Results Information

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Customer ID: Adrian MI Residents
Project #: P-473
Sample Date Received: 12/19/18
Date of Analysis: 12/19/18 - 12/21/18
Sample Type: 2 Tap Water
Analysis Type: Cyanobacteria ID
Microcystin Toxin Gene ID

Sample Information:

Two tap water samples designated as Sample D and Sample E collected on December 17, 2018 were submitted for analysis. Sample collection locations are listed below:

Sample D – [redacted]
Sample E –

Analysis Description:

The analysis of the submitted water samples involved determining the presence/absence of Cyanobacteria and the Microcystin producing toxin gene in each of the two samples using real time PCR analysis.

Procedure: DNA Extraction

500 ml of each submitted water sample was vacuum filtered through a 0.45 micron filter membrane that proceeded to capture any microorganisms present in the water sample. Following vacuum filtration, the individual filter membranes were removed from the filter column and microorganism DNA was extracted from each filter membrane using the Generite DNA EZ ST1 DNA extraction kit. The DNA extractions were used in all subsequent analysis.

Procedure: PCR Analysis Cyanobacteria, Microcystin Producing Toxin Gene

Qualitative PCR analysis to determine the presence/absence of Cyanobacteria and the Microcystin producing toxin gene incorporated oligonucleotide primers that target specific DNA marker sequences. The PCR targets and the location of the specific DNA markers for each target are listed in the table below:

Target	Location of DNA Marker
Cyanobacteria Genera	Cyanobacteria nif Gene
Microcystin Toxin Gene	Microcystin Synthetase A Gene

Real Time PCR analysis was performed using the Stratagene Mx 3005P Real Time PCR System instrument. PCR analysis incorporated oligonucleotide primers that target specific marker DNA sequences unique to Cyanobacteria and the Microcystin producing toxin gene. Appropriate positive and negative control samples were included in PCR experimentation. Replicates of each sample were run for quality assurance purposes and to confirm the observed results.

Following the PCR procedure, the PCR products were run on a 1.5 % agarose gel at 100V for 60 minutes, after which the agarose gel containing the PCR products was visualized under UV light and examined for PCR amplification products that represent the amplification of the target DNA markers.

Results:

Results of the qualitative PCR testing indicate the presence (positive) or absence (negative) of the specific DNA markers for Cyanobacteria and the Microcystin producing toxin gene through PCR amplification of the specific DNA marker sequence.

A sample testing positive for a specific DNA marker confirms the presence of the organism or toxin gene in the sample. A sample testing negative for a specific DNA marker confirms the absence of the organism or toxin gene in the sample.

Sample	Cyanobacteria	Microcystin Toxin Gene
D - [redacted]	negative	negative
E - [redacted]	negative	negative

These results were verified by Tom Prychitko, Laboratory Director for Helix Biological Laboratory.