



# Efficacy of “One Drop” Disinfectant for Inactivation of Indicator Microorganisms

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## ABSTRACT

Water is often microbially contaminated and must be disinfected at point of use to be made safe to drink. Chlorine is widely used for drinking water disinfection but often rejected by users due to taste and odor problems and toxic disinfection by-products. As an alternative, One Drop is an aqueous solution of natural ionic minerals, including silver, gold, aluminum and copper believed to have microbicidal properties when added in small quantities to contaminated water, yet is harmless to humans and does not affect taste and odor. As little as one drop of the solution per liter of water may inactivate microbes. One Drop may also interact with negatively charged microbes to facilitate settling or removal by filtration. One Drop was tested to reduce turbidity and inactivate indicator microorganisms in raw surface water. One-liter volumes of raw water containing 0, 1, 2, or 4 drops of One Drop were measured for turbidity and concentrations of added *E. coli* B, *Klebsiella terrigena*, spores of *Bacillus subtilis* variety niger, and male-specific coliphage MS-2 initially and after 10, 30, 90, and 240 minutes. Additionally, indigenous heterotrophic plate count (HPC) bacteria and indigenous spores were measured. We compared simple addition to water, as suggested by the manufacturer, with a standard coagulation/flocculation, or jar test method. There were no changes in turbidity with increasing drops of One Drop but appreciable reductions of some microbial indicators. Reductions of  $>6\log_{10}$  ( $>99.9999\%$ ) were achieved for *E. coli* B and *Klebsiella terrigena* and  $>3\log_{10}$  ( $>99.9\%$ ) were achieved for HPC bacteria and MS-2. Reductions generally increased over time and with increasing number of drops added. Microbe reductions with One Drop were more rapid and extensive than natural reductions without One Drop. The addition method suggested by manufacturer produced more extensive reductions than that by the jar-test method. We conclude that One Drop reduces concentrations of some representative indicator microorganisms in raw water and may serve as an effective and low-cost means of household or other point-of-use water treatment.

## INTRODUCTION

- Cost-effective and simple physical and chemical point-of-use drinking water disinfection technologies have been developed, evaluated and applied in household use in developing countries,
- Current options, such as boiling, chlorine, sunlight, filtration and combined chemical coagulant-flocculant-disinfectant, have limitations and deficiencies
- Physical methods do not provide residual disinfectant
- Chlorine may be rejected due to taste and odor and is ineffective against *Cryptosporidium parvum*
- Combined coagulants-flocculant-disinfectant products are more costly, cumbersome to use and have not been sustainable in field practice.
- One Drop is an ionic, aqueous solution that includes silver, copper, aluminum, zinc and gold as natural mineral ions.





- Manufacturer information recommends 1-4 drops of the solution to treat one liter of water, depending on initial quality.
- This study tested the efficacy of One Drop to inactivate indicator microbes and reduce turbidity in raw water

## OBJECTIVES

- Test the efficacy of 1, 2, and 4 drops of One Drop for reduction of indicator microbes in raw water at 10, 30, 90, and 240 minute contact times.
- Test the efficacy of One Drop to reduce water turbidity.
- Compare the manufacturer dosing method with a standard coagulation/flocculation.

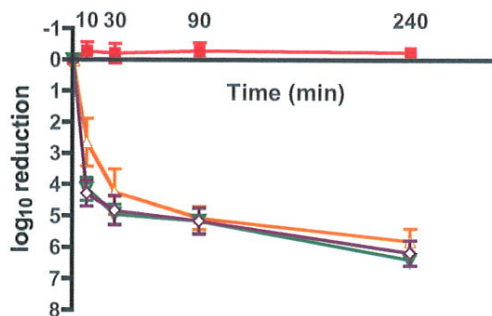
## METHODS

- Add 0, 1, 2, and 4 drops of One Drop to each of 4 one-liter volumes of raw water (23-25oC) spiked with E. coli B, K. terrigena, B. subtilis variety niger spores or coliphage MS-2
- Assay bacteria by membrane filtration at 10, 30, 90, and 240 minutes of contact time.
- Assay MS-2 coliphage by the Single Agar Layer Method (EPA 1601) at 10, 30, 90, and 240 minutes of contact time.
- Also assay for B. subtilis variety niger spores, indigenous heterotrophic plate count (HPC) bacteria, and indigenous bacterial spores.
- Measure turbidity after the manufacturer's suggested treatment method of adding, mixing and then static holding to a standard jar-test coagulation-flocculation-sedimentation.
- Compare manufacturer's treatment to jar-test treatment for the reduction of microorganisms

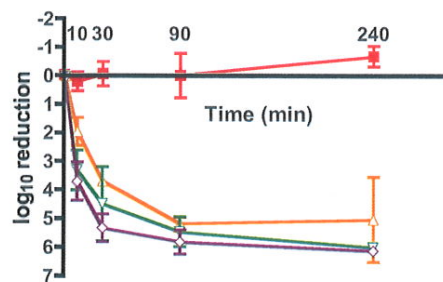
## RESULTS

Figure 1: Arithmetic Mean Log<sub>10</sub> Reductions and their Standard Deviations by One Drop at t = 10, 30, 90, and 240 minutes

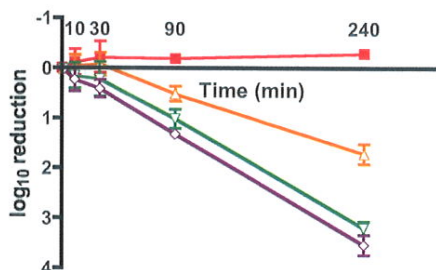
### A. E.coli B



### B. K. Terrigena

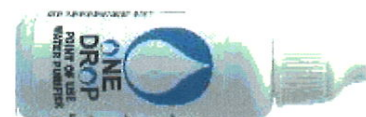


### C. MS-2



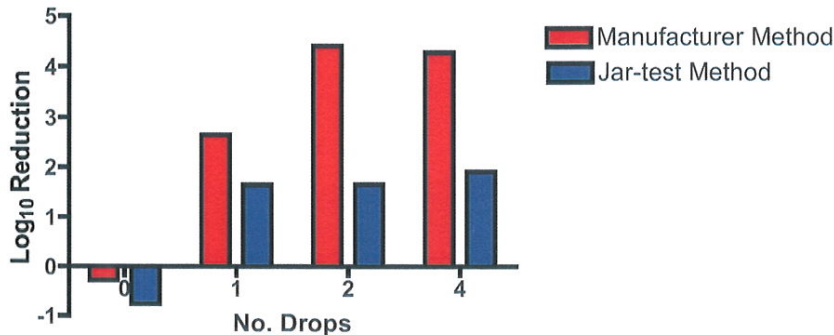
### Dosing-levels

- 0 drops
- ▲ 1 drop
- ▼ 2 drops
- ◇ 4 drops





**Figure 2: E. coli B log<sub>10</sub> reductions using two One-Drop dosing methods**



- Appreciable reductions were achieved for bacterial indicators (4 drops, 240 minutes):
  - E. coli B: >6 log<sub>10</sub> (99.9999%)
  - K. terrigena: >6 log<sub>10</sub> (99.9999%)
  - HPC bacteria: >3 log<sub>10</sub> (99.9%) (data not shown; detection limit of assay)
  - MS-2: >3.5 log<sub>10</sub> (99.97%)
- Reductions for vegetative bacteria and coliphage MS-2 generally increased over time and with increasing number of One Drop drops added to raw water
- Microbe reductions were more rapid extensive in One Drop-treated than in control natural water samples lacking One Drop
- Bacterial spores (indigenous or added Bacillus subtilis variety niger) were not reduced by One Drop treatment (data not shown)
- One Drop did not significantly affect turbidity in raw water samples when tested with either the manufacturer’s application method or the jar-test method (data not shown)
- E. coli B reductions were more extensive using the manufacturer’s method than the jar-test method
  - More replicate experiments are needed to determine if these differences are statistically significantly different (see Figure 2)

### SUMMARY AND CONCLUSION

- One Drop treatment reduced concentrations of indicator microorganisms in raw water
- One Drop treatment achieved the WHO and US EPA bacterial reduction target of 6 log<sub>10</sub> for E. coli B and K. terrigena (4 drops, 240 minutes)
- MS-2 coliphage reduction was >3.5 log<sub>10</sub> for 4 drops of One Drop at 240 minutes contact
- A longer contact time will likely achieve the US EPA and WHO virus reduction target of 4 log<sub>10</sub> for viruses; work is planned to document this level of performance
- One Drop appears to be an effective and low-cost means of disinfecting drinking water at the household level or other point-of-use, at least for bacteria and viruses.

### ACKNOWLEDGEMENTS

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# Efficacy of "One Drop" Point-of-Use Chemical Disinfectant to Inactivate Waterborne Pathogens and Indicator Microorganisms



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## ABSTRACT

In many regions of the world, microbial contamination of drinking water requires point-of-use disinfection in order to make the water safe to drink. Among chemical treatments available, chlorine remains the most widely used to disinfect drinking water. Although chlorine's strong oxidizing properties make it effective at inactivating waterborne microorganisms, it is often rejected by users due to taste and odor problems and toxic disinfection by-products. As an alternative, One Drop is an aqueous solution of natural ionic minerals, including silver, gold, aluminum, zinc and copper believed to have microbiocidal properties when added in small quantities to contaminated water. As little as one drop of the solution per liter of water may inactivate enteric microbes. One Drop may also interact with negatively charged microbes to facilitate settling or removal by filtration. In such small quantities, One Drop is harmless to humans and does not affect taste or odor, providing an advantage over other means of chemical disinfection. One Drop was tested for its ability to inactivate pathogenic microorganisms in raw surface water. Different volumes of One Drop (0, 1, 2, or 4 drops) were added to one-liter volumes of raw surface water spiked with *Salmonella typhimurium* WG-45 and *Vibrio cholerae* as representative bacteria pathogens. Concentrations of each microbe were measured both initially and after 10, 30, 90, and 240 minutes at ambient (room) temperatures of 23-25°C. There were appreciable reductions of both *S. typhimurium* WG-45 and *V. cholerae*, at >7.8 and >4.7 log<sub>10</sub>, respectively. Reductions generally increased over time and with increasing number of drops added. Microbe reductions with One Drop were more rapid and extensive than natural reductions in the same test water without One Drop. MS-2 coliphage reductions of >4.4 log<sub>10</sub> were also achieved. We conclude that One Drop reduces concentrations of important pathogenic bacteria and perhaps viruses in raw water and may serve as an effective and low-cost means of household or other point-of-use water treatment.

## INTRODUCTION

- Cost-effective and simple physical and chemical point-of-use drinking water disinfection technologies have been developed, evaluated and applied in household use in developing countries
- Current options, such as boiling, chlorine, sunlight, filtration and combined chemical coagulant-flocculant-disinfectant, have limitations and deficiencies
- Physical methods do not provide residual disinfectant
- Chlorine may be rejected due to taste and odor and is ineffective against *Cryptosporidium parvum*
- Combined coagulants-flocculant-disinfectant products are more costly, cumbersome to use, and have not been commercially sustainable in field practice
- One Drop is an ionic, aqueous solution that includes silver, copper, aluminum, zinc and gold as natural mineral ions
- Manufacturer information recommends 1-4 drops of the solution to treat one liter of water, depending on initial quality
- This study tested the efficacy of One Drop to inactivate *Salmonella typhimurium* WG-45, *Vibrio cholerae*, and viral indicator MS-2



## OBJECTIVES

- Test the efficacy of 1, 2, and 4 drops of One Drop for reduction of pathogenic microbes in raw surface water at 10, 30, 90, and 240 minute and 24 hour contact times
- Test the efficacy of 1, 2, and 4 drops of One Drop for reduction of viral indicator MS-2 in raw surface water at 10, 30, 90, and 240 minute and 24 hour contact times

## METHODS

- Add 0, 1, 2, and 4 drops (1 drop = 57 µl) of One Drop to each of 4 one-liter volumes of raw surface water spiked with *S. typhimurium* WG-45, *V. cholerae*, or coliphage MS-2 at ambient (23-25°C) temperature.
- Raw water quality: pH = 6.8 - 6.9; turbidity = 4.8 - 5.5 NTU; total organic carbon = 6.2 - 7.0 mg/l.
- Assay bacteria by spot plating (*V. cholerae*) or by membrane filtration (*S. typhimurium* WG-45) at 10, 30, 90, and 240 minutes and 24 hours of contact time
- Assay MS-2 coliphage by the Single Agar Layer Method (EPA 1601) at 10, 30, 90, and 240 minutes and 24 hours of contact time

\*This research was supported by The Water Development Program.

\*We thank Douglas Wait, Lindsey Witmer, and Katherine Pierson for their laboratory assistance.

## RESULTS

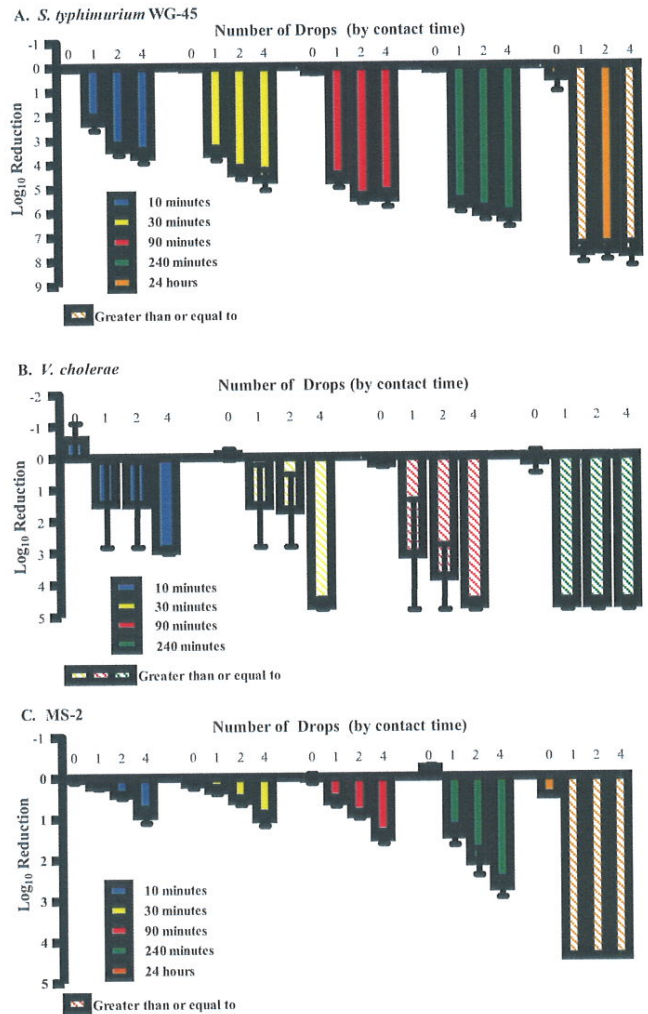


Figure 1: Arithmetic Mean Log<sub>10</sub> Reductions and their Standard Deviations by One Drop at t = 10, 30, 90, and 240 minutes and 24 hours

- Appreciable reductions were achieved for pathogenic bacteria and coliphage MS-2:
  - *S. typhimurium* WG-45: >6.1 log<sub>10</sub> (99.99992%) at 2 drops, 240 minutes
  - *V. cholerae*: >4.7 log<sub>10</sub> (99.998%) at 4 drops, 30 minutes
  - MS-2: >4.4 log<sub>10</sub> (99.996%) at 1 drop, 24 hours
- Reductions for pathogenic bacteria and coliphage MS-2 generally increased over time and with increasing number of One Drop drops added to raw surface water
- Microbe reductions were more rapid extensive in One Drop-treated than in control natural water samples lacking One Drop

## SUMMARY & CONCLUSIONS

- One Drop treatment reduced concentrations of pathogenic and indicator microorganisms in raw surface water
- One Drop treatment achieved the US EPA bacterial reduction target of 6 log<sub>10</sub> for *S. typhimurium* WG-45 (2 drops, 240 minutes)
- One Drop treatment achieved >4.7 log<sub>10</sub> reduction for *V. cholerae* (4 drops, 30 minutes) which may meet the US EPA bacterial target; this cannot definitively be concluded due to lower than desired bacterial concentrations in initial test water
- One Drop treatment achieved the US EPA viral reduction target of 4 log<sub>10</sub> for MS-2 coliphage reduction (1 drop, 24 hours)
- One Drop appears to be an effective and low-cost means of disinfecting drinking water at the household level or other point-of-use, at least for bacteria and viruses