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## A QUANTITATIVE APPROACH TO DIGITAL IMAGE ANALYSIS WITH APPLICATIONS IN THE STUDY OF BIOFILMS USING CONFOCAL MICROSCOPY

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Biofilms are formed by bacterial colonies encapsulated in an extra-cellular polymeric substances matrix. They form in short time if environmental conditions are favorable. The study of biofilms is facilitated by advances in microscopy, such as the scanning confocal laser microscopy (SCLM) technique used in conjunction with analytical imaging, digital analysis, and semi-automated image processing. Various approaches were developed based on SCLM to assess the structure of biofilms and spatial variability within biofilms.

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The purpose of this study is to utilize digital images of biofilms from SCLM used in conjunction with fluorescent lectin probes to develop a tool that will ideally determine or at least estimate the volume occupied by bacteria in a certain region of the space. If the average volume of a bacterium is known, then an estimation of the number of bacteria can be performed easily. Applications may include the study of biofilm formation and growth, bacterial colonization, and determination of enzymatic activities.

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## **Confocal Microscopy: A** Particular Type of Remote Sensing

The novelty of our approach consists of recognizing that confocal microscopy is a particular type of remote sensing and treating digital images obtained using confocal microscopy in a similar manner with aerial images

Five Sections through a Biofilm, Taken at 2 µm Distance



## Jsing GIS to Track Changes

If the bacterial images are obtained correctly, then a similar approach can be used to determine, for example, how enzymatic activities within a biofilm are influenced by various environmental conditions or stressors.

Using SCLM techniques in conjunction with digital image processing techniques, remote-sensing techniques, or Geographical Information Systems to investigate the bacterial biofilms can be used to develop numerous applications, such as:

- Quantitative studies of biofilm formation
- Quantitative studies of bacterial colonization
- · Determination of enzymatic activity
- · Quantitative studies of biofilm growth and decay
- · Quantitative studies of the formation of dental plates
- · Quantitative studies of pipe corrosion due to biofilm formation
- Quantitative studies of nosocomial infections