

# FECAL COLIFORM POLLUTION TESTING - NORTH CREEK AND HICKS LAKE



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## TOTAL COLIFORM, FECAL COLIFORM, AND *E. COLI*

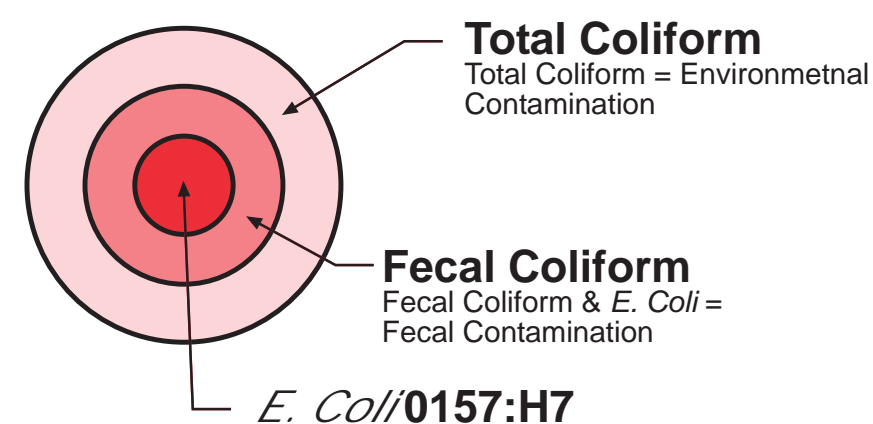


Figure 1, After Washington State Dept. of Health, 2007.



After Washington State Dept. of Ecology, 2002

## BACKGROUND

Fecal Coliforms are thermotolerant bacteria capable of growing at an elevated temperature of 44.5°C. This group of organisms includes species such as: *Klebsiella pneumoniae* and *Citrobacter freundii*, but are primarily represented by *Escherichia coli*. *E. coli* is known as the 'best coliform indicator of fecal contamination' (Hach 2007) This suggest surface waters with elevated Fecal Coliforms has the potential risk of containing harmful pathogens. (Hach Company, 2007)

Poor water quality due to Fecal Coliform and *E. coli* contamination prevents recreational use and poses a threat to public and environmental health. High bacteria levels also lead to low dissolved oxygen levels that make sustainability hard for aquatic species (Meehan, M., & Kalenius, S. 2004, June).

Washington Administrative Code 173-201A-200 regulates the use of fresh waterbodies when Fecal Coliform levels exceed the state standards for defined waters. Secondary Contact Recreation, waters with limited use, must meet the following criteria: 'Fecal Coliform organism levels must not exceed a geometric mean value of 200 colonies/100mL, with not more than 10 percent of all samples (or any single sample when less than ten sample points exist) obtained for calculating the geometric mean value exceeding 400 colonies/100mL.

## SITE INFORMATION

**HICKS LAKE (Figure 2c)**  
Hicks Lake, also known as Lake Garrett, resides in Lakewood Park in Seattle, Washington. This 4-acre lake sitting on three aquifers is fed by springs, but is heavily influenced by surface run-off, neighborhood septic tanks, and the local high school athletic field. Because Hicks Lake has no natural outlets, water overflow is pumped out of the lake to nearby Salmon Creek during heavy rain events. Hicks Lake has not been used for recreational purposes since 1975 due to poor water quality. This presents concerns when contaminated lake water is pumped into Salmon Creek which feeds directly into the Puget Sound. (King County Department of Natural Resources and Parks, 2005.)

**NORTH CREEK (Figure 2a,b)**  
North Creek has a 30 square mile basin running from Everett to the Sammamish River in Bothell. High Fecal Coliform contamination is the result of failing septic systems, waste from pets, livestock and wildlife. Since it is a primary salmon habitat, its contamination is a great concern for the City of Bothell (Meehan, M., & Kalenius, S. 2004).

## TESTING PROCEDURES

Our laboratory testing was conducted at the Washington State Department of Health Public Health Laboratories in Shoreline, Washington. We performed two procedures using the Millipore m-ColiBlue24® Broth and Idexx Colilert®. Each sample was run in parallel at two different temperatures, 35°C and 44.5°C, to determine Total Coliform, Fecal Coliform, and *E. coli* contamination levels.

### Idexx Colilert® Methodology:

Colilert uses a 'Defined Substrate Technology' which allows for simultaneous detection of Total Coliforms and *E. coli*. Two nutrient indicators, ONPG and MUG, are used to detect metabolic activity of the enzymes  $\alpha$ -galactosidase and  $\alpha$ -glucuronidase, respectively. After adding the Colilert reagent to a 100mL sample and incubating for a 24 hour period at 35°C, detection of Total Coliforms can be observed by a color change from clear to yellow. Samples which turns yellow are subjected to long-wavelength UV light (362nm) which will fluoresce when *E. coli* is present. Figure 3 (bottom).

### Membrane Filtration Methodology using Millipore m-ColiBlue24®:

Millipore m-colibblue24® is a lactose based media containing inhibitors which allows for simultaneous detection of Total Coliforms and *E. coli*. Water samples are vacuum filtered through 47mm membrane filters with 0.45 mm porosity trapping coliforms on the filter surface. The filter is then transferred into a petridish containing an absorbent pad soaked in the m-ColiBlue24 media and inverted for incubation. After a 24 hour incubation at 35°C, red and blue colonies will develop. Total Coliforms are determined by adding the total number of red and blue colonies. *E. coli* is determined by adding the total number of blue colonies. (Hach Company, 2007, Millipore Company, 2007) Figure 3 (top).

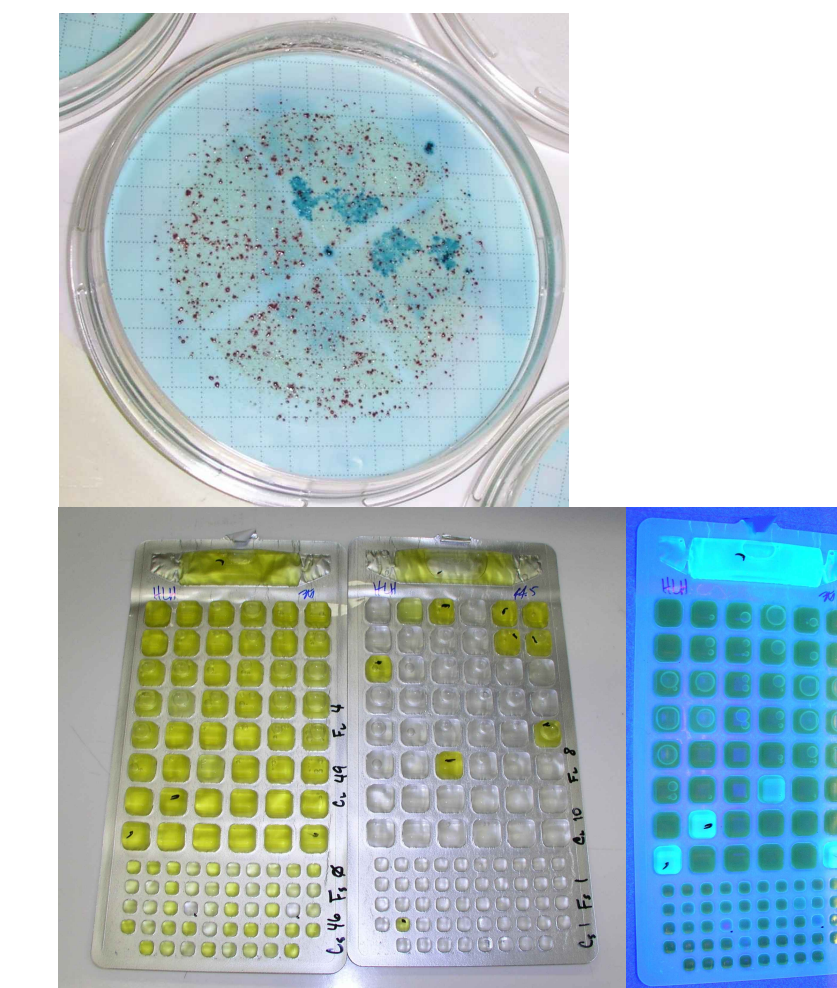


Figure 3, m-ColiBlue24 Broth and Colilert testing media. (Melanie Panoke, 2007)

## ANALYSIS

- H1: *E. coli* is an equivalent indicator of water quality compared to Fecal Coliform testing.  
**Conclusion:** *E. coli* is an indicator of Fecal Coliform contamination. However, results show evidence that levels of *E. coli* is not directly proportional. Figure 5.
- H2: The Colilert test will give equivalent results to the m-ColiBlue24 Broth test for the same samples.  
**Conclusion:** Colilert and m-colibblue24 do not produce comparable results, but do provide trends of excessive coliform contamination. Colilert can produce most probable number values for a 100mL volume without the need for further dilutions. M-ColiBlue24 may require additional dilutions depending on the coliform density of each sample. Confluent growth and excess debris trapped on the filter may interfere with the interpretation of results. Figure 6.
- H3: Coliform levels will be lower downstream in North Creek than upstream due to vegetative filtering.  
**Conclusion:** Results prove Fecal Coliforms slightly decline downstream in North Creek. However, additional testing would be required to demonstrate statistically. Figure 4.
- H4: Fecal Coliform in Hicks Lake will exceed State standards.  
**Conclusion:** Results prove Fecal Coliforms consistently exceed the 'Secondary Contact Recreational Fecal Coliform Value of 200 Colonies/100mL.' (WAC-173-201A-200, ) Figure 6.

## HYPOTHESES

- H1: *E. coli* is an equivalent indicator of water quality compared to Fecal Coliform testing.
- H2: The Colilert test will give equivalent results to the m-ColiBlue24 Broth test for the same samples.
- H3: Coliform levels will be lower downstream in North Creek than upstream due to vegetative filtering.
- H4: Fecal Coliform in Hicks Lake will exceed State standards.

### North Creek Site A



### North Creek Sites B and C



### Hicks Lake Sample Sites

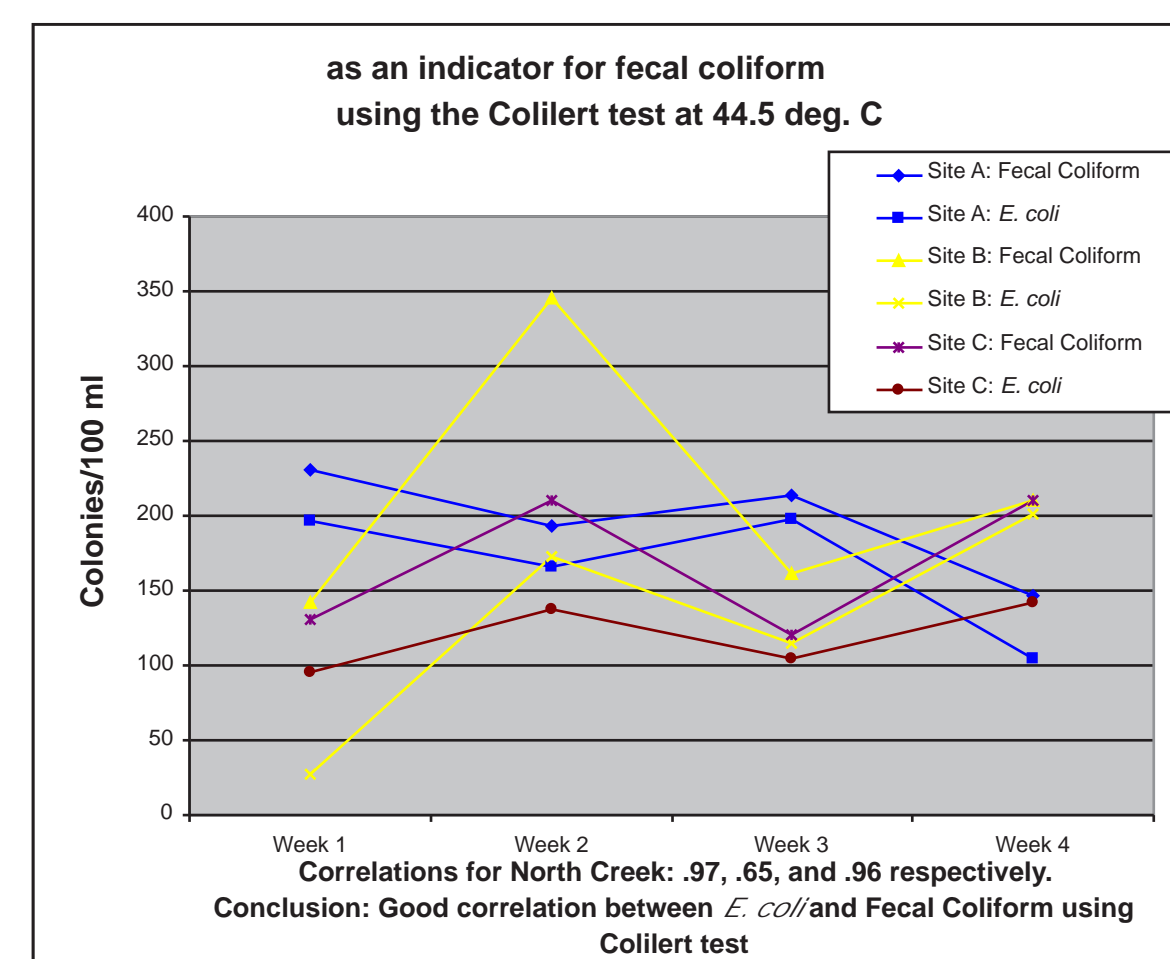
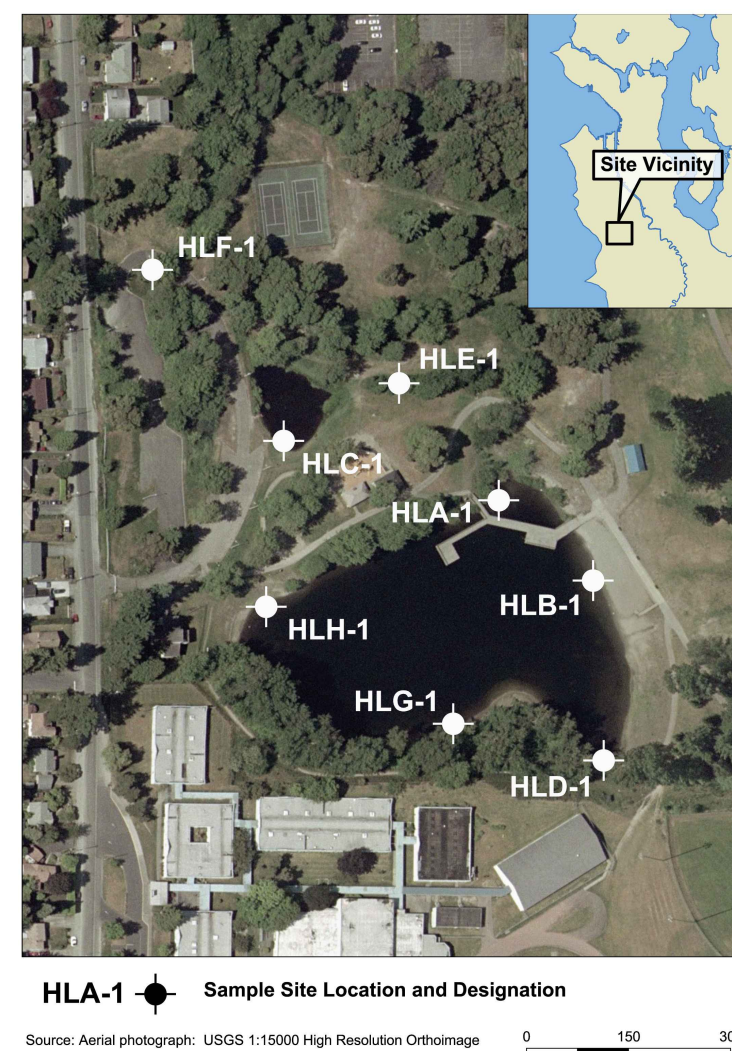


Figure 4, Comparison of Fecal Coliform and *E. coli* levels in North Creek samples using the Colilert method at 44.5°C.

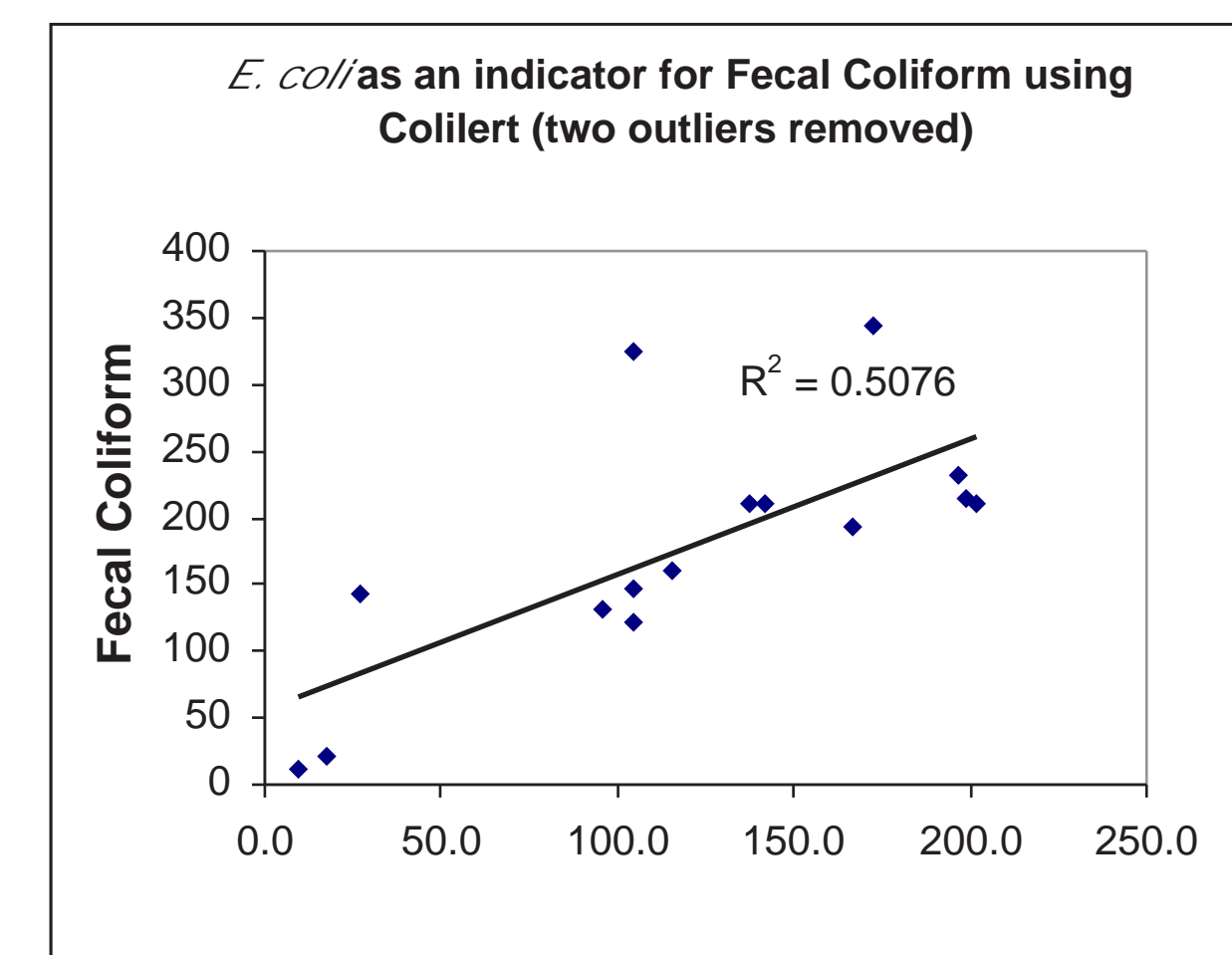


Figure 5, Statistical correlation between *E. coli* and Fecal Coliform in terms of their use as biological indicators of water quality.

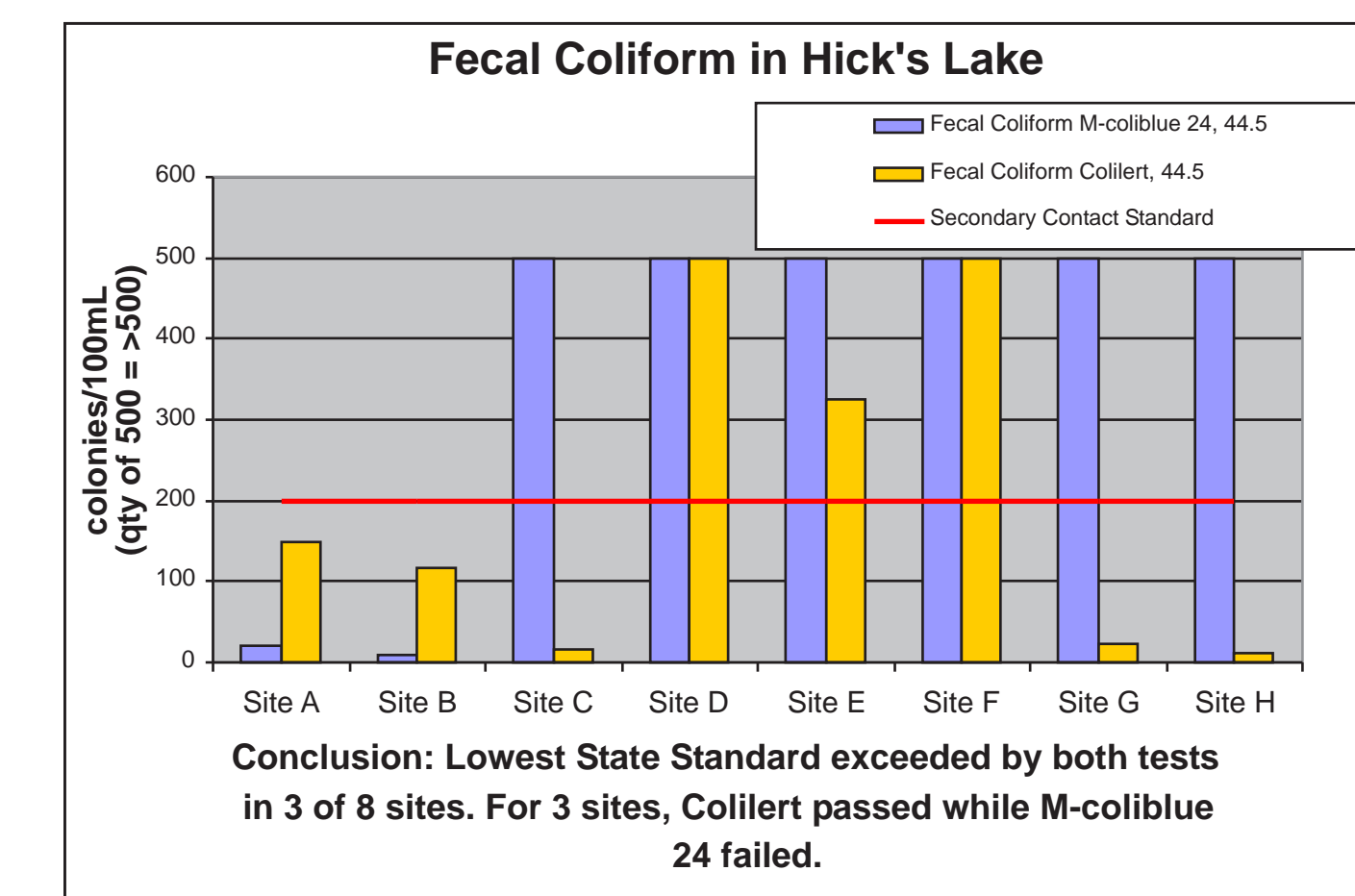


Figure 6, Fecal Coliform levels in Hicks Lake.