Challenges in implementing new technology for beach water quality monitoring: Lessons from a California demonstration project

John F. Griffith and Stephen B. Weisberg

ABSTRACT

Beach water quality is currently monitored using culture methods that require 24 hours, but quantitative polymerase chain reaction (qPCR) can be performed in two hours. Efforts to date have focused on technology development, with less attention to practical challenges of method implementation. Here we present a demonstration project in which qPCR was used to make health protection decisions at nine beaches. Project goals were to assess success in technology transfer, consistency of results between culture-based and qPCR methods and success in implementing health protection decisions the same day the samples were collected. Technology transfer went smoothly, as laboratories routinely produced amplification efficiencies >90%, a high degree of repeatability between replicates, and results comparable to that of an experienced reference laboratory. qPCR methods slightly overestimated levels of Enterococcus compared to culture-based methods and some samples were rejected due to PCR inhibition. However, a stakeholder task force did not consider these an impediment to method adoption, because they were outweighed by the value of providing same-day results. Providing warnings by noon was challenging, but achieved by limiting the number of sites and using electronic means for communicating warnings. The task force concluded that capital and training costs were a smaller impediment to method adoption than the expectation for more monitoring, as there is no benefit to rapidity if a result is extrapolated across a week. Cost and

temporal logistics are likely to limit initial use of rapid methods primarily at beaches that are heavily used and/or have the most variable water quality.

INTRODUCTION

Beach water quality monitoring is presently conducted employing culture-based methods to measure fecal indicator bacteria (FIB). These methods typically require at least 24 hours for sample processing, which is too slow to protect swimmers from exposure to waterborne pathogens. As a consequence, the majority of water contamination events have dissipated by the time results become available and a warning sign is posted at the beach (Leecaster and Weisberg 2001).

The BEACHES Act of 2000 requires the United States Environmental Protection Agency (USEPA) to address the time lag between sample collection and public notification of poor water quality by developing more rapid methods that can provide results the same day samples are collected. After much research, the USEPA is poised to promulgate new criteria for beach water quality and certify quantitative polymerase chain reaction (qPCR) as the rapid measurement method (Boehm et al. 2009). The agency has conducted numerous epidemiological studies to support these criteria and published findings that demonstrate a relationship between levels of *Enterococcus* measured by qPCR and gastrointestinal symptoms in swimmers (Wade et al. 2006, 2008, 2010).

California has been supportive of USEPA efforts to develop rapid methods and even has its own legislation requiring development of rapid beach water quality measurements methods (Nakano 2001). Numerous rapid method evaluation studies (Griffith *et al.* 2009, Noble *et al.* 2010) previously conducted in California and epidemiological studies have or are being conducted at several locations to provide information on how qPCR performs at California beaches (Colford *et al.* 2007).

Despite the tremendous efforts put forth to evaluate and validate the technological efficacy of using qPCR for beach water quality monitoring, little has been done to examine the practical challenges involved in implementing a beach monitoring program once the method has been approved and becomes available for use by water quality monitoring agencies. These challenges include transferring new technology to water quality laboratories, developing a laboratory certification program, and communicating results to health care agencies in time to protect swimmers from exposure to contaminated water.

Recognizing the need to explore issues surrounding implementation of new technology for beach water quality monitoring, the Commission of the Southern California Coastal Water Research Project Authority (SCCWRP) formed a task force to determine the efficacy of implementing qPCR for measurement of *Enterococcus* at southern California beaches. The task force was comprised of a wide range of stakeholders, including: a wastewater treatment facility laboratory manager; a water quality monitoring laboratory director; federal, state, and local regulators; state and county health officials; a City Manager; and an environmental non-government organization (NGO; Table 1).

The task force met for over a year, examining data from both method evaluation and epidemiological studies. In May 2010, the task force recommended implementation of a demonstration project designed to determine the efficacy of using qPCR for *Enterococcus* as a substitute for currently approved culture-based measurement methods. This article describes the results of the demonstration project and highlights insights that were gained and areas of uncertainty that will need to be addressed before large scale implementation of qPCR for beach water quality monitoring can proceed.

Methods

The demonstration project took place from July 6 to August 31, 2010 in Orange County, California. Orange County was selected because the local health department was familiar with California's efforts to develop rapid methods for beach water quality monitoring and was supportive of using the new methods for making health protection decisions.

Nine beach sites impacted by non-point sources of fecal contamination were selected for inclusion in the project (Table 2). Locations were chosen based on several criteria: 1) a large population of beachgoers; 2) a history of poor water quality; 3) proximity to the laboratories analyzing the water samples; and 4) the ability to install a remotely operated electronic sign to display water quality information.

Eight sites were high-energy open coast beaches with breaking waves. Three of these were located at Huntington State Beach, upcoast of the Santa

Table 1. Members of Task Force.

Wastewater Treatment Facility Laboratory Manager: Dr. Mas Dojiri, City of Los Angeles
Water Quality Monitoring Laboratory Director: Charles McGee, Orange County Sanitation District
County Beach Water Quality Program Manager: Larry Honeybourne, Orange County Public Health Care Agency
State Department of Public Health: Gary Yamamoto, Division of Drinking Water and Environmental Management
State Water Resources Control Board: Darrin Polhemus, Director of Water Quality Division
Regional Water Quality Control Board: Michael Lyons, Los Angeles Regional Water Quality Control Board
US EPA Headquarters: Shari Barash, Office of Science and Technology
Environmental NGO: Dr. Mark Gold, Heal the Bay
City Manager: David Kiff, City of Newport Beach

Ana River, where flushing of bird droppings and decaying organic material from tidal mud flats have been hypothesized to contribute to poor beach water quality (Grant et al. 2001). Nearshore sand bars at this beach cause swells to break extremely fast and hollow, making it a world famous surfing venue. An additional three open coast sites were located at another famous surfing beach, just south of the entrance to Dana Point Harbor, at Doheny State Beach. Sites at this location are impacted by flow from San Juan Creek during wet weather, but the creek is ephemeral and hydrologically disconnected from the ocean by a large sand berm that routinely forms during the summer months and was present during the entire study period. Very long-breaking waves and plunging breakers characterize surf at this location. The last two open coast sites were at Newport Beach on opposite sides of the entrance to Newport Harbor. The northern of these sites was located adjacent to Newport Pier and is characterized by spilling breakers, while the southern site (Corona del Mar State Beach) is semi-protected by the harbor jetty and sometimes subject to poor water circulation depending on the direction of the swell. The ninth site was a popular bathing beach located in calm water inside Newport Bay and characterized by eutrophic conditions, poor water circulation and lapping waves.

Three microbiology laboratories participated in the project: Orange County Sanitation District (OCSD) in Fountain Valley, CA; Orange County Public Health Laboratory (OCPHL) in Newport Beach, CA; and South Orange County Wastewater Authority (SOCWA) in Dana Point, CA. Each laboratory had a different level of familiarity with the qPCR methodology. OCPHL was the most experienced, as their technicians routinely ran qPCR assays for clinically relevant microbes. OCSD had the next most experience, having participated in a method evaluation study two-years ago in which they ran qPCR for *Enterococcus* side-by-side with culture methods, but not having much experience subsequently. SOCWA was the least familiar with the method with no one on staff who had previously performed qPCR. This broad range of experience allowed us to assess the ease with which technology transfer could take place to end users with varying levels of expertise.

Six weeks prior to starting the demonstration project, the laboratories were trained on the qPCR method for Enterococcus using the method described by Noble et al. (2010), with the exception that salmon testes DNA, rather than Lactococcus lactis was added to each sample post-filtration. The salmon DNA served as an inhibition control and a separate qPCR reaction specific for this target was run separately from the assay for Enterococcus (Converse 2009). Each laboratory was provided a qPCR Training Manual and Standard Operating Procedure for performing the method. They also received one day of classroom training on qPCR theory and laboratory practices, followed by three days of supervised hands-on training performing the assay in the laboratory. Technicians were evaluated based on their ability to produce a standard curve from reference material used to calibrate the assay,

Agency	Latitude	Longitude	Description Newland Street, Huntington State Beach		
OCSD	33.64170N	-117.98126W			
OCSD	33.63838N	-117.97541W	Magnolia Street, Huntington State Beach		
OCSD	33.63311N	-117.96602W	Brookhurst Street, Huntington State Beach		
OCPHL	33.60563N	-117.92425W	15th/16th Street, Newport Beach		
OCPHL	33.59315N	-117.87556W	Corona del Mar State Beach		
OCPHL	33.61595N	-117.89215W	Newport Dunes, Newport Bay*		
SOCWA	33.46139N	-117.68928W	North Beach, Doheny State Beach		
SOCWA	33.46171N	-117.68454W	Mid North Beach, Doheny State Beach		
SOCWA	33.46195N	-117.68252W	San Juan Creek Ocean Interface, Doheny State Beach		

Table 2. Water quality sampling sites

* Upper Newport Bay: calm waters

in this case, pre-enumerated *Enterococcus* on frozen membranes. To be deemed proficient, technicians were required to produce a standard curve with greater than 90% amplification efficiency. Once technicians had demonstrated proficiency and were able to perform the assay without assistance from the training staff, the qPCR equipment was transferred to their respective laboratories for two days of additional on-site training using environmental samples collected from their routine monitoring sites.

Equipment provided varied from lab to lab, but included at minimum a BioRad CFX 96 thermocyler (BioRad, Hercules, CA) and Biospec Mini-Beadbeater-16 (Biospec Products, Bartlesville, OK). In the case of SOCWA, which had never performed PCR prior to this study, additional help was provided to prepare the laboratory, such as installation of a UV3 PCR Workstation (UVP, Ontario, CA) and -20°C and -80°C freezers. SOCWA was also provided with common laboratory equipment needed to perform PCR, but not typically found in laboratories using only culture-based methods, such as micropipettes, an electronic repeating pipettor, bench-top centrifuge, microfuge, tube racks, and all necessary consumable items.

Four weeks prior to the scheduled start of the demonstration project, the laboratories began collecting and analyzing samples from the test sites. Each laboratory processed samples from three sites (Table 2). Samples were collected Monday through Friday between 7:00 and 8:30 a.m. at ankle to shin depth on an incoming wave in sterile 1.5 liter polypropylene bottles and transported to the laboratory on ice. In order to expedite arrival of the samples at the laboratory, arrangements were made to intercept the person collecting routine samples and bring the desired samples back to the lab or to make a separate sample collection trip.

One hundred ml of sample water was analyzed in duplicate using qPCR, but results were not reported to the health department. EPA Method 1600 (Messer and Dufour 1998), and Enterolert[™] (Idexx, Westbrook, ME) were also performed in duplicate using volumes appropriate for each method. During this non-reporting period and throughout the remainder of the study, performance of the test laboratories was continuously monitored. Each sample collected by the test laboratories was split and two additional qPCR assays performed in duplicate by experienced technicians at SCCWRP's laboratory in Costa Mesa, CA. The second qPCR assay performed by SCCWRP was the TaqMan method for *Enterococcus* developed by U.S. EPA and described by Haugland *et al.* (2005). Results for both assays were calculated using the comparative C_T method as per Noble *et al.* (2010) and expressed as cell equivalents (CE). Samples for which the C_T value for the salmon DNA assay was greater than 1.6 C_T higher than expected were considered inhibited. If diluting the sample to reduce the concentration of inhibitory substances did not resolve the inhibition, results from that sample were considered unreliable and excluded.

On June 29, 2010, the task force met to review data produced by the labs during the four-week proficiency-testing period. The task force examined the data in four ways before deciding to go forward with the project. The first was to examine the amplification efficiencies produced by the labs from the calibrator material. Second, they looked at repeatability between replicates. Third, they compared results between those produced at the application laboratories and those produced by the more experienced technicians at SCCWRP. Finally, they compared results between qPCR and traditional culture-based methods to ensure they would not be making substantially different decisions, only more rapid ones, using the new methods. Based on the recommendation of the task force, the health department began using qPCR results to make beach management decisions on July 6, 2010. The project ran for 41 days, with each lab processing samples from three sites Monday - Friday, by which time the application laboratories had processed a total of 123 samples each.

The health department chose a conservative approach to beach management decisions, posting health warnings only when both qPCR replicates exceeded cell equivalents to the existing California numerical single sample standard of 104 colony forming units (CFU) per 100 ml of water. Beach postings were not removed based on qPCR results. Instead, health officials chose to keep postings in place until levels of *Enterococcus* were shown to be below standard using EPA Method 1600. When no qPCR results were available due to inhibition, the health department relied on the last available data.

Results of beach water quality testing were rapidly communicated to the public in three ways. The first was through large LCD displays that were installed at the beach and could be immediately updated by the health department from their offices through a cellular phone connection. The LCD screens were mounted on permanent booths where parking fees were collected at the entrances to Huntington and Doheny State Beaches, and on Newport Beach Pier. The signs displayed a map of the beach with the water quality sampling sites indicated by green, yellow or red dots. A green dot indicated that levels of Enterococcus were below the water quality standard at that site. A yellow dot indicated that the health department had posted a warning sign at that location because levels of *Enterococcus* were above the state standard. Red dots were reserved for beach closures due to an actual or suspected raw sewage spill. The second method of communicating results was through a posting to the health department website. The third method of communication was to "tweet" results to subscribers via Twitter. Each method of communication reached a different audience. The signs reached potential swimmers as they entered the beach. The website reached those who may have been considering a trip to the beach and might be concerned about water quality. The "tweets" presumably reached the most frequent beach users as they would have had to subscribe to the service and would be receiving information on almost a daily basis.

RESULTS AND DISCUSSION

Transfer of the qPCR technology to the three laboratories was successful. Throughout the study, the labs continued to produce amplification efficiencies greater than 90% (Figure 1), indicating that their overall proficiency in performing the method remained high. Results produced by the laboratories on replicate filters from the same sample were also highly repeatable, especially for values that exceeded the regulatory action threshold of 104 CE/100 ml (Figure 2). Further, results from split samples run by the relatively inexperienced technicians at the test labs compared favorably with those produced by experienced qPCR users at SCCWRP (Figure 3).

Although the laboratories demonstrated their ability to successfully learn and perform qPCR, the method overestimated levels of *Enterococcus* compared to EPA Method 1600 (Figure 4). There are several reasons why overestimation may have occurred, the most likely of which is that qPCR measures a different endpoint than does EPA 1600. qPCR measures all enterococci caught on the membrane, whether live, dead, stressed, or growing. As long as the cellular membrane remains intact,



Figure 1. Amplification efficiencies produced by the water quality laboratories during the Demonstration Project.

their DNA will be counted. In contrast, EPA 1600 measures only enterococci that grow and produce a colony with a blue halo after 24 hours.

Relying on qPCR data for beach management decisions led to 19 additional beach posting events (false positives) during the study that would not have occurred using EPA Method 1600. This accounted for about 7% of the total samples and was higher than the approximately 2% difference we saw in posting decisions between the two culture-based methods (data not shown). In contrast, qPCR failed to detect only one sample that was above the standard as measured by EPA 1600. The task force had indicated that false negative results were their biggest concern because swimmers would be given a false sense of security and could unwittingly be exposed to



Figure 2. Repeatability between replicate filters from the same sample processed by the water quality laboratories using qPCR (r²=.60).



Figure 3. Comparison of qPCR results produced by water quality laboratories vs. SCCWRP ($r^2 = 0.38$).

contaminated water. While the task force expressed a need to further investigate the reasons for the false positives relative to culture methods, they did not consider the increased number an impediment to adopting the method, expressing the view that the need to provide same-day results trumped the small incremental increase in beach postings.

Another area of technical concern was the number of samples for which there was no result due to PCR inhibition. Overall, 14% of all samples were excluded from the study because the C_T value of the salmon sperm control was greater than the 1.6 cycles allowed. While the health department was able to fall back on the culture method for this study, this would not have been the case if qPCR had been adopted as the sole measurement method. In this case, the task force recommended that additional research be performed to identify ways to correct or eliminate the effects of inhibition. Avenues explored by others include adding a DNA purification step to remove inhibitors (Noble et al. 2010) or using a separate internal control to estimate the extent of PCR inhibition and calculate a "corrected" value (Haugland et al. 2005). In this study, we used the salmon sperm internal control as pass/fail only and chose not to purify DNA due to the added time and variability inherent in this additional step.

One of the critical questions addressed during the demonstration project was whether the new methods could be implemented in a manner that allows for same-day health warnings. While qPCR can theoretically be accomplished in about two



Figure 4. *Enterococcus* values produced by water quality labs for qPCR vs. EPA Method 1600 ($r^2 = 0.46$, p <0.003). (Reference lines indicate California single-sample water quality standard for *Enterococcus*).

hours, this does not include the time for many other steps in the process, such as sample collection, sample preparation, data analysis, quality assurance checks, communication of results to the health officer and time to place signs at the beach. These additional steps take about seven hours under present procedures, exclusive of the time for sample processing, meaning that if sampling was initiated at 7 a.m., warnings using the new rapid methods would not be issued until around 4 p.m.

Prior to initiating the study, the task force identified that a warning issued in the afternoon, after beachgoers have already been exposed to the water, has little benefit relative to one issued the next morning using present culture-based methods. A focal point of the demonstration project was adapting the warning systems to ensure that health warnings were issued by noon of the same day. Modifications of every step in the process were necessary to achieve same-morning warnings.

Modification of the sample collection routine provided the greatest opportunity for reducing the time until a warning is issued. Each of the laboratories collects samples at between 12 and 20 sites, which typically take about four hours because of the transportation time among sites. Starting sampling earlier provides some relief, but only saves about an hour because samplers are unwilling to deploy in the dark for safety reasons. Instead, sampling was reduced to about an hour by limiting the number of sites in the demonstration project and using a second sampling crew for these beaches. Reducing the number of sites was a compromise to achieve the time limitations, but it led the beach managers to recognize that the number of beaches at which they may use rapid methods is limited. They felt they could prioritize use of rapid methods at the most heavily used beaches and/or at those beaches with the most variable water quality. Beaches with a long history below the water quality standards provide less urgency for use of rapid methods. In this way the laboratory can limit resources for additional samplers to high priority beaches, leaving lower priority beaches to be sampled later in the morning or by another individual on a less aggressive time schedule.

The time limitation also led to a number of modifications to laboratory procedures, starting with method selection. Our original intent was to use the Haugland et al (2005) Tagman-based method, but we used the Noble et al (2010) Scorpion-based method instead because we found that the former uses a slower chemistry and requires a longer cycling time, which takes about 45 additional minutes. Similarly, we would have preferred to use a DNA extraction kit to minimize inhibition, but could not afford the extra 45 minutes and new source of error introduced by the multiple pipetting steps (Noble et al. 2010). With these two adjustments, we found that we could achieve a two-hour processing time, but even then only because the technician was processing a limited number of samples and that all preparation steps were completed prior to samples arriving at the laboratory. This said, by the end of the study, technicians were confident they could have processed several additional samples in time to report results as long as they received them at the same early hour as those analyzed for this project.

After samples were processed, communication with the beach manager was expedited by automating the data analysis and quality assurance by writing macros in Microsoft Excel that required that the technician only copy and paste raw data from the thermocycler software into a spreadsheet. Calculations were then performed automatically and values that fell outside of predetermined quality assurance parameters flagged by the software. The larger speed improvement was in developing tools for communicating the health department decisions to the beachgoer in electronic ways that had not been used previously. Historically, warnings were accomplished by either the health department calling one of the sampling laboratories to go back to the beach site to place physical signs at the locations of concern or calling lifeguards at the site to do the same, if lifeguards were present at that beach. These communication steps often added more than an hour. While these mechanisms were still used during the demonstration project, augmentation with the three electronic means of communicating a decision as described above greatly enhanced the speed with which information was disseminated to the beachgoing public.

The cumulative effect of these changes was that warnings were issued often as early as 11:30 a.m. and consistently by 12:30 p.m. at all of the demonstration beaches (Table 3). The few times this was missed were due to extraneous events, such as the sampling vehicle getting stuck in the sand or a staff meeting that called people away from the laboratory for an hour. Warnings around noon were not early enough to save inlanders a trip to an affected beach, but the task force found it acceptable because the majority of beachgoers typically don't arrive until about noon and the warnings were soon enough to keep most of them out of the water at affected sites. Mostly, though, the demonstration project illustrated how challenging it is to achieve warnings by noon and how method modifications that reduce sample processing time further, or automation that allows sample processing to be achieved in the field, are at a premium if rapid methods are to receive widespread adoption.

The task force also reviewed whether cost would be an impediment to method adoption based on what they learned during the demonstration project and considered four types of cost. The first was the per-unit cost for processing samples, which everyone

Table 3. Average time by lab for each step from sample initiation to data transmittal to Health Care Agency.

Sequence	OCPHL	OCSD	SOCWA
Sampler Out	7:00	6:56	7:04
Sampler Return	8:04	7:44	7:48
qPCR Plate In	9:52	9:40	9:30
qPCR Plate Out	10:49	10:58	10:36
Data Reported to HCA	11:22	11:07	10:55

agreed would be a huge impediment if it required an ongoing budget increase. However, per unit cost does not seem likely to increase over present methods. While there is some potential additional cost for separate sample collection teams to address the speed issue, personnel time in the laboratory during the demonstration project was less than for EPA 1600. Cost for expendable supplies averaged about \$30 per sample during the demonstration project compared to less than \$10/sample for traditional methods, but the manufacturers have affirmed that cost of expendables will drop to about \$5 per sample once they are in full production runs.

The second cost is that associated with start-up, including capital equipment, laboratory improvements and training. Our demonstration project experience was that it cost about \$100K to initiate qPCR at a laboratory, though it could be less depending on existing laboratory equipment. Capital costs for a thermocycler and other specialty equipment was around \$60K, though one of the laboratories needed new equipment costing around \$30K that might already be available in larger laboratories. Training time averaged about two person-months, involving one week of hands-on training for each employee and about three weeks of trial implementation before the laboratory staff felt they were proficient enough for routine application. The feedback we received, though, is that these onetime expenditures to upgrade laboratory capability are easier to achieve than an increase in ongoing cost. Perhaps EPA can establish grant programs for such one-time costs after they adopt new criteria based on qPCR.

The third type of cost is for confidence building, as there was a general feeling that there would be a period, probably as long as an entire sampling season, in which both new and traditional methods would be used simultaneously. Some of this is to develop confidence that any differences from management actions taken using previous methods do not result from idiosyncrasies of the new methods or because of laboratory competence. The task force also suggested that simultaneous processing would also be desirable to establish a new baseline for trend analysis and to link to previous regulatory actions based on historical methods, such as total maximum daily load allocations. Although this is also likely a one-time cost, it is of concern because it essentially doubles operating costs for a season, which is likely

to be substantially greater than the one-time capital start-up costs.

The biggest cost impediment is the expectation for more monitoring. The task force's perspective was that a rapid method doesn't help much if you only sample one day a week and then extrapolate that result to the rest of the week. With adoption of rapid methods, laboratories that are struggling to maintain existing programs will be asked to sample multiple days per week. Even more difficult will be the expectation to sample on weekends, when the most swimmers are present, but which typically entails overtime costs. The task force also identified concern that adoption of rapid methods will bring with it the expectation of adaptive sampling, in which a sample greater than typical will require a confirmatory sample(s) that afternoon and additional costs as staff are asked to stay later in the day. Clearly this will increase public health protection as the most egregious problems are investigated immediately, but the laboratories are not presently staffed for this.

Although it is external to the laboratories, another cost that could serve as an impediment is that for establishing a certification program. When laboratories begin performing a new method, they must be certified to ensure that the setup and training steps were effective. Certification will control for many of the factors that could alter results from one laboratory to another, such as sterility of the sample processing environment, storage of reagents, pipette calibration, or dilution preparation. This cost would most likely be borne by the states, most of which are presently under financial pressures that could make establishment of a new certification program problematic.

Epilogue

The demonstration project taught us that the biggest impediments to adoption of qPCR will be logistics and cost, not shortcomings of the underlying technology or challenges with technology transfer. We also learned that while the community is enthusiastic to adopt qPCR, they are unlikely to apply it at all beaches initially.

Interestingly, we learned from our discussions with the laboratories that while the faster processing time was their apparent motivation for adopting qPCR, they are thinking of speed as a Trojan horse for incorporating new technology into the laboratory. The concept of warnings the same morning as sampled is a salable concept, but the greater incentive for adoption may be the ability to use qPCR for source identification. Many of the most promising source identification techniques are based on qPCR and adaptation for their measurement mostly entails a simple exchange of primer-probe sets to a different target. Source identification is appealing because it puts the laboratories in better position to solve the problems that they are presently only identifying using more general assays such as those for Enterococcus. Moreover, the United States Environmental Protection Agency has committed to publishing a rapid method (e.g., qPCR) along with the new criteria it will recommend in 2012 and it has been suggested that they establish different thresholds for beaches without apparent human fecal sources (Boehm et al. 2009), for which source identification technology will be needed.

Initial adoption of qPCR will also serve to establish a market for molecular technologies that will encourage additional innovation. For instance, training and certification challenges can be minimized by automation. Automation may also lead to field deployable devices that can be run by lifeguards or even ones that measure and continuously telemeter data. These are merely engineering adaptations, but ones that rely on commercial investment. Commercial entities are most likely to make this investment following enthusiastic reception of the qPCR technologies by an array of users, such as occurred during the demonstration project.

LITERATURE CITED

Boehm, A.B., N.J. Ashbolt, J.M. Colforld, L.E. Dunbar, L.E. Fleming, M.A. Gold, J.A. Hansel, P.R. Hunter, A.M. Ichida, C.D. McGee, J.A. Soller and S.B. Weisberg. 2009. A sea change ahead for recreational water quality criteria. *Journal of Water and Health* 7:9-20.

Colford, J.M., T.J. Wade, K.C. Schiff, C.C. Wright, J.F. Griffith, S.K. Sandhu, S. Burns, M. Sobsey, G. Lovelace and S.B. Weisberg. 2007. Water quality indicators and the risk of illness at beaches with non-point sources of fecal contamiation. *Epidemiology* 18:27-35.

Converse, R.R. 2009. Bridging the gap between molecular and traditional assessments of recreational

water quality. Ph.D. Dissertation, University of North Carolina. Chapel Hill, NC.

Griffith, J.F., Y. Cao, C.D. McGee and S.B. Weisberg. 2009. Evaluation of rapid methods and novel indicators for assessing microbiological beach water quality. *Water Research* 43:4900-4907.

Grant, S.B., B.F. Sanders, A.B. Boehm, J.A. Redman, J.H. Kim, R.D. Mrse, A.K. Chu, M. Gouldin, C.D. McGee, N.A. Gardiner, B.H. Jones, J. Svejkovsky, G.V. Leipzig and Brown, A. 2001. Generation of *Enterococci* bacteria in a coastal saltwater marsh and its impact on surf zone water quality. *Environmental Science and Technology* 35:2407-2416.

Haugland, R.A., S.C. Siefring, L.J. Wymer, K.P. Brenner and A.P. Dufour. 2005. Comparison of *Enterococcus* measurements in freshwater at two recreational beaches by quantitative polymerase chain reaction and membrane filter culture analysis. *Water Research* 39:559-568.

Leecaster, M.K. and S.B. Weisberg. 2001. Effect of sampling frequency on shoreline microbiology assessments. *Marine Pollution Bulletin* 42:1150-1154.

Messer, J.W. and A.P. Dufour. 1998. A rapid, specific embrane filtration procedure for enumeration of *Enterococci* in recreational water. *Applied and Environmental Microbiology* 64:678-680.

Nakano, F. 2001. Assembly Bill 639. State of California. Sacramento, CA.

Noble, R.T., A.D. Blackwood, J.F. Griffith, C.D. McGee and S.B. Weisberg. 2010. Comparison of rapid quantitative PCR-based and conventional culture-based methods for enumeration of *Enterococcus* spp. and *Escherichia coli* in recreational waters. *Applied and Environmental Microbiology* 76:7437-7443.

Wade, T.J., R.L. Calderon, E. Sams, M. Beach, K.P. Brenner, A.H. Williams and A.P. Dufour. 2006. Rapidly measured indicators of recreational water quality are predictive of swimming-associated gastrointestinal illness. *Environmental Health Perspectives* 114:24-28.

Wade, T.J., R.L. Calderon, K.P. Brenner, E. Sams, M. Beach, R. Haugland, L. Wymer and A.P. Dufour. 2008. High sensitivity of children to swimmingassociated gastrointestinal illness. *Epidemiology* 19:375-383. Wade, T.J., E. Sams, K.P. Brenner, R. Haugland, E. Chern, M. Beach, L. Wymer, C.C. Rankin, D. Love, Q. Li, R. Noble and A.P. Dufour. 2010. Rapidly measured indicators of recreational water quality and swimming-associated illness at marine beaches: a prospective cohort study. *Environmental Health* 9:66.

ACKNOWLEDGMENTS

The authors wish to thank the task force members and the following for their assistance to project success: Denene Blackwood, Yiping Cao, Reagan Converse, Darcy Ebentier, Rachel Noble, Meredith Raith, Orange County Sanitation District, Orange County Public Health Care Agency, South Orange County Wastewater Authority, Idexx Corp. Funding for this project was provided by the American Recovery and Reinvestment Act of 2009 and the California State Water Resources Control Board.