BACTERIAL COLONY COUNTER: MANUAL Vs AUTOMATIC

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Abstract— **Bacterial colony counter is used in many areas. They are used in veterinary practice to prevent the spread of infectious diseases such as influenza. They are used as well as in the pharmaceutical industry, to prevent contamination of products with harmful bacteria. These existing bacterial colonies counters count bacterial colonies manually which is very hectic and time consuming process. An increased area of focus in Microbiology is the automation of counting methods. Counting of bacterial colonies is complex task for microbiologist. We proposed a method to count these colonies to save time with accurate results and fast delivery to customers.**

Keywords- Bacterial colony, corner detection, image processing, thresholding, automation

I. INTRODUCTION

Bacterial colony is defined as a cluster of bacteria derived from one common bacterium. Many biological procedures depend on an accurate measure of the bacterial colonies and other organisms. The details of such colonies are a slow, tedious task. When counts are made by more than one technician, wide variations are frequently noted.

Accurate colony counting, to a large extent, depends on the ability to "see" colonies clearly, whether viewed by the naked eye or by a mechanized instrument. Colony morphology is a result of the individuality of the augmentation medium and other ecological conditions. To enhance visibility and counting accuracy of colonies, it is good practice to make use of measures that structure colonies that are counted easily by their improved size, shape, distribution and contrast.

The counting of bacterial colony is usually performed by well-trained technicians manually. However, this manual counting process has a very low throughput, and is time consuming and labor intensive in practice. To provide consistent and accurate results and improve the throughput, the existing colony counter devices and software were then developed and commercialized in the market.

In this paper, we propose a fully automatic colony counter and compare its performance with manual counting of bacterial colonies.

II. THE CONVENTIONAL METHOD

The Conventional methods may utilize both direct and indirect methods of counting colonies. The traditional plate count method is considered an indirect method, desirable due to its low cost. Some other examples of technologies that may be used for quantification of microbial growth include: ATP Bioluminescence, Impedance/Conductivity, Spiral Plating, Membrane Filtration, Direct Epi-fluorescent Filter Microscopy, Membrane Laser Scanning Fluorescence Cytometry, Fluorescence Flow Cytometry, and Latex Agglutination.

III. THE AUTOMATED APPROACH

An increased area of focus in Microbiology is the automation of counting methods. Several obstacles need to be addressed for methods that count colonies present, for example on Petri plates. These obstacles include: how to handle confluent growth or growth of colonies that touch or overlap other colonies, how to identify each colony as a unit in spite of differing shapes, sizes, textures, colors, light intensities, etc.

Shen Wei-zheng and WU Ya-chun[1] developed a new automatic colony counting system, which makes use of image-processing technology to feasibly count white bacterial colonies in clear plates according to the RGB color theory. It has been proved that the method greatly improves the accuracy and efficiency of the colony counting and the counting result is not affected by shape or size of the colony.

Chengcui Zhang and Wei-Bang Chen[2] proposes a fully automatic colony counter and compare its performance with Clono-Counter, an existing automatic colony counter reported by Niyazi et al. This proposed method can significantly reduce the manual labor by automatically detecting the dish/plate region and extracting and counting colonies

Wei-Bang Chen & Chengcui Zhang[3] proposed a fully automatic yet cost-effective bacterial colony counter. In this paper, the proposed method can recognize chromatic and achromatic images and thus can deal with both color and clear medium. In addition, the proposed method is software-centered and can accept general digital camera images as its input. The counting process includes detecting dish/plate regions, identifying colonies, separating aggregated colonies, and reporting colony counts. The proposed method is robust and efficient in terms of labor-and time-savings.

Hong Men, Yujie Wu, Xiaoying Li, Zhen Kou, Shanrang Yang [4] proposed a method of rapid determination of Heterotrophic bacteria in industrial cooling water is developed.

Wen-Lin Liu, and Chi-Bang Chen et. al[5] introduce a fully automatic bacterial colony counter. In this paper, proposed method can recognize chromatic and achromatic images and can deal with both color and clear medium. In addition, proposed method can also accept general digital camera images as its input. The whole process includes detecting dish/plate regions, identifying colonies, separating aggregated colonies, and finally reporting consistent and accurate counting results.

Sigal Trattner, Hayit Greenspan[6]has been developed an automatic tool to identify microbiological data types using computer vision and statistical modeling techniques. The statistical methodology presented in this paper, provides for an automated, objective and robust analysis of visual data, along with the ability to cope with increasing data volumes.

The general process followed can be summarized as:

- Colonies are grown on a Petri plate.
- The growth is epi-illuminated (one of the already proposed techniques), such that the reflection of the concentrated light coming from the halogen lamp is directed towards the image-sensing device. (Petri plate edges may be covered or blocked to prevent unwanted reflection.)
- Images are collected using digital (scanner/camera/web-cam) or video equipment.
- The collected images are digitized on a computer utilizing a image processing software package that has programming capabilities (note: the system works with any of the software packages with these capabilities)
- The digitized picture is processed using the multi-threshold segmentation procedures described to separate and detect the colonies present.
- The separated colonies are counted in a final processed image using a conventional single-threshold segmentation procedure.

IV. Proposed System

Our proposed method can reduce the manual labor by automatically detecting the colonies and count of those colonies efficiently. Bacterial colony counting is tedious and laborious work because these colonies are not easily seen by naked eyes.

Goal is to develop software to save time with accurate results and fast delivery to customers. There are so many devices to count these bacterial colonies but these devices ranges about 50,000 to 70,000 according to the Indian currency, that's why these devices are not so much efficient for daily use.

This proposed research work will count the colonies after 6 to 8 hours priori, saving a lot more time and this work will more efficient because market range for this is about 10,000 only as compare to prior systems.

Intense Testing is required before actual installation, on different images of filters of types:

- Images in which size & shape of bacterial colonies vary.
- Images containing very dense bacterial colonies.
- Images containing different types of bacterial colonies on same filter.

There are lots of sample of bacteria for which the proposed method will efficiently work. Some of the samples images are(fig 1.1):

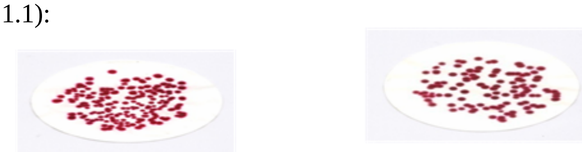


Figure 1.1: Sample Input Image

There are various devices available in market to count these are various devices available in market to count these colonies but those devices are very costly. We can design an automated bacterial colony counter which used many image processing algorithms such as grayscaling, thresholding, filtering etc. to count these colonies efficiently.

A. Block Diagram

To count the bacterial colonies, the block diagram for proposed method is given below (Figure 1.2):

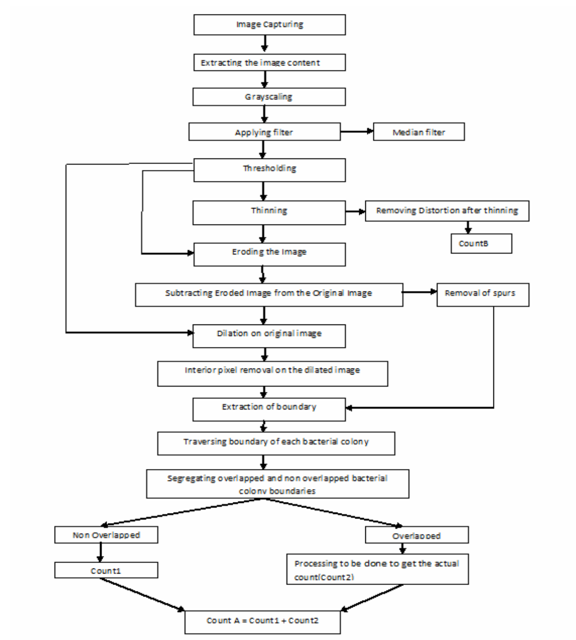


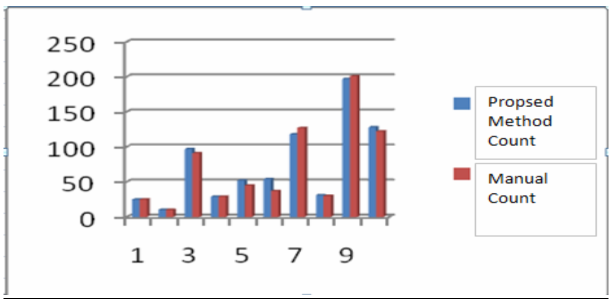
Figure 1.2: Block Diagram of Proposed Method

V. Result

Out of 50 samples on which algorithm were tested, following are 10 samples with variations in contrast, density, color and noise, following output occurred as shown in TABLE 1.1 & GRAPH 1.

Table 1.1: Proposed Method Count & Manual Count

Image	Proposed Method Count	Manual Count
1	25	25
2	10	10
3	97	91
4	29	29
5	52	45
6	54	37
7	118	127
8	31	30
9	197	201
10	128	122



VI. Conclusion

Bacterial colony in simple words is a group or cluster of bacteria derived from one common bacterium. We can design an automated bacterial colony counter which used many image processing algorithms such as grayscaling, thresholding, filtering etc. to count these colonies efficiently.

Graph 1.1: Proposed Method Count versus Manual Count

Hence, Images with high contrast and low or medium density give accurate or near to accurate count (99-100%). Images with low contrast or high density give less accurate count (95-98%).

The reason thereof is that in case of low contrast after thresholding shape of colony/colonies gets distorted which leads to appearance of high curvature points along the boundary. These high curvature points (corners) get accumulated in count result.

References

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