

Detection of Sharp Contour of the element of the WBC and Segmentation of two leading elements like Nucleus and Cytoplasm

Prof. Samir K. Bandyopadhyay

Sr Member IEEE

Professor, Department of Computer Science & Engineering,
University of Calcutta, 92 A.P.C. Road,
Kolkata –700009, India.
skb1@vsnl.com

Sudipta Roy

M.Tech student of Department of Computer Science & Engineering,
University of Calcutta, 92 A.P.C. Road,
Kolkata –700009, India.
scse.roy@gmail.com

ABSTRACT

Assessment of blood cells smear is a normally experimental test these days and the haematologists are spent most of the time or paying attention on white blood cells (WBCs) merely. To analysis, identification and diagnosis digital image processing techniques be able to help us. Based on the circumstances, amount and condition of the white blood cells infection like sensitive leukemia is detected. The major intention of this paper is to detect sharp contour of the element of the WBC and segment to its two leading elements like nucleus and cytoplasm. The segmentation is conducted by means of a planned segmentation framework that consists of an incorporation of quite a lot of digital image of each element of white blood cells. The differential counting and detecting of white blood cell and segmentation of nucleus and cytoplasm provides incredibly useful information to pathologist for identification and treatment of many diseases manually counting, detecting and segmentation of white blood cell and its elements is a tedious, time-consuming and vulnerable to inaccuracy procedure due to the boring nature of this process and that is why an automatic system is most wanted in this automatic process, segmentation and classification of white blood cell are the most vital stages. Segmentation of WBC from other blood elements is achieved using various image processing techniques. Applying threshold and a new algorithm on the microscopic image resulted in producing appropriate segmentation of the all component WBC blood elements.

Keywords:

Contours Detection, Image Analysis, Image Segmentation, morphological analysis, White Blood Cells

1. INTRODUCTION

The recognition accuracy largely depends on subjective factors like experience and fatigue due to human tiredness. With the need for quality results, there arose a necessity for the automation of the whole process to reduce the burden on haematologists and to accurate results in significantly short period of time therefore new approach of automated detection is shown here. The count and shape, lineage and maturity level of white and red blood cells could aid in the diagnosis of diseases that range from inflammatory to leukemia. Important information for correct patient diagnoses by Peripheral or marginal blood cell differential counting and therefore the microscopic review is effort exhaustive and requires a extremely trained or qualified expert or professionals. Blood cell images consist of red and white blood cells and also some platelets spread across the whole images. Therefore for the most part research in leukemia WBC elements recognition and segmentation of nucleus is the important course of action in which the ultimate objective is to take out all the WBCs from a complex haphazard background and then segment the WBCs into their components, such as the nucleus and cytoplasm. In the past when automated system are not used very much digital image processing techniques have helped to analyze the cells that lead to more accurate, standard, and remote disease diagnosis systems but there are a small amount of technical hitches to extracting the information from WBCs due to the spacious variation of cells in edge, shape, position and size. However there are various problems because of illumination is not fair. The image contrast sandwiched between cell boundaries and the surroundings varies depending on the situation during the image capturing process. If the nucleus size increased then lymphocytes that become large it is called Reactive lymphocytes. The nucleus of a reactive lymphocyte can be round, elliptic, indented, cleft or folded. The cytoplasm is often

abundant and can be basophilic. Normal lymphocyte and abnormal lymphocyte are shown in figure 1 and figure 2.

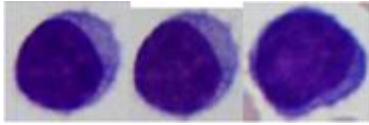


Figure 1. Normal Lymphocyte

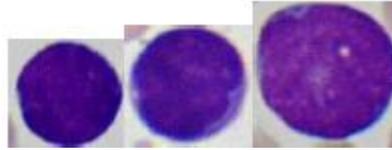


Figure 2. Abnormal Lymphocyte

This study is focusing on WBC segmentation and detection using microscopic images by the digital microscope. So our main goal is to identify by contour wise WBCs elements and segment and detect each component of WBC and nucleuses and cytoplasm which has developed by means of digital image processing. The intention of the current learning is to build up an automatic tool which can identify, detect, segment and classify the white blood cells namely, lymphocytes, monocytes, basophil, eosinophil and neutrophil in digital microscopic images. Segmenting and classifying WBC was shown to be a difficult task due to various reasons including cell touching, close cell/background intensities. In many of the researches presented in literature automatic cell segmentation was avoided to decouple the error due to segmentation with that of classification have done manual segmentation for all the acquired images to individually get WBC.

2. REVIEW WORKS

Automated detection of the abnormalities in medical images is an important and sometimes necessary procedure in medical diagnostics, planning, and treatment [1]. The recognition accuracy largely depends on subjective factors like experience and fatigue due to human tiredness. With the increasing demand for more number of such examinations along with the need for quality results, there arose a necessity for the automation of the whole process. This not only reduced the burden on haematologists but also yielded accurate results in significantly short period of time. An automated diagnosis system will alleviate the workload and the influence of subjective factors. Automated detection involves removal of red blood cells and platelets from the background. A standard image binarization technique, namely, Otsu [2] Image binarization is a very important step in image processing and pattern recognition applications, previously, most proposed methods followed the traditional manual makeover, i.e., detecting a cell, extracting its features, classifying the cell, and then updating the count [3-6]. Even though several attempts have been made to solve the blood cell counting, they are applied to peripheral blood only. The counting problem in

WBC is much more difficult due to the high density of cells. A projected structure for segmenting white blood cells is used by means of incorporation of concepts in digital image processing [7]. The nucleus segmentation part measurement is based upon morphological analysis and the cytoplasm segmentation is based on pixel strength threshold. In the case of edge detection we apply several different type of edge detection technique for blood cell boundaries have been extraction one of them is canny edge detection [8] by using this technique, edges taking place in the images would not be missed and there would be no responses to non-edges. Instead of considering each pixel, we consider a group of connected pixels called a patch. The fuzzy clustering of pixels provides the over segmentation in which several patches are generated. These patches are then combined to form two segments of nucleus and non-nucleus regions depending upon their similarities. Ngoc-Tung Nguyen, Anh-Duc Duong, and Hai-Quan Vu gained a high result in splitting cell images with lower complexity and time-performing [11]. These results demonstrate that the proposed technique is able to perform well with variable degrees of overlapping. Even better, distorted cells as well as their different size can be exactly split by this method. Fabio Scotti proposes a method [12] to enhance the microscope images by removing the undesired microscope background and a method for the robust estimation of the mean cell diameter and a new fully self-adaptive segmentation strategy to robustly identify white cells permitting. This method is easily extracting the features of white cells for subsequent automatic diagnosis of blood diseases (i.e., the Acute Leukemia). Nicola Ritter, James Cooper in ACSC [13] presented an unsubstantiated blood cell segmentation algorithm for images which taken from peripheral blood smear slides. Unlike preceding algorithms the scheme is rapid and totally automated, finds every single items cells, cell fragments and cell groups that do not overlap the border image identifies the each points inside to each WBCs elements and finds an correct one pixel extensive border for each object then disconnect the objects that just touch and has been exposed to work with a large collection of red blood cell

morphologists. S. Savkare and S. P. Narote in International Journal of Computer Science and Security proposed automated method of segmentation and classification of cell is simple [14] automatic approach is used Otsu Thresholding on gray image and green channel of the blood image for cell segmentation, watershed transform is used for separation of touching cells, color and statistical features are extracted from segmented cells and SVM binary classifier is used for classification of normal and parasite infected cells. Cell segmentation is a difficult crisis due to the complex nature of the cells and the ambiguity present in microscopy. Manual methods for this intention are burdensome, highly subjective and imprecise, thus requiring automated methods that complete this task in an objective and very efficient way. There are three types of cells in normal human blood: red cells, leukocyte or white cells and blood platelets. Generally, red cells are simple and similar. While white cells contain nucleus and cytoplasm and there are different types of them. White cells are categorized into five groups: neutrophil, eosinophil, basophil, monocyte and lymphocyte. The texture, colour, size and morphology of nucleus and cytoplasm make differences among these groups. In our paper we are considering only the nucleus. In blood smear, number of red cells is many more than white cells. P.S.Hiremath, Parashuram Bannigidad and SaiGeeta in IJCA proposed an automated image segmentation and classification of electron microscope images and extracting geometric features of leukocyte cells. The experimental results are compared with the manual results obtained by pathologies which is more reliable and computationally less expensive.

3. METHODOLOGY

First of all some image noise reduction techniques to removal of noise of noise by grayscale conversion, in this process conversion takes input colour image and convert into grayscale image which is done by forming a weighted sum of each three (RGB) component, such as a gray value equals to $0.2989 * R + 0.5870 * G + 0.1140 * B$, eliminating the saturation and hue information while retaining the luminance and the image returns a grayscale colour map. In next we have calculate the size of the matrix (as all image store in matlab are in the matrix form) in rows and column. Converts grayscale image data matrix into a double precision array, double may be overloaded for any object when it makes sense to convert it to a double-precision value, if value of matrix is already a double precision array, double need not double precision. Double precision due to fix the image size this means do not degrade the image size. Scan left to right top to bottom whole image

each row and column check if double precisionized pseudorandom value is greater than 255 then set those position of matrix is 255 and if double precisionized pseudorandom value is less than 0 then set those position of matrix is 0. Then give a cut-off value in which above the cut off value passes and then scan again from left to right and top to bottom and add matrix element with allowed value which is given calculated by average of maximum and minimum value, it is a methodology of sub-suppression of hazy images. Then we apply a technique to convert it in to binary image by giving a threshold value. After this a problem is that most intense part of the pixel which are not bright (white) are turned as black that is why we reversed the pixel value 0 as 1 and 1 as 0, the make our aim correct that only more intense part are glow brightly with pixel value 1. Scanning performed here also left to right and top to bottom. Then morphological analysis is applied which is a technique of image processing based on shapes. The value of each pixel in the output image is based on a comparison of the corresponding pixel in the input image with its neighbors. By choosing the size and shape of the neighborhood, you can construct a morphological operation that is sensitive to specific shapes in the input image. Here we apply two matlab command of morphological operation is `strel` and `imerode`. Morphologic operations are especially suited to the processing of binary images and greyscale images. In the basis of morphological operation we can determine contour and segment of the nucleus. To detect only the portion of WBC elements small area removal is applied [15-19].

Algorithm:-

Input: A color sample (blood) image.

Output: Contour and segmented WBC elements image.

Step 1: Converts input colour image in to grayscale image which is done by forming a weighted sum of each three (RGB) component, eliminating the saturation and hue information while retaining the luminance and the image returns a grayscale colour map.

Step 2: Calculate the size of the matrix which returns the number of rows and columns in separate output variables x and y .

Step3: Converts grayscale image data matrix into a double precision array, which store in d .

Step 4: Multiply a matrix which returns x -by- y matrix containing pseudorandom value, the size x , y is nonnegative integers (negative integers can be treated as 0) with square root of a constant and return to T , calculate $r = d + T$.

Step 5: For $i = 1$ to x do

```

Step 6:   For i = 1 to y do
Step 7:   If ( r(i,j) > 255) then
Step 8:   r (i,j) = 255
Step 9:   EndIf
Step 10:  If ( r(i,j) < 0 ) then
Step 11:  r ( i,j) = 0
Step 12:  EndIf
Step 13:  EndFor
Step 14: EndFor
Step 15: Calculate max and min value of r into m1
and m2, average a = (m1+m2)/2 .
Step 16: Initialize q1=s1=q2=s2=p=counter=0, cut =
0.5.
Step 17: Returns an absolute value corresponding
to the (a-p) and store it to the variable z.
Step 18: If (z >= cut) then
Step 19: counter=counter + 1
Step 20: For i=1 to x do
Step 21:   For j=1 to y do
Step 22:   If (r(i,j) >= a)then
Step 23:   q1=q1 + r(i,j)
Step 24:   s1=s1+ 1
Step 25:   EndIf
Step 26:   If (r(i,j) < a)
Step 27:   q2=q2+r(i,j)
Step 28:   s2=s2+ 1
Step 29:   EndIf
Step 30:   EndFor
Step 31: EndFor
Step 32: Set the variable b1 = (q1 / s1), b2 = (q2 /
s2), p = ((b1 + b2) / 2).
Step 33: Repeat step 17 and set a=p.
Step 34: EndIf
Step 35: Initialize all element of a matrix zero store
it into R.
Step 36: For i =1 to x do
Step 37:   For j=1 to y do
Step 38:   If(r(i,j) >= a)
Step 39:   R(i,j) = 1
Step 40:   EndIf
Step 41:   EndFor
Step 42: EndFor
Step 43: Reverse the matrix element by c = (1 - R).
Step 44: After converting binary image show more
intensity and more area by creates a flat, disk-
shaped structuring element, where radius
nonnegative integer parameter by strel command.
Step 45: Erodes the grayscale binary, or packed
binary image, returning the eroded image from
tep44 function by imerode command.
Step 46: Removes from a binary image(step45) all
connected components (objects) that have fewer
than P pixels, producing another binary image This
are done by the determination of connected
components, computation the area of each
components, removal small objects then get the
ultimate output image.
    
```

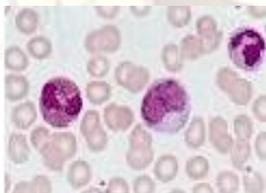


Figure 3. Blood Image

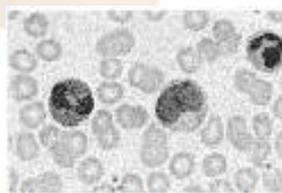


Figure 4. Grayscale Image



Figure 5. Threshold segmentation



Figure 6. Contour of blood smear Image



Figure 7. Canny edge of blood Image



Figure 8. Contour of WBC elements



Figure 9. Segmented WBC elements

The part of the (WBC) images that has the white blood cells element has more intensity than that of other portion of image, other things is that highest intensity area is the nucleus and we can make our assumptions about the type of the white blood cells, these are the basic things which we considered. Different type white blood cell has different type of nucleus with different type of lobe, detecting contour and segmenting there nucleus we can determine each element of the WBC. A sample smear image is shown in figure 3 and a grayscale conversion of original image is shown in figure 4 which is due to the noise reduction from image. Figure 5 shows the threshold segmentation from grayscale image and also it is a technique to convert binary image which helps us applying morphological operation. Applying morphological operation we get contour of blood elements image which is actually done by less strel value, disk type of strel is apply here and output is shown in figure 6. If we compare with the canny edge detection technique our algorithms give better result because

canny shows all edges of blood cell element but our goal is to detect only WBC elements though some red and platelet are detected in our algorithms but overall consideration our algorithms for edge detection of WBC elements give better result. Small area remove strategy is due to the extraction of only white blood cells. Contour of WBC elements without any other blood element is shown in figure 8. we can detect white blood cells from this contour because different WBC element has different type of lob or nucleus. Segmentation shown in figure 9 in which we extract nucleus from its each element. Thus from figure 8 and figure 9 we can conclude that there are neutrophil, eosinophil, lymphocyte, in the right side neutrophil, left side eosinophil and the middle one is lymphocyte. In normal lymphocyte ratio of cytoplasm and nucleus is almost same but abnormality is due to the growth of nucleus and reduces cytoplasm namely reactive lymphocyte, thus segmentation of nucleus of a lymphocyte is shown in figure 10.



Figure 10. Normal lymphocyte segmentation

Some Other Result For Different Input Blood Images

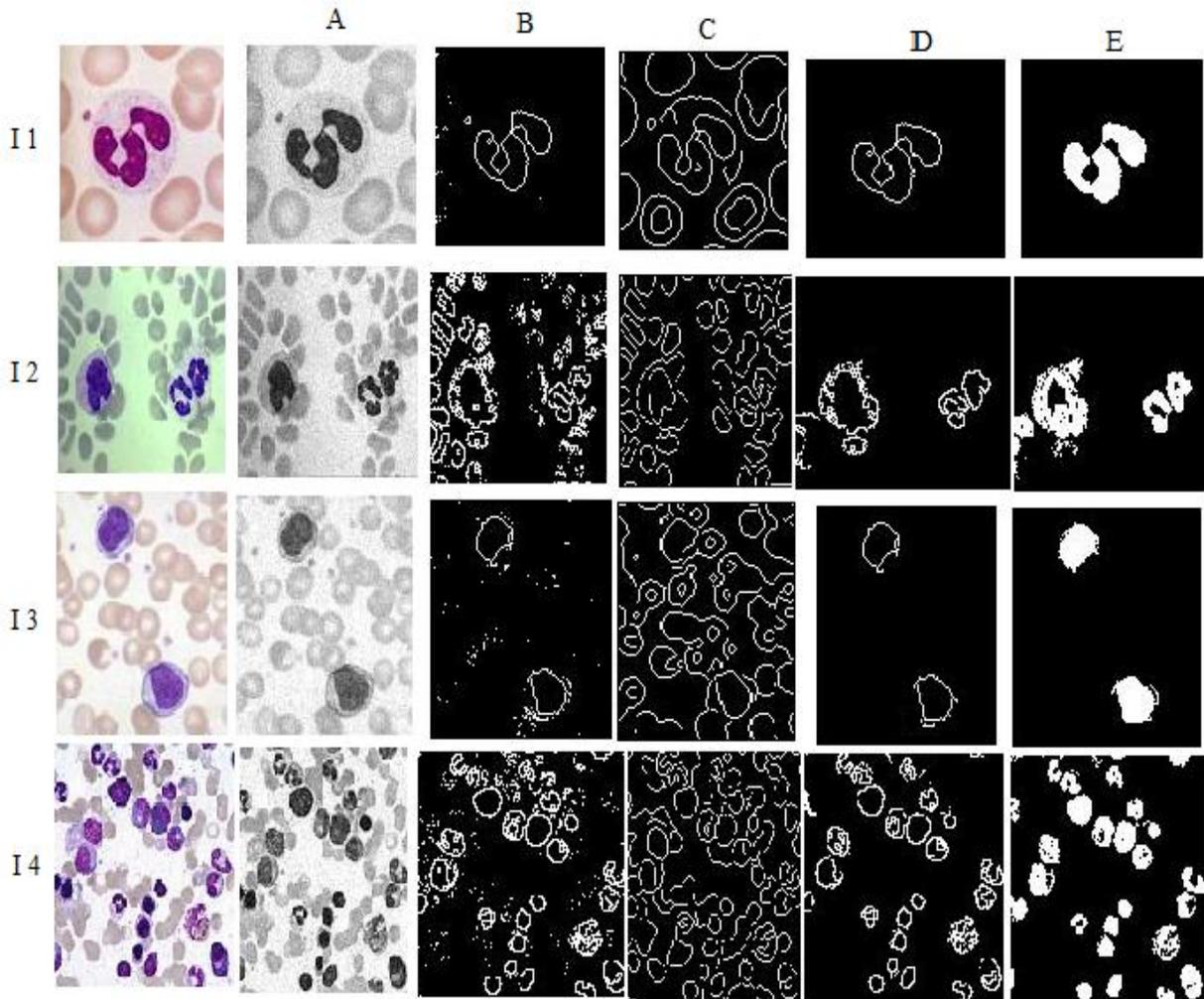


Figure 11. (I 1, I 2, I 3, I 4) are the four input images, (A, B, C, D, E) are the different type of output such that A is the grayscale conversion of original smear blood image, B is contour image of WBC's elements of our proposed scheme, C is the canny edge detection output, D is the contour image of only WBC's element suppressing other element of blood, E is the segmented nucleus of the each WBC's element.

4. CONCLUSIONS

We have presented a method for WBC segmentation into nucleus and cytoplasm which helps in the diagnosis of several diseases. Our method successfully segments WBC images into nucleus, cytoplasm and background. Another important contribution is the use of a scale-space toggle operator to regularize the image contours, leading to a better segmentation without leaking. The errors that do occur are often caused by noise within a cell. The trace “wants” to curve in towards the cell, and a noisy cell surface will create enough gradient information for it to do so. This kind of error makes up the majority of the incorrectly traced contours. The image simplification performed by the multi-scale operator also filters

out the noise present in some images. The nucleus information will be important to classify similar shapes from different maturation levels, where the only difference between classes is the proportion between nucleus and cytoplasm areas. The proposed method is more reliable, computationally less expensive and the proposed algorithms are better to detect the cells.

5. FUTURE WORK

Future work will focus on classification of the resulting contours, as well as improving culling measures to filter out more incorrect contours. In order to find more than just the topmost cells, we envision a multi-pass approach, first marking the top contours, and then repeating the process to find

lines that end at previously found cells. The usability of the minimally obscured contours will have to be tested in further stages of the project. These apparently good results are, for a large part, due to specific properties of our problem domain. This way is more precisely accuracy, although it is still not an optimum solution and sometimes it might need more time on obtaining these results.

6. REFERENCES

- [1] RitterNand Cooper J (2007) "Segmentation and border identification of cells in images of peripheral blood smear slides", In Proceedings of the Thirtieth Australasian Conference on Computer Science 62:161–169
- [2] Otsu, N., "A Threshold Selection Method from Gray-Level Histograms," IEEE Transactions on Systems, Man, and Cybernetics, Vol. 9, No. 1, 1979, pp. 62-66.
- [3] M. Beksac, M. S. Beksac, V. B. Tipi, H. A. Duru, M. U. Karakas, and A. N. Cakar, "An Artificial Intelligent Diagnostic System on Differential Recognition of Hematopoietic Cells From Microscopic Images," Cytometry, vol. 30, pp. 145-150, 1997.
- [4] H. Harms, H. Aus, M. Haucke, and U. Gunzer, "Segmentation of Stained Blood Cell Images Measured at High Scanning Density With High Magnification and High Numerical Aperture Optics," Cytometry, vol. 7, pp. 522-531, 1986.
- [5] J. Park, and J. Keller, "Fuzzy Patch Label Relaxation in Bone Marrow Cell Segmentation," IEEE Intl Conf on Syst, Man,Cybern, pp. 1133-1138, Orlando, FL, 1997.
- [6] S. S. S. Poon, R. K. Ward, and B. Palcic, "Automated Image Detection and Segmentation in Blood Smears," Cytometry, vol. 13, pp. 766-774, 1992.
- [7]FarnooshSadeghian, ZaininaSeman, Abdul RahmanRamli, BadrulHisham Abdul Kahar, and M-IqbalSaripan "A Framework for White Blood Cell Segmentation in Microscopic Blood Images Using Digital Image Processing", springerlink.com. Biological Procedures Online, Volume 11, Number 1.
- [8] Canny J (1986) "A computational approach to edge detection" IEEE Trans. Pattern Anal Mach , pp. 679–698.
- [9]http://en.wikipedia.org/wiki/Reactive_lymphocyte
- [10] NiponTheera-Umpon "Patch-Based White Blood Cell Nucleus Segmentation Using Fuzzy Clustering" Ecti Transactions On Electrical Eng, Electronics, And Communications Vol.3, No.1 February 2005.
- [11] Ngoc-Tung Nguyen, Anh-Duc Duong, and Hai-Quan Vu "Cell Splitting with High Degree of Overlapping in Peripheral Blood Smear" International Journal of Computer Theory and Engineering, Vol. 3, No. 3, June 2011.
- [12] FabioScotti "Robust Segmentation and Measurements Techniques of White Cells in Blood Microscope Images" IMTC 2006 - Instrumentation and Measurement Technology Conference Sorrento, Italy 24–27 April 2006.
- [13] Nicola Ritter, James Cooper "Segmentation and Border Identification of Cells in Images of Peripheral Blood Smear Slides" Australian Computer Society, 2007.
- [14] S. S. Savkare and S. P. Narote "Automatic Detection of Malaria Parasites for Estimating Parasitemia"International Journal of Computer Science and Security (IJCSS), Volume (5), Issue (3), 2011.
- [15] P.S.Hiremath, ParashuramBannigidad and SaiGeeta "Automated Identification and Classification of White Blood Cells (Leukocytes) in Digital Microscopic Images" IJCA Special Issue on "Recent Trends in Image Processing and Pattern Recognition" RTIPPR, 2010.
- [16] Prof. Samir K. Bandyopadhyay" An Edge Detection Algorithm for Human Knee Osteoarthritis Images" Journal of Global Research in Computer Science, 2 (4), April 2011.
- [17] Jiann-Shu Lee, and Yi-Nung Chung "Integrating Edge Detection And Thresholding Approaches To Segmenting Femora And Patellae From Magnetic Resonance Images " Biomedical Engineering applications, Basis & Communications Vol. 17 No. 1 February 2005.
- [18]Sezgin, M., Sankur, B.,"Survey over Image Thresholding Techniques and Quantitative Performance Evaluation" , Journal of Electronic Imaging 13(1), pp. 146–165, January, 2004.
- [19]Prof. Samir K. Bandyopadhyay, "Method for blood cell segmentation" Journal of Global Research in Computer Science, pp. Volume 2, No. 4, April 2011.