Assessing filtration efficacy for *Cyclospora* control (AFECCT)

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Summary

Reducing contamination of irrigation water with foodborne pathogens, such as the parasite *Cyclospora cayetanensis*, should mitigate risk. A promising means to safeguard produce entails filtering irrigation water. To accelerate progress, we are studying filtration efficacy using surrogate parasites (of poultry) that closely resemble *Cyclospora* but which are far more abundant and safer to study. By loading hundreds of thousands of such *Eimeria* parasites (with defined bacterial loads resembling common field conditions), we are finding that filters succeed in preventing passage of almost every parasite. In the coming months, we will evaluate viability of parasites recovered from the filtrate, filter performance under actual field conditions, and filter performance when challenged by necessarily limited supplies of *Cyclospora* itself.

Objectives

1. Determine filtration *efficacy* for *Eimeria*.
2. Determine how well *Eimeria* models *Cyclospora* undergoing filtration.
3. Evaluate filter performance in field deployments.

Methods

Figure 1 depicts the project's experimental design. We filtered deionized water (4 L) inoculated with *Eimeria tenella* (5×10^5 oocysts) and rifampicin-resistant *E. coli* TVS 353 (1×10^5 CFU/mL) at 0.5 L/min through a sand pre-filter (100% sand) and then a filter (50% zero-valent iron, 50% sand by volume) with 2-L pore volume (liquid capacity). An additional 6 L of water transited the filters before backflushing with 4 L of water. Using continuous flow centrifugation, *E. tenella* oocysts were concentrated from effluents and from the backflush, and then quantified using microscopy. *E. coli* were quantified using direct plate counts. *E. coli* and *E. tenella* abundance in influent, effluent, and backflush were compared using one-way ANOVA ($p < 0.05$) over four replicate experiments.

Results to Date

Early experiments indicate that zero-valent iron (ZVI) filtration reduces *E. tenella* in effluents by at least 98% (**Figure 2A** and **Figure 3A**). All but 17% were recovered in the backflush, indicating that most did not penetrate, or permanently bind, the sand pre-filter. Additional studies will determine if filters containing mixtures of ZVI and sand bind parasites more tightly.

E. coli levels were significantly ($p < 0.05$) reduced by over 97% in both the effluent and backflush (**Figure 2B** and **Figure 3B**).

Benefits to the Industry

Growers and grocers will benefit if these laboratory results prove applicable and sustainable in field settings. If true, physical filtration mitigates foodborne risks posed by both bacterial and parasitic organisms. Industry will benefit from future studies identifying the means to reduce, or render harmless, parasites released in the backflush. Industry also will benefit from accelerated progress in other CPS-funded projects (PIs Scott Lenaghan and Lia Stanciu) on improving parasite detection and mitigation, using surrogate *Eimeria* parasites supplied by our team.

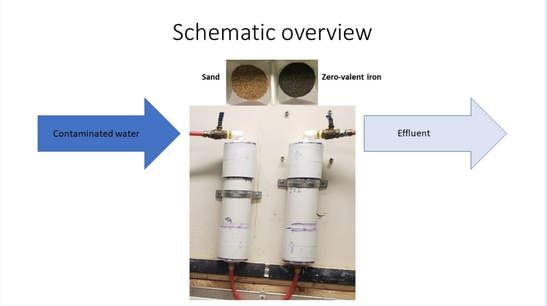

Figure 1. Schematic overview of experiments.
Water containing half a million oocysts of *Eimeria tenella* and 150,000 colony forming units of *E. coli* TVS 353 was filtered through a pre-filter of sand and then a filter containing 50% sand/ZVI. Bacteria and parasites were then enumerated in the effluent, and in the backflush.

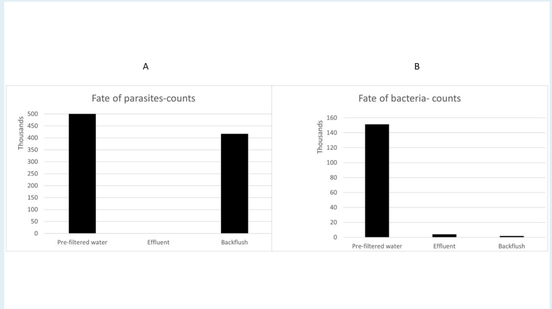

Figure 2. Substantial impact of filtration on parasite and bacterial abundance.
Filtration removed almost all parasites (A) and bacteria (B) from the effluent. Fewer than 300 (of 500,000) parasite oocysts transited the filters. More than 80% of inoculated parasites were recovered from the backflush (A), indicating that they were physically obstructed from penetrating the sand pre-filter. A smaller proportion of inoculated bacteria were recovered from the backflush, suggesting they were more tightly bound, or killed, by contact with the filters.

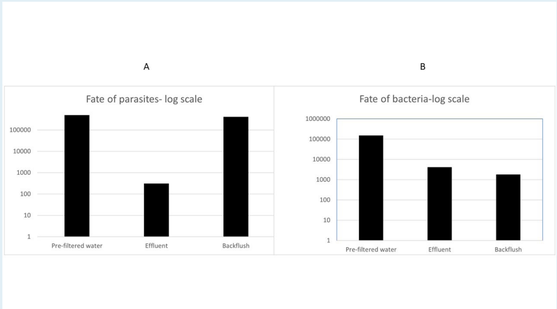

Figure 3. Substantial impact of filtration on parasite and bacterial abundance.
The same results as Figure 2, depicted on a log scale.