

AN EFFECTIVE AUTOMATIC RECOGNITION METHOD OF SICKLE CELLS IDENTIFICATION IN THE BLOOD SMEAR

Abstract

Red blood cells carry oxygen and transport it to all body parts through a connective tissue known as blood. One of the abnormalities of RBC is anemia, known as sickle cell, and it is one kind of blood disorder. An RBC usually is in a disk shape, but it changes its disk shape due to improper hemoglobin protein content to form a sickle cell structure, and it is a disease. In this disease, the red blood cell shape is changed to crescent-shaped, called sickle-shaped. In India, specifically in Chhattisgarh, it is a deliberate problem of disorder very much common among backward classes, including scheduled tribes and scheduled castes. This disease is the highest percentage in the surname Sahu, Dewangan, Gond, Mahar, Halba, and Kurmi. Among various methods of blood cell count, manual counting is the most commonly used method. But manual counting mainly depends on the technical person. Therefore, an image processing-based effective object detection method to automatically detect and count the sickle-shaped cells present in the blood smear is proposed in this paper. The proposed method gives 99.96% accuracy for sickle cell counting compared to manual and other existing counting methods.

Keywords: Sickle cell, image processing, and feature extraction

Author

Dr. Amit Kumar Biswas
Department of Computer Applications
Bhilai Institute of Technology
Chhattisgarh, India.

I. INTRODUCTION

Blood has many types of cells that are connective. Therefore, the number, as well as shape analysis of red blood cells (RBC), can provide important information which provides a facility for recognizing some illnesses such as Sickle cell anemia etc. In this disease, the body transforms the hemoglobin protein in RBC, through which the oxygen circulates in the whole body. The source of these abnormalities is genetic, which generates a deformed crescent-shaped or sickle-shaped RBC. Sickle-shaped cells are sticky and rigid, forming a cluster that gets stuck in the blood vessels and clogs the blood flow. Different method has been developed over time for RBC and sickle cell recognition and counting.

For separating WBC from the RBC threshold method is used after getting the segmented image for feature detection circular Hough transform approach is most commonly used [1]. Detection of Abnormal blood cells findings in Human RBC for the diagnosing sickle cell anemia by using image processing [2] and another method can detect overlapped sickle cells[3].Automatic counting of the red blood cell and sickle cell using edge detection [4], watershed segmentation, and Hough transform for circular overlapping detection and counting are used [5]. In addition, the clustering-based segmentation technique is used to identify sickle-shaped cells and RBC cells present on microscopic slides [6].Automatic detection of sickle cells from the blood cells by separating the RBC from the digital image of blood cells [7].

One proposed elliptical matching approach to detect elliptical and circular objects [8]. Another researcher used morphological operations to properly segment and detect the cells, and then the artificial neural network was applied to separate the overlapped cells [9]. A semiautomatic technique for overlap cell segmentation process [10]. This technique needed user interaction to see the beginning points before the death penalty, the k-means algorithmic rule to spot the borders of the cells. Proposed Hough Transform-based automatic processes of segmentation, recognition, and calculation of the RBC in the microscopic images have been developed [11]. Used the Hough transform technique for RBC counting and the morphological approach for segmentation [12]. Proposed an automated detection system to count the fetal and maternal RBC [13]. Proposed a method to automatically classify the RBC into overlap, normal and abnormal clusters[14]. Presented the segmentation of blood cells by using the watershed, edge detection, laplacian of Gaussian and Otsu thresholding approaches and then detecting the abnormality of the blood cells as sickle cells by labeling method [15]. Automatic counting method of RBC and WBC on the grayscale image by thresholding[16].

This paper proposed an algorithm of feature extraction and morphological operation to highlight and distinguish the sickle cell from the normal red blood cell image. The following section 2 explains the proposed method. Section 3 discusses the experimental setup. The result in section 4 discusses the experimental results. In conclusion, section 5 details how the automatic method is better than other existing methods.

II. PROPOSED METHODOLOGY

The image processing technique has developed an automated process to recognize and count sickle-shaped cells in blood smears. The proposed study is an application area of image processing that helps to make the task easy, refined, and in a form that has been applied to achieve the desired result quickly and accurately.

1. Proposed algorithm

The working steps of the proposed method are as follows:

Step 1: Input a microscopic digital image.

Step 2: A pre-processing operation to convert the digital image to a grayscale image and then enhance the grayscale image by fuzzy enhancement process[17].

Step 3: Converting the fuzzy enhances grayscale image to a binary image.

Step 4: The next pre-processing operation, perform the filling process to fill all the hole regions.

Step 5: The next pre-processing operation, perform the noise removal process to remove the unwanted regions like dots and small regions.

Step 6: Apply the morphological operation as an erosion followed by dilation to remove the connected sickle and/or red blood cells.

Step 7: Detect and recognize the different regions.

Step 8: Extract the features of the recognized regions to separate the regions into the red blood cells and the sickle cells.

Step 9: Count the sickle cells and total number of cells.

The above processing steps of the proposed automatic system are integrated into six major processes shown in Figure 1.

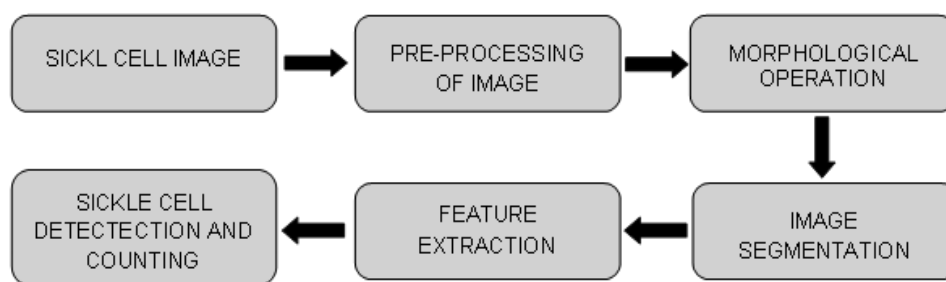


Figure 1: Sickle Cell Detection and Counting Steps

The above six major processes, shown in Figure 1, are described in the following subsections:

2. Sickle Cell Image: The glass slides of the blood sample smear taken in the laboratories are used as an image for processing. The microscope is attached to a computer to transfer the microscopic images and convert them into digital images using a digital camera. The picture quality and the resolution of the generated digital image of blood samples mainly depend on the type of digital microscope used. Figure 2 shows a digital blood sample image.

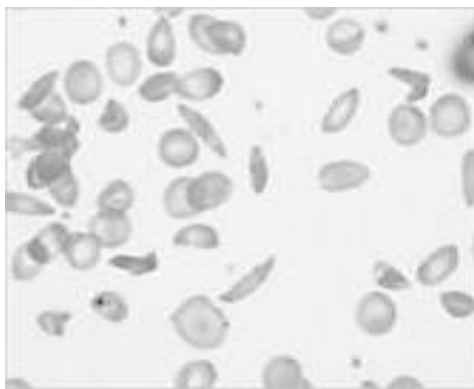


Figure 2: Original Microscopic Image

3. Pre-processing of Image: The second process, which removes the noise from the image, is known as the preprocessing step, and the outputs are shown in Figure 3(a)-(e). This process is divided into six different sub-processes. The colored image is converted into a grayscale (0-255) gray level in the first sub-process, shown in Figure 3(a). In the second sub-process, the brightness of an image is adjusted. In the third sub-process, produce the complement image of the previous sub-process image shown in Figure 3(b), in which subtract each pixel value from the maximum pixel value of that image, and the difference value of that pixel is used as the output image for the fourth sub-process. The fourth sub-process enhances the image's contrast, as shown in Figure 3(c). The fifth sub-process discarded the object present at the image's border, which does not contain any information, is shown in Figure 3(d). Finally, the binary image for the blood smear is generated in the sixth sub-process, as shown in Figure 3(e).

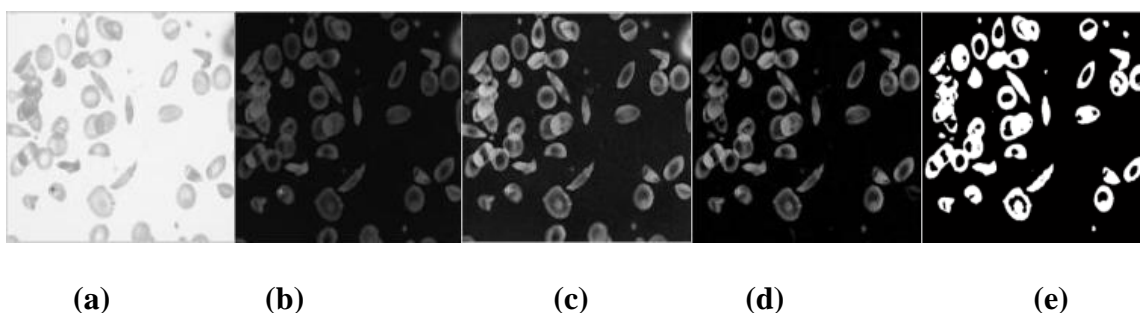


Figure3: Preprocessing Process:(a) Grayscale image, (b) Complemented image, (c) Contrast-Enhanced image, (d) Clear Border, and (e) Binaries image

4. Morphological Operation of the Image: For the proper segmentation of the preprocessed image, a morphological operation is applied to fill up the holes of the binary image. This process integrated three sub-processes: the opening of the binary image, which is nothing but erosion, followed by dilation, shown in Figure 4(a). The opening image then used a process to remove the connected object (sickle cell) shown in Figure 4(b). This image is then applied to the branch point's morphology to separate the objects that touch through the boundary shown in Figure 4(c).

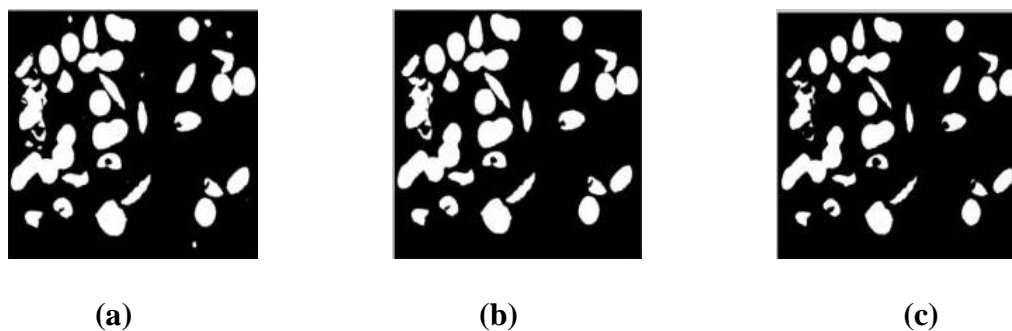


Figure 4: Morphological operations: (a) Hole Filling (b) Noise removal, (c) Objects Separation

- 5. Image Segmentation:** This process segmented the different regions shown in Figure 5. First, the threshold value is used for pre-segmentation. Then the boundary tracing method is used for further segmentation, which is done by labeling all pixels with some numerical values.

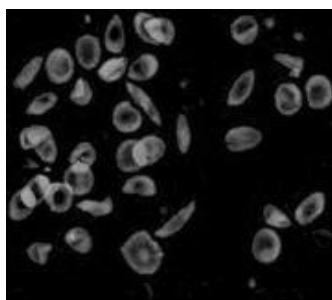


Figure 5: Segmented regions

- 6. Feature Extraction:** The primary and geometric feature extraction process has been applied to the segmented image. It is used to recognize and count sickle-shaped cells automatically. Table 1 shows the basic features of different shape recognition, and Table 2 shows the geometric features applied to separate the sickle cell shape from RBC.

Table 1: Basic Features of Shape Recognition

Feature	Description
Area	1. Scalar value 2. The actual number of pixels in the region.
Centroid	1. Vector of 1xQ 2. Specifies the center of mass of the region
Perimeter	1. Scalar value 2. Calculate the distance (in pixels) around the boundary of the region.
Eccentricity	1. Scalar value 2. Determine the eccentricity of the ellipse.

Table 2: Different Geometric Features for Shape Recognition

Feature	Description	Formulas
Shape Geometric Factor (SGF)	Calculate Circularity and Elongation for Each Cell.	$\frac{MajorAxisLength}{MinorAxisLength}$
Form Factor (FF)	Used to differentiate Circular and non Circular object	$\frac{4\pi * Area}{perimeter^2}$
Roundness Factor (RF)	Calculate the shape of the object.	$\frac{4 * Area}{\pi * MajorAxisLength}$

According to table 1 and table 2, I computed SGF, FF, and RF for the image given in figure 2. The computed values in the label form are shown in figure 6 (a)-(c). From figure 6(a), it is clear that the SGF values of sickle cells lie between 2 and 5 and from 6(b), the values of FF lie between .4 and .7. Similarly, from figure 6(c), the roundness values RF of sickle cells lie between 13 and 25. From the results of SGF, FF and RF parameters, it is concluded that apply these ranges of parameters can effectively separate the sickle cells from the RBC.

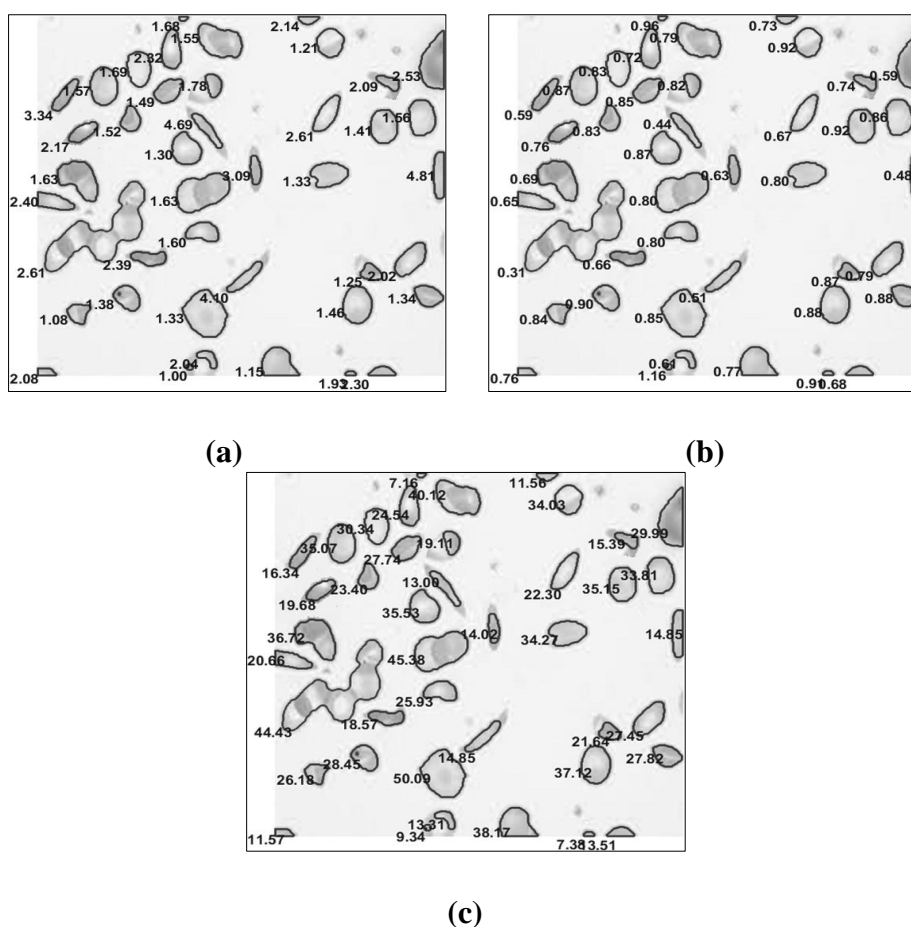


Figure 6: Geometric Features: (a) SGF, (b) Form Factor, and (c) Roundness Factor

- 7. Sickle Cell Detection and Counting:** Finally, applied the detection process to identify the sickle cell shapes and then counted them using the logical comparison operation based on various features. Figure7 shows the detected sickle cells in the red color boundary.

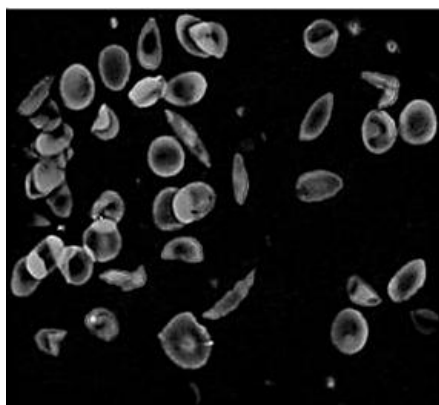


Figure7: Sickle Cell Detection

III. EXPERIMENT

All the experiments have been performed on the machine of Intel® M CPU @ 1.60 GHz with 2GB RAM under the Windows 1064-bit operating system using the Image Processing Toolbox of MATLAB 10R. For the performance evaluation, the proposed method has been implemented on the five blood sample images of different patients. The microscopic sample images of five different patients named as P1, P2, P3, P4, and P5 are depicted in Figure8(a)-(b), respectively.

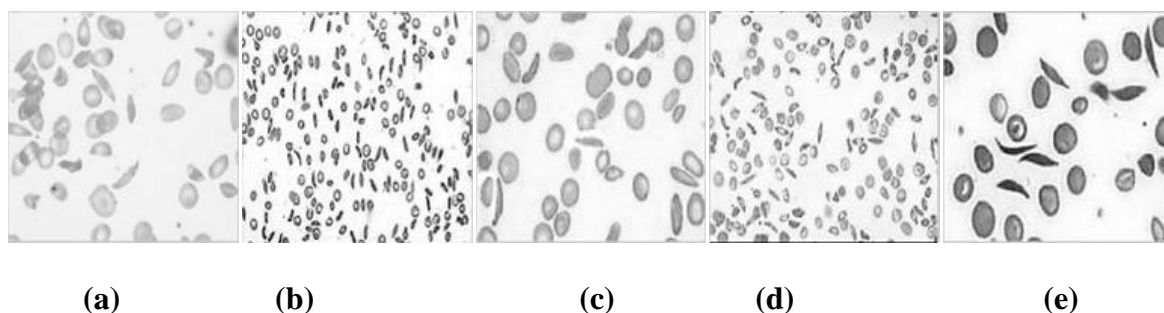


Figure 8: Sickle Cell Images: (a) P1, (b) P2, (c) P3,(d) P4, and (e) P5





Various calculated values of different feature extraction parameters for the RBC and sickle cells are shown in Table 3. Based on these features, it has been found that the feature parameters such as perimeter, area, major-axis-length, minor-axis-length, and roundness depend on the size of the cell object in the image. So it requires a manual setting for the specific size of the cell to classify the sickle cells and the red blood cells efficiently.

Table 3: Feature Extraction for Counting Number of Sickle Cell

Features	Red Blood Cell	Sickle Cell
Perimeter	400.54	376.49
Area	78354	4837.7
Eccentricity	0.67	0.93
Major-Axis-Length	141.11	134.37
Minor-Axis-Length	130.48	57.69
SGF	1.08	3.32
FF	0.93	0.56

The feature parameters FF, eccentricity, and SGF calculated values precisely classify the sickle cell and red blood cells. Table 4 shows the calculated values of the different feature parameters of the four sickle cell images depicted as four columns. From Table 4, it is clear that the value of FF is greater than 0.3, eccentricity is more significant than 0.9, and SGF is greater than 2 of all the sickle cell images, which are in the range discussed above. Thus, for the exact classification of RBC from Sickle cells, these parameters can be used to efficiently separate the sickle cells from the RBC, which can help detect and count sickle cells from the digital image of blood cells.

Table 4: Feature Extraction for Counting Number of Sickle Cell

Parameters	Sickle Cell Image			
				
Perimeter	58.25	106.55	112.29	63.76
Area	137.00	429.00	717.00	120.00
Eccentricity	0.96	0.96	0.92	0.98
Major Axis	27.04	48.10	48.65	30.77
Minor Axis	7.48	14.26	19.02	6.12
SGF	3.61	3.37	2.56	5.02
FF	0.51	0.47	0.71	0.37

IV. RESULTS AND DISCUSSIONS

The experiments have been performed on sickle cell images of five patients named P1, P2, P3, P4, and P5. The experiment results are divided into three parts, which are shown

in Table 5. The first part displays the information on sickle cells only. The second part displays the total number of cells, and the third part displays information about the detection accuracy of sickle cells.

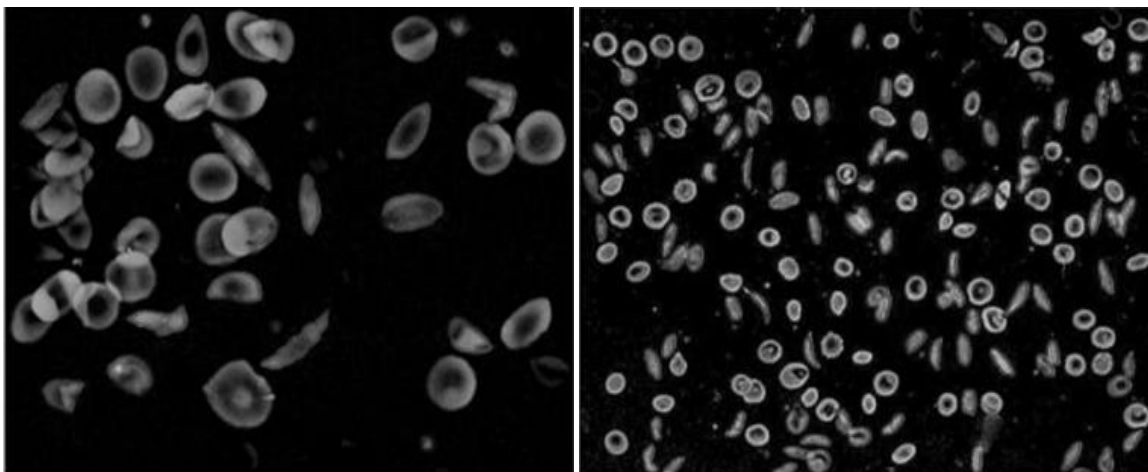
The experiment results depicted in Table 5 show comparative outcomes achieved after implementing the existing method (Kothari *et al.*, 2009) on the proposed and manual counting methods. The experiment results have included sickle cell and total cells (including sickle cells and RBC cells). From the results, it has been clear that the proposed automatic system accurately counts sickle cells and the total number of cells compared to the existing method (Kothari *et al.*, 2009) and manual counting. The average counting accuracy of (Kothari *et al.*, 2009) and sickle cells is **77.46%** and **99.96%** respectively.

Table 5: Detection Accuracy of Sickle-Shaped Cells

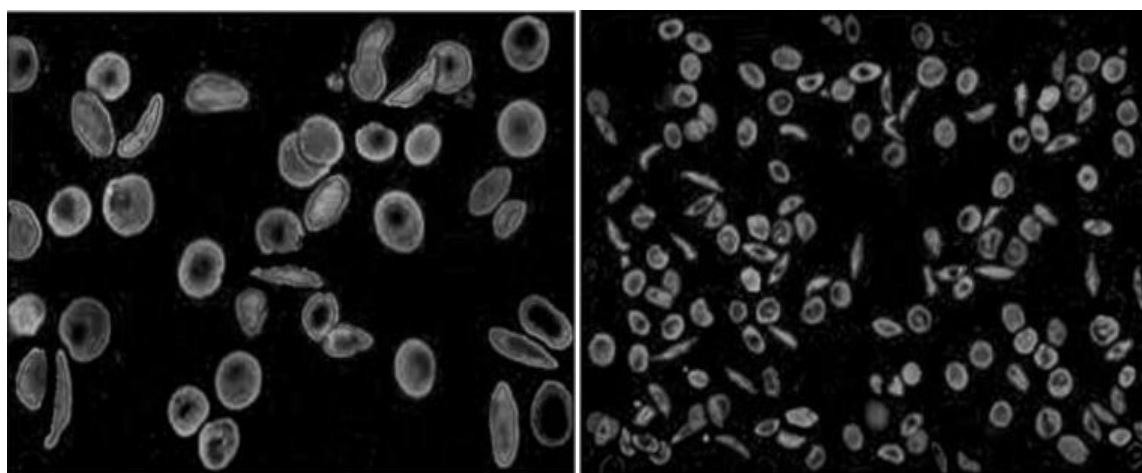
Image Name	No. of Sickle Cells			Total No. of Cells			Detection Accuracy (in %)	
	Manual Counting	[10]	Proposed Method	Manual Counting	[10]	Proposed Method	[10]	Sickle Cells
P1	13	10	13	27	19	25	76.92	100
P2	75	60	73	147	105	145	80.00	99.97
P3	19	15	18	37	26	36	78.95	100
P4	60	48	58	130	93	125	80.00	99.96
P5	7	5	8	23	16	23	71.43	99.85
Average							77.46	99.96

The existing method [10] achieved an average efficiency of **77.46%** to detect sickle-shaped objects because this approach has used segmentation based on a line of the object. Therefore, due to this approach, it is clear that it has not been suitable, especially for sickle-shaped blood cells or overlapping sickle cells in several cases. While the proposed study applied the fuzzy enhancement process to highlight the blood cells, including sickle cells, efficiently and achieved the accuracy of detecting sickle cells is **99.96%**.

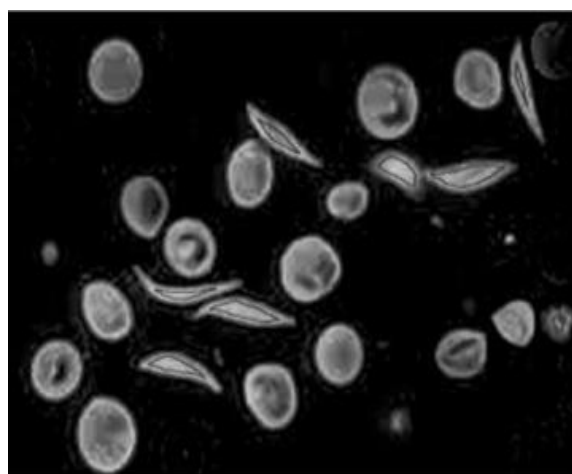
Figure 9 shows the detected sickle cell images of five patients, P1 to P5. The processed images show how the proposed (automatic counting of sickle cells) method is as accurate as the manual calculation method. For example, in Figure 9(a), the total sickle cells are 13, the same as manual counting. Similarly, in Figure 6(b), sickle cells are 73, which is close to the manual counting of 75.



(a) Total Cells = 25 and Sickle Cells = 13 (b) Total Cells = 145 and Sickle Cells = 73



(c) Total Cells = 36 and Sickle Cells = 18 (d) Total Cells = 125 and Sickle Cells = 58



(e) Total Cells = 23 and Sickle Cells = 08

Figure 9: Processed Image: (a) P1, (b) P2, (c) P3, (d) P4, (e) P5

V. CONCLUSION

It proposed an automatic sickle detection approach to detect and identify the normal blood cells and sickle-shaped cells contained by a cell cluster. The proposed method is completely automated and uses different geometric features as a parameter to adequately identify sickle-shaped cells. This method implemented a new preprocess fuzzy enhancement approach to highlight the blood cells efficiently and accurately segment the sickle-shaped blood cells. The results achieved are outstanding after applying my proposed method because it is efficient to find accurate sickle cells by the proposed. The proposed method has a similar effect to the manual counting method and is better than the existing one. The proposed method correctly detected and counted a maximum of the desired sickle-shaped cell with acceptable error. It is also concluded that the specialist can be used to evaluate the patient's actual clinical situation but for diagnostic support only.

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