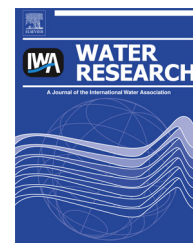




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Bacteria and virus removal effectiveness of ceramic pot filters with different silver applications in a long term experiment

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ABSTRACT

In 2012 more than 4 million people used a ceramic pot filter (CPF) as household water treatment system for their daily drinking water needs. In the normal production protocol most low cost filters are impregnated with a silver solution to enhance the microbial removal efficiency. The aim of this study was to determine the role of silver during the filtration and subsequent storage. Twenty-two CPFs with three different silver applications (non, only outside and both sides) were compared in a long-term loading experiment with *Escherichia coli* (K12 and WR1) and MS2 bacteriophages in natural challenge water under highly controlled laboratory circumstances. No significant difference in Log Removal Values were found between the filters with different silver applications. The results show that the storage time in the receptacle is the dominant parameter to reach *E. coli* inactivation by silver, and not the contact time during the filtration phase. The hypothesis that the absence of silver would enhance the virus removal, due to biofilm formation on the ceramic filter element, could not be confirmed. The removal effectiveness for viruses is still of major concern for the CPF. This study suggests that the ceramic pot filter characteristics, such as burnt material content, do not determine *E. coli* removal efficacies, but rather the contact time with silver during storage is the dominant parameter to reach *E. coli* inactivation.

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1. Introduction

1.1. Ceramic pot filters

Ceramic Pot Filters (CPF) are widespread around the world as point-of-use water treatment systems. In 2009 there were 35 ceramic pot filters factories in 18 countries worldwide with a monthly production of 20,175 filters (Rayner et al., 2013). It is estimated that in 2012 more than 4 million people use their more than 700,000 ceramic pot filters (Fig. 1) as a household water treatment system for their daily drinking water needs.

The ceramic filter systems are the most effective household water treatment system to reduce illness compared to other systems such as Biosand, Solar Disinfection (SODIS) and chlorination, especially on the long term, as stated in a meta-study by Hunter (2009). An overview of the bacterial and viral testing of low-cost Ceramic Pot Filters (CPF), as presented by Simonis and Basson (2011), gives an average Log Reduction Value (LRV) of 2.0 (99% reduction) for *Escherichia coli* over the listed 15 laboratory and field studies. This complies with the performance target indicated as 'Protective' as set by the WHO (WHO, 2011) for bacteria. The removal effectiveness of the CPF is good and is one of the reasons why these filters are so widespread around the globe.

Yet, the variability in the documented performance of the filters with regards to *E. coli* removal is large as Simonis and Basson (2011) report a LRV range between 0.9 and 6.8. Although efforts have been made to come to standardized procedures by the 'Ceramics Filter Manufacturing Working Group' (Lantagne et al., 2009; Rayner et al., 2013), a recent overview of the current practices shows 'that manufacturing processes vary widely both between and within factories, including the consistency of materials, manufacturing methods, and quality control practices' (Rayner et al., 2013).

Virus removal by CPF has been tested with Bacteriophages such as MS2. Van Halem found LRVs for MS2 of 0.6–0.9 after 5 weeks, which increased to 1.1 to 1.8 after 13 weeks (Van Halem et al., 2007) with silver impregnated filters. Tests with deionised water showed a low LRV by CPF of 0.21 and 0.45 for MS2 Bacteriophages (Salsali et al., 2011). Others (Brown and Sobsey, 2010) using rain water and drinking water found a LRV of 1.2

for MS2 on the long term (after filtering 100 L). In a study using virus sized microspheres of 0.02 and 0.1 μm the LRV was highly variable ranging between 0.43 and 2.4 (63–99.6%) for six filters (Bielefeldt et al., 2010). Virus removal efficiency of ceramic pot filters does not meet the WHO standards for being 'Protective' (LRV ≥ 3 ; WHO, 2011). No critical parameter is yet found to enhance the virus removal efficiency.

Different factors in the production process and performance assessment influence the reported removal effectiveness (Table 1). The local craftsmanship and materials, typical for the production process of these ceramic pot filters, have an inherent variability in itself. The type of clay used, burnt materials as poreformer, temperature and place in the kiln, the way and type of silver that is applied, wet or dry season and many more parameters all have their influence on the performance of the filter. Another variable is the way the filters are used. Some families treat mostly rain water, others treat local surface water with a high turbidity which can have a direct effect on performance and on cleaning practice. Finally the research method used to assess the performance can have a large impact on the reported efficacy. The type of bacteria and viruses, their culture techniques, the number of duplicates and laboratory or field conditions all influence the performance of the filter and the accuracy of the assessment. The variability creates concern about the consistency of the removal effectiveness of the CPF. For a water treatment technique which is used to supply more than 4 million people such uncertainty should not be acceptable. It also shows that suggestions for optimization are only valuable when they are based on thorough and solid research, since it might influence the water supply of millions of people.

1.2. Role of silver

In this study the role of silver, as additive to enhance bacterial disinfection, is investigated, since the influence of silver impregnation on the microbial performance is still poorly understood. In some studies the (re)application of silver has an immediate effect on the removal efficiency with regards to *E. coli* (Bielefeldt et al., 2009). The research and modelling by Bielefeldt et al. (2009) showed that bacterial efficacy increases

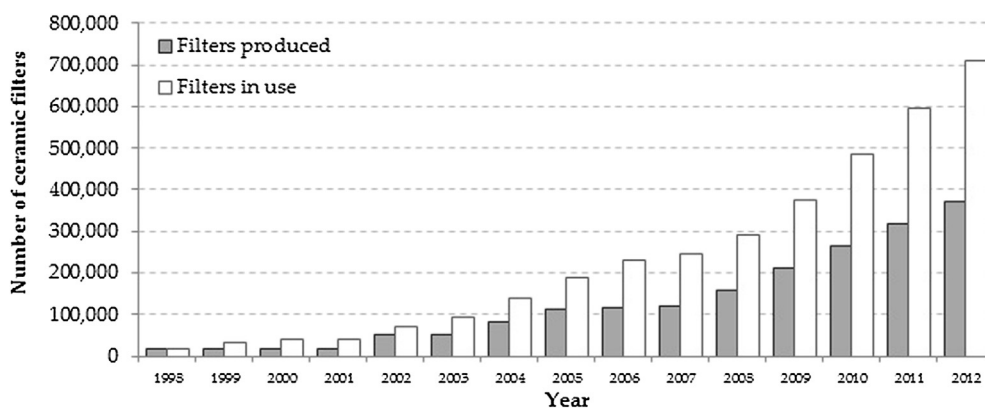


Fig. 1 – Estimation of the total number of ceramic pot filters worldwide, based on information provided by Rayner et al. (2013) and assuming 5 new factories per year since 2009 with each 1000 filter pots per month production capacity, a failure rate of 12% at the factory (Rayner et al., 2013) and a disuse rate of 2% a month at the users (Brown, 2007).

Table 1 – Possible influencing parameters on the removal effectiveness of the ceramic pot filters.

	Possible influencing parameters
Raw materials	Clay, amount and particle size of rice husk,
Production process	laterite, silver nitrate solution
	Temperature, place in kiln, dry or wet season,
Use	pressure, amount of silver used, the way silver is applied
	Water quality (turbidity, pH, temperature, concentration bacteria, viruses), cleaning
	frequency and protocol, throughput, age
Research setup and procedure	E. coli and virus type, concentration of spiked water, analysis method, residence time, temperature, laboratory or field study

with higher silver impregnation. Yet, with subsequent batches of water loaded onto the filters, the microbial effectiveness diminished (Bielefeldt et al., 2009). In other research (Oyanedel-Craver and Smith, 2008; Brown and Sobsey, 2010) a comparison showed no significant difference between filters with and without silver impregnation. A recent study with Nicaraguan filters without silver show an average LRV of 2.1 for *E. coli*, suggesting that colloidal silver may not be necessary for the filters to remove bacteria effectively (Clark and Elmore, 2010).

In previous studies (Van Halem et al., 2007; Van Halem et al., 2009) it was hypothesized that the removal of viruses could be enhanced when no silver is applied. Without silver a biofilm can develop, creating a surface or filter cake layer that can remove MS2 bacteriophages. This hypothesis was formulated based on a higher LRV for viruses by Nicaraguan filters without silver compared to similar impregnated Nicaraguan filters.

The aim of this study was to determine the role of applied silver during ceramic pot filtration and subsequent storage in the plastic receptacle. In this study non-silver filters, filters with silver impregnation on the outside of the filter element and filters impregnated with silver on both sides were compared in long-term loading experiments with *E. coli* (K12 and WR1) and MS2 bacteriophages in natural challenge water. Additionally, batch experiments were performed to investigate the potential inactivation of *E. coli* during contact time in the receptacle.

2. Materials and methods

2.1. Filter production and silver application

The performance of 22 filters was evaluated in this study. Four of these filters served as a reference and were produced at the full scale RDIC factory in Cambodia, following the standard

production process, as described by Brown and Sobsey (2010). Standard RDIC mix consists of 30 kg clay, 9.7 kg rice husks, 1 kg of laterite and 14.5 L of water per batch of six filters. The rice husks are finely ground and sieved at a screen size of <1 mm.

The other 18 filters were produced using a small-scale gas fired temperature-controlled kiln at the production site of RDIC (Gensburger, 2013). With this smaller kiln batches of six ceramic filters could be produced under highly controlled conditions while using exactly the same local materials as standard produced filters. These filters were produced with three different rice husk contents, namely 9.7 kg, 12 kg and 14 kg per batch. All filters were shipped to The Netherlands for the experimental work.

Silver was applied in three different ways:

- i) Ten filters, including the four RDIC reference filters, were painted with AgNO₃ on the inside and outside of the filters as it is commonly done;
- ii) Six filters were only painted with AgNO₃ on the outside of the filter;
- iii) Six filters were not impregnated with silver.

The silver was applied following the standard RDIC silver impregnation protocol: filter elements were painted with 0.00215 M reagent-grade AgNO₃ with 200 mL solution on the inside, and 100 mL on the outside of the element. The silver

Table 2 – Characteristics of the filters used in this study. Filters were selected from six different batches based on availability, silver impregnation and after passing a soak-and sound test, to find cracks in the material. The* indicates that the filters were made using the gas fired temperature-controlled kiln instead of the full-scale kiln of the RDIC factory. The flow rates and amount of rice husk listed here show that the scope of this research was wider, yet this article focuses only on the role of silver.

AgNO ₃ application	Rice husk per batch (kg)	Initial flow rate (L h ⁻¹)	Batch number	Filter number
Both sides	9.7	2.4	RDIC	I
Both sides	9.7	3.1		II
Both sides	9.7	2.6		III
Both sides	9.7	2.4		IV
Both sides	9.7	2.2	B7*	P1
Both sides	9.7	2.6		P5
Both sides	12	11.3	B25*	P3
Both sides	12	10.7		P4
Both sides	14	19.1	B26*	P3
Both sides	14	11.6		P4
Outside	9.7	2.3	B7*	P3
Outside	9.7	3.3		P4
Outside	12	5.9	B18*	P1
Outside	12	6.9		P4
Outside	14	14.4	B17*	P1
Outside	14	15.0		P4
None	9.7	5.3	B23*	P1
None	9.7	5.5		P2
None	12	11.5	B25*	P1
None	12	12.7		P2
None	14	21.0	B26*	P1
None	14	19.3		P2

nitrate solution used originated from the RDIC factory, where the Microdyn silver-based disinfectant with 3.2% AgNO_3 and 0.6% $\text{Cu}(\text{NO}_3)_2$ by mass is applied (Brown and Sobsey, 2010). This silver nitrate solution was also used in other studies (Brown and Sobsey, 2010; Lantagne, 2001; Fahlin, 2003; Van Halem et al., 2007; Bloem et al., 2009). An overview of the filters with the different characteristics is shown in Table 2.

During the experiment the leaching of silver was monitored and silver analyses were done with an Inductively Coupled Plasma Mass Spectrometer (Thermo scientific X-series) using the NEN-EN-ISO 17294-2 method, with a lower detection limit of $0.1 \mu\text{g L}^{-1}$.

2.2. E. coli and MS2 stock preparation

The *E. coli* (WR1) and *E. coli* (K12) bacteria were used for spiking the raw water. The WR1-type was chosen since it is commonly used for research and routine quality control of drinking water in Dutch laboratories. The K12-type was selected, since this type of *E. coli* was used in several other studies with CPF (Van Halem et al., 2007; Oayandel and Smith, 2008). For both *E. coli* types stock was prepared by growing overnight in Peptone water at 25°C . The concentrated solution of *E. coli* (WR1) was diluted with sterile skimmed milk and immediately stored until use at -80°C . The solution of *E. coli* (K12) was prepared the night before the day of spiking the water. The MS2 stock was generated following the procedure according to ISO 10105-1.

The filters were spiked six times: four times with *E. coli* (week 1, 3, 6 and 7) and two times with MS2 (week 10 and 16). A 210 L vessel was filled with surface water and stirred (150 RPM) to prevent settling. On the testing day, the required amount of *E. coli* or MS2 stock was mixed in the water while the water was stirred for 15 min (150RPM). The filter elements were all emptied beforehand of the remainder of the water load of the day before, and filled with the spiked water. The first 2 L throughput of each filter was collected and discarded, to displace any remaining unspiked water in the filter pores. Directly thereafter a 250 mL sample was collected in a sterile bottle prepared with 0.5 mL Sodium thiosulfate ($0.06 \text{ M Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$) and Nitrilotriacetic acid (NTA) ($0.13 \text{ M C}_6\text{H}_6\text{NNa}_3\text{O}_6 \cdot \text{H}_2\text{O}$) solution. The samples were refrigerated at a temperature of $2-8^\circ\text{C}$ before processing on the same day.

E. coli analyses were performed according to NEN-EN-ISO 9308-1 (ISO, 2000) using membrane filtration and incubation on Lauryl sulfate agar (Oxoid). MS2-bacteriophages analyses were performed according to NEN-EN-ISO 10705-1 (ISO, 1995). MS2 bacteriophages were enumerated on tryptic yeast glucose agar (TYGA) using the double agar layer technique.

2.3. Water quality

For this study natural challenge water from the canal Schie was used, a water body which flows through the city of Delft, with an average Total Suspended Solids (TSS) of 14.9 mg L^{-1} (STD = 6.1), temperature of 9.2°C (STD = 5.7; 16°C at the start and 7.5°C at the end of the study due to seasonal variation), a pH of 7.9 (STD = 0.1) and a conductivity of 0.85 mS m^{-1} (STD = 0.10). Water quality was measured at OW062-002

(Kruithuisweg, Delfland Water Board). This water contained on average 94 (19–290) $\text{CFU} \cdot 100 \text{ mL}^{-1}$ *E. coli*.

2.4. Batch experiments

Batch experiments were conducted to quantify the deactivation of *E. coli* in relation to different contact times. In this experiment 4 L brown glass bottles were filled with surface water filtrate of non-silver ceramic filters, simulating the water quality in the storage. An AgNO_3 solution was added and the solutions were spiked with a suspension of *E. coli*. Three silver concentrations were tested <0.001 (as reference), 0.021 and $0.083 \mu\text{mol}$. The batch experiment was done in duplicate for both *E. coli* K12 or WR1. During the experiment 5 samples were taken after 0–360 min of contact time.

2.5. Loading experiment

All filter elements were placed in a plastic receptacle (22 L), which was customized with a valve fitted in the bottom to minimize the dead volume of water inside the receptacle (Fig. 2). Receptacles were rinsed after valve placement. No chlorine was used to prevent inactivation by any residuals. As a control, before the start of the spike test reference samples were taken from the receptacles with chlorine free tap water to see whether contamination of the receptacle had occurred.

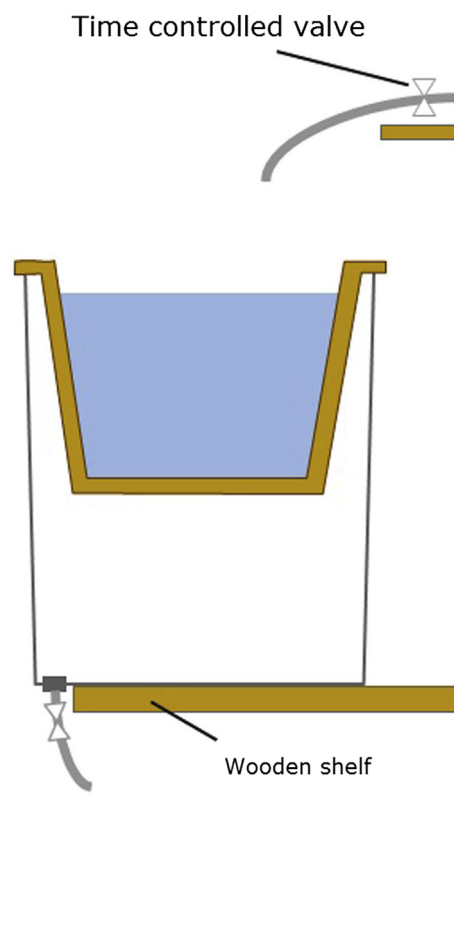


Fig. 2 – Schematic drawing of research setup of filters.

100 mL samples resulted in an average of 16 (STDEV = 23) CFU·100 mL⁻¹ (data not shown), indicating that these numbers would only create bias if the LRV values would be higher than 3 since the spiked influent concentrations varied between $8 \cdot 10^4$ – $2 \cdot 10^7$ CFU 100 mL⁻¹.

The flow rate of the filters was measured using the same measuring protocol as at the RDIC factory. As a constant head permeability test, the throughput was measured twice while keeping a constant water level using a volumetric measuring beaker (2 L) and the average of the two measurements was taken as flow rate. When the flow rate dropped below 1 L h⁻¹ the filter was emptied and scrubbed with a hand brush three times inside and one time outside using chlorine free tap water. After scrubbing, the flow rate of the scrubbed elements was determined again.

3. Results

3.1. Silver leaching and inactivation during batch tests

The results of silver leaching out of the filter elements as a function of the amount of filtered water are shown in Fig. 3. The markers in the figure differ for the three ways of silver application (both sides, only outside and no silver), the latter once serving as a reference to check natural die-off without silver addition. The results show that for filters impregnated with silver the leaching of silver starts at 0.093–0.232 μmol (10–25 μg L⁻¹) and quickly diminishes. There is no significant difference in the silver leaching between the filters with only silver on the outside of the pot and the filters impregnated on both sides.

The range of silver concentrations found was used in the batch experiment to quantify the deactivation of *E. coli* in relation to different contact times. In Fig. 4 the results are shown for both *E. coli* species (WR1 and K12) with three silver concentrations (<0.001, 0.021 and 0.083 μmol). It is evident that with increasing silver concentration the inactivation rate λ increases. Even with a silver concentration of 0.021 μmol the LRV is 1.0 after 180 min contact time and 2.1 after 280 min. With 0.083 μmol Ag the LRV is 1.6 after 180 min, and the *E. coli* analyses were negative (<1 CFU mL⁻¹; LRV > 2.5) after 280 min.

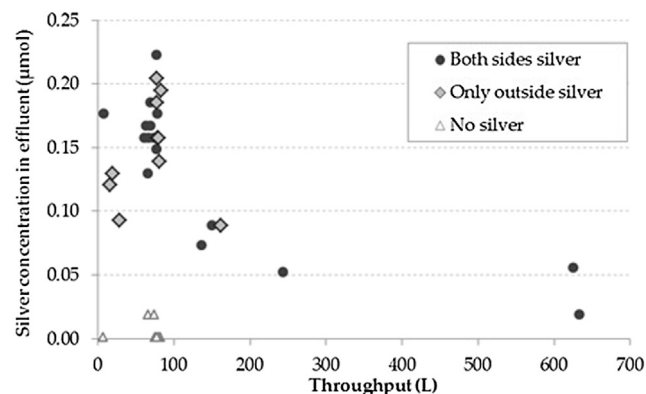


Fig. 3 – Silver leaching of the filter pots with different types of silver application.

The WR1 species at <0.001 μmol Ag appeared to have strong inactivation after 90 min. However, since the consecutive samples resulted in a constant inactivation curve, the sample after 90 min was considered an outlier. For K12 the initial concentration at t = 0 was rather low at 8 CFU mL⁻¹, which makes the results less reliable and the maximum LRV to determine 0.9. Nonetheless it can be seen that for the silver concentrations of 0.021 and 0.083 μmol the inactivation rates are similar compared to the WR1 species. At <0.001 μmol the K12 species did not give consistent results. Both *E. coli* species seem to react similar to the influence of silver in these specific batch experiments; in the remaining figures no speciation will be made between the two species.

3.2. Silver inactivation during storage

The silver inactivation during filtration and storage was investigated in a long-term loading experiment, with 16 ceramic pot filters (all except the non-silver filters). In order to differentiate between silver activation during filtration in the ceramic filter element and subsequent storage in the plastic polypropylene receptacle, samples were taken at two different intervals: (1) 660 min after filtration (overnight in receptacle) and, (2) <5 min storage time. Results in Fig. 5 clearly show that with a contact time overnight the LRV is up to 2 log values higher than when the water is directly sampled after filtration.

3.3. Silver inactivation during filtration step

The silver inactivation during the filtration step was determined by comparing the results of the filters with the same storage time (<5 min) and the three different silver applications (none, outside, both sides). Fig. 6 shows that there is no significant difference between the three categories (One-way ANOVA test; Tukey–Kramer method; significance level α = 0.05). This is a strong indication that during the filtration through the ceramic element the inactivation by silver present on ceramic material itself does not play a dominant role. This implies that the measured removal effectiveness, between LRV 0.6 and 3.1, are not a result of silver inactivation, but can be designated to the physical removal mechanisms of the filters such as size exclusion and tortuosity.

3.4. Interaction silver on removal effectiveness MS2

The results of removal effectiveness tests for MS2 bacteriophages are shown in Fig. 7, samples were taken with a storage time <5 min. The boxplot diagram shows the results of two testing days, after 391 L and 632 L throughput. It can be seen that the effectiveness of the filter elements is slightly but not significant diminishing over the amount of water treated (throughput 632 L). The LRVs for MS2 are lower than the LRVs for *E. coli*. The removal varies between a LRV of 0 and 1.4, with an average of 0.6 (n = 22; STD = 0.3).

The results in Fig. 8 show that no relation between different ways of silver application and the LRV for MS2 bacteriophages was found (One-way ANOVA test; Tukey–Kramer method; significance level α = 0.05). The hypothesis that the absence of silver would enhance the virus removal cannot be confirmed. The found removal effectiveness for MS2 is on

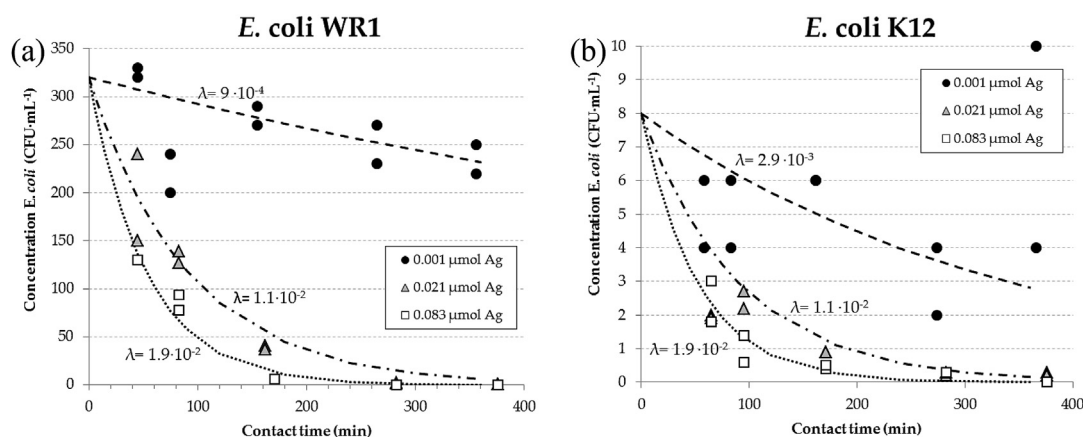


Fig. 4 – Results of the batch experiment, with the influence of different silver concentrations on the die-off rate of the *E. coli* WR1 (a) and K12 (b) at a temperature of 20 °C.

average LRV = 0.6 (STD = 0.3); measured after 391 L and 632 L throughput).

4. Discussion

The results show that during the filtration phase the inactivation by silver does not play a dominant role. No significant difference is found between the filters with or without silver, similar to results reported in other studies (Oyanedel-Craver and Smith, 2008; Brown and Sobsey, 2010).

Yet, the removal efficiencies found in this study are quite low (LRV = 1.2 (STD = 0.6); measured over first 300 L, average of all filters) compared to others studies with filters without silver, where LRVs around 2 were measured (Van Halem et al., 2007; Oyanedel-Craver and Smith, 2008; Brown and Sobsey, 2010; Clark and Elmore, 2011). This might be partly due to the difference in the use of naturally present *E. coli* (Clark and Elmore, 2011) and lab grown bacteria used in this study. In natural waters bacteria may be aggregated, attached to larger particles or encapsulated in flocs, with the result that mechanisms such as size exclusion and tortuosity are more effective.

Another aspect is the influence of temperature on the inactivation of *E. coli*. The temperature of the water (7.5–16 °C) is relatively low compared to tropical conditions in the target areas of the CPF. Other disinfection reactions, such as with ozone and chlorine, are known to follow the Arrhenius equation, which implies that with a difference of 10 °C the log inactivation doubles (Hunt and Mariñas, 1997; Larson and Mariñas, 2003). It is reasonable to assume that for the reaction with silver there is a similar influence, therefore at higher temperatures the LRV might be significantly higher under tropical conditions.

The influence of silver during storage is found to be crucial. The results show that even concentrations as low as 0.0021 μmol Ag in stored water can cause a LRV >2.5 after approximately 5 h storage time. This can explain the higher LRVs found by Van Halem et al. (2007) compared to this study. In this study the storage time was minimized to less than

5 min, while the samples in Van Halem's study were taken overnight resulting in a storage time of more than 8 h. Nevertheless, questions remain since Brown and Sobsey (2010) report no difference between silver/non-silver, while the contact time was 5 h.

It has been suggested before that the aspect of safe storage is an inherent quality of the ceramic pot filters. We believe that this is one of the main reasons behind the success of the pot filter: it ensures safe storage, prevents recontamination and provides long-time inactivation by silver during storage. This finding implies as a practical consequence that it is beneficial to look for ways to ensure an extensive storage time. Furthermore it creates chances for high flow rates pots with lower clay:rice husk ratios, since the inactivation during the filtration step is not the domination mechanism. Yet, the filtration step is still essential for the removal of suspended solids, the removal of aggregated bacteria attached to larger particles and pathogens which are less sensitive to inactivation by silver.

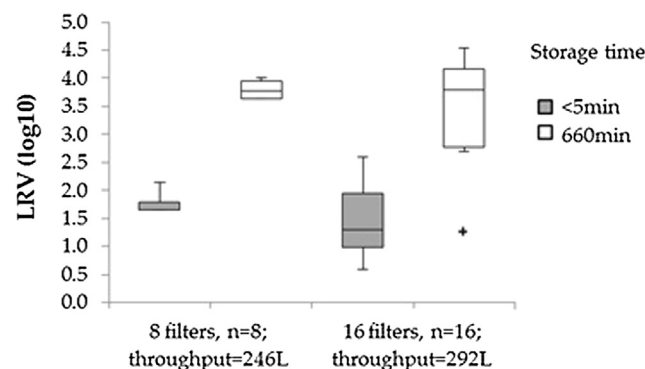


Fig. 5 – Comparison LRV of *E. coli* by silver in two independent experiments with two different storage times. Box plots show minimum, first quartile, median, third quartile, and maximum. Plus symbol indicate outlier. *n* corresponds with the number of measurements on which the boxplot is based.

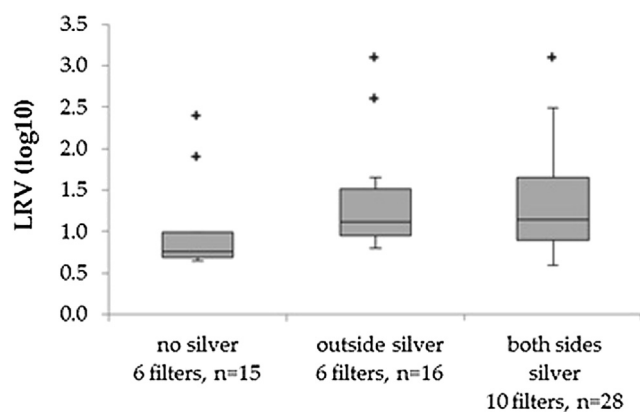


Fig. 6 – Comparison between LRV for *E. coli* with minimal storage time with the three different ways of silver application (non, outside and both sides). Box plots show minimum, first quartile, median, third quartile, and maximum. Plus symbols indicate outliers. *n* corresponds with the number of measurements on which the boxplot is based.

The found removal effectiveness for MS2 bacteriophages, as indicator mechanism for viruses, is still too low and does not meet the desired standard of $LRV \geq 3$ (WHO, 2011). A hypothesis that in natural waters the removal MS2 bacteriophages is better, as stated by Salsali et al. (2011), could not be confirmed. An improved removal by the formation of a biofilm in the non-silver pots was not found. Here the low temperatures of the natural challenge water (9.5 °C) might have created sub-optimal circumstances for growth of a biofilm. Typically the surface waters in the target areas of the CPF have a higher turbidity with higher amount of suspended solids, which could have an additional effect for viruses may attach to suspended organics and other particles, making the filtration process more effective. It is suggested that future research should be conducted with higher water temperatures and with varying amounts of suspended solids.

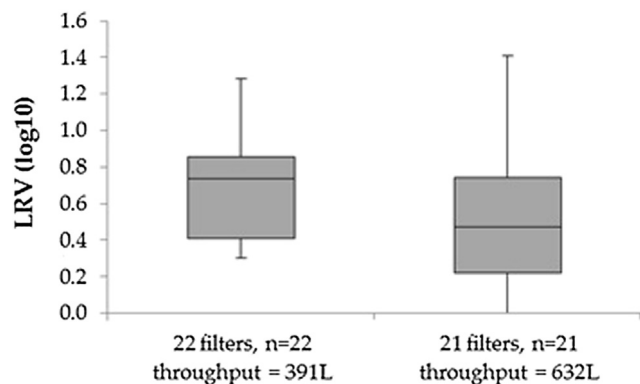


Fig. 7 – The LRVs for MS2 bacteriophages in two independent subsequent testing days. Box plots show minimum, first quartile, median, third quartile, and maximum. *n* corresponds with the number of measurements on which the boxplot is based.

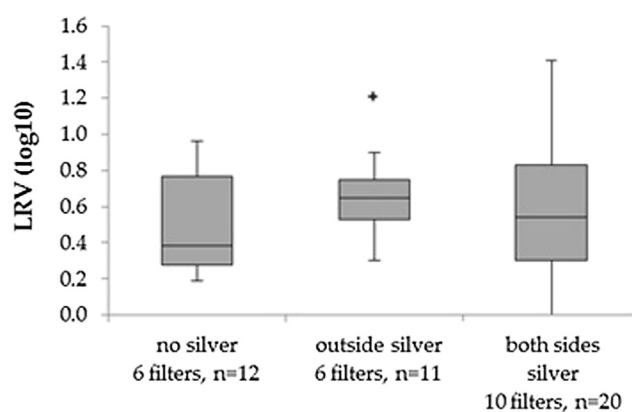


Fig. 8 – Comparison of difference silver application and the influence on the removal effectiveness of MS2 bacteriophages. Box plots show minimum, first quartile, median, third quartile, and maximum and one outlier. *n* corresponds with the number of measurements on which the boxplot is based.

Furthermore, the influence of a longer storage time on virus inactivation by silver, similar to the influence on *E. coli*, was not determined in this study and is suggested to be explored in future research. So far no production variable was found in this study to enhance the virus removal efficiency. Hence, the removal effectiveness for viruses is still of major concern for the CPF.

5. Conclusions

In this study the influence of silver on microbial removal efficiencies during ceramic pot filtration and storage was investigated. It was found that the storage time in the receptacle is the dominant parameter to reach *E. coli* inactivation, and not the contact time during the filtration phase. Even with rather low concentration of silver in the receptacle a storage time of about 4–5 h resulted in a Log Reduction Value of 2. The hypothesis that the absence of silver would enhance the virus removal could not be confirmed, potentially due to the low natural challenge water temperatures in this study.

The main strength of ceramic pot filters – its local production – sets at the same time its biggest challenge: how to ensure a standardized ceramic element quality? Although the filter was proven effective in practice, there is potential room for improvement. This study shows that the contact time with silver is the main factor for *E. coli* reduction by ceramic pot filters as they are currently produced. Other (pathogenic) microorganisms may be less susceptible to silver, as shown for MS2 phages. *E. coli* is therefore not a suitable indicator to test non-chemical efficiency of silver coated filter pots.

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