
Funded by Arizona Water Institute

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Rose Valley Water System, Peoria, AZ, ca. 2002

- RVW Tank #1
- RVW Tank #2
- RVW Tank #3
- RVW Well #2
- Small tank not in use since June 2002

POE 002
3. Inside Tank No. 1
Rose Valley Water System, Peoria, AZ, Tank No. 3; ca. 2002

- Ladder
- Tank Wall #3
- Float for water level gage
- Floor on Tank #3
Project Hypotheses

• Biofilms in drinking water systems provide a substrate for growth and survival of *Naegleria fowleri*

• Factors that reduce the accumulation, survival and spread of *N. fowleri* include:
  – Chemical and biological factors
  – Plumbing construction materials
  – Operational practices
Research approaches

- Test for presence of *N. fowleri* in samples from drinking water systems before and after an operational practice
- Test for survival and growth of *N. fowleri* on lab scale pipe loop biofilm accumulation
- Evaluation of selected chemical and biological parameters as indicators of *N. fowleri*
Water System Selection

• City of Peoria volunteered to serve as the chlorinated groundwater source (pop. 145,000)

• U of A Maricopa Agricultural Center (MAC) volunteered to serve as the unchlorinated groundwater source (pop. 100)
MAC Valve Section Sample Location
MAC Valve Section Biofilm Sample
MAC Hydrant Flush Operational Practice (OP)
City of Peoria Water Reservoir
Biofilm Sample from Water Reservoir, Before Operational Practice
Biofilm/Sludge from Water Reservoir Bottom, Before OP
A total of 28 (8 40-L water and 20 biofilm) samples were collected and processed.

- All samples were analyzed for amoebic activities using a viability assay.
- Amoebic activity was observed in two of 8 water samples and 14 of 20 biofilm samples.
- 31% of the sampling locations that originally had amoebic activity showed no such activity after the best operational practices such as flushing.
- For confirmation, DNA has been extracted from the positive samples and stored at -80 C for a molecular analysis. Preliminary results are inconclusive.
N. fowleri spiked: 3.5x10^7 TCID_{50}^1

Estimated number of N. fowleri in flowing water: 10^3 TCID_{50}/mL

Weekly water sampling from both pipes until no amoebic activity is observed (natural die-off, >4.5 log_{10})

Biofilms from both pipes were assayed for amoebic activity at the end of simulator run

1 Tissue Culture Infective Dose per ml

Slide provided by Morteza Abbaszadegan, Ph.D., Director, NSF Water Quality Center
5-month old biofilm samples from newly installed pipe segments were collected and analyzed for amoebic activity.

Slide provided by Morteza Abbaszadegan, Ph.D., Director, NSF Water Quality Center
Slide provided by Morteza Abbaszadegan, Ph.D., Director, NSF Water Quality Center
Trophozoites

Cyst

Amoeba

Slide by Hodon Ryu, Ph.D.
Amoebic activity was observed in both water and biofilm samples after 5-month simulator run.

*N. fowleri* was observed in biofilm after 5 months

This supports the hypothesis that biofilm can sustain *N. fowleri*
Chemical and Biological Parameters

- Temperature
- pH
- Dissolved oxygen
- Total chlorine
- Free chlorine
- Dissolved organic carbon (DOC)
- Biodegradable dissolved organic carbon (BDOC)
- Total nitrogen
- Total ammonia
- Ammonium
- Nitrite
- Nitrite
- Heterotrophic plate count
Chemical and Biological Field Sample Results

- **DOC**
  - Lower in unchlorinated system
  - Lower with more residence time/stagnation
  - Operational practices had insignificant effect
- **BDOC**
  - Lower in unchlorinated system
- **HPC**
  - Higher in unchlorinated system
  - Higher with residence time/stagnation
  - Lower after flushing
- **Cl residual**
  - Lower with residence time/stagnation
- **NO\textsubscript{3}**
  - Lower in unchlorinated system
  - Lower after flushing in both systems
## Chemical and Biological Pipe Loop Results

<table>
<thead>
<tr>
<th>PVC</th>
<th>VS.</th>
<th>Cast Iron</th>
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<tbody>
<tr>
<td>• DOC lower over time</td>
<td>• DOC lower over time</td>
<td>• DOC lower over time</td>
</tr>
<tr>
<td>• BDOC lower over time</td>
<td>• BDOC lower over time</td>
<td>• BDOC lower over time</td>
</tr>
<tr>
<td>• HPC higher</td>
<td>• HPC lower</td>
<td>• HPC lower</td>
</tr>
<tr>
<td>• NO$_3$ lower</td>
<td>• NO$_3$ higher</td>
<td>• NO$_3$ higher</td>
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<tr>
<td>• Biofilm thickness less/even</td>
<td>• Biofilm thickness greater/uneven</td>
<td>• Biofilm thickness greater/uneven</td>
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<tr>
<td>• Biofilm bacteria higher/unit area</td>
<td>• Biofilm bacteria lower/unit area</td>
<td>• Biofilm bacteria lower/unit area</td>
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Significant Findings

- *N. fowleri* colonized biofilms in the laboratory setting
- Amoebic activity was present in 70% of water distribution biofilm samples
- Amoebic activity was present in 25% of water distribution water samples
- Amoebic activity increases with:
  - Lack of chlorine
  - Stagnation/lack of flushing
  - Low DOC
  - Low BDOC
  - Higher HPC
Recommendations

• Work towards elimination of the main causes of bacteriological growth in finished water. The primary sources of bacteria/biofilm growth are BDOC and ammonia.

• Additional studies on amoebic activity are needed through samples of several different water systems.
QUESTIONS AND DISCUSSION

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