



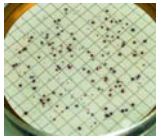
Development of Molecular Methods to Monitor Waters for Threats to Human Health

Kelly Goodwin, Chris Sinigalliano, David Wanless

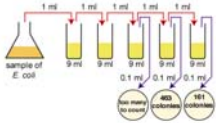


Monitoring programs seek improvements

...because traditional fecal indicator assays are:



- Too Slow
- Difficult to meet hold time requirement (6 hrs)
- No ability to source track (in time or space)
- No pathogen detection
- Axioms of indicator concept are sometimes violated; consequences to human health are unclear



The first critical step is to concentrate the sample & extract nucleic acids



A) filter water to concentrate cells



B) extract genomic DNA (or RNA)

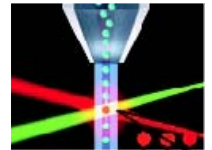
The next step is to test various approaches of molecular detection



qPCR
(quantitative)



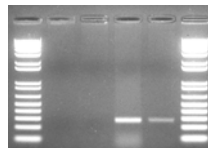
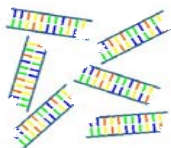
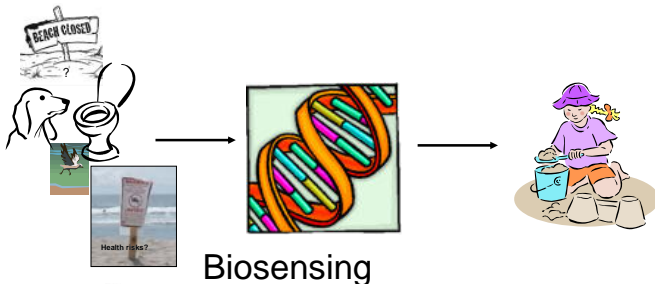
electrochemistry
(hand-held)



Luminex
(high throughput)



We seek to improve monitoring of microbial contaminants using molecular biological techniques



Targets

- Enterococcus* spp.
 - Escherichia coli* *Shigella* spp.
 - Bacteroides Fragilis Group
 - Bacteroides distasonis*
 - Enterococci human marker
 - Bacteroides human marker
 - Bacteroides dog marker
 - E. coli* O157:H7
 - Campylobacter jejuni*
 - Salmonella* spp.
 - Staphylococcus aureus*
 - adenovirus
- standard fecal indicators
- alternative fecal indicators
- source tracking markers
- pathogens
(intestinal & dermal bacteria and virus)