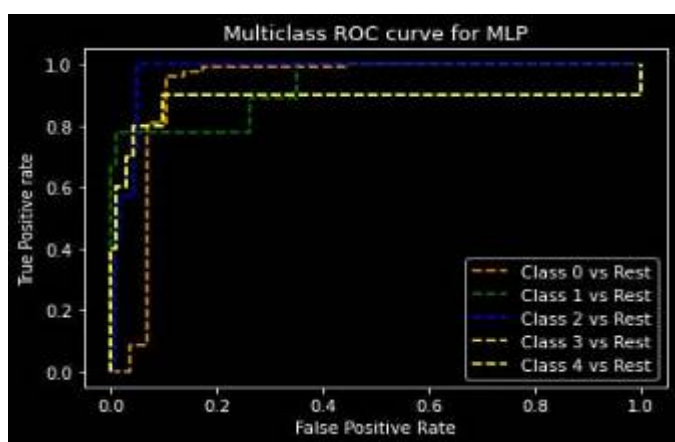
Chapter 2 discusses about the techniques used in WBC analysis and also different approaches that can employed in convolutional neural networks.

Chapter 3 describes the methods used in the analysis of WBC and the design of convolutional neural network used in the thesis. Chapter 4 showcases the results of the thesis and comparison of the results from different techniques used in the thesis.

Chapter 5 discusses what each result shown in the above section mean and also the possible limitation in the thesis.

Chapter 6 a conclusion to the thesis and also possible works that can be done in the future.

The user-friendly GUI provides a wizard-style configuration, scoping the trigger and measured signals. Like every vector network analyzer, it has built in calibration. The GUI has an option for verbose commenting at every measurement start.



The image processing data flow from the instrument is shown in figure 4. The auto focus camera takes the images of the white blood cells in the particular sample at different focal lengths. In the end, the 37 images at the different focal lengths are stacked one upon the other as shown in figure 5.

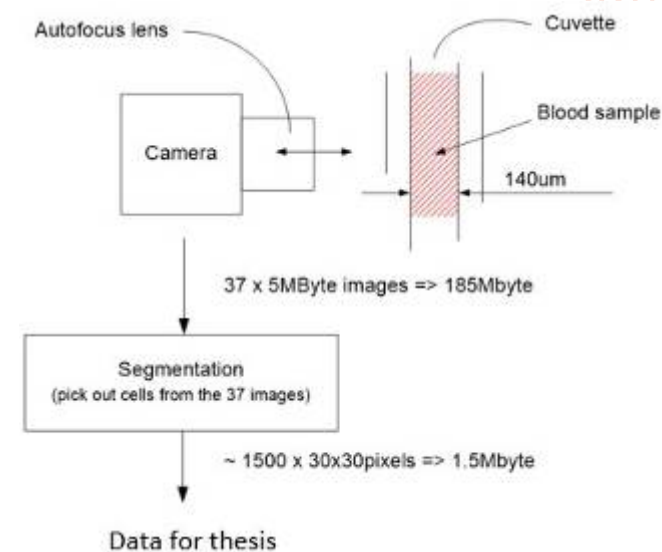
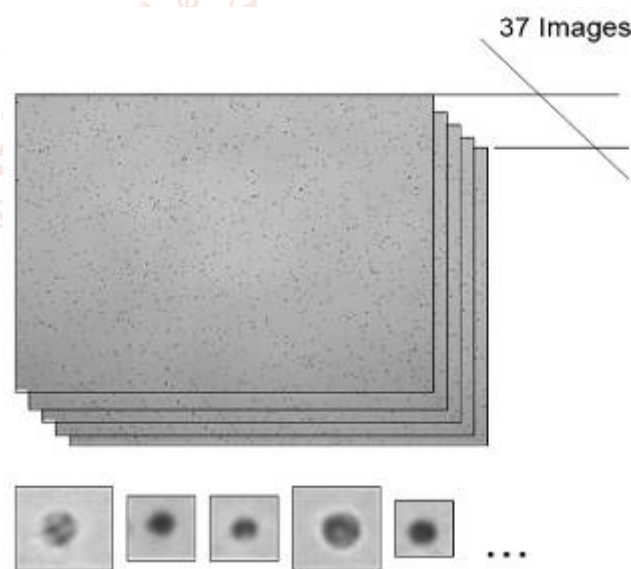
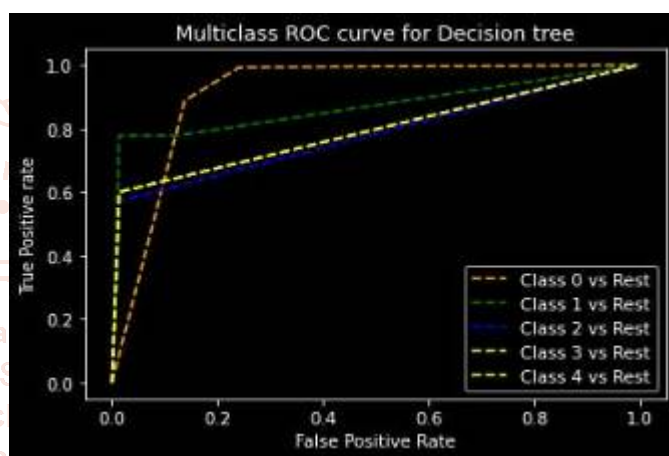


Figure 4: Flow diagram of WBC image processing inside HemoCue^R WBC DIFF System

data collection

The data used in this project was manually labelled by experts, looking at each image and identifying whether it was single, double, triple or trash cell, and identifying cell type. These manually classified images were used for training the different models. 442, 130 WBC images were received, and each such image is a stacked TIFF image with 46 different data planes, out of which 37 images corresponds to the previously described focus planes, see figure 5. Nine additional images, developed by the company, were added to the TIFF stack and they contain processed information regarding the identified WBC. Details of top five image planes in the image stack are: Image plane 1 is a combination of the segmented image and the binary raw image. Image plane 2 is the focused grayscale WBC image.



For every identified WBC in the sample there is one image plane that corresponds to the best-focused image. For all of the identified WBCs in a given blood sample all 37 image planes are stored with an indication of the best-focused plane, as illustrated in figure 6. For the final representation of a WBC, an additional number of planes are created (see next section).

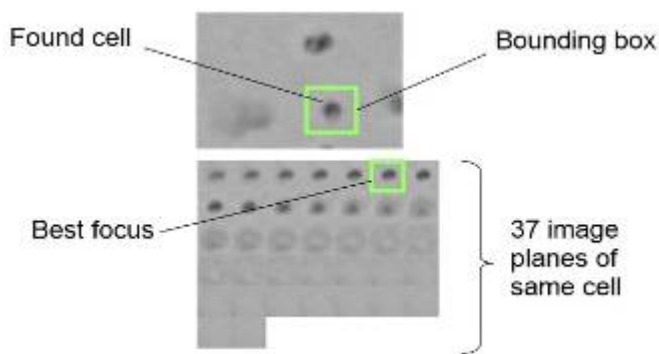


Fig 8 Calculated SSR antenna surface phase distribution

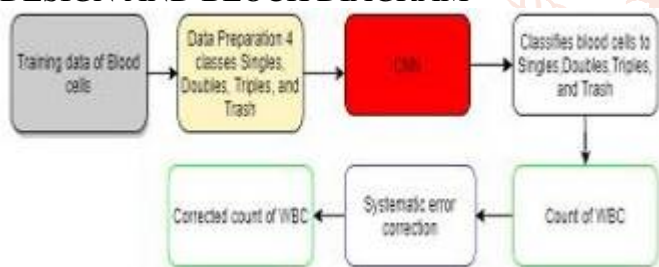
Image plane 3 is the binary mask for the focus WBC image. Image plane 4 is the binary mask for the min-image objects used for labeling of the objects. The surrounding objects are removed. Image plane 5 is the binary mask for the min-image objects used for labeling of the objects. The

Table ¹ shows the details of how ^{442, 130} images are used in for different tasks. In the table the word ‘samples’ refer to the blood samples taken by the company for analysis. Each sample contains images of single cells, double cells, triple cells and trash. The images of single cells further have the labels of monocytes, lymphocytes, neutrophils and eosinophils. The images that are used in systematic error correction is not used training the model. Images in systematic error correction part is first classified into single cells, double cells, triple cells, and trash using our trained algorithm and the total count of WBC is calculated. Total count of WBCs for the same set of images used in systematic error correction is calculated using the Sysmex XN-100 analyser, which is taken as the reference. The images in verification samples are used for final testing our final model. Hence, these images are not used in training of the models.

TYPE	DATA	IMAGES
	Train	17551
WBC identification	Validation	3683
(Single, Double, Triple, Trash)	Test	3692
Total data for WBC identification		25106
	Train	5459
WBC differential	validation	1157
classification(Single cells)	Test	1157
Total data for WBC identification		7773
Systematic error correction	Samples (¹²⁰)	204654
Verification	Samples	204777
Total thesis data		442130

Table 1: Description of the data received.

DESIGN AND BLOCK DIAGRAM



When the images of single, doubles, triples and, trash are observed, identification of a single cell amongst others is easier. The identification doubles, triples and, trash require the neural network to learn more features. Also, it is important to achieve high accuracy in classification of single cells since they are further used in differential classification of WBC. Also, authors Rifkin and Klautau in [15] have shown that the OVA approach have given better results in a multi-class classification problem. Hence, we are separating singles and classifying them against the rest of the three classes by grouping the double cells, triple cells and, trash as one class.

This design approach involves two different convolutional neural networks. One convolutional neural network is built for the classification of single white blood cells and others. The other neural network is built for the classification of doubles, triples, and trash. Further on the test samples these two neural networks work together to give the final count of the WBC. Figure 20 depicts the design of this approach.

Systematic error correction

There is a pessimistic impact on the total count of WBC that is calculated. This impact is due to the loss of data that occurs when the cell falls around the edges in an image from a sample. Images with the incomplete image of a cell is not considered for the total count of WBC which impacts the final count of WBC calculated from our algorithm. Hence, we are proposing a systematic error correction method for the total count of WBC.

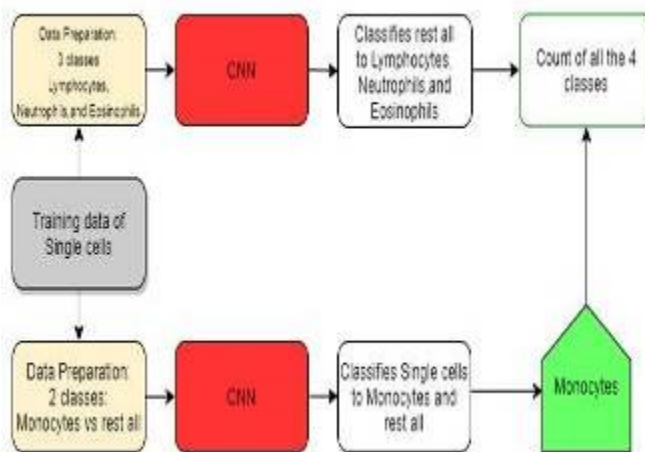
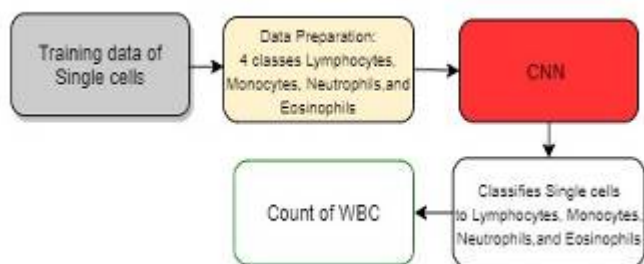
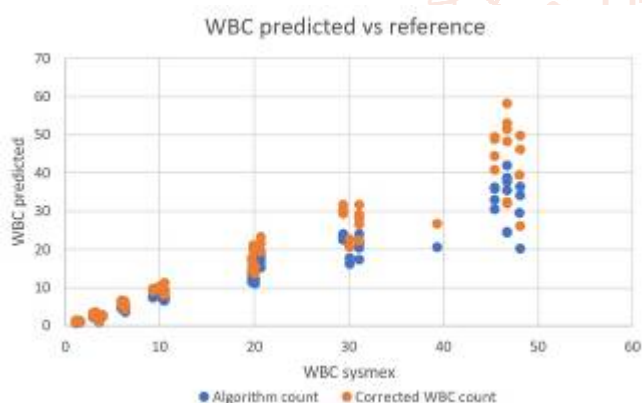
A part of the data set (around 120 samples) is taken and the total count of WBC in those samples is

calculated using an analyzer "Sysmex XN-1000" from Sysmex Corp. This count is taken as the reference. The total WBC count for the same samples is calculated using our proposed algorithm. The resulting scatter plot (figure 20) can then be used to derive a correction that depends on the estimated WBC count.

The systematic error correction is found by fitting a polynomial, see equation 2, to the scatter plot. A third degree polynomial equation gave us better count which is near to the reference count. A scatter plot of predicted WBC count against the reference WBC count before and after systematic error correction is plotted as shown in figure.

WBC differential classification

In this part, different types of white blood cells are classified. There are five major types of white blood cells. Basophils are a type of WBC whose presence in the human body is only around 0.5% to 1%. This poses a challenge to collect sample data from them. Hence, the four classes that we are classifying are - Neutrophils (figure 11), Lymphocytes (figure 13), Monocytes (figure 14), and Eosinophils (figure 12).



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