



Investigation of E|Line Water Quality Test kit

Objectives:

1. To evaluate the sensitivity of **E|Line Water Quality Test kit** to detect known concentrations of representative waterborne bacteria.
2. To investigate the sensitivity of **E|Line Water Quality Test kit** in the detection of representative biofilm bacteria.

Materials and Methods:

Bacterial suspensions of commercial *Staphylococcus aureus* ATCC # 25923, *Escherichia coli* ATCC # 25922, and *Pseudomonas aeruginosa* ATCC # 49129 were prepared by aerobically culturing bacteria in 10 mL of trypticase soy broth at 37°C for 24 hours. Working concentrations were calculated based on the measured optical density of each bacterial culture using the Spectronic 2000 spectrophotometer (Thermo Fisher Scientific, Inc). Stock cultures were diluted to 10¹, 10², and 10³ colony forming units (cfu)/mL using sterile saline. **E|line Bacteria Test Vials** (Germiphene Corporation) were filled with 1 mL aliquots of *S.aureus* and *E.coli* dilutions. Similarly, *P.aeruginosa* dilutions were added to the **E|line Biofilm Test Vials** (Germiphene Corporation). Bacterial test solutions were removed from test vials after 25 minutes of exposure and then incubated in the **E|line Dry Block Incubator** (Figure 3) at 37°C for 18-24 hours. Vials were visually inspected for internal color changes immediately following incubation. Control specimens were created using sterile saline and evaluated in the same manner as test specimens.

Results:

Test Bacteria

Stock concentrations were consistent with typical 24-hour cultures for each microorganism (Table 1). No abnormalities in growth rate or contamination were detected.

Test Vials

After 18-24 hour incubation, all bacterial test vials exposed to 10¹ (n=3) and 10² cfu/ mL (n=3) bacterial solutions (*S. aureus* and *E. coli*) remained in their original green state and therefore tested negative for containing 200 cfu or more (Table 2). Positive results were observed for the test specimens exposed to representative microorganisms at 10³ cfu/mL concentration (n=3). These results were easily detectable by the distinct color change from green to yellow

Table 1. Stock concentrations of test bacteria.

Microorganism	Concentration (cfu/mL)
<i>S.aureus</i> ATCC # 25923	1.9 x 10 ¹⁰
<i>E.coli</i> ATCC # 25922	9.6 x 10 ⁸
<i>P.aeruginosa</i> ATCC # 49129	4.6 x 10 ⁸

Table 2. Bacteria test vial results.

Concentration (cfu/mL)	# Positive tests
<i>Staphylococcus aureus</i>	
Sterile saline	0/3
10 ¹	0/3
10 ²	0/3
10 ³	3/3
<i>Escherichia coli</i>	
Sterile saline	0/3
10 ¹	0/3
10 ²	0/3
10 ³	3/3

Figure 1: Bacteria test vials

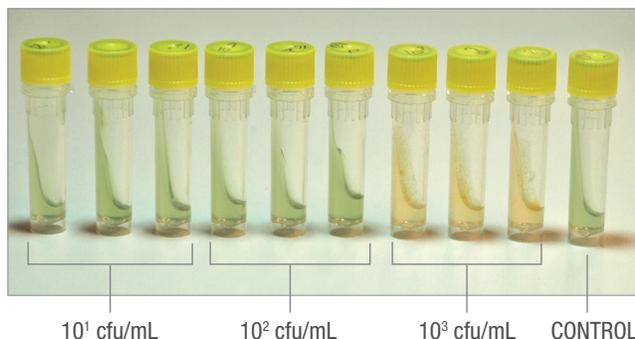
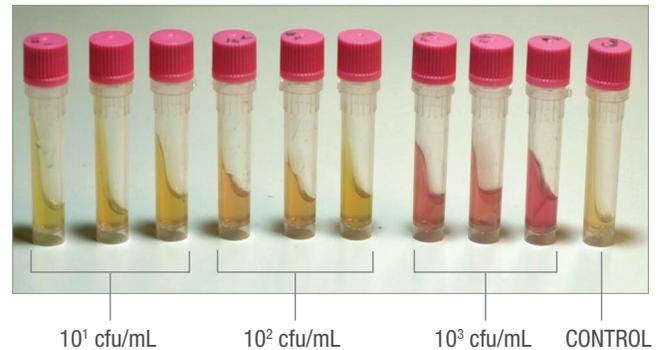


Table 3. Biofilm test vial results.

Concentration (cfu/mL)	# Positive tests
<i>Pseudomonas aeruginosa</i>	
Control	0/3
10 ¹	0/3
10 ²	0/3
10 ³	3/3

Figure 2: Biofilm test vials.



(Figure 1). Biofilm test results demonstrated the same level of accuracy in detecting representative microbes as the bacteria test vials (Table 3). Results were negative, and no color change was observed when the biofilm test vials were challenged with the two lowest concentrations of *P. aeruginosa*, 10¹ and 10² cfu/mL. When exposed to the highest challenge concentration, a pink color resulted indicating a positive test result (Figure 2). All control specimens containing sterile saline produced negative results.

Summary:

Within days of installing a new dental unit, biofilms begin to form in the lumens of waterline tubing and bacterial levels reached 50-100 times the amount recommended by the American Dental Association (<200 cfu/mL). Commercial waterline treatments can remove biofilms and maintain acceptable microbial levels if used properly and monitored frequently. However, our research has found that many practices are unknowingly using treatments incorrectly and waterline microbial levels are alarmingly high. The *E|line bacteria and biofilm test system* offers dental healthcare practitioners a rapid, reproducible, and easy way to monitor the efficacy of their waterline treatment system.



Figure 3: E|line Dry Block Incubator.