New tools for analysis and detection of water pollution

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The Indian economy has been growing at ~ 8% during the last decade. The service sector has contributed immensely to this growth and continues to grab a large share of the gross domestic product (GDP). However, the agricultural sector is still the biggest employer in India and supports the livelihood of more than half the population. Given the dependence of such a large proportion of Indians on the four-month monsoon season – the storage, conservation and protection of water is of paramount importance for the economy as well as health of the nation.

The Intelligence Bureau, in its latest report, warned Indian security agencies of possible terrorist attacks on Indian dams and their water. The report highlighted that the current security measures and available gadgets (such as walkie-talkie sets) at most dam sites are grossly inadequate and outdated. The threat concerns not only the structure of the dams but also the water they store; there is a significant risk of poisoning of the waters by anti-social elements. Therefore, there is a need to develop new technologies that can efficiently monitor these waters.

Several methods can be implemented for surveillance and protection. Sensors, sonar and underwater cameras may be considered for this purpose, in addition to the divers who can periodically check underwater for any unwanted activity. The use of sensors to detect pollutants and threats in water is one of the efficient methods in environmental analysis. In addition to providing information about water quality, they can also detect the presence of various chemical and biological contaminants in water. There are, however, some major limitations in detection technologies that have prevented the use of sensors. These sensors need to be effective in reporting the pollutants in water and also be selective and specific for biotargets. Moreover, such sensors have to be cost-effective to be widely employed throughout the country. The electronic sensors that are currently available in the market are complex, labour-intensive, require high maintenance and are expensive; these factors have impeded their widespread adoption.

Recently, researchers at the University of Kentucky, UK have developed new bacterial spore-based sensing systems for analysing water pollutants[1]. A biosensor is an analytical device comprising a biological sensing component coupled to a transduction element, which produces a measurable signal in response to an environmental change or a target analyte. The new biosensors that have been developed are bacteria-based and will glow in the presence of specific pollutants. The main advantage here is their specificity and sensitivity to the target.

The new technology also addresses the problem of storage and transport of such cell-based sensors, which has been an Achilles heel for quite some time. The effective storage of bacterial biosensing systems requires the preservation of viability, activity and analytical performance of cells. The ability to keep reporter bacteria at ambient temperatures for long duration without special requirements, while maintaining their analytical characteristics, is still a major challenge.

The system developed by Date *et al.*[3] has been highlighted by the US magazine *Popular Science* in February 2010 as 'one of the top 25 technologies that will transform our crumbling infrastructure'. The researchers have used a unique property of the bacillus species to sporulate; this bacterium produces an endospore when starved of nutrients, which can survive in a metabolically dormant state for hundreds, if not thousands, of years without losing viability. The genetic information of the sensor is stored in the bacterial DNA of such bacteria. The bacteria can be revived quickly for testing, with a small amount of nutrients.

The new system addresses some of the major concerns that have inhibited the use of biosensors for on-field applications. These sensors are cheap, easy-to-use, sensitive, selective, easy-to-store and require no maintenance. In developing countries, such a technology can instantly improve the ability to monitor waters at a low cost. In fact China, during the Olympic Games held there last year, implemented the whole-cell biosensor technology for detection of pathogens in drinking water.

These sensors have also been integrated onto microfluidic platforms and chips, and have proved their ability to detect analytes of interest in various matrices such as serum and real samples[2]. The authors[2] have evaluated the applicability of these spore-based sensing systems for direct detection of analytes in environmental and biological samples. Spores were able to germinate in the presence of the sample matrix; the minimum time required for them to germinate and generate vegetative sensing cells – that are able to elicit a measurable response to target analytes – was ~ 2 h. Of the two spore-based sensing systems selected to detect model analytes in real samples, one was able to detect arsenic concentration as low as 1×10^{-7} M in freshwater and serum samples, and the other could sense down to 1×10^{-6} M of zinc in serum.

The achieved limits of detection confirm the usefulness of these systems in that physiological zinc serum concentrations are known to be ~ 10 μM, whereas the arsenic standard for drinking water has been set by the US Environmental Protection Agency at 10 parts per billion, which corresponds to 7.7×10^{-8} M of arsenite. The complete assays could be performed directly from dormant spores to obtain quantitative results from samples in 2.5 h for both arsenic and zinc. Furthermore, the assay is inexpensive, simple to carry out and offers unique advantages in the incorporation of these systems into portable analytical platforms such as microfluidic devices that are employed for on-site analysis.

The sensors have been shown to be effective in detecting biotargets in various river and lake waters[3]. Date *et al.*[3] demonstrated the feasibility of incorporating these dormant sensing systems on a centrifugal compact disk (CD)-like platform provided with multiple microfluidic architectures, and the potential of this integrated system for on-field analysis of real samples such as freshwater and human serum. The spore-based sensing systems were adapted to CD microfluidic platforms, where the mixing of fluids is achieved by centrifugal pumping. The fluids from the reagent reservoirs are pumped to the detection chamber at the edge of the platform when the capillary force is overcome by the centrifugal force generated by the spinning of the CD. The analytical signals that are produced are then measured by coupling the CD with a fibre optic-based detection system.

Employing this technology, the authors[3] were able to demonstrate the applicability of these spore-based sensors to such platforms and their efficiency in terms of assay time and sensitivity to analytes in real samples. The major advantages that are offered by such miniaturized sensing systems are the ease and rapidity of analysis and transportability and preservation of assay kits. The combination of microfluidic platforms and spores addresses the portability and storage issues of whole-cell sensing systems, thus facilitating their use for on-field applications. The possibility of performing automated on-site measurements using inexpensive and small, disposable platforms that are easy to use has huge potential in medical and environmental applications. The integratibility of these sensors will quickly enable their use as cheap water-quality testing kits for the home.

As the field of sensing evolves with time, our waters can be secured more efficiently and effectively (in terms of cost), with positive impacts on the health and well-being of the country. For this, investment in development of such technologies is paramount. The new advances in such diagnostic technologies will have lasting impacts in the fields of environmental science and biomedicine. It is envisioned that such spore-based sensing systems as developed by Date *et al*.[3] can be packaged with required reagents, thus generating micro-total analytical systems that would serve as viable tools for environmental and clinical on-site analyses. The diagnostics chips visualized by Date *et al.* will revolutionize detection in the field of medicine, and would be particularly beneficial in developing countries where appropriate storage and transport facilities are insufficient or unavailable.

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2. Date, A., Pasini, P., Sangal, A. and Daunert, S., *Anal. Chem.*, 2011, **82**(14), 6098–6103.
3. Date, A., Pasini, P. and Daunert, S., *Anal. Bioanal. Chem.*, 2011, **398**, 349–356.

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