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QUANTITATIVE ANALYSIS OF BACTERIAL BIOFILMS USING DIGITAL IMAGE TECHNIQUES

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Background

- **Biofilms are formed by bacterial colonies encapsulated in an extra-cellular polymeric substances matrix.**

- **Form in short time if environmental conditions are favorable.**

- **The study of biofilms is facilitated by advances in microscopy, such as the 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.**

Our Study

- Scanning confocal laser microscopy is used to acquire digital images to assess the structure of biofilms and spatial variability within biofilms.

- Various substances are stained using specific biochemical markers such as the fluorescent lectin probes.

- Need to develop a tool to determine the volume occupied by bacteria in a certain region of the space and estimate of the number of bacteria.

- Use analytical imaging techniques, digital analysis, and semi-automated image processing.

Methodology

Use SCLM in conjunction with:

- Digital image processing techniques

- Remote-sensing techniques

- Geographical Information Systems
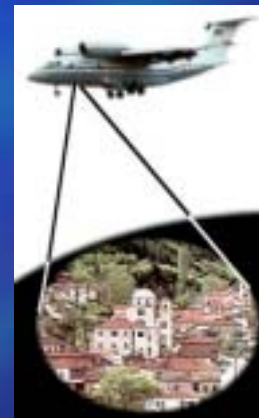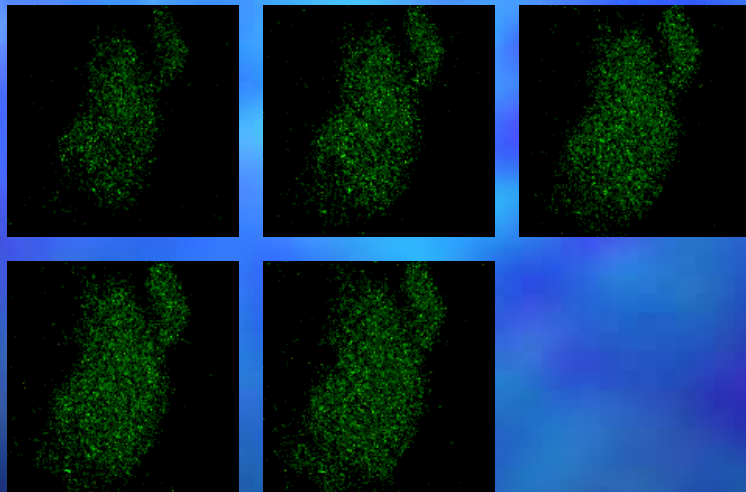
Methodology (Continued)

Figure 1.
Five Sections through a Biofilm, Taken at 2 µm Distance

Methodology (Continued)

- **Determine the area occupied by bacteria using image processing techniques.**

- **Based on the area, determine the volume; various approximations may be used.**

Example: $V = d \times \sum_{i=1}^{p} A_i$, **where:**

- A_i **is the area occupied by bacteria within each of the p photos**

- **d is the distance between the photos**

- **If the average volume of a bacterium is known, then an estimation of the number of bacteria can be performed easily.**

Applications

- **Quantitative studies of biofilm formation and growth**

- **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**

Example

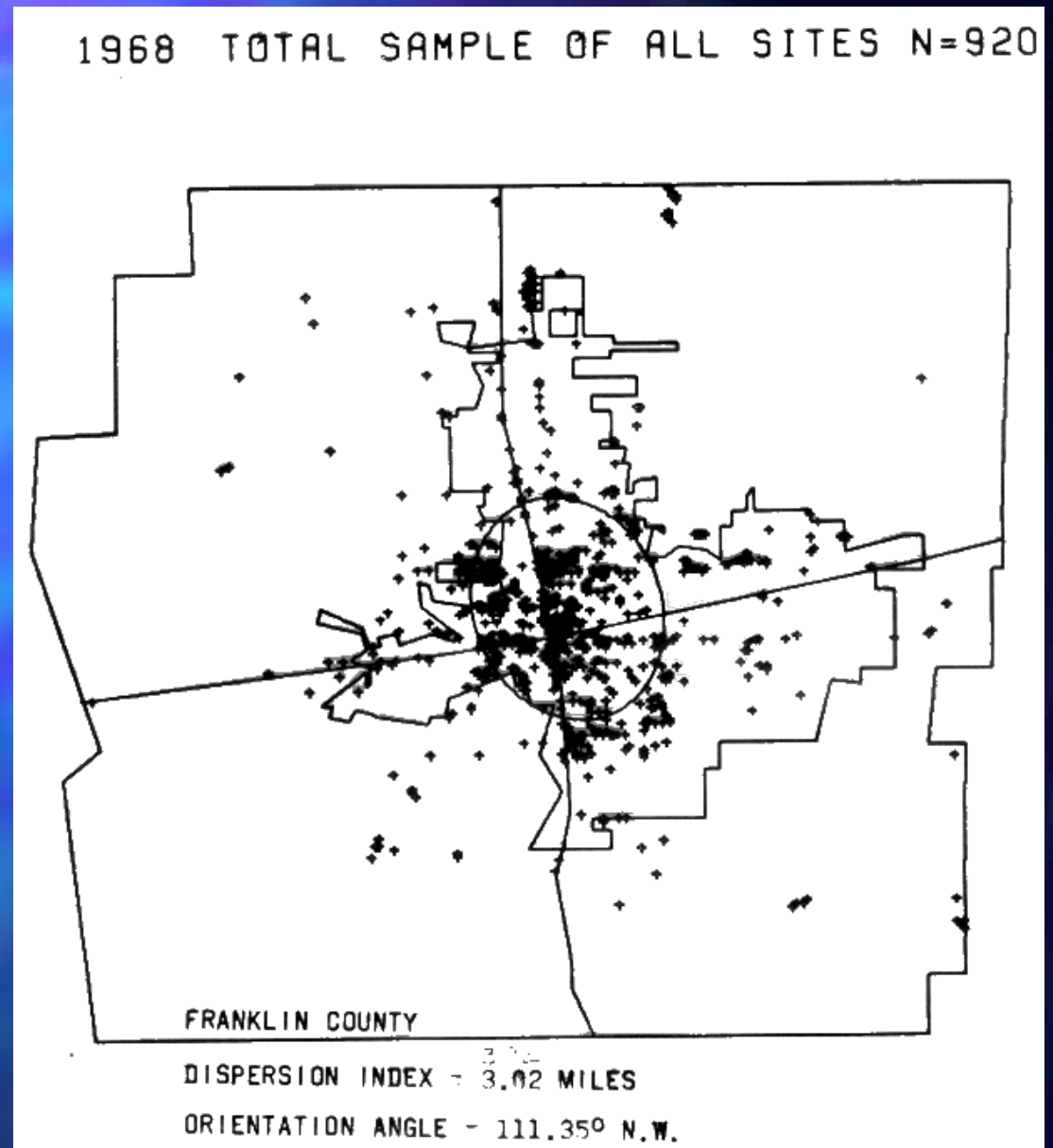**Dissertation: Cowen DJ, 1968**

1954 TOTAL SAMPLE OF SITES N=819

FRANKLIN COUNTY
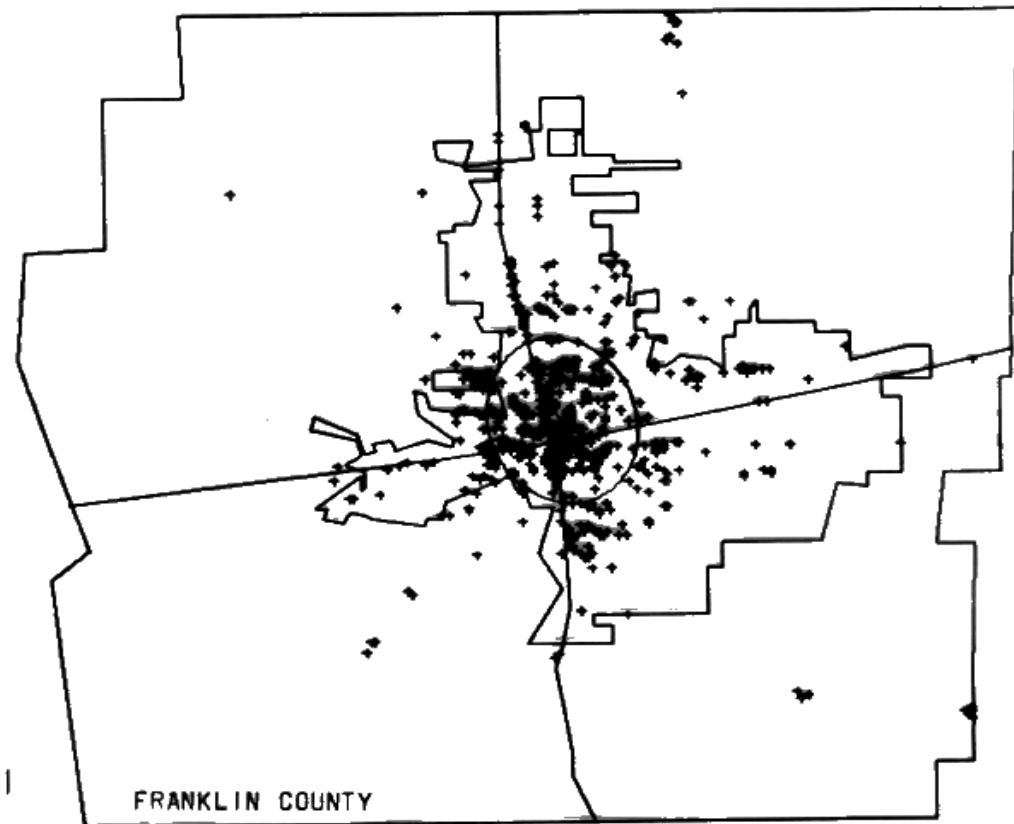
DISPERSION INDEX = 3.05 MILES
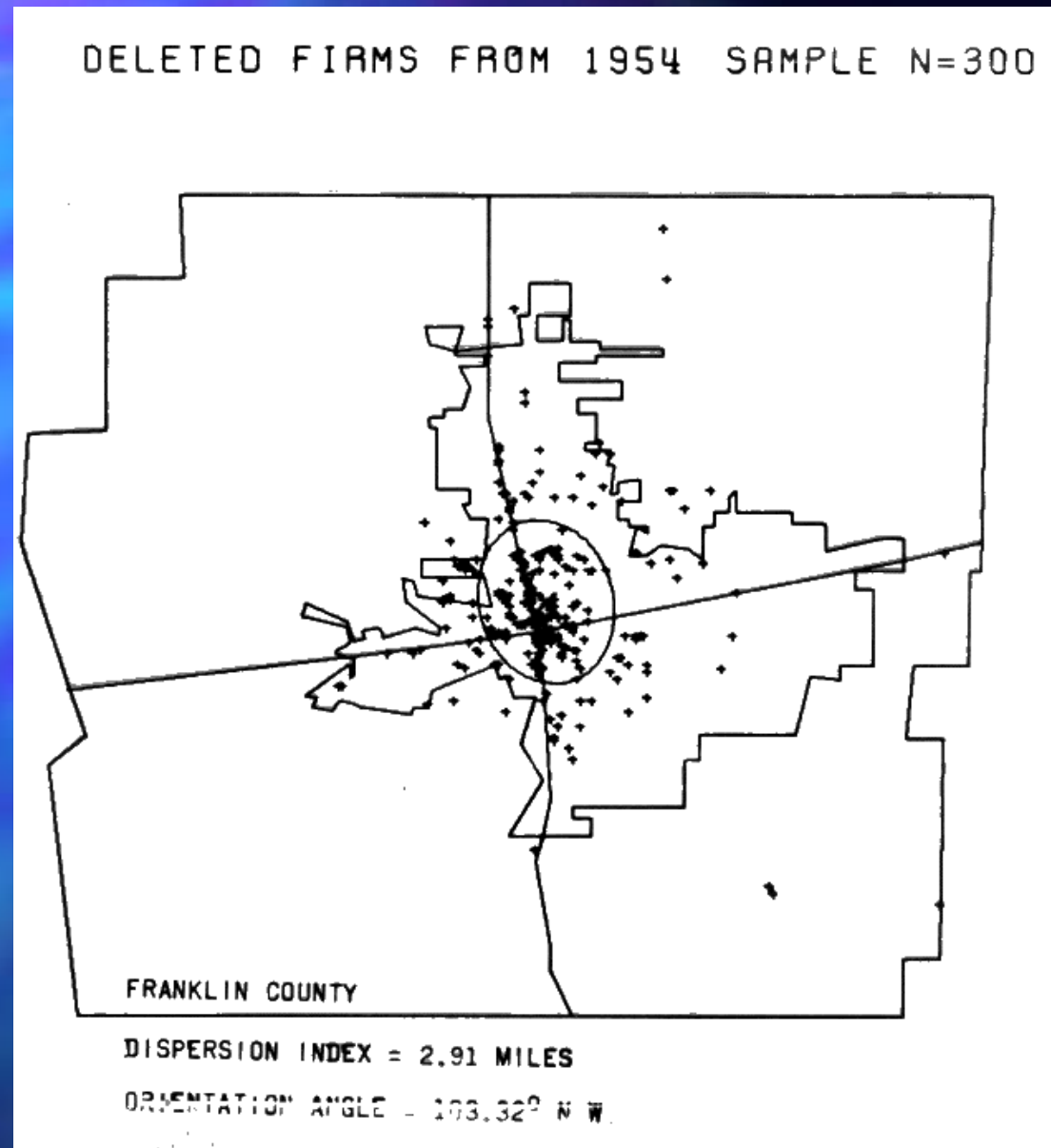
ORIENTATION ANGLE = 104.79° N.W.

Example (continued)

Dissertation: Cowen DJ, 1968

1968 TOTAL SAMPLE OF ALL SITES N=920

FRANKLIN COUNTY

DISPERSION INDEX - 3.02 MILES

ORIENTATION ANGLE - 111.35° N.W.

Example (continued)

Dissertation: Cowen DJ, 1968

DELETED FIRMS FROM 1954   SAMPLE N=300

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DISPERSION INDEX = 2.91 MILES

ORIENTATION ANGLE – 103.32° N W.

Final Remark

The objective of this project is to apply the approach presented in the previous example to the study of phenomena related to bacterial biofilms.

Any Questions?