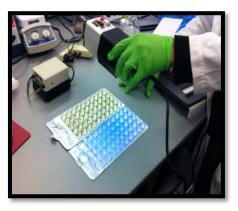
The Efficacy of LifeStraw® Water Filters in Filtering *Enterococci* from Various Water Samples





By William Echavarria¹, Mauricio Gonzalez², Nazish Nawaz¹ and Kathleen Nolan¹,

¹St. Francis College, Brooklyn, NY, ²New York Harbor School, Governors Island, NY

Rationale:

Enterococci, contained in normal human fecal flora, can be dangerous to humans if ingested in large quantities. LifeStraw® water filters (by Vestergaard) have been shown to filter water that might be contaminated with Enterococci and other bacteria. In this study, samples of undiluted marine and fresh water were filtered using a LifeStraw® water filter in an attempt to test the efficacy of these filters.. Tap water was used as a negative control. Test samples included brackish water from the Brooklyn Bridge Park (East River) and a fresh water sample from the Spuyten Duyvil Pond in the Bronx. Unfiltered samples tested positive with an Enterolert® detection kit. Samples filtered through the LifeStraw® water filter yielded the same results as the negative control, as did water from an aquarium containing zebrafish. Therefore, we have shown that LifeStraw® water filters are an efficacious way to filter water contaminated with Enterococci. Next steps include testing additional water samples from various locations with the LifeStraw®.

Null Hypothesis:

There is no difference between LifeStraw post filter water quality and pre-filtered water in counts of *Enterococcus*.

Materials:

- 1 Sterile 10 ml pipette
- 1 Safety Pipette Filler Bulb
- 1 1-liter beaker
- 1 LifeStraw® filter
- 100mL Distilled water
- 2 100mL samples of marine water
- 2 100mL samples of fresh water
- 4 IDEXX Quanti-trays Most Probable Number (MPN)
 - **Swimming Suitability:** The average (based on the log mean) MPN/100 mL* of at least 5 evenly spaced samples in a 30-day period should not exceed 1,000 for total coliform, 200 for fecal coliform, 126 for E. coli, or 33 for Enterococcus.
 - **Drinking water requirements**: The EPA has set the following maximum contaminant levels (MCLs) on treated drinking water:

"For systems that analyze at least 40 samples per month, no more than 5% of the samples may be total coliform positive. For systems analyzing fewer than 40 samples per month, no more than 1% of the samples may be total coliform positive" (AWWA, 1990b).

- 1 Iron to seal Quanti-tray
- 4 IDEXX Enterolert reagents*
- 1.41 ± 0.5 °C incubator
- 1 Long-wave UV Lamp (365-366 nm).
- 4 Sterile transparent non-fluorescent 100 ml vessels
- 4 50 ml sterile tubes

* Toxicity -

There are two items with a threat of toxicity in this experiment – the enterococcus bacteria and the reagent. The reagent is hazardous to human health when inhaled, swallowed, injected or if it should come in contact with the skin. If this bacteria are accidentally swallowed, this could be harmful because this bacteria is usually found in human and in animal intestines but in a very low level. If this bacteria is present and increases, then it could attack tissue cells and even take over the good flora's tasks. Inhaling this bacteria would lead to similar problems as if would be

swallowed. In skin contact, it's harmful mostly if you have a cut or if your hands or whatever part of your body that became in contact is place in your mouth.

Safety Measures

To avoid inhaling the reagent and any bacteria, one must open the reagent container away from one's body and directly into the sterile vessel.

To avoid swallowing the sample one must shake the bottle slowly and always from one's body.

To avoid injection or contact with skin, wear gloves, goggles and a lab coat at all times.

Methods:

- 1) We added the unfiltered water samples and tap water to the 100 ml sterile vessel (one sample at a time, not all on the same day).
- 2) Then we added Enterolert reagent and shook the bottle for 60 seconds until the reagent had dissolved in the water.
- 3) Next we poured the solution into the Quanti-Tray (the best way to open the tray is by pushing the top of the Quanti-Tray while forming your hand into a complete "U"- shape you push the tray into your "U" shaped hand. Pull the top part away from the plastic site and avoid touching the inside of the tray. Then iron the tray in a slow and steady motion as this will seal the tray, making sure that the iron is hot; otherwise it will not work.
- 4) We placed the tray in the 41°C incubator for 24 hours and the results were read within 24 to 28 hours. If not, then the results would not have been accurate.
- 5) After the incubation we placed the tray under the UV lamp and looked for a light blue color which meant that the solution was positive for enterococcus bacteria.

- 6) We next filtered the water samples (one at a time, not all on the same day) by pouring 50 ml at a time into the top of the Lifestraw and letting it drip via gravity into a second 50 ml tube (2 tubes were collected in all for each sample.)
- 7) We then repeated Steps 1-5 with our filtered water samples.

Results

The tap water negative control did not show any growth, and the Brooklyn Bridge Park marine water sample turned blue, indicative of *Enterococcal* growth, so we felt confident enough to proceed and test the LifeStraw[®]. The LifeStraw[®] was able to successfully filter both the marine and fresh water samples (Brooklyn Bridge Park and Spuyten Duyvil Pond). The aquarium water with the zebrafish did not depict *Enterococcal* growth, probably because of the tank filter. We did not dilute the marine water (possibly to be done in a future experiment) but, since the tray turned completely blue, the *Enterococci* were too numerous to count. The undiluted pond water, however, supported less *Enterococci*, which we were thus able to count.

Conclusion:

Our result showed that the LifeStraw filters out *Enterococcus* bacteria which makes us reject our null hypothesis. Our results were conclusive and based on the data collected and the samples that were done in the lab. We came to this conclusion by comparing our sample water with our control water and our filtered water.

Future Research Needs

In water, *Enterococci* are used as indicators of environmental contamination, because they are found in high concentrations in feces, and exposure to *Enterococci* is linked to adverse health effects in swimmers. LifeStraws® safely filter waters that have been contaminated with *Enterococci* and would then allow someone to drink potentially contaminated water using the Lifestraw filter. A method to detect sources of *Enterococci* found in surface waters would be beneficial. This could be in the form of a molecular microbial source tracking tool, analogous to the tools used to track sources of *Bacteroidales*. Researchers have just recently started to use *Enterococci* on hands as indicators of hand hygiene. Additional studies that link *Enterococci* density on skin to hand hygiene practices (like hand washing) and health outcomes such as respiratory disease and gastrointestinal illness will further lend credence to their use as hygiene indicators.

Literature Cited section:

Radimersky, T.; Frolkova, P.; Janoszowska, D.; Dolejska, M.; Svec, P.; Roubalova, E.; Cikova, P.; Cizek, A.; Literak, I. (2010) Antibiotic resistance in faecal bacteria (Escherichia coli, Enterococcus spp.) in feral pigeons.(Clinical report) Journal of Applied Microbiology, Vol.109(5), p.1687(9).

Ferretti, James A.; Tran, Hiep V.; Cosgrove, Elizabeth; Protonentis, John; Loftin, Virginia; Conklin, Carol S.; Grant, Robert N. (2011) Comparison of Enterococcus density estimates in marine beach and bay samples by real-time polymerase chain reaction, membrane filtration and defined substrate testing; Marine Pollution BulletinVol.62(5), pp.1066-1072.

Suter, Elizabeth; Juhl, Andrew R.; O'mullan, Gregory D. (2011) Particle association of Enterococcus and total bacteria in the lower Hudson River Estuary, USA.(Report) Journal of Water Resource and Protection (JWARP), Vol.3(10), p.715(11).