Microbes in deionized water: Implications for maintenance of laboratory water production system

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Conflict of Interest

The authors declare no conflict of interest.

Author's contributions

Wenfa Ng conceived the idea, designed and performed the experiments, analyzed the data, and wrote the abstract. Yen-Peng Ting mentored W. Ng in the research, analyzed the data, and wrote the abstract.

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Abstract

Microbes, with their diverse metabolic capabilities and great adaptability, occupy almost every conceivable ecological niche on Earth – thus, could they survive in the oligotrophic (i.e., nutrient-poor) deionized (DI) water that we use for our experiments? Observations of white cauliflower-like lumps and black specks in salt solutions after months of storage in plastic bottles prompted the inquisition concerning the origin and nature of the "contaminants." Hypothesising that the "contaminants" may be microbes from DI water, a series of growth experiments was conducted to detect and profile the microbial diversity in fresh DI water - produced on a just-in-time basis by a filter-cum-ion-exchange system with tap water as feed. While microbes could also be present on the surfaces and headspace of the unsterilized polyethylene bottles, investigating whether microbes are present in freshly produced DI water provides a more stringent performance test of the production system. Inoculation of DI water on R2A agar followed by multi-day aerobic cultivation revealed the presence of a wide variety of microbes (total viable cell concentration of ~10^3 colony forming units (CFU) per mL) with differing pigmentations, growth rates as well as colony sizes and morphologies. Additionally, greater abundance and diversity of microbes was recovered at 30 ^{o}C relative to 25 and 37 ^{o}C; most probably due to adaptation of microbes to tropical ambient water temperatures of 25 to 30 ^{o}C. Comparative experiments with tap water as inoculum recovered a significantly smaller number and diversity of microbes; thereby, suggesting that monochloramine residual disinfectant in tap water was effective in inhibiting cell viability. In contrast, possible removal of monochloramine by adsorption onto ion-exchange resins – and thus, alleviation of a source of environmental stress - might explain the observed greater diversity and abundance of viable microbes in DI water. Collectively, this study confirmed the presence of microbes in fresh DI water – and suggested a possible source for the "contaminants" in prepared salt solutions. Propensity of microbes for forming biofilm on various surfaces suggested that intermittent flow in just-in-time DI water production provided opportunities for cell attachment and biofilm formation in the system during water stagnation, and subsequent dislodgement and resuspension of cells upon water flow. Thus, regular maintenance and cleaning of the production system should help reduce DI water's microbial load. Additionally, simple and low-cost culture experiments on agar medium can provide a qualitative and semi-quantitative estimate of microbial diversity and viable cell concentration in DI water, respectively; which, along with regular monitoring of water resistivity or conductivity, comprise a trio of tests useful for detecting possible contamination or, deterioration of DI water's chemical and microbiological quality.

***Keywords*:** viable-but-not-cultivable; biofilm; microbial ecology; tap water; deionized water; chlorine residual; nutrient-poor; water quality; disinfection; bacteria

Subject areas: Microbiology; Environmental Sciences; Ecology; Biodiversity;