

Threat Assessment of Free-Living Amoeba in Drinking Water

By Kelly A. Reynolds, MSPH, PhD

In November 2013, the Department of Health and Hospitals issued a state-wide emergency rule in Louisiana with new disinfectant mandates for the control of *Naegleria fowleri* in drinking water. *Naegleria* in domestic water supplies are easily killed by chlorination; however, when residual disinfectant levels decrease, the organisms can proliferate. *Naegleria*, however, are not the only amoeba of concern and thus, new research is focused on assessment of the threat of other amoeba commonly isolated from drinking water supplies.

Louisiana's mandate

Following the death of a four-year-old boy who was playing on a slip 'n slide wetted with municipal tap water that was later found to be contaminated with the free-living amoeba (see On Tap, October 2013) and two more deaths following neti-pot use from Louisiana tap water supplies (including a 51-year-old woman and a 28-year-old man), the state of Louisiana decided to implement a new rule for drinking water treatment. Specifically, the mandates require higher disinfectant levels of 0.5 mg/L of chloramine/chlorine residual and for utilities to increase their water quality monitoring by 25 percent. These levels are higher than current US federal standards.

Today, 1,296 of Louisiana's 1,369 municipal water systems (nearly 95 percent) are in compliance with the rule; however, 73 systems were issued notices of violation earlier this year.¹ While noncompliance does not necessarily mean a contaminated water supply, conditions of increased temperature, biofilm formation and dead zones in the distribution system are conducive to the growth and survival of free-living amoeba (FLA), even in the presence of detectable chlorine. A residual of 0.5 mg/L, therefore, is the minimum concentration for control, with a preferred level of 0.75 mg/L or higher to reduce risks. Periodic system flushing is also essential to purge dead-end zones and reduce free-living amoeba in the system. Public water utilities do not routinely test for FLA in their systems. Monitoring POU supplies, however, particularly during warm summer months and in regions of high temperatures, is recommended as a best-practice approach.

Free-living amoeba in the environment

Free-living amoebae are naturally present in soil and water environments. There are more than 11,300 species of amoeba currently identified, with only a few known to be hazardous to humans.² Amoebic infections are still rare and typically due to forced water exposures into the nasal cavity. Thus, most infections are from recreational water exposures but in the last decade, the fatal cases in Louisiana and Arizona from drinking water sources have caused concern. The route of infection from tap water remains via the nasal passage, where exposures have

occurred following bath, neti-pot and other environmental routes, but not via ingestion.

Water commonly contains FLA, including genera of *Acanthamoeba*, *Harmannella*, *Naegleria* and *Vahlkampfia*.³ Species known to cause illnesses in humans include *Acanthamoeba*, *Naegleria fowleri* and *Balamuthia*, which cause swelling of the brain with a high rate of fatalities (up to 99 percent). *Acanthamoeba* is also associated with severe eye infections that can lead to blindness. FLA are commonly isolated from rivers, groundwater and other water reservoirs at rates of 100, 82 and 100 percent, respectively.² Concentrations of FLA per liter have ranged as high as 90,000 in rivers and 3,000 in groundwater.⁴ Feeding on bacteria and algae, they are found in higher numbers close to biofilm environments and correspond to increased algal concentrations in water reservoirs.

Hundreds of FLA have been isolated from drinking water distribution systems and have been shown to increase in concentration at increasing distances from the treatment plant.² Growth in household plumbing has also been identified, suggesting the need for treatment at the point of use as a best management approach.

Some species of FLA are highly susceptible to chlorination while others are more resistant. *Acanthamoeba*, for example, was found to survive a two-hour exposure to 100 mg/L of chlorine and thus, would not be controlled by current drinking water disinfectant treatments alone. This organism is also resistant to UV light and thermal treatment strategies. A combination of treatments, which include the use of filtration for physical removal of the amoeba, are expected to be the most broadly effective. Controlling the proliferation of FLA is an important strategy to minimize health effects, which is best accomplished by reducing organic matter and biofilms in the treated water distribution and storage system.

FLA as bacterial hosts

Other than health risks associated from direct exposure and infection, waterborne amoeba can serve as protective vessels for pathogenic bacteria, such as *Legionella*, that hide inside amoeba hosts to effectively avoid inactivation from disinfectants or other water treatment works. A significant association has been established between FLA and the presence of pathogenic *Legionella* and *Mycobacterium* species.² *Legionella* are known to rapidly recolonize domestic water systems, sometimes immediately after halting the use of increased disinfectants, which may be due to the constant supply of the bacteria from FLA hosts already established in the system. FLA also harbor viruses and protect them from disinfectants, which has been shown to occur with giant mimiviruses (see On Tap, February 2014). The human

health implications of these organisms are currently unknown; however, mimiviruses have been associated with the incidence of rheumatoid arthritis.

One study in France focused on genomic characterization of the FLA population in a drinking water network.³ Monitoring of three sites over a period of four months revealed distinct diversity of amoeba and particularly, amoeba serving as bacterial hosts. Previously, 102 bacteria have been identified as being able to survive and grow in FLA, with 27 being suspected pathogens.⁵ The France study identified 54 types of bacteria within amoebic hosts, 21 of which were not previously recognized as intracellular organisms. *Hartmannella* was identified as the most dominant genera, although *Naegleria* were found in a few drinking water samples (4.4 percent of more than 15,000 sequences generated) and *Acanthamoeba* at a rate of 14.4 percent in one site. FLA and FLA-associated bacteria were highly diverse across sites.

The reason for such variability is unknown but may provide clues in future studies to help predict FLA risks and identify more effective management approaches. Total FLA counts may offer a tool for water quality monitoring even in the absence of the deadly strains.

Conclusion

While *Naegleria fowleri* has been clearly identified as a known human health hazard, the impact of other free-living amoeba persisting in water supplies is of interest. FLA not only serve as protective reservoirs for bacterial and viral pathogens, allowing for their survival, but also aid in the multiplication of the organisms by providing additional food sources in the protective environment. The organisms may also increase in virulence during their intracellular camp out.

Current federal standards for drinking water treatment do not necessarily control FLA and many public utilities are

examining practices for increased control. Given the variability of FLA across different drinking water systems and the potential for colonization on water components throughout distribution, the final barrier for control is at the consumer's tap. POU filters rated for cyst removal are expected to be effective for removal of most FLA and should be applied for risk reduction.

References

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About the author

◆ Dr. Kelly A. Reynolds is an Associate Professor at the University of Arizona College of Public Health. She holds a Master of Science Degree in public health (MSPH) from the University of South Florida and a doctorate in microbiology from the University of Arizona. Reynolds is WC&P's Public Health Editor and a former member of the Technical Review Committee. She can be reached via email at reynolds@u.arizona.edu



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