AUTOMATED BLOOD GROUP DETERMINATION USING IMAGE PROCESSING TECHNIQUES WITH INTEGRATION OF RASPBERRY PI

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Abstract: **Blood group detection is a vital procedure during conducting any transfusion of blood during critical transplants or emergencies. Currently, blood group determination tests are performed under standard laboratory conditions by experienced technicians. A novel method employing the technique of Image processing is used in this research paper to deduce blood group with less consumption of time and elimination of personnel error. Clear pictures of the blood samples which are tested are taken from the laboratory. These pictures of the blood samples are captured after the addition of commercial antigen reagents, and are processed using image processing techniques such as Preprocessing, Thresholding, Morphological operations, HSL Luminance plane extraction and Quantification. The blood group is determined by looking for agglutination in the sample image on reaction with a specific reagent, and calculating the intensity variations of the pixels.**

This procedure can find its application as a self-testing device that does not require any technician supervision, thus reducing the clinical exposure of patients, and reducing human-to-human contact which is of utmost significance in situations where these could be further detrimental to the health of the patient like in the ongoing COVID-19 pandemic.

Keywords — Blood phenotype, Agglutination, Antigen, Image processing, OpenCV, Raspberry Pi, Pandemic.

I. INTRODUCTION

A. Blood and It's Types

A blood group which is also called blood type is the categorization of blood depending on the antigens. These antigen particles are present on red blood cells' (RBC) surface. These antigens may consist of proteins or carbohydrates or glycoproteins or glycolipids. Detection of blood group is done by studying the antigens and antibodies in the blood. Antibodies which are also called immunoglobulin are proteins which are produced inside the body that helps in fighting against foreign particles. These are called antigens. Antigens that cause disease or infection include bacteria, viruses or fungi. When infection causing particles i.e. antigens enters the human body, the immune system is activated to develop antibodies to fight against the antigens or infection causing particles. These antibody fixes to antigen surface to deactivate them so the antigens would not affect other body activities. Blood grouping systems are:-

- ABO blood group system
- Rh blood group system

The **ABO blood group system** is the most rudimentary and basic blood group system used during human blood transmission. Anti-A, anti-B antibodies are usually grouped under IgM i.e. Immunoglobin M. ABO blood group system is done to check if the patient has A blood group or B blood group or AB blood group or O blood group. These are the four prime blood groups are distinguished on the basis of A & B Antigens, whether or not present on the surface of the red blood cells (RBC), i.e :

- A blood group contains the antigen A on red cells and antibody B in the plasma.
- B blood group contains only the antigen B on red cells and antibody A in the plasma.
- AB blood group contains both antigens A and antigen B on red cells and none of the antibodies in the plasma.
- O blood group contains no antigens on red cells and contains both antibody A and antibody B in the plasma.

Antigen A and antigen B are present in AB group but no antibodies. As there are no antibodies they are compatible with any group of blood. Hence they are called as universal recipient. O blood have none of the antigens, which makes their blood resistant to be agglutinate with any antibodies, Hence they are called as universal donor.

The **Rh blood group system** determines whether or not the Rh antigen is present in the blood. This is usually called Rh factor. These are present on the cell membranes of the erythrocytes.
When a person with Rh negative types is transfused with Rh positive type, it causes a fatal risk. There will be no noticeable effects the first time of transfusion. But the immune system of a human retaliates to the foreign Rh antigen. Then it initiates in producing Anti-Rh antibodies. And now if Rh-positive blood is transfused a second time after the formation of antibodies, and these react with the Anti-Rh, triggering them to form agglutination or clumping. The resulting destruction of the red blood cells, called hemolysis causes major illness and can even lead to death.

B. Blood Phenotypes

Blood group is determined by the presence or absence of three antigens A, B and O on the surface of red blood cells. Below are the pictorial representation of all the different blood groups.

1) O positive group

The below figure shows a sample of O+ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 1: O+ve blood group

2) O negative group

The below figure shows a sample of O-ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 2: O-ve blood group

3) A positive group

The below figure shows a sample of A+ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 3: A+ve blood group

4) A negative group

The below figure shows a sample of A-ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 4: A-ve blood group

5) B positive group

The below figure shows a sample of B+ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 5: B+ve blood group

6) B negative group

The below figure shows a sample of B-ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 6: B-ve blood group

7) AB positive group

The below figure shows a sample of AB+ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 7: AB+ve blood group

8) AB negative group

The below figure shows a sample of AB-ve blood group when tested with Anti-A, Anti-B and Anti-D respectively.

Figure 8: AB-ve blood group

II. LITERATURE REVIEW

A. Paper Review

Determination [1] of the blood group was done by converting the image into the grey Scale and then local binary pattern (a computer vision technique) was used with Nearest Neighbor search for classification. The image [2] acquired from the camera from the slide was involved in processing on MATLAB software using several image processing techniques. As in, Pre-processing techniques, Thresholding, Morphological operations, HSL Luminance plane, Quantification. Later from the processed image, the pixel deviations were compared to determine the agglutination. This paper [3] uses an image of centrifuged blood sample and the reagents. Then the morphological operations are done and to classify pattern matching and geometric matching is used. In [4] the system is developed in Visual studio using the techniques of image processing and open CV methods. The user interface is developed using open CV and the implementation of identification of the blood group is processed using image processing techniques. Determination [5],[6] was done using the principle of spectrophotometry. The sample was placed under a monochromatic light and the optical densities obtained from each of the samples were compared and the agglutination was found out.

B. Existing Methodology

In the current existing system of blood group determination, a manual method by adding commercial reagents or monoclonal antibodies which are also called antisera, such as anti-A, anti-B, anti-D to the blood sample. The blood samples are divided into three parts and each reagents are added. After some time, it is observed whether agglutination has taken place or not.. On this basis, blood group can be distinguished by the lab technician manually.

Say, for example, the subject with B antigen when mixed with anti-B antibodies, clumping will be observed. If there are no clumping for anti- A or anti-B reagents, then it is said to be blood group O. A course of tests using various types of antibody solutions can be used to distinguish the blood groups. When the subject has to undergo a blood transfusion, the recipient blood will be tested with the donor blood sample that contains both ABO and Rh antigens. If there are no visible reactions, donor blood with can be safely tranfused. If there are any agglutination reaction observed, it indicates that is is not safe to use that particular donor blood for. Although it must be noted that, only experts can surely classify the blood type by seeing at the agglutination process alone.

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III. PROBLEM STATEMENT

A. Improvement of Accuracy

The conventional methods performed in the laboratory have chances of false classification due to human errors, but in a computerized method, once the algorithm is fed to the system the chances of errors are significantly reduced.

B. Determination and Classification of Blood Type.

When the blood sample is left to react with the antisera Anti-A, Anti-B and Anti-D, clumping can be observed where contrasting antibodies are present. The blood group is detected by the algorithms by extracting features from the images of these agglutinated or non-agglutinated sample.

IV. OBJECTIVE

- Determine the A, B, AB, O blood group of the sample.
- Perform the Rh phenotyping for detecting the presence of antigens in the blood sample.
- Develop a portable electronic design for self-test of blood grouping without any contact from any of the technicians, so as to adhere to all the non-contact protocols of the ongoing COVID-19 pandemic.
- Reduced number of clinical visits as simple diagnostic tests such as blood group detection can be performed from the home of the patient.

V. METHODOLOGY

A. Proposed Methodology

Two sets of images are considered, Set One contains images of the blood sample with the added reagents taken from database and Set Two consists of images of real blood samples taken from the laboratory. In both the sets, the sample blood is divided into 3 parts and each is mixed with one of the three antibody reagents- anti-A, anti-B for A group, B group, O group of blood grouping and Anti-D for Rh blood grouping. To the fourth part of the sample the Control Reagent is added. The Control Reagent is a qualitative negative control for the primary antibodies. This ensures a negative or null response to the test. It is used to test the validity of an experiment. Images of these mixtures is captured using a camera. These images are then fed to the Raspberry-Pi module for image processing. The image processing techniques employed for classification are Preprocessing, Thresholding, Morphological operations, HSL Luminance plane extraction and Quantification.

The functions of each of these can be elucidated as:

- **Color plane extraction** is performed to extract the maximum value in RGB color plane, as the image has different foreground colors.
- **Thresholding** is a uncomplicated image segmentation method of image. If the image intensity is more than the set value, all those pixels are converted or replaced with black pixel.
- **Morphological operation** includes pre- and post-processing operations which are erosion dilation, granulometry and morphological filtering etc.
- **HSL** where stands for Hue, S stands for Saturation and L stands for Luminance. Hue is a degree of color shown aroun the wheel of colors, saturation are set as a percentage of color saturated. The luminance plane is extracted from the image during this process.
- **Quantification** is the main technique to determine the blood group. The intensity is measured only at the area of interest region of the image.
- Both extremities of pixel intensity values are determined by plotting **histograms** for the images. And area, mean and standard deviations are calculated for the obtained intensity values. Occurrence of agglutination is identified by standard deviation values.
- The surface examined for full image is expressed in the form of percentage which is area.
- The average value of the pixel is mean.

B. Block diagram

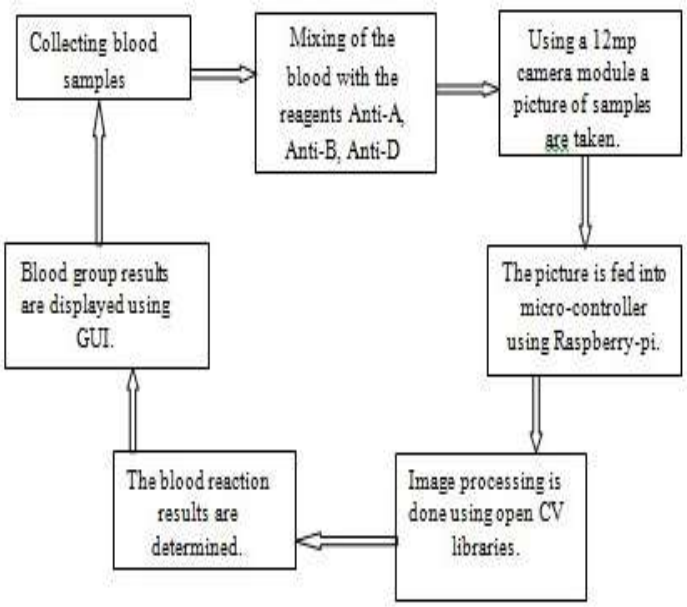

Figure 9: Block Diagram

C. Raspberry Pi Setup

Raspberry Pi was installed with Raspbian OS and it was displayed virtually using VNC server. Raspberry pi and VNC server was connected using a unique IP address.

Figure 10 : Raspberry Pi connection

VI. OUTCOMES

The images of the four sub-samples from Set One before processing can be seen in Fig.10.

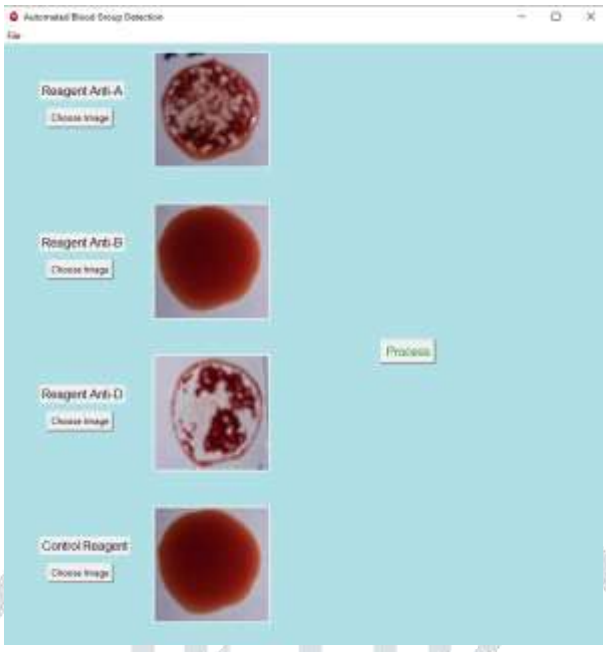

Figure 11: Sub-samples before processing

The green plane of the captured images is then extracted for all four sub-samples. The results of this stage, can be seen in Fig.11.

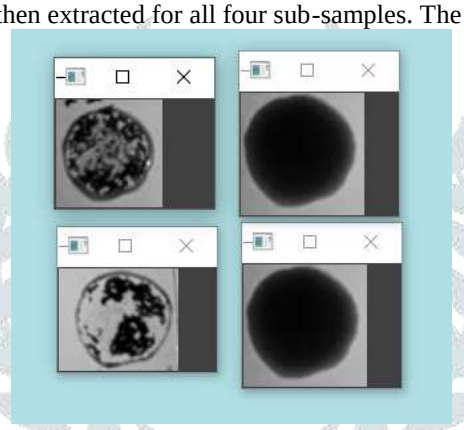

Figure 102: Green Plane extraction

Automatic thresholding is a widely used image processing technique. It is used to extract relevant information which is encoded into pixels of the image also significantly minimizing the background noise in the image. This is achieved before converting the original gray scale image to binary by employing a feedback loop for optimized threshold value The objective of this is to divide the images into two distinct parts, namely the foreground and background. This is done in a step-by-step manner using the following algorithm:

- Initial value of threshold is assigned. Conventionally, mean of the 8-bit value of sample image is used.
- Bifurcate the sample image into two regions;

    a) The background region is formed by the pixels whose values are either equal to or less than the threshold.

    b) The foreground is formed by the pixels whose values are higher than the assigned threshold value.

- Calculate the mean of the pixel values for the two new regions formed.
- Enumerate an updated threshold value by calculating the average of the means.
- If the difference between the two threshold values falls below a pre-defined limit, the process is over. Otherwise, the process must be repeated by applying the updated threshold to the image.

On performing auto-thresholding on the samples, the following results are obtained.

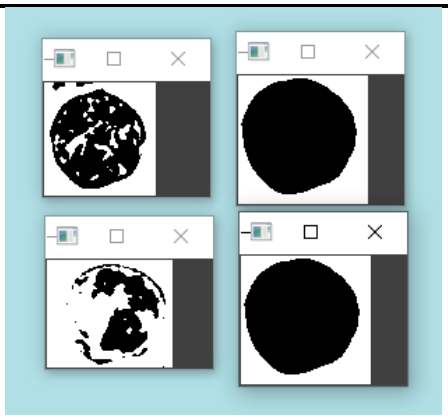

Figure 13: Auto thresholding

In auto-thresholding, the threshold values of image pixels are same for all. But, in Adaptive thresholding, the threshold value is enumerated for all the sub-regions which are smaller. So, different regions have different threshold values. This more intricate rendition of thresholding can assimilate dynamic lighting conditions in the image. The results after performing adaptive thresholding on the samples can be seen in Fig.13 below.

Figure 14:Adaptive thresholding

As a next step, Morphological operations are performed on the image. The main objective of performing morphological operations is to give a structural skeleton to the input image. This is used to create an output image which is of the same structure. In the output image, the value of every pixel is contingent on a collation of the neigbouring pixel in the input image and it's surrounding pixels as well. The most fundamental and eminent morphological operations are said to be dilation and erosion. Dilation is done by adding pixels to the boundary of objects in the image, while erosion removes pixels from the boundaries of objects. The number of pixels that must be appended or discarded from the objects is determined by the shape and length of the structuring element. The outputs of the morphological operations and advanced morphological operations can be seen in Fig.14 and Fig.15 respectively.

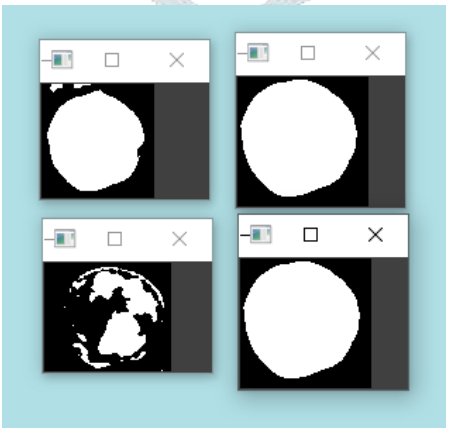

Figure 15: Morphological Operations

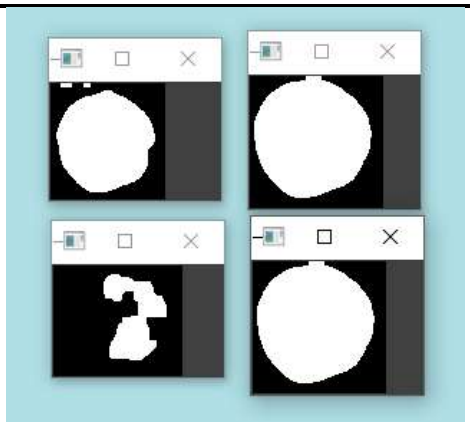

Figure 16: Advanced Morphological Operations

Moving ahead, a histogram is plotted. In image processing, the histogram of an image refers to, plotting of intensity values of each pixels of the image on a graph. Meaning, it is a graph showing the number of pixels in an image at each different intensity value found in that image. The histograms for the image intensity values can be seen in Fig.16.

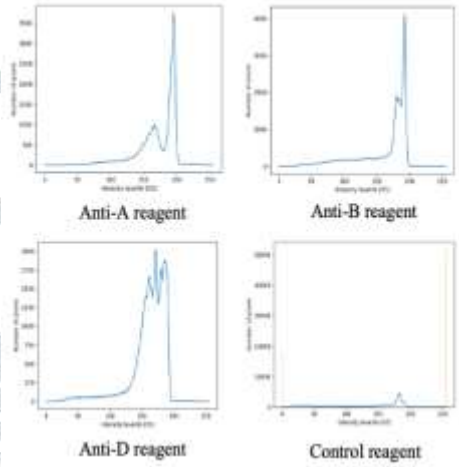

Figure 17: Histograms of pixel intensity values

As a next step, Quantification is performed. This is the primary technique to ascertain the blood group. It quantifies the intensities of pixels which are present only in the region of interest. Area, mean, standard deviation, minimum and maximum values of pixel intensity are deduced. Occurrence of agglutination is identified by standard deviation values. The output from this stage can be seen in Fig.17.

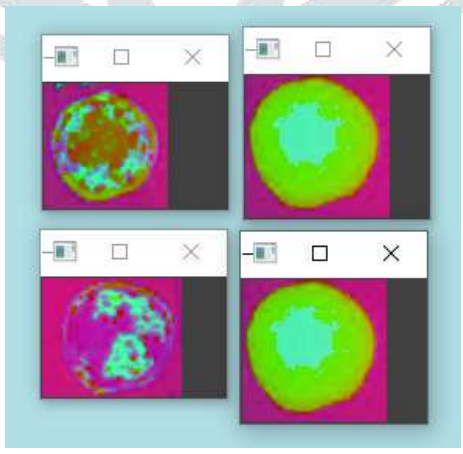

Figure 18: Quantification

The processor is then trained to compare the pixel intensity values and give the results.

Accurate determination of the blood group with Rh factor is performed by integrating image processing to Raspberry Pi. The results of the sub-samples after image processing can be seen in Fig.18 below.

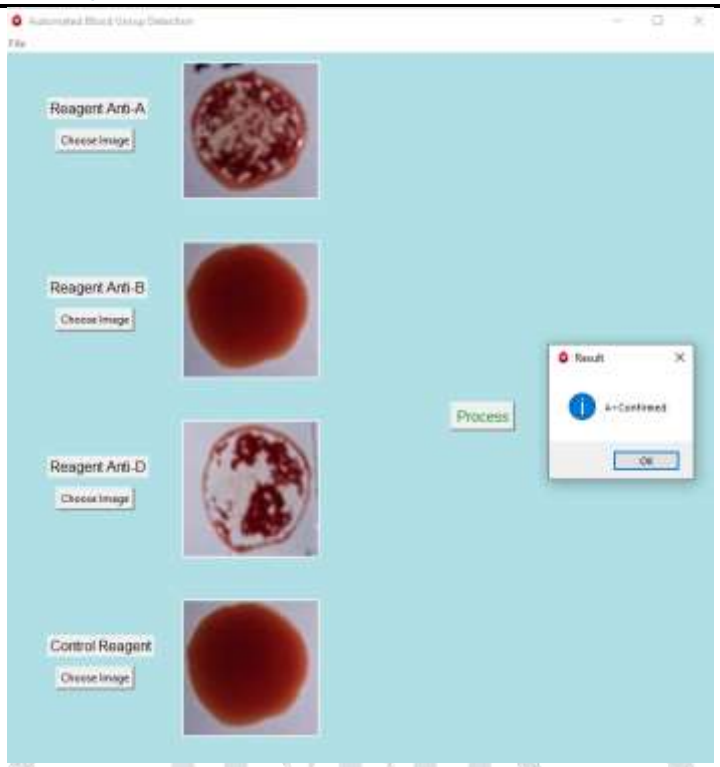

Figure 19: Sub-samples after processing

After processing, it is found that the sample is A positive.

This process is then repeated on another sample, the sub-samples before image processing can be seen in Fig.19.

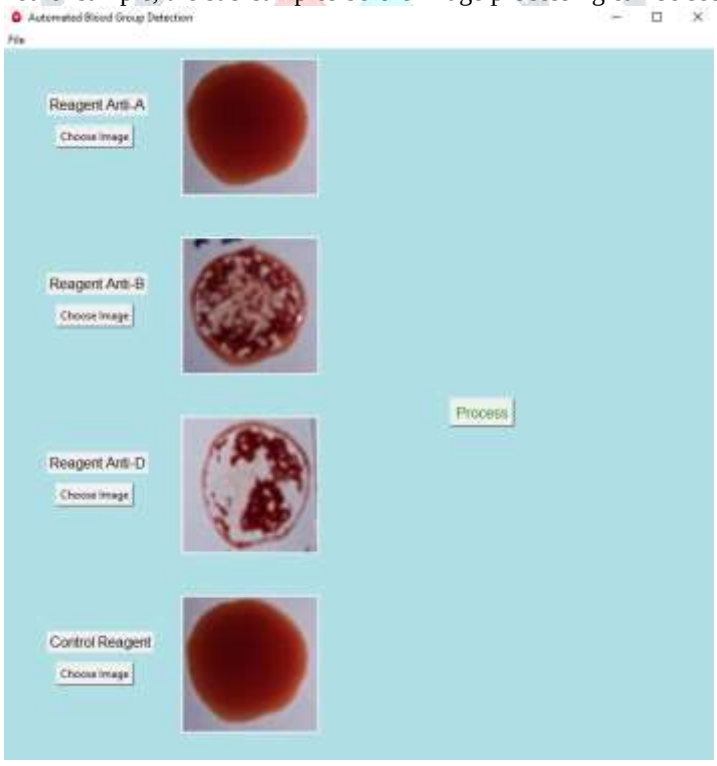

Figure 20: Sub-samples before processing

The sub-samples after processing can be seen in Fig.20.

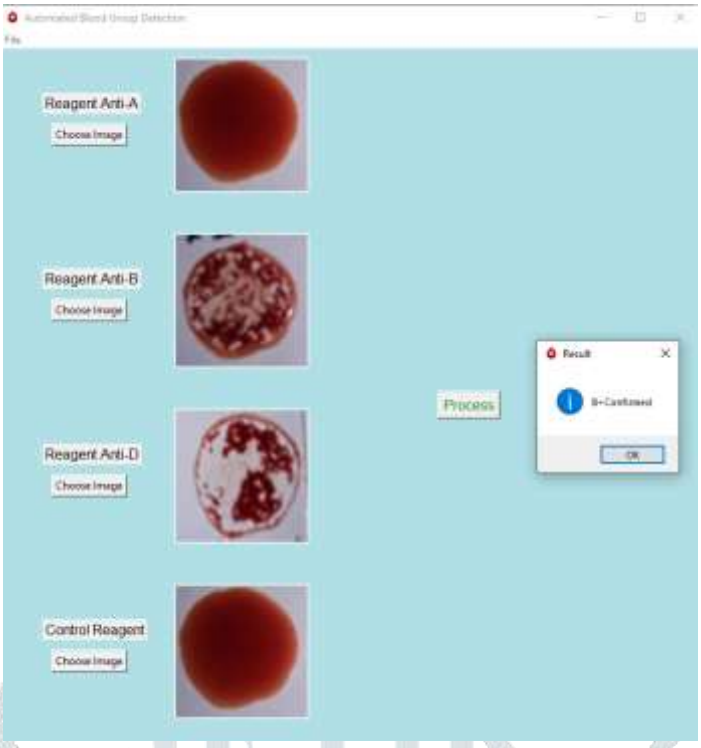

Figure 21: Sub-samples after processing

From the above image, it can be confirmed that the sample was B positive, and similarly other blood groups are also confirmed.

Correspondingly, the same process in carried out on samples from Set Two, outputs of which can be seen below.

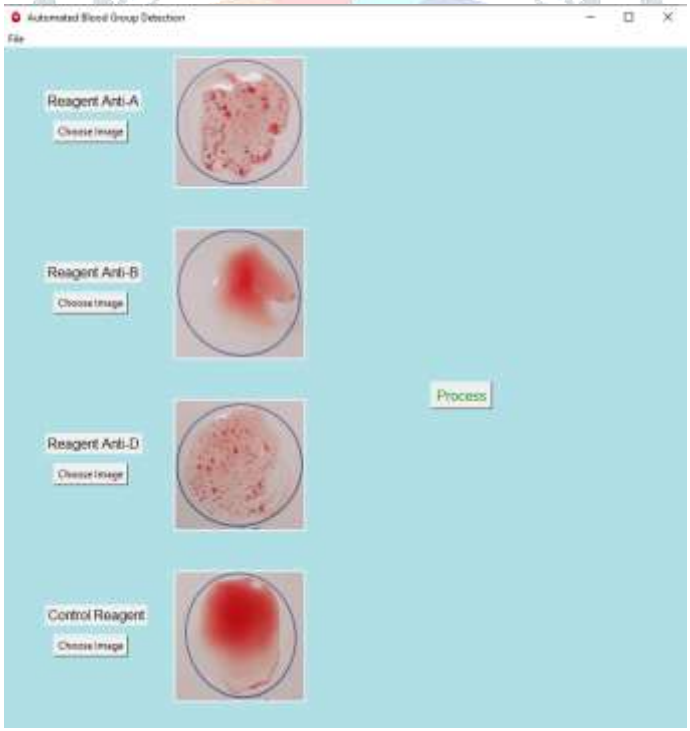

Figure 22: Sub-samples before processing

The output after processing of the samples can be seen in Fig. 29.

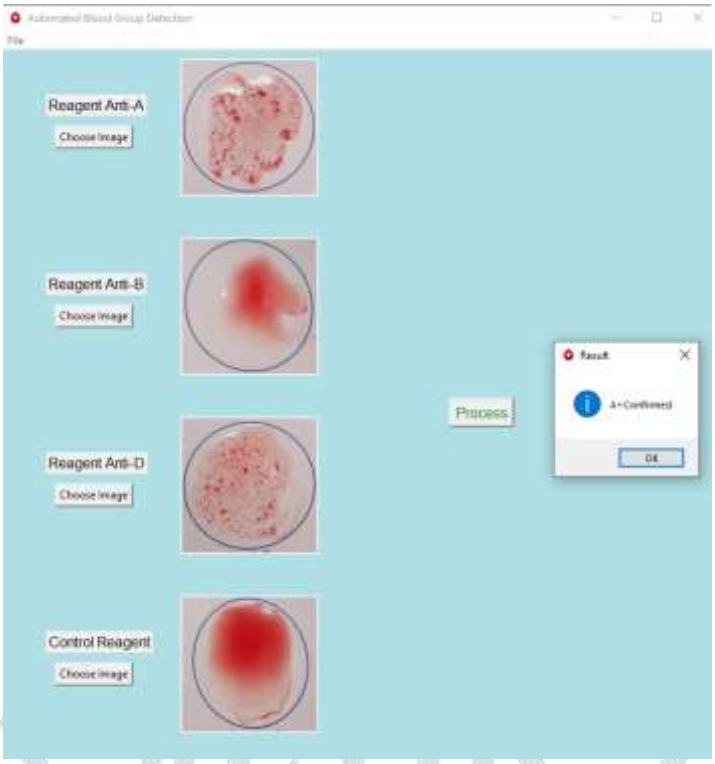

Figure 113: Sub-samples after processing

From the above image it can be observed that the given sample is A positive. The above steps are then repeated on other samples, and all the different blood groups are confirmed in a similar manner.
This method provides the benefits of less time consumption and decreased involvement of human resources compared to the traditional method of detection allowing non-contact blood grouping, which could be vital in curbing the spread of contagious diseases such as the COVID-19 pandemic.

VII. RESULTS AND TABLE

The images of both set one and set two produced different standard deviation values. These standard deviation values were analyzed and particular threshold values were set accordingly. The below represented table 1 gives all the specific details of these values.

Set no.	Standard Deviation Values				Threshold Values
	Anti-A	Anti-B	Anti-D	Control	
Set One	35.720	53.760	23.447	53.760	40
Set Two	577.916	593.391	570.359	670.394	580

Table 1 : Standard Deviation and Threshold Values

I. CONCLUSION

The proposed system enables to perform blood grouping test outside the conventional pathological laboratory and this system does not require any trained technician for conduction of the test. It is a simple and averts common problems such as reagent dilution, blood separation and incubation. It saves both money and time. The results obtained are more accurate without any chances of human errors. Quick results lead to improved quality of healthcare services. From the above mentioned advantages and features, this system results to be highly effective and allows better health service quality, bringing forth a novel proposition for the blood grouping.

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