MiSAC matters
MICROBIOLOGY IN SCHOOLS ADVISORY COMMITTEE
50th Anniversary Articles

'I'm pickin' up good vibrations': how nanoscale vibrations and advances in microscopy are helping us in the fight against bacteria

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The emergence of antibiotic resistant bacteria has been identified as a serious global threat to health. Antibiotic resistance occurs when an antibiotic has lost its ability to effectively kill bacteria or control their growth. There are many reasons attributed to the emergence of antibiotic resistant bacteria, the so called 'superbugs', including widespread misuse of antibiotics, poor infection control and hygiene as well as the absence of new antibiotics being discovered. Therefore, we find ourselves in a race against time to come up with novel ways to combat this global emergency.

Recent advances in microbiology and microscopy may offer new solutions to help tackle this problem. In the last few years, researchers have discovered that bacterial cells vibrate however, until recently, these tiny motions have been difficult to detect. Due to the advancement of techniques such as atomic force microscopy (AFM), we can now detect and quantify these bacterial motions at the nanoscale and it appears that these tiny vibrations are linked to the metabolic state of the organism.

AFM is arguably one of the most versatile and powerful microscopy techniques for studying samples at the nanoscale. It can be used to capture high-resolution images and also measure sub-nanonewton forces. The AFM uses a cantilever (which essentially acts as a 'force sensor') with a small tip at the end (**Figure 1**) to image samples and measure forces. During imaging, the AFM tip is brought into contact with the sample and scanned across the sample in a raster fashion (*i.e*. the AFM tip is scanned across the sample line-by-line). Deflection of the cantilever, as it tracks across the sample, is recorded using an optical lever detection system (**Figure 2**) and this deflection signal is used to produce images (**Figure 3**) or quantify single cell vibrations.

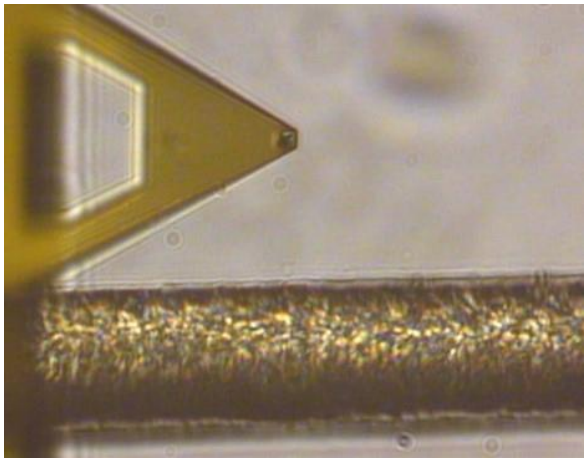

Figure 1. AFM triangular cantilever, highlighting the AFM tip (10-20 nm diameter at the apex). Human hair included for size comparison. Image taken using an inverted optical microscope (x60 mag).

The ability to sense vibrations is offering novel ways to study bacteria and may offer new means for more rapid detection of antimicrobial resistance to antibiotics. For example, conventional antimicrobial susceptibility testing (AST) is usually carried out in clinical microbiology laboratories and can take days to complete. However, using AFM it now appears possible to determine the effects of antibiotics on bacteria within minutes. This is achieved by attachment of the bacteria to the cantilever and recording any changes in cantilever deflection following

antibiotic treatment. Under normal conditions, the bacteria attached to the cantilever will vibrate (caused by metabolic activity within the cells) and this will result in very small but detectable fluctuations of the highly sensitive cantilever. Following treatment with antibiotics, any changes in cantilever deflection (*i.e*. dampening of the cantilever motions) can be attributed to bacterial vibration and thus to changes in metabolic activity.

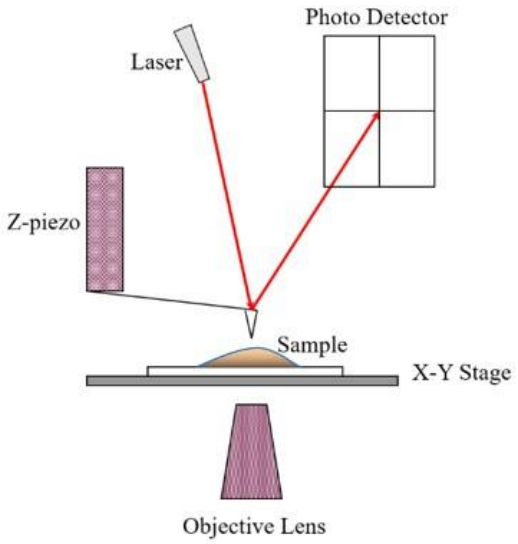

Figure 2. Schematic of AFM. A laser beam is reflected off the backside of a cantilever and onto a photo detector. Small deflections of the cantilever (e.g caused by bacterial vibrations) are detected by the movement of the laser spot on the photodetector. This signal is fed into a digital signal processor to quantify cantilever deflection.

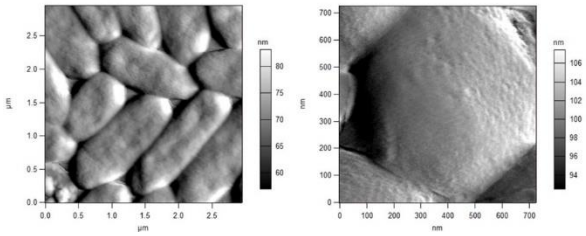

Figure 3. AFM contact mode image showing *Pseudomonas aeruginosa* cells (left) and *Staphylococcus aureus* cell (right) showing differences in morphology and surface structure.

Using this method, research is showing that we can now detect metabolic activity in low levels of bacteria and quantitatively screen their response to antibiotics. This novel approach has the potential to provide improved AST thus potentially enabling healthcare providers to offer more effective and timely treatment regimes.

AUTHOR PROFILE

Dr Mark Murphy is Senior Lecturer in Biomedical science and Biochemistry at Liverpool John Moores University and programme leader for the Industrial Biotechnology Masters course. His research is focused on the application of atomic force (AFM) and confocal microscopy in the study of mechanical forces influencing cell structure and behaviour. More recently, he has focused on the application of nanoscale vibrations to control microbial growth and biofilm formation; the key goal being to exploit the ability of microorganisms to sense and respond to external physical cues for medical and biotechnological applications.