Chapter 8

A Sample Application for Computer-Based Bacteria Counting by Using Watershed Transform

Merve Nur Onal
Afyon Kocatepe University, Turkey

Nesibe Cekmez
Afyon Kocatepe University, Turkey

Gokhan Akarca
Afyon Kocatepe University, Turkey

ABSTRACT

Bacteria counting process and bacterial counting by eye can be performed by manual counting in which a specialist counts the bacteria with naked eye or the automatic counting that utilizes computer-based techniques. The counting process becomes exhausting, long, and incorrect when the counting is performed by specialist. Therefore, there is a need for an efficient and more reliable method for the bacteria counting process performed automatically. In this chapter, a semi-automatic method has been designed for bacterial counting from the images of petri plates and photomicrograph. For this purpose, the image processing procedure has been applied to the images. In the image processing procedure, watershed transform was used to separate the adjacent bacteria and thus a more accurate counting was performed. The efficiency of the presented method has been shown for many different images, and the counting results of the proposed method have been compared with counting results of an expert. The method has been shown to be a successful and promising work for bacterial counting.
INTRODUCTION

Bacteria are a single-celled microscopic organism. They form a separate living category between plants and animals. Bacteria can be found in nature, in soil, in air, in water, in plants, in animals and even in humans. Bacteria are small enough not to be seen with the naked eye in sizes between 1 and 6 microns. Bacteria multiply by division. In addition to the harmful bacteria that make the disease, there are also types of bacteria that are useful. Generally, bacteria can have classified as disease causing bacteria, yeast bacteria, rotting bacteria, and nitrogen bacteria. They can live in very hot environments where other creatures cannot survive or even under very high pressure. They corrupt the structure of dead organisms and allows them to come to a state where organic materials are used by other organisms. These are useful aspects of bacteria. Bacteriologists report that there are more than 100 species of bacteria and classify them according to type, structure, growth, culture environment and staining type. Bacteriology is the field of researching bacteria in microbiology. Examination of microorganisms, production and staining in different culture environment are included in bacteriological studies. Experts working on this field are called bacteriologists.

Bacterial counting is one of the most basic and frequent applications. Not only to demonstrate efficacy of disinfectants, but also to medical examinations, as well as evaluations for food and drug safety. Bacteria counting with this much precaution is usually done by manual counting of well trained technicians. For this reason, manual numbering is very time-consuming and labor intensive in practice. In addition, manual counting procedure is an error-prone method especially when counting on large number of plates and taking into account the different results available to different technicians. One of the most important reasons for the different count results that can be obtained by different technicians is the indistinguishable bacterial overlap (Talas, 2017).

In recent years, computer vision and image processing techniques have been used in many areas. One of these is the colony counting and the analyzing. Over the past 10 years, high-tech image analysis techniques and software applications with studies on artificial intelligence have accelerated histologic cell analysis, accelerating with studies on artificial intelligence in general. Especially the development of histologic markers enables the same image and many different structures to be visible at the same time, enabling the groups to perform studies in the field of medical image processing. Analysis of medical images includes many methods such as image acquisition, image reconstruction, image derivation, image compression and storage, image analysis and image-based recognition. All of these methods can be used to simplify operations that are challenging in cell and bacterial counting stages.

In the earlier stages of studies on tissue fragments was usually performed using the thresholding method which is a very popular method in this area. Thresholding is basically performed at the pixel level on the gray level images. It is aimed to make it possible to distinguish between the background and the desired object to be seen (Otsu, 1979). In many studies done on this area, good results were obtained with a reasonable threshold value selected according to the gray level (Kittler & Illingworth, 1986). There are also empirical methods to choose the threshold value. Since these methods are able to perform cell counting purposes, experiments can be made to increase the number (Markiewicz, 2003). Semi-automatic systems provide interactive processing by allowing counting at the user selected threshold interval by examining pixels in the spatial plane (Colley et al., 1989).

In the literature there are several studies on computer based microorganism counting. One of these studies Gilchrist E. et.al. had given Spiral Plate Method for bacterial determination in 1973. In this study a method for determining the number of bacteria in a solution via a machine which deposits a known