Impact of Antibiotics on Denitrifying Biofilm Bacteria
Jeremiah Rocha, Julie Swierenga, Professor David Wunder, Calvin College, Summer 2014

Introduction
Nitrate contamination of drinking water supplies, partially attributable to agricultural runoff, is a prevalent problem. Nitrate presents public health concerns, especially for pregnant mother and infants. Low µg/L concentrations of antibiotics have also been found in surface waters worldwide. This research focuses on how low concentrations of antibiotics affect the ratio of biological denitrification in biofilm bacteria used to treat drinking water supplies.

Objective
To understand the impact of low concentrations of antibiotics on denitrifying bacterial biofilm used in water treatment processes by assessing the presence of relevant enzymes, denitrification rates, and bacterial viability.

Methods
Experimental Set-Up
- Continuous-feed Rotating Annular Bioreactors (CFRABs) were run for 7 days to simulate biofiltration of drinking water.
- Mineral medium, acetate, nitrate, and antibiotics were fed to the CFRAB which was seeded with biofilm.

Viability Analysis
- Effluent water was collected daily
- Samples of effluent were collected before and after each nitrate sweep
- All samples were analyzed for nitrate via Ion Chromatography

Denitrification Analysis
- Ion Chromatography
- Effluent water was collected daily
- Samples of effluent were collected before and after each nitrate sweep
- All samples were analyzed for nitrate via Ion Chromatography

DNA Analysis
- Biomark Fluidigm Real-time PCR
- DNA was extracted from the biofilm using the MoBio Power Biofilm DNA Extraction Kit
- Sample DNA was analyzed using the Biomark Fluidigm Real-time PCR for different gene markers present in denitrifying bacteria: 16S, nosZ, nirS, nirK, norB, nrfA, napA, and narG

Conclusions
- Low concentrations of antibiotics (3.44 µg/L and 34.4 µg/L) decreased the utilization of nitrate in a denitrifying biofilm by approximately 50% when present in water.
- Biomass in the reactor showed an increase with the introduction of low and high µg/L concentrations of antibiotics.
- Some denitrification genes were present in the biofilm DNA and were affected by antibiotic concentration.

Acknowledgments
Jansma Family Research Fund
Mr. Bob De Kraker, Calvin College
Mr. Rich Huisman, Calvin College
Mr. Phil Jasperse, Calvin College
Ms. Lori Keen, Calvin College
Mr. Randall Delong, Calvin College
Mr. Scott Prentice, Calvin College

References