Rapid Microbiological Monitoring Methods

A Fact Sheet from the Southern California Coastal Water Research Project



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Beach water quality monitoring safeguards public health.

How is beach safety monitored?

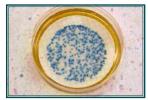
Rather than testing beach water for every pathogen (disease-causing microorganism), public health officials test the number of "fecal indicator bacteria", which has a strong correlation with the presence of pathogens. Traditional methods culture (or grow) bacteria from a water sample within a liquid media or petri dish, so that they can be counted. This takes 18-96 hours.

What's the problem?



qPCR = quantitative polymerase chain reaction

Current beach monitoring methods are too slow to keep pace with changes in the environment. According to recent studies, most sources of contamination are intermittent and last less than one day. Thus, contaminated beaches may stay open to swimmers while samples are being processed, and return to safe conditions by the time warnings are issued the next day.



Rapid methods could improve the accuracy and speed of beach warnings.

Why use rapid methods?



Tests are complete in 2-4 hours, allowing same-day posting of beach water quality information. qPCR, the most advanced rapid method to date, shortens testing time by detecting and quantifying DNA from the microorganisms of interest, rather than waiting for them to grow.

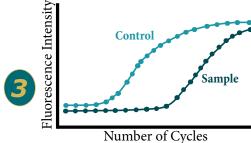
How does it work?

DNA replicates itself as part of many normal biological processes, and qPCR imitates and speeds up this process

in the laboratory.







Step 1: Bacteria in the water sample are captured on a filter and mixed with reagents that simulate normal DNA replication.

Step 2: Fluorescent probes that adhere to the new DNA copies are added, and the sample is subjected to repeated heating and cooling cycles to double the amount of DNA every few minutes.

Step 3: A computer records the amount of fluorescence in real time to estimate how much DNA, and how many cells, were present in the original sample.

Over a decade of SCCWRP research has gone into the development and implementation of rapid monitoring methods in California.



Epidemiology studies

At four southern California beaches, SCCWRP gathered data on the actual incidence of illness in beachgoers at the same time water samples were taken and tested using different methods. qPCR was more accurate than traditional methods at predicting

health risk considering the time that test results became available.



Method refinement

SCCWRP has evaluated a number of

method alternatives for qPCR, and worked to understand the chances of false positive test results owing to detection of DNA from non-viable bacteria. Importantly, they have also studied how often and where water samples contain substances that inhibit DNA replication, and the effectiveness of different internal controls to account for this inhibition.



Demonstration project

SCCWRP trained three Orange County water quality laboratories that routinely conduct beach monitoring to perform qPCR, and assessed their proficiency.

After a trial period, they used this qPCR data to issue same-day beach advisory decisions using electronic signs at several locations over eight weeks. The demonstration showed the impor-

tance of timing and practical logistics. Overall, it went well, though there was some concern related to the increased number of beach warnings based on qPCR, and the number of samples with no qPCR result due to substances in the water that interfered with the reaction.



Next generation

SCCWRP is continuing work to increase speed and automation of rapid methods. Future goals include development of portable devices that will

cut down processing time, and the ability to permanently mount a qPCR machine in the water at the beach for continuous automated measurements.



Nationally, the US Environmental Protection Agency (EPA) is working to authorize use of qPCR for beach monitoring by 2012.