Slow sand filters: a biological treatment method to remove plant pathogens from nursery runoff^ $^{\rm \odot}$

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Abstract

Slow sand filters (SSF) are an effective technology, capable of developing highquality water from untreated sources including irrigation runoff. The sand serves as a substrate on which a microorganism community grows. This microbial community can breakdown a wide range of pollutants including plant pathogens. This report reviews results on the removal of *Phytophthora* spp., *Fusarium oxysporum*, and tobacco mosaic virus. We were interested in the capacity of these filters to remove different kinds of plant pathogens from captured irrigation run off. Our experiments removed *P. capsici* after the microbial community was established (2 weeks) and after a simulated 7-day pump failure in previously established SSFs. However, SSFs did not remove *F. oxysporum* after 7 weeks. In our tests, the SSFs were also able to remove tobacco mosaic virus from inoculated runoff water after 6 to 9 weeks of exposure.

INTRODUCTION

Captured runoff may contain plant pathogens and it is necessary to remove them prior to reuse for irrigation to prevent disease spread. Treatment can include chemical compounds, such as chlorine, radiation (from UV light), thermal (using heat from steam or other sources) and biological treatment methods such as slow sand filters (SSF). These filters have been use for a very long time, originally to produce drinking water and in the last decade or two are being used in horticultural production at an increasing rate.

Slow sand filters are a biological treatment method that is simple to set up, requiring little or no chemical or energy inputs. As its name implies, flow rates are slow. For each square foot of sand bed surface, 0.06 to 0.2 gal of water can be treated per min. So a round tank that is 12 ft in diameter can treat about 10,000 gal per day. Any container that holds sand and water can be used: steel water tanks, septic tanks, or earthen lined reservoirs. At the bottom of the container is a manifold of pipes to collect the treated water (Figure 1). The manifold is buried in coarse gravel to facilitate collection of the treated water. Above the gravel are several layers of sand, graduating from coarse at the bottom to fine at the top, to prevent the filtration sand from entering the gravel layer. Finally, at the top, is the bed of sand. A pump may be necessary to move the treated water into or out of the sand filter as it may not be possible to rely on gravity for both inflow and outflow. While filtration takes place particulates may clog the filter; this needs to be prevented with sedimentation ponds and other pre-filtering treatments.

The sand serves as a substrate on which a community of microorganisms grows and water should flow continuously through the sand bed for optimizing treatment volume. Key characteristics of slow sand filters include:

- Round grains, uniform size of about 0.3-0.6 mm. Uniformity is important to maintain water flow through the sand. Sharp sand can pack and restrict flow, so round grains are necessary
- One meter deep sand bed. Maintenance can require removal of a few centimeters of the top layer of sand. When 0.5 m of sand is left after many "cleanings", the sand bed should be rebuilt.
- One meter deep water head above the sand. These filters are gravity driven, so the water head is needed to push the water through the bed.

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- Flow control. To obtain most efficient treatment, flow needs to be controlled to balance water quality and flow. Slow rates improve treatment, but reduce the volumes of water treated.
- Recommend two filters. While one filter is being serviced the other can remain in operation.

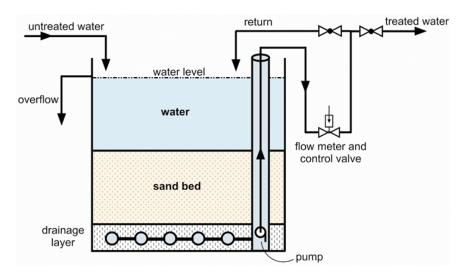


Figure 1. The slow sand filter (SSF) consists of a drainage layer that includes a pipe manifold assembly buried in pea gravel to collect treated water. Several layers of sand of gradually decreasing size cover the pea gravel so the SSF sand doesn't become incorporated in the gravel. The manifold is connected to a pump that moves the filtered water to storage. If topography enables it, the system may be entirely or partially gravity driven. Flow control is key for effective treatment.

To study how SSFs work in removing plant pathogens, three sets of experiments were conducted: (1) treatment performance, (2) pathogen switch, and (3) virus removal. The first set of experiments was used to determine the time required for treatment to occur. Although it is a biological treatment method, it is not necessary to inoculate the filters. Because the sand in a new filter is essentially sterile it takes time for the microorganism community to develop in response to the pollutants present. In the second set, it was not known if a sand filter established exposed to one pathogen can remove another type of pathogen if it suddenly appears. It was also not known if treatment would be compromised when water supply to a SSF system was shut down inadvertently, mimicking a pump failure. In the third experiment, since there was little information on the ability of SSFs to remove plant pathogenic viruses, work was done to assess this.

MATERIALS AND METHODS

Treatment performance

Slow sand filters were constructed using 4-in. PVC pipe that included sampling valves located just above the sand bed (unfiltered), at 20 cm intervals down the depth of the bed and below the sand bed (filtered) (Figure 2). Flow rates were set at 150, 250, and 500 L m⁻² h⁻¹. The recommended flow rate of 150 L m⁻² h⁻¹ corresponded to a flow of 20 mL min⁻¹ retrieved from the column. Irrigation runoff was generated by irrigating plants on a tray in a greenhouse (Harris and Oki, 2009). Captured runoff water was inoculated with *Phytophthora capsici* zoospores and then provided to the SSF columns. Water samples were collected every 5 days beginning on the day that water was introduced to the SSFs. The water samples were then analyzed for the presence of *P. capsici* colony forming units (CFUs) using culture media that selects for *Phytophthora* and *Pythium*.

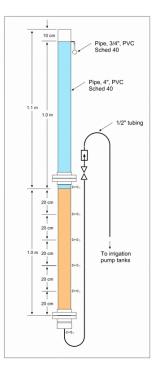


Figure 2. Slow sand filters for experimentation were constructed of 4-in. PVC pipe and included valves that enabled collecting samples above the sand bed, at 20-cm intervals down the sand bed, and below the bed.

Pathogen switch

Two sets of columns were set up, one set exposed to *P. capsici* inoculum in runoff water and the other exposed to *Fusarium oxysporum* f. sp. *lycopersici* for 6 weeks (Lee and Oki, 2013). Then, the pathogen inoculum for each treatment was switched (Figure 3). To simulate a system failure after 12 weeks, the pumps supplying water to the columns were turned off for 7 days, then restarted, and allowed to run for 6 more weeks. The entire experiment lasted a total of 19 weeks. Flow rates were set at 20 mL min⁻¹. Water samples were collected from above (unfiltered) and below (filtered) the sand bed and analyzed on culture plates to determine pathogen concentration and were also tested by bioassay on tomato and pepper plants to test for *F. oxysporum* and *P. capsici*, respectively.

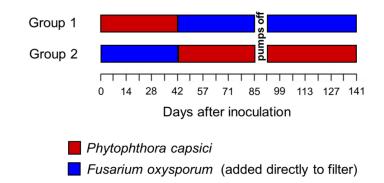


Figure 3. Two sets of slow sand filters (SSF) were set up. One set was initially exposed to *Phytophthora capsici* for 6 weeks then *Fusarium oxysporum*. The other SSF set was the opposite: initially exposed to *Fusarium* then *Phytophthora*. In addition, after 12 weeks the pumps were shut off for 7 days to simulate a pump failure.

Virus removal

Sand filters assembled in the same manner previously described were installed in the greenhouse at the UC South Coast Research and Extension Center in Irvine, California (Mathews et al. in preparation). A suspension of tobacco mosaic virus was introduced and mixed into the water above the sand beds. Samples of water were collected weekly from above and below the sand bed for 12 weeks and were analyzed by ELISA and bioassays. Bioassays used leaves of *Nicotiana glutinosa* and *Chenopodium quinoa* and whole plants of *N. tabacum* 'Turkish' and *N. benthamiana*. A pilot study utilized a single filter and a subsequent study involved three filters. The leaf assay for TMV was not conducted in the second study.

RESULTS

In all of the flow rate treatments in the first set of experiments, pathogen concentration declined steadily over time. Complete removal was apparent after 15 to 21 d and continued through the 30-d duration of the experiment (Figure 4).

In the pathogen switch study, during the first 6-week test period of the *P. capsici* was not detected in the samples collected at Week 2 and thereafter, but *F. oxysporum* was always recovered from the SSFs. When the pathogens were switched, those filters initially exposed to *F. oxysporum* were able to immediately remove *P. capsici* but *Fusarium* was always recovered from the SSFs. After the pumps were shut off for 7 d and then restarted, *P. capsici* was not detected, but *F. oxysporum* was recovered from treated water.

Although this is mainly an aerobic biological, interrupting water flow for 7 d did not diminish the ability of the filters to remove *P. capsici* when water flow was resumed. So it appears that these systems are resilient when experiencing flow interruptions caused by pump failures, for example, as long as the biological layer does not dry out.

In the virus removal pilot study that utilized a single filter, TMV was not detected in the samples collected at Week 9 and later. In the subsequent experiment using three filters, the virus was not detected in samples collected from Week 6 and later.

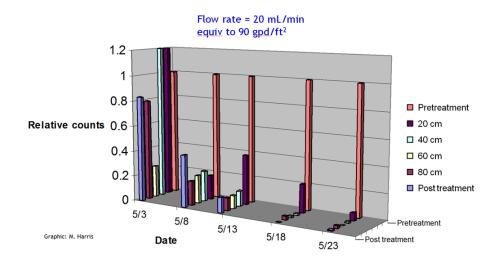


Figure 4. Recovery of pathogens measured as CFUs. Counts are relative to the pretreatment amounts. Removal occurred after 15 d in this experiment conducted in May. In the cooler fall season, treatment took 21 d to appear.

DISCUSSION

All of the treatment flows resulted in removal of *P. capsici* after about 14-21 d. But it was difficult to maintain the desired flow with SSFs running at the highest flow rate. Although pathogen removal is possible at greater than recommended flow rates, more frequent maintenance is necessary. Further study is needed to determine if the maintenance frequency is a factor of either the volume of water treated or the time interval between maintenance events.

The pathogen switch study showed us how these filters perform in the removal of a pathogen when the microorganism community is allowed to develope when exposed to another. The selection of the pathogens was based on their cellular composition. Specifically, the cell walls of *Phytophthora* are composed of β 1,3 glucans, whereas those of *Fusarium* are of chitin, so it was posited that the microorganism communities that mitigated each of the pathogens would be distinct. Although the filters were not able to remove *E oxysporum*, they were immediately able to remove *P. capsici* when it was introduced, but the opposite was not the case. This may suggest that organisms that can mitigate *P. capsici* are also present in the treatment of *F. oxysporum*, but organisms that can remove *P. capsici* may not be involved in *F. oxysporum* removal. Since *F. oxysporum* wasn't removed in this study, this is only speculation.

The experiments inoculating SSF with TMV are the first demonstrations of the removal of this virus using slow sand filters. Since TMV is so robust, there is a very high probability that most other plant pathogenic viruses can also be removed from captured irrigation runoff.

CONCLUSIONS

Slow sand filters are effective in removing a wide range of plant pathogens from water molds to viruses. As with other reports, we weren't able to remove *Fusarium* from the water. However, there are other reports indicating that removal can be attained after a long exposure or with pretreatment using chitin. Although these systems have high initial costs and can consume a large area, there are no other chemical or energy inputs required other than lifting the water into the filter and periodic cleaning.

ACKNOWLEDGEMENTS

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