

## Multi-resolution studies of biofilms for bioremediation - Collaborative Research Network in Imaging and Visualisation

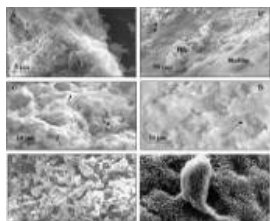
### Multi-resolution studies of biofilms for bioremediation

Bacteria grow on surfaces as layers called biofilms. Individual bacteria are too small to be clearly seen using a normal microscope but by using advanced microscopic techniques it is possible to see individual cells clearly. The electron microscope causes damage to the cells because the electron beam needs to operate in a vacuum and the cells need to be dried. However, using a new method called the environmental scanning electron microscopy (ESEM) we see a layer of a polymer with individual cells projecting through it. It is possible to see individual bacteria (1-2 microns in length) just as they occur in nature. In fact, because each individual cell is surrounded by a layer of hydrated polymeric material, this obscures a lot of its surface detail. When a biofilm is viewed using ESEM we can see this surface layer with bacterial cells jutting through it but largely hidden from view.

Biofilms are not just flat layers. They comprise a basal layer with long stacks of cells growing out from it. Water can flow between the stacks, suggesting a primitive circulatory system and biofilms have been likened to a tissue structure. A confocal microscope can image in three dimensions, allowing us to build up a picture of the 3-D structure of the stacks and channels.

By growing the biofilm on a sponge it is possible to make a spongy, porous material which readily transports liquids through the pores to all parts of the biofilm. Water is visualised by using magnetic resonance imaging (MRI), which uses the signal from protons in the water to build up a 3-D image. The limit of resolution of MRI means that it is not possible to see the stacks and channels visible by confocal microscopy (up to 100 microns) but at the 100 micron level images can be obtained using MRI and confocal microscopy to cross-calibrate each other. In this way the depth of the hydrated biofilm can be measured.

The major benefit of MRI is that it is possible to image flows in real time and space. For a system which is becoming progressively blocked, the flow will be going faster through the smaller spaces which remain unblocked and this will show up in the images. By processing MRI data it is possible to obtain quantitative information which helps us to design better bioreactors for harnessing the properties of the bacteria for useful activities. For example, these activities can be applied in the removal of pollutants from industrial flows.



#### Biofilm and calcium phosphate coatings on the surface of polyurethane foam and Ti-discs

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