Physical sciences / Nanoscience and technology / Nanoscale devices / Biosensors
[URI /639/925/927/59]
Biological sciences / Biological techniques / Nanobiotechnology / Biosensors
[URI /631/1647/350/59]

Subject area: NANOMECHANICAL SENSORS

Title: Measuring a response in blood serum

Standfirst: Nanomechanical cantilevers can determine the concentration of active drugs in human serum.

F. Huber, H.P. Lang, and Ch. Gerber

The recent increase in the number of bacteria that are resistant to drugs, such as methicillin-resistant *S. aureus* (MRSA) and vancomycin-resistant Enterococci (VRE), represents a significant threat to public health. According to a report by the Centers for Disease Control and Prevention [1], for example, 2 million people are infected and 23,000 die each year in the United States from bacteria that are resistant to antibiotics; many more deaths occur as the result of complications associated with other conditions caused by an antibiotic-resistant infection. This increase in drug-resistant bacteria combined with a decline in the development of antibiotics, means that drug resistance is an impelling global concern [2]. One innovative way to try to address these clinical problems and develop effective therapies is to use mechanical signals, rather than chemical or electrical signals, to explore novel antibiotic therapies. Nanomechanical sensors based on microcantilevers provide a technology platform for label-free and sensitive detection of biomolecular interactions [3] on a solid surface. However, the development of a new drug requires a detailed understanding of its therapeutic effects in a complex environment, such as in a patient's blood. Writing in *Nature Nanotechnology*, Joseph Ndieyira, Rachel McKendry and colleagues now show that arrays of nanomechanical cantilevers can characterise the mechanical response of the bacterial cell wall to antibiotics while taking into account binding to other molecules in solutions that reduce their potency [4].

Nanomechanical sensors work by measuring the bending of a cantilever that is generated by molecular binding/adsorption taking place on the surface of the cantilever (Figure 1). The cantilever bending is a result of surface stress due to factors such as electrostatic and van der Waals forces, as well as conformational changes of molecules and molecular layers upon a binding event. The main advantage of the microcantilever technique is the possibility

to measure forces at the nanoscale, and the versatility of cantilever sensors in life science applications has been previously demonstrated through the detection of RNA [5], proteins [6] and microorganisms [7]. Spurious effects, such as nonspecific interactions and thermal drift, are cancelled out by calculating the differential response of a probe and a reference cantilever.

The researchers – who are based at University College London, Jomo Kenyatta University of Agriculture and Technology, the University of Queensland and the University of Cambridge – investigate the interactions of drug molecules that are in solution with strongly and weakly interfering ligands such as the proteins in human serum [4]. Some of these components of blood can influence the activity of drugs by binding antibiotics and thereby lowering the concentration of the active ingredient. Therefore, the interactions between the drug and competing ligands affect the dosage needed for effective treatment, and an in-depth understanding of these processes is essential for evaluating the effectiveness of new antibiotics, as well as for therapeutic drug monitoring. Furthermore, the researchers find that the additional presence of analogous bacterial targets in solution enhances the surface binding activity of antibiotics such as vancomycin. This could boost the efficiency of drugs in killing bacteria. The method could therefore also be used to evaluate new drugs and investigate resistance to antibiotics.

Ndieyira and colleagues use nanomechanical cantilever arrays to explore the mechanisms of antibiotic interactions with the bacterial cell wall and the blood/serum constituents at the same time. The binding of an antibiotic in solution to bacterial cell wall components causes surface stress changes, which illustrates the importance of nanomechanics in antibiotic action on bacterial cells [8]. Surface receptors on the cantilevers also probe the impact of blood serum on the activity of antimicrobials, allowing the correct dosage for a patient to be determined. For each new antibiotic drug that is developed, the influence of serum albumin and other proteins must be investigated to evaluate its effect. The nanomechanical technique allows antibiotic activity to be analysed quickly and could therefore help refine dosage prescriptions.

This nanomechanical technology has a number of advantages over other methods such as radioactive or fluorescence labelling in immuno assays, quartz crystal microbalance (QCM) [9] or surface plasmon resonance (SPR) [10], including label-free detection of ligands in solution, and screening of multiple receptor-ligand interactions in parallel and under identical conditions. Ndieyira and colleagues show that methods like SPR are two orders of magnitude less sensitive compared to the cantilever approach in the detection of antibiotics

in solution. Importantly, the researchers demonstrate that distinct changes of surface mechanics are drug-specific, and that competing ligands in solution play a fundamental role in modulating these mechanical properties. Moreover, they developed a conceptual framework that allows quantitative judgments to be made in understanding the mode of action of antibiotics in a complex environment.

The direct nanomechanical quantification of dosage effectiveness in clinical studies is a prerequisite for personalized healthcare. Additionally, it provides insight into a cure designed to optimize treatment outcomes, particularly in a combinational therapy, acting as a platform for the early detection of infectious diseases and therapeutic monitoring applications against multidrug resistant pathogens. The work of Ndieyira and colleagues represents a new approach towards determining the role of chemistry and mechanics in membrane-bound receptor and protein assays.

F. Huber, H. P. Lang, and Ch. Gerber are at the Swiss Nano Institute, University of Basel, Klingelbergstrasse 82, 4056 Basel, Switzerland.
e-mail: christoph.gerber@unibas.ch

References

[1] Antibiotic resistance threats in the United States 2013 by the Centers for Disease Control and Prevention, http://www.cdc.gov/drugresistance/threat-report-2013/pdf/ar-threats-2013-508.pdf
[2] Laxminarayan, R. *et al*. *Lancet. Infect. Dis*. **13**, 1057-1098 (2013)
[3] Fritz, J. *et al. Science* **288**, 316-318 (2000)
[4] Ndieyira, J. W. et al. *Nature Nanotech*. **XX**, XXX-XXX (2014)
[5] Huber, F., Lang, H. P., Backmann, N., Rimoldi, D., Gerber Ch. *Nature Nanotech*. **8**, 125-129 (2013)
[6] Buchapudi, K. R., Huang, X., Yang, X., Ji, H.-F., Thundat, T. *Analyst* **136**, 1539-1556 (2011)
[7] Longo, G. *et al. Nature Nanotech*. **3**, 522-526 (2013)
[8] Ndieyira, J. W. *et al*. *Nature Nanotech*. **3**, 691–696 (2008)
[9] Wang, D. *et al. Anal. Chem*. **84,** 7008-7014 (2012)
[10] Hoa, X. D., Kirk, A. G. and Tabrizian M. *Biosens. Bioelectron.* **23**, 151-160 (2007)

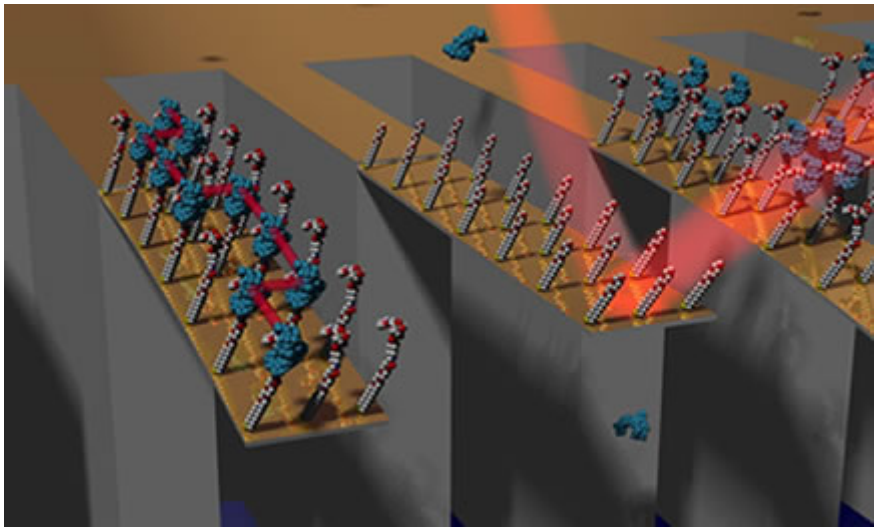

Fig. 1 Working principle of nanomechanical microcantilevers. The gold coated surface of some of the cantilevers is functionalized with receptor model molecules of Lysine-D-Alanine-D-Alanine (first and third cantilevers from the left) [4], whilst the other cantilever are functionalized with the reference molecule triethylene glycol (second from left), which does not allow binding of the ligand. Antibiotic molecules are depicted in blue, floating freely in solution and bound to the receptor molecules. The dark red line on the surface of the first cantilever shows the force distribution on the cantilever surface, which causes the cantilever to bend. Bending is analyzed using a laser beam (shown in bright red, deflecting from the middle cantilever), and a position sensitive detector (not shown). Cantilevers are measured sequentially.

The picture is taken from: http://www.bio-nano-consulting.com/NewsAndEvents/Archive%202009/Rachel%20wins%202.aspx
Please adapt the image or produce a new one similar to this. Add more free ligands (in blue) in solution.