

# Efficacy of Peragreen 5.6% (Peracetic Acid) Against *E. coli* in Irrigation Water (Part 2)

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#### Purpose

In a previous study conducted by Enviro Tech Chemical Services, it was determined that 2-ppm peracetic acid (PAA) from Peragreen 5.6% was able to reduce *E. coli* from 3088 MPN/100-mL to non-detect (ND) within 5-minutes and 5-ppm PAA was able to reduce *E. coli* from 3088 MPN/100-mL to ND within 2-minutes<sup>1</sup>. The purpose of this study is to determine the lowest contact time it takes for 2- and 5-ppm PAA to reduce *E. coli* to ND in irrigation water.

## **Materials and Methods**

E. coli Culture

A freeze-dried pellet *E. coli* (ATCC® 25922, Bactrol™ Plus) was dissolved in 10-mL of Brain Heart Infusion (BHI) Broth (Hardy Diagnostics Cat. No. K25). Next, five aliquots were taken and plated on individual 5% Sheep Blood Tryptic Soy Agar (blood agar) plates. The plates were incubated at 37°C for 24-hours. Next, three, 10-µL inoculation loops of bacteria was aseptically transferred from the blood agar plate to 2500-mL of Modesto city water (irrigation water). The *E. coli* solution culture was mixed to ensure homogeneity.

## Control Sample Analysis

One 100-mL sample of the untreated inoculated water was aseptically transferred to a sterile jar containing sodium thiosulfate neutralizer. Idexx Colilert-18 powder reagent was then added to the jar and agitated until dissolved. The sample was then transferred to an Idexx Quanti-Tray/2000, heat sealed, and placed in an incubator at  $35.0 \pm 0.5$ °C.

<sup>&</sup>lt;sup>1</sup> Donabed, Joseph Efficacy of Peragreen 5.6% (Peracetic Acid) Against E. coli in Irrigation Water October 2, 2019 <a href="https://envirotech.com/wp-content/uploads/2016/01/Peragreen-5.6-Irrigation-Water-Efficacy-Report-E.-coli-1.pdf">https://envirotech.com/wp-content/uploads/2016/01/Peragreen-5.6-Irrigation-Water-Efficacy-Report-E.-coli-1.pdf</a>



This sample would serve as the undiluted and untreated control sample for *E. coli*. Next, a 1 mL aliquot of the untreated inoculated water was transferred to a sterile jar containing 99 mL of sterile reverse osmosis (RO) water and sodium thiosulfate neutralizer. Idexx Colilert-18 powder reagent was then added to the jar and agitated until dissolved. The sample was transferred to an Idexx Quanti-Tray/2000, heat sealed, and placed in an incubator at  $35.0 \pm 0.5$ °C. This sample would serve as the 1:100 diluted and untreated control sample for the study.

Next, the *E. coli* stock solution was divided into three individual, 1000-mL samples. The individual samples would be treated with 2-, and 5-ppm active PAA from Peragreen 5.6%.

### **PAA Treatment**

The *E. coli* inoculated water samples were treated with a nominal 2- and 5-ppm PAA from Peragreen 5.6% by dosing each sample with 32- and 80- $\mu$ L, respectively. The solutions were mixed via magnetic stir bar at 200-rmp to simulate in-line mixing. After 1, 2, 3, 4, and 5-minutes of contact, 100-mL samples of the treated water were aseptically transferred to individual sterile jars containing sodium thiosulfate neutralizer. Idexx Colilert-18 powder reagent was then added to the jar and agitated until dissolved. The sample was then transferred to an Idexx Quanti-Tray/2000, heat sealed, and placed in an incubator at 35.0  $\pm$  0.5°C then enumerated.

## Results

<u>Table 1</u> lists the *E. coli* counts in the control (untreated) sample as well as the 2-ppm PAA treated samples after 1, 2, 3, 4, and 5-minutes of contact.

Description	E. coli (MPN/100-mL)	% Reduction
Control (Untreated)	2247	NA
2-ppm PAA, 1-minutes	45.2	98.0
2-ppm PAA, 2-minutes	22.3	99.0
2-ppm PAA, 3-minutes	2.0	99.9
2-ppm PAA, 4-minutes	ND	>99.9
2-ppm PAA, 5-minutes	ND	>99.9



<u>Table 2</u> lists the *E. coli* counts in the control (untreated) sample as well as the 5-ppm PAA treated samples after 1, 2, 3, 4, and 5-minutes of contact.

Description	E. coli (MPN/100-mL)	% Reduction
Control (Untreated)	2247	NA
5-ppm PAA, 1-minutes	5.1	99.8
5-ppm PAA, 2-minutes	ND	>99.9
5-ppm PAA, 3-minutes	ND	>99.9
5-ppm PAA, 4-minutes	ND	>99.9
5-ppm PAA, 5-minutes	ND	>99.9

## Conclusions

- The untreated control sample contained an E. coli count of 2247 MPN/100-mL
- Treatment with a nominal 2-ppm PAA from Peragreen 5.6% for 4-minutes decreased the *E. coli* count to ND which equates to a >99.9% reduction
- Treatment with a nominal 5-ppm PAA from Peragreen for 2-minutes resulted in no viable *E. coli* (ND) which equates to a >99.9% reduction.

It should be noted that while laboratory studies are often representative, full-scale application performance may differ due to the presence of variables that cannot be replicated in the laboratory.