BIOSENSORS

New leverage against superbugs

As the evolution of new strains of bacteria that are resistant to antibiotics continues, a nanomechanical approach to understanding the interactions between them could help efforts to develop new antibiotics.

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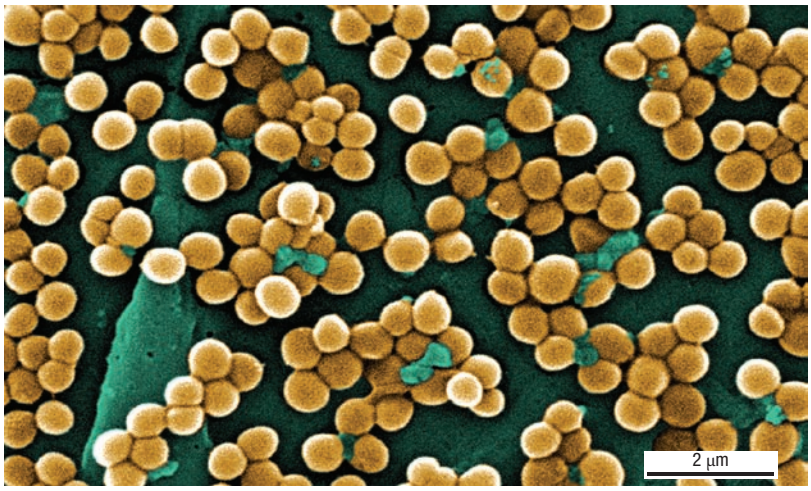

Figure 1 Scanning electron micrograph of MRSA. The use of nanomechanical detection to explore how superbugs like MRSA interact with antibiotics could lead to the development of new antibiotics.

Ever since the first therapeutic use of penicillin as an antibiotic in the early 1940s, humanity has thought that life-threatening bacterial infections were a problem of the past. However, bacteria have begged to differ, and by 1950 almost half of all hospital samples of the bacteria *Staphylococcus aureus* were resistant to penicillin, although drug-resistant bacteria have been mostly kept at bay since then because of progress in the development of other antibiotics.

Various mechanisms exist for bacteria to acquire and spread resistance. Some, like *Mycobacterium tuberculosis*, develop mutants to survive, whereas others, such as *S. aureus,* acquire resistance by exchanging genes or genetic elements with other bacteria of the same species. It is also possible for bacteria to transfer these resistance genes to other species, and this shuffling of genes within a particular species of bacteria, and between different species, has led to the arrival of so-called multi-drug-resistant strains of bacteria. Indeed, the use of 'broadband' antibiotics in hospitals was intended to enforce a germ-free environment but, sadly, it has had the opposite effect of unintentionally favouring strains of bacteria that are resistant to antibiotics.

These 'superbugs' include methicillin-resistant *S. aureus* (MRSA; Fig. 1), which has resulted in an increase in post-surgical infections caused by *Staphylococci*[1–3]. Another threat is vancomycin-resistant *Enterococci* and, in particular, the possibility that the gene responsible for this resistance could be transferred to other species of bacteria, including MRSA. This actually happened in 2002 but, luckily, very few cases of infections have been reported so far[4].

The recent decline in new antibiotics either under development or approved by the Food and Drug Administration in the US, and similar bodies elsewhere in the world, combined with the increase in bacterial resistance to the last lines of antibiotic defence, in particular to vancomycin and methicillin, makes the situation worse for the foreseeable future. Life-threatening bacterial infections could be back with a vengeance unless new approaches for faster detection of new antibiotics are developed.

On the *Nature Nanotechnology* website today, Rachel McKendry of the London Centre for Nanotechnology (LCN) and co-workers from the UK, Kenya and Australia describe a new approach for investigating antibiotic reaction mechanisms that could speed up the development of new antibiotics[5]. McKendry, Joseph Ndieyira, Moyu Watari and colleagues used microcantilever arrays to explore the mechanisms of antibiotic interactions with mucopeptides — components of bacterial cell walls — down to a sensitivity of 10 nM, and at clinically relevant concentrations in blood serum.

It had previously been shown[6] that biochemical interactions between a receptor coupled to one side of a microcantilever and a ligand in solution causes a compressive surface stress, resulting in bending of the cantilever that can be detected by deflection of a laser beam. This nanomechanical approach to detection has multiple advantages over other techniques: in particular, it is possible to detect ligands in solution in a single step without the use of fluorescent or radioactive labels (as is required in many biochemical detection methods), and also without the use of external probes.

Moreover, the use of arrays of cantilevers, each coated with a different receptor, enables multiple ligands to be detected (or multiple receptor–ligand interactions to be explored) in parallel under identical conditions. The use of one or more reference cantilevers in the array — that is, cantilevers that have been passivated to prevent ligands binding to them — enables differential measurements and quantitative determination of the binding constants for various interactions (see Fig. 1a of the article

by McKendry and colleagues[5]). It has been known since the mid-1980s that bacterial resistance in *Enterococci* can arise because a single hydrogen bond is deleted[7] from the binding pocket (see Fig. 1c in ref. 5), and this very subtle effect has now been clearly recognized in experiments using cantilever technology for the first time.

McKendry and co-workers investigated in a quantitative way the interactions of the antibiotic vancomycin with cantilevers that had been coated with an amino acid sequence (lysine-D-alanine-D-alanine) that occurs naturally in mucopeptides in the cell walls of bacteria. Vancomycin binds to the carboxy-terminus of a mucopeptide containing this amino acid sequence, hampering cell-wall synthesis by introducing weak points into the wall and eventually leading to the death of the bacterial cell[7–9]. These measurements are compared with measurements made with cantilevers coated with a mucopeptide from bacteria that are resistant to vancomycin. The data shows that vancomycin has different binding constants and causes different surface stress with each mucopeptide.

The team suggest that changes in the surface stress cause mechanical disruption of both the bacterial membrane and the cell wall, which eventually leads to the destruction of the bacteria. The observed compressive stress on the surface of the cantilever is interpreted as a product of a local chemical binding factor and a geometrical factor describing stress transduction as a collective phenomenon. Therefore, for mechanical disruption to occur, a relatively large fraction of the surface needs to be covered with vancomycin to establish connectivity between the weak points in the cell wall.

Investigating the mechanical influence of the antibiotic on the bacterial cell wall, as well as measuring binding properties, could result in the development or discovery of more potent antibiotics. And given the ability of bacteria to continually evolve to remain resistant to antibiotics, it will be necessary to understand the interactions between antibiotics and bacteria at the most fundamental level if we hope to keep superbugs at bay.

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