

# Removals of *Cryptosporidium parvum* oocysts and *Cryptosporidium*-sized polystyrene microspheres from swimming pool water by diatomaceous earth filtration and perlite-sand filtration

Ping Lu, James E. Amburgey, Vincent R. Hill, Jennifer L. Murphy, Chandra L. Schneeberger, Michael J. Arrowood and Tao Yuan

## ABSTRACT

Removal of *Cryptosporidium*-sized microspheres and *Cryptosporidium parvum* oocysts from swimming pools was investigated using diatomaceous earth (DE) precoat filtration and perlite-sand filtration. In pilot-scale experiments, microsphere removals of up to 2 log were obtained with 0.7 kg-DE/m<sup>2</sup> at a filtration rate of 5 m/h. A slightly higher microsphere removal (2.3 log) was obtained for these DE-precoated filters when the filtration rate was 3.6 m/h. Additionally, pilot-scale perlite-sand filters achieved greater than 2 log removal when at least 0.37 kg/m<sup>2</sup> of perlite was used compared to 0.1–0.4 log removal without perlite both at a surface loading rate of 37 m/h. Full-scale testing achieved 2.7 log of microspheres and oocysts removal when 0.7 kg-DE/m<sup>2</sup> was used at 3.6 m/h. Removals were significantly decreased by a 15-minute interruption of the flow (without any mechanical agitation) to the DE filter in pilot-scale studies, which was not observed in full-scale filters. Microsphere removals were 2.7 log by perlite-sand filtration in a full-scale swimming pool filter operated at 34 m/h with 0.5 kg/m<sup>2</sup> of perlite. The results demonstrate that either a DE precoat filter or a perlite-sand filter can improve the efficiency of removal of microspheres and oocysts from swimming pools over a standard sand filter under the conditions studied.

**Key words** | *Cryptosporidium parvum*, diatomaceous earth, perlite, polystyrene microspheres, precoat filter, swimming pool

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## INTRODUCTION

Numerous outbreaks of cryptosporidiosis have been reported to be associated with swimming pools in the United States (USA), United Kingdom (UK), Australia, and other countries over the past 20 years (Lisle & Rose 1995; Puech *et al.* 2001; Karanis *et al.* 2006; Hlavsa *et al.* 2011; Lu *et al.* 2013). In August 1988, the first outbreak of cryptosporidiosis associated with a swimming pool in the UK was recognized (Joce *et al.* 1991). In 1992, a large increase in the number of cases of cryptosporidiosis was reported in the USA (McAnulty

*et al.* 1994). Due to *Cryptosporidium* oocysts being extremely tolerant to free chlorine at levels typically used in treated recreational water venues, this pathogen poses a significant risk to bathers, as exemplified in large outbreaks in New York in 2005 (more than 4,000 cases of cryptosporidiosis), in Utah in 2007 (nearly 2,000 cases), and in the Dallas, TX area in 2008 (at least 378 cases) (Rolfs *et al.* 2008; Cantey *et al.* 2012).

The Centers for Disease Control and Prevention (CDC) recommends that swimming pools in the USA maintain a

free chlorine residual of 1–3 mg/L for effective disinfection (CDC 2012). At 1 mg/L of free chlorine, achieving a 3 log (99.9%) inactivation of *Cryptosporidium* could require almost 11 days assuming a 3 log Ct at pH 7.5 [free chlorine concentration (C) × exposure time (t)] of 15,300 mg/L·min (Shields *et al.* 2008). Thus, other treatment techniques (e.g., filtration, UV irradiation) are necessary to reduce *Cryptosporidium* in treated recreational water venues under normal operating conditions. However, swimming pool sand filters in the USA are not very effective at removing *Cryptosporidium parvum* oocysts and *Cryptosporidium*-sized (4.5 µm) microspheres (<63%, or 0.43 log) (Amburgey 2011; Lu & Amburgey 2016).

Media such as diatomaceous earth (DE) and perlite can be used to precoat filters and improve filtration efficacy. DE is a naturally occurring, soft, siliceous sedimentary rock and can be applied to filters in the form of a very fine white to off-white powder. Perlite is a naturally occurring amorphous volcanic glass that, when heated sufficiently, has the unusual property of greatly expanding in volume with a concomitant drop in density (10- to 35-fold). While often used in horticultural and insulation applications, perlite is also widely used as a filter aid. Amburgey *et al.* (2012) reported that DE and perlite precoat filters demonstrated pathogen removal efficiencies of 2.3 to 4.4 log (or 99.4–99.996%) in a small-scale 946 L model swimming pool.

Intentionally interrupting the water flow in a precoat filter by turning off the filter pump allows the precoat media and collected contaminants to detach from the filter septum and become intermixed. When the filter is restarted, the media reattaches to the filter septum, forming a new (intermixed) matrix. This process can lengthen the media lifespan. However, because interruption of water flow changes the media and pathogen distribution within the filter, it might impair pathogen removal or facilitate the release of pathogens previously trapped in the filter. The effects of flow interruption have not been sufficiently studied (Amburgey *et al.* 2012).

Additional research is needed to optimize DE filter performance in removing *Cryptosporidium* oocysts in pilot- and full-scale pools. Parameters to be investigated included water flow rate, mass/depth of DE, and operational practices like filter flow interruption.

The current study investigated *Cryptosporidium*-sized microsphere removals with different amounts of precoat (i.e., DE or perlite) in a pilot-scale swimming pool in order to determine

an appropriate amount of precoat for full-scale pool operation. *Cryptosporidium*-sized microsphere and *C. parvum* removals by DE filtration and perlite-sand filtration in a full-scale swimming pool were evaluated to verify the filtration performance. The impact of flow interruption on DE filter performance was evaluated in pilot-scale and full-scale pools to determine its impact on the removal of *Cryptosporidium* from the pool.

## MATERIALS AND METHODS

### Experimental setup

#### Pilot-scale

The pilot-scale trials were conducted in Charlotte, NC, USA. Figure 1 illustrates the setup of the 5.5 m<sup>3</sup> swimming pool used in pilot-scale trials, including the DE precoat filter (FNS Plus 24, Pentair, Sanford, NC, USA), perlite-sand filter (Triton II TR 40, Pentair, Sanford, NC, USA), and supporting control and monitoring equipment (CAT 5000, Hayward, Rockville, MD, USA). Pool water was pumped through the filter and circulated at different rates by a centrifugal pump (Y-2051.0185, Speck Pumpen, Jacksonville, FL, USA). Pool water pH and oxidation reduction potential (ORP) were monitored using sensors that were mounted in a bypass line and connected to a controller. Local tap water was supplemented with NaHSO<sub>4</sub>, CaCl<sub>2</sub>, NaHCO<sub>3</sub>, Ca(OCl)<sub>2</sub>, to adjust pool water to alkalinity of 100 mg/L as CaCO<sub>3</sub>, hardness of 200 mg/L as CaCO<sub>3</sub>, free Cl<sub>2</sub> of 2 mg/L, and pH of 7.5. Acid and chlorine were fed into the pool to maintain the water chemistry by pumps (gala 1601PVT200UDO12100, ProMinent, Heidelberg, Germany; SVP4, Stenner Pump Company, Jacksonville, FL, USA).

#### Full-scale

The full-scale trials were conducted at the Lonza research facility in Conley, GA, USA. Figure 2 illustrates the experimental setup of the two 37.9 m<sup>3</sup> (10,000 gallon) swimming pools, including the DE precoat filter, perlite-sand filter, and supporting control and monitoring equipment. A chemical feed controller (Siemens Strantrol; System 3i) monitored pool water pH and ORP. Filter pressure was indicated by a

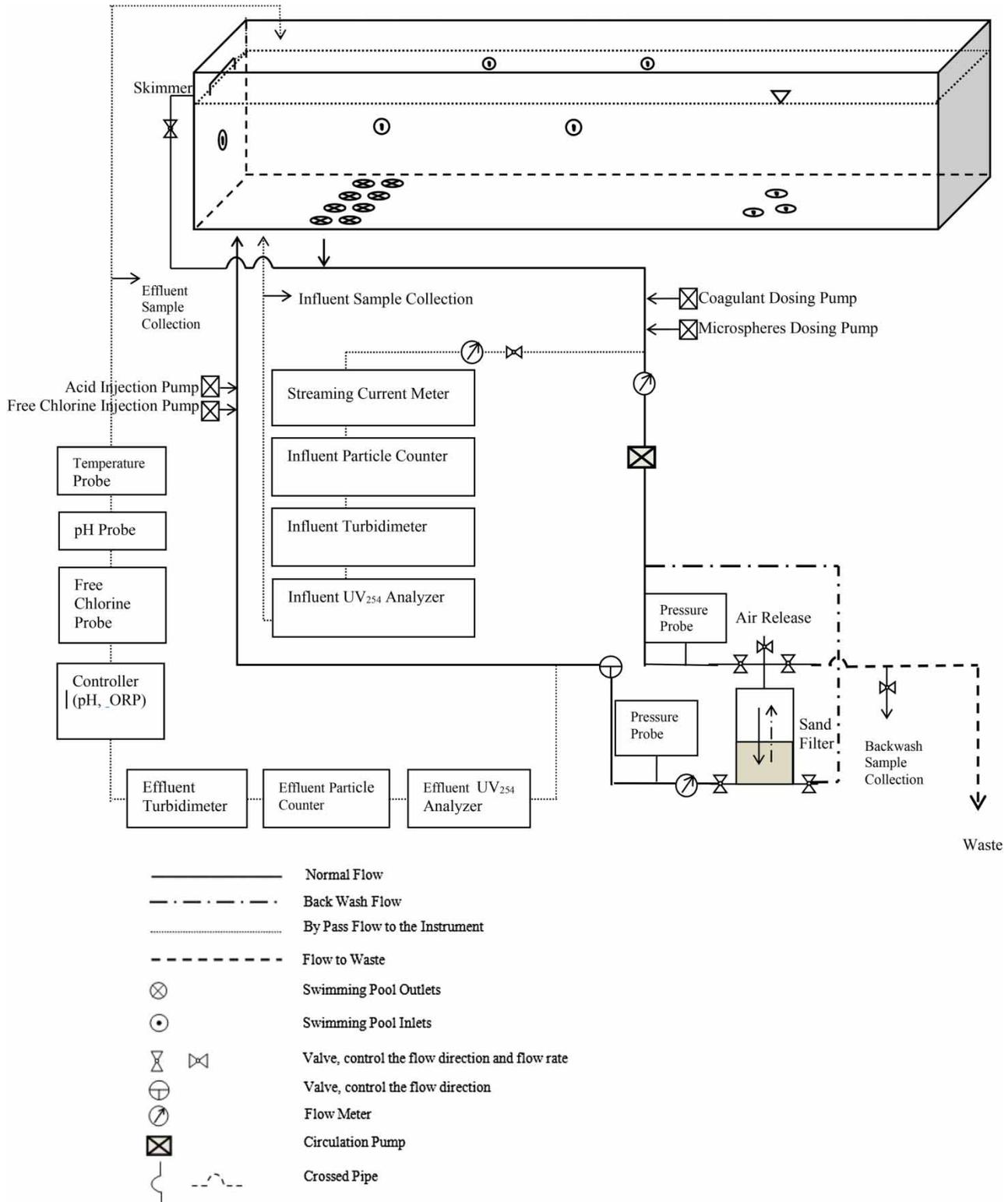
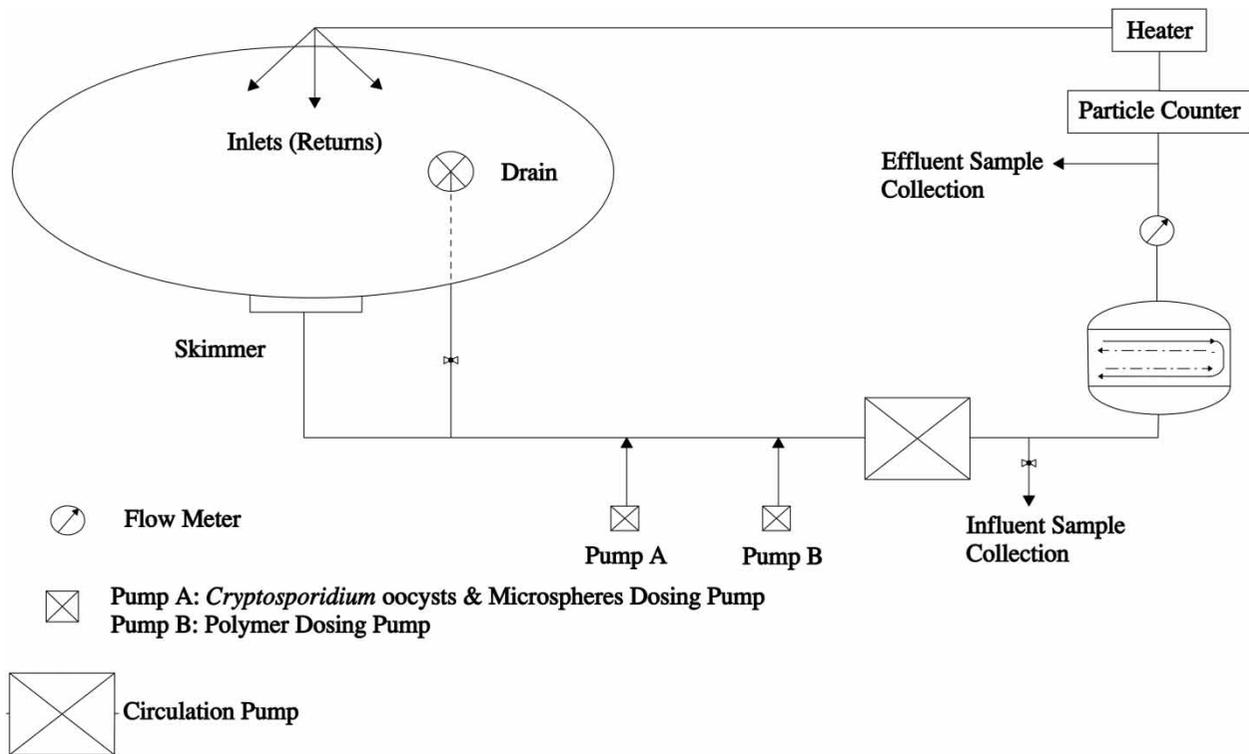


Figure 1 | Pilot-scale swimming pool setup (Lu & Amburgey 2016).



**Figure 2** | Full-scale swimming pool setup.

mechanical pressure gauge. Local tap water was supplemented with  $\text{NaHSO}_4$ ,  $\text{CaCl}_2$ ,  $\text{NaHCO}_3$ ,  $\text{Ca}(\text{OCl})_2$ , to adjust pool water to alkalinity of 84 mg/L as  $\text{CaCO}_3$ , hardness of 247 mg/L as  $\text{CaCO}_3$ , free chlorine of 2–3 mg/L, and pH of 7.3–7.5. Free chlorine concentration (Standard Method 4500-Cl F: DPD Titration; APHA 2005) and pH (Mettler Toledo 7 Easy pH Meter) were measured manually and adjusted by the controller as needed (APHA 2005).

For pilot-scale study, duplicate experiments were conducted with triplicate samples (50 mL influent and 500 mL effluent samples were collected); for full-scale study, triplicate samples were collected (500 mL influent and 1 L effluent samples were collected).

### ***Cryptosporidium*-sized microspheres and *C. parvum* oocysts**

#### ***Cryptosporidium*-sized microspheres**

Fluoresbrite™ Carboxylate YG polystyrene microspheres (Polysciences, Inc., Warrington, PA, USA) with diameter

of 4.5  $\mu\text{m}$  were used as a model particle surrogate for *C. parvum* oocysts in both the pilot- and full-scale studies because they are virtually identical to *Cryptosporidium* oocysts in size, shape, and density. For microsphere analysis, influent and effluent samples were mixed by vortexing and hand shaking for at least two minutes each before analysis. Samples were passed through 3.0  $\mu\text{m}$  pore size polycarbonate track-etched filters. Each polycarbonate filter was mounted on a glass microscope slide with polyvinyl alcohol-DABCO solution, covered with a glass cover slip, and enumerated under an epifluorescence microscope (Lu & Amburgey 2016).

#### ***C. parvum* oocysts**

Heat-inactivated *C. parvum* oocysts were also used in full-scale experiments. In the perlite study, oocysts produced in experimentally infected bovine calves were purified from calf feces by sucrose and cesium chloride gradients and stored in phosphate-buffered saline (PBS) with antibiotics and 0.01% Tween 20 for approximately 7.5 months. In the DE study,

oocysts were purchased from Waterborne, Inc. (New Orleans, LA, USA) and stored at 4 °C in PBS with antibiotics for approximately four months. Oocysts in both studies were heat-inactivated at 55 °C for 1 hour prior to seeding.

For *C. parvum* analysis, filter influent and effluent samples were concentrated using two centrifugation steps at 4,000 × g at 4 °C for 15 minutes followed by two microcentrifugation steps at 15,000 × g at 4 °C for 5 minutes. All bottles and tubes were rinsed with a Tween 80/Antifoam A solution (0.01% and 0.001% [v/v], respectively) between transfers to prevent oocysts from sticking. The entire resuspended pellet was transferred onto one or more *Crypto* SuperStick™ slide wells (Waterborne™, Inc., New Orleans, LA) and stained with Easy Stain™ (TCS Biosciences Ltd, Buckingham, UK) per the manufacturer's instructions. *C. parvum* oocysts were detected and enumerated using an epifluorescence microscope under 200× magnification (USEPA Method 1623, 1999) (USEPA 1999).

## DE experimental procedure

### Pilot-scale

DE density was 320 kg/m<sup>3</sup>, and the permeability was 3.6 μm<sup>2</sup>. The filter surface area was 2.23 m<sup>2</sup>. The amount of precoat was defined as the mass of DE loaded divided by the filter surface area. A DE slurry was prepared by mixing dry DE media into water at a volumetric ratio of 1:2 and mixed well in a 10 L plastic container. The slurry was delivered through a skimmer, circulated through the system, and collected on the filter septum resulting in final amounts of 0.5, 0.7, and 1.0 kg-DE/m<sup>2</sup>. Pool water was pumped through the filter and circulated at filtration rates of 3.6 m/h, 5 m/h, and 6 m/h.

To assess pilot-scale DE filtration efficacy, microspheres (final concentration of 150 microspheres/mL in the pool water) were fed over 60 minutes directly before the filter pump with a peristaltic pump (Model 505 Di, Watson Marlow, Wilmington, MA, USA). Triplicate samples were collected over the 60-minute time period (at 10 min, 30 min, and 50 min), and triplicate samples were taken each time to assess variability. Flow interruption was conducted by restarting the filter after a 5-minute, 15-minute, or 30-minute interruption of filter flow. Influent samples were

collected in 50 mL polypropylene centrifuge tubes, and effluent samples were collected in 500 mL glass bottles. All samples were stored at 4 °C and analyzed within 48 hours.

### Full-scale

The filter surface area was 2.2 m<sup>2</sup>. Dry DE media was delivered through a skimmer, circulated through the system, and collected on the filter septum, resulting in final density of 0.7 kg DE/m<sup>3</sup>. Pool water was pumped through the filter and circulated at a filtration rate of 3.6 m/h.

To assess full-scale DE filtration efficiency, approximately 2.5 microspheres/mL (10<sup>8</sup> total) microspheres and 1.5 oocysts/mL (6 × 10<sup>7</sup> total) were then mixed and seeded to the pipe directly before the circulation pump over a 60-minute interval (as shown in Figure 2). Triplicate samples were collected 30 minutes after seeding began. Influent (500 mL) and effluent (1 L) samples were collected in glass bottles containing Tween 80 (0.01% [v/v]), Antifoam (0.001% [v/v]), and sodium polyphosphate (0.01% [v/v]), to prevent oocysts from sticking to surfaces. Flow interruption was conducted by restarting the filter after a 15-minute interruption of filter flow and then collecting influent and effluent samples as described above. All samples were stored at 4 °C prior to analysis.

## Perlite-sand experimental procedure

### Pilot-scale

Experiments were carried out using 30 cm of sand with or without different amounts of perlite added on top of the sand. Table 1 shows sand and perlite characteristics as

**Table 1** | Filter media and filter operating details

	Sand	Perlite
Effective size (d <sub>10</sub> , μm)	485, 0.49	16.9, 0.18 <sup>a</sup>
d <sub>10</sub> /d <sub>90</sub>		
Uniformity coefficient (UC, d <sub>60</sub> /d <sub>10</sub> )	1.50	2.98 <sup>a</sup>
Filter bed depth (cm)	30	–
Filter surface area	0.018 m <sup>2</sup> (0.196 ft <sup>2</sup> )	0.018 m <sup>2</sup> (0.196 ft <sup>2</sup> )

<sup>a</sup>Source: IIG 2011.

well as filter operation details. The filter surface area was 0.018 m<sup>2</sup>. Each perlite slurry was prepared by mixing perlite dry media with water at a volumetric ratio of 1:2 in a 10 L plastic container. The perlite slurries were delivered through a skimmer, circulated through the system, and collected on the filter surface resulting in final amounts of 0.24, 0.37, 0.49, 0.61 kg-perlite/m<sup>2</sup>. Pool water was pumped through the filter at a filtration rate of 37 m/h.

To assess pilot-scale perlite-sand filtration efficiency, microspheres (influent concentration of 150 microspheres/mL in the pool), were fed over 60 minutes directly before the circulation pump. Microspheres were seeded 15 minutes before each sample collection in order to ensure microspheres could be present in the filter effluent during sample collection. The theoretical hydraulic detention time of the piping and filtration systems was approximately 3 minutes.

### Full-scale

Perlite (Tech-Flo 2000X/SwimBrite, IIG, LLC, Brunswick, GA, USA) permeability was 3.21 μm<sup>2</sup>, and the particle size range was between 2 and 200 μm. A control experiment with the sand filter was conducted with sand media only. Dry perlite media were delivered through a skimmer, circulated through the system, and collected on the sand filter surface resulting in a final amount of 0.5 kg-perlite/m<sup>2</sup>. Pool water was pumped through the filter at a surface loading rate of 34 m/h.

To assess full-scale perlite-sand filtration efficiency, approximately 2.5 microspheres/mL (10<sup>8</sup> total) microspheres and 1.5 oocysts/mL (6 × 10<sup>7</sup> total) were then mixed and seeded to the pipe directly before the circulation pump over a 60-minute interval (as shown in Figure 2). The remaining experimental procedures were as described in 'Full-scale' in the subsection 'DE experimental procedure'.

### Data analysis

Box plots are used to present the *C. parvum* oocyst and microsphere removal efficiencies. The box plots depict removal data in five metrics: the smallest observation (or sample minimum), lower quartile, median, upper quartile, and largest observation (or sample maximum). Removal of

microspheres through each filtration process was expressed either as percent removal or in terms of the logarithmic reductions (base 10). Log<sub>10</sub> removals are presented as the log<sub>10</sub> of the ratio of the influent and effluent concentrations. Log removals that incorporated non-detects (i.e., no particles detected in filtrate) are prefixed with the '>' symbol and based on a calculated value that assumed one particle was present in the sample volume analyzed. A two-tailed t-test was performed to compare removals of *C. parvum* oocysts and microspheres before and after flow interruption (Microsoft Excel 2010, Redmond, WA, USA). The t-test significance level applied was α = 0.05. In addition, analysis of variance (ANOVA) was performed to compare microsphere removals by different amounts of DE (Microsoft Excel).

## RESULTS AND DISCUSSION

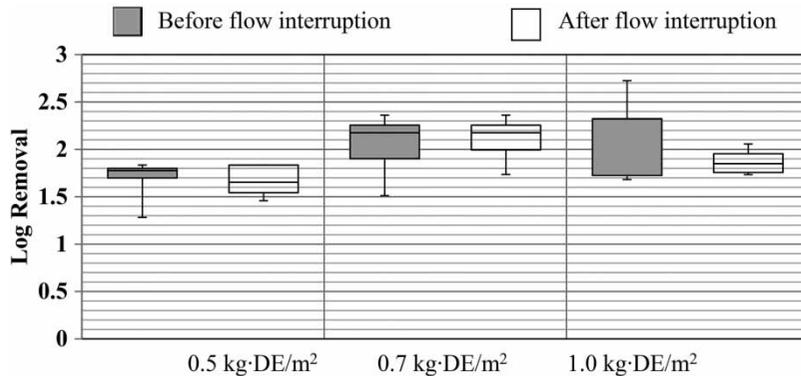
### Pilot-scale results

#### DE

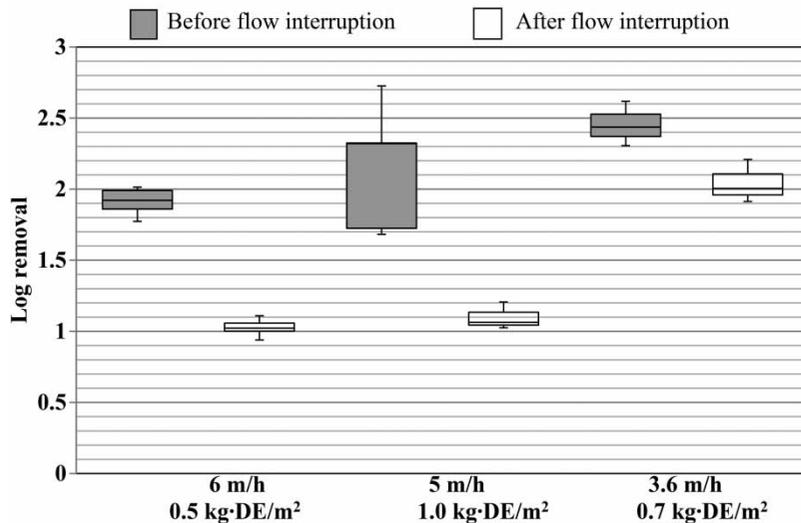
Figure 3 shows microsphere removals (before and after flow interruption) with a 5 m/h filtration rate and different amounts of precoat (DE). Removals of microspheres from filters operated at 5 m/h averaged 1.7 ± 0.18 log (97.0% to 98.7%) for 0.5 kg-DE/m<sup>2</sup>, 2.1 ± 0.16 log (98.9% to 99.5%) for 0.7 kg DE /m<sup>2</sup>, and 2.2 ± 0.39 log (98.5% to 99.7%) for 1.0 kg DE /m<sup>2</sup> prior to flow interruption. ANOVA results show that the removal with 0.5 kg DE /m<sup>2</sup> was significantly lower than the other DE amounts.

Removals of microspheres after flow interruption with a 5-minute stop were 1.7 ± 0.16 log (97.1% to 98.6%) for 0.5 kg DE/m<sup>2</sup>, 2.1 ± 0.21 log (98.7% to 99.5%) for 0.7 kg DE/m<sup>2</sup>, and 1.8 ± 0.17 log (97.7% to 98.9%) for 1.0 kg DE/m<sup>2</sup>. No significant differences were observed between the removals before and after flow interruption at a filtration rate of 5 m/h with a 5-minute interruption of the filter flow for the different amounts of precoat as determined by a paired t-test ( $p = 0.36$  for 0.5 kg DE/m<sup>2</sup>,  $p = 0.38$  for 0.7 kg DE/m<sup>2</sup>, and  $p = 0.06$  for 1.0 kg DE/m<sup>2</sup>).

Figure 4 shows removals before and after 15-minute flow interruption and different amounts of precoat at varying filtration rates. Significant differences in microsphere



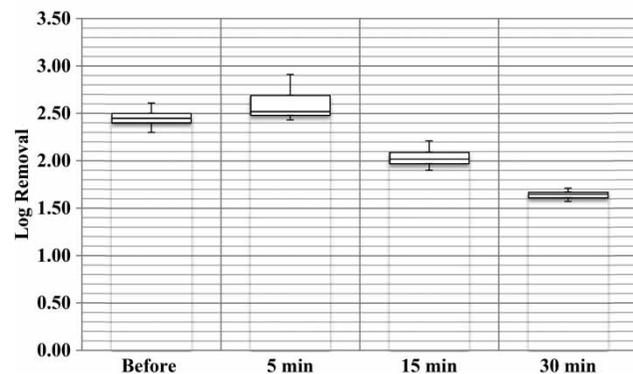
**Figure 3** | Microsphere removals by DE filtration before and after flow interruption with different amounts of precoat, a 5-minute filter stop, 5 m/h; duplicate pilot-scale experiments with triplicate samples.



**Figure 4** | Microsphere removals by DE filtration before and after filter flow interruption with different amounts of precoat and different filtration rates of 6 m/h, 5 m/h, and 3.6 m/h, and a 15-minute filter stop; duplicate pilot-scale experiments with triplicate samples.

removals were obtained before and after 15-minute flow interruptions at filtration rates of 3.6 m/h, 5 m/h, and 6 m/h ( $p < 0.01$ ).

Figure 5 shows microsphere removals before and after flow interruption with different time intervals using 0.7 kg DE/m<sup>2</sup> DE at 3.6 m/h. Microspheres were seeded in four consecutive cycles in this experiment without replacing the media. The number of microsphere seeding cycles might have impacted DE filter performance and will be discussed later. Removals were  $2.5 \pm 0.15$  log (99.6% to 99.8%) before flow interruption and were  $2.6 \pm 0.28$  log (99.5% to 99.7%) after a 5-minute flow interruption. Microsphere removals decreased to  $2 \pm 0.15$  log (98.6% to 99.3%) after



**Figure 5** | Microsphere removals by DE filtration before and after filter flow interruption (5-minute, 15-minute, and 30-minute), 0.7 kg DE/m<sup>2</sup>, filtration rate of 3.6 m/h; duplicate pilot-scale experiments with triplicate samples.

a 15-minute flow interruption and decreased to  $1.6 \pm 0.06$  log (97.1% to 97.8%) after a 30-minute flow interruption. A t-test indicates removals were significantly decreased after 15-minute and a 30-minute flow interruption ( $p = 0.01$  for 15-minute stop and  $p < 0.0001$  for 30-minute stop). These results indicate that the longer flow interruption times were associated with lower removals for the same filtration rate and amount of precoat media.

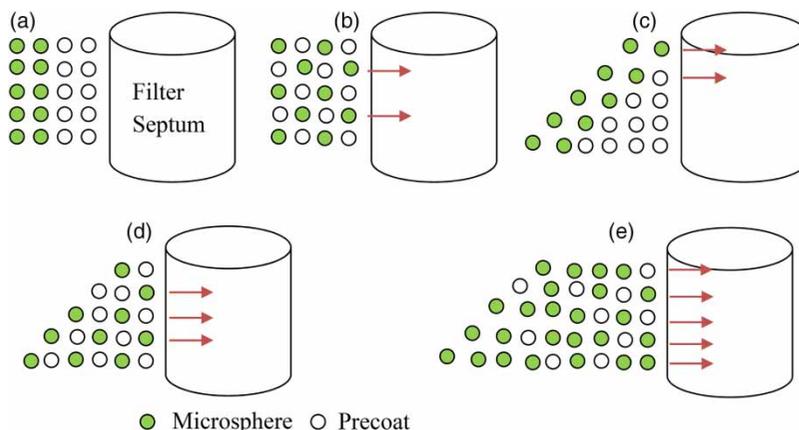
Several conceptual distributions of precoat media and microspheres before and after flow interruption are shown in Figure 6. The precoat is shown as evenly distributed on the filter septum before flow interruption and was recoated after flow interruption. DE filter performance was potentially influenced by all of these contributions. Microspheres are removed by surface blockage as shown in Figure 6(a). Precoat and microsphere mixtures led to microspheres passing through the filter septum more easily as shown in Figure 6(b). Uneven coating of precoat can occur after flow interruption as shown in Figure 6(c), which allows microspheres to pass through DE filters in locations with less or no precoat on the filter septum. Precoat and microsphere mixture as well as uneven coating combined to reduce filter performance as shown in Figure 6(d). Finally, the number of microspheres seeded in multiple flow interruption cycles prior to each round of sample collection impacted DE filter performance by resulting in higher microsphere concentrations in the filter relative to the amount of precoat as shown in Figure 6(e). DE filters and regenerative media filters that use either DE

or perlite could be impacted by the processes shown in Figure 6 each time flow is interrupted through the filter.

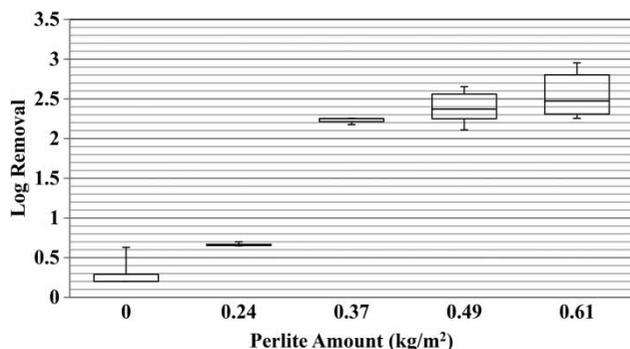
### Perlite-sand

Microsphere ( $4.5 \mu\text{m}$ ) removals by a sand filter were 0.1–0.4 log (20% to 63%) without perlite. When perlite was added at  $0.24 \text{ kg/m}^2$ ,  $0.37 \text{ kg/m}^2$ ,  $0.49 \text{ kg/m}^2$ , and  $0.61 \text{ kg/m}^2$  onto the surface of the sand media, microsphere removals increased significantly (Figure 7). Perlite depth was approximately 0.30 cm, 0.45 cm, 0.60 cm, and 0.70 cm for  $0.24 \text{ kg/m}^2$ ,  $0.37 \text{ kg/m}^2$ ,  $0.49 \text{ kg/m}^2$ , and  $0.61 \text{ kg/m}^2$ , respectively. Microsphere removals were equal to or greater than 2.2 log (99.4%) when the precoat amount was  $0.37 \text{ kg-perlite/m}^2$  or greater. Similar results were previously reported where 1.7–3 log (98% to 99.9%) of  $5 \mu\text{m}$  microspheres were removed by a perlite-sand filter with  $1.2 \text{ kg-perlite/m}^2$  in a 757 L (200 gal) swimming pool with a filtration rate of 37 m/h (Amburgey 2011).

Perlite-sand filtration provided roughly a 2 log improvement in the removal of microspheres as compared to the high-rate sand filtration control. The increased removal could be attributed to the small size of perlite (and the correspondingly small pores). The effective size of the sand was more than 28 times larger than that of the perlite. The removal of microspheres in a precoat media filter occurs by straining through the pores in the filter bed, or by sedimentation of microspheres in the media pores (Letterman & Yiacoumi 2010).



**Figure 6** | Distribution of precoat and microsphere before and after DE filter flow interruption: (a) before flow interruption, (b) after flow interruption – mixture of precoat and microsphere, (c) after flow interruption – uneven coating, (d) after flow interruption – combination of (b) and (c), (e) after flow interruption – three microsphere seeding cycles.



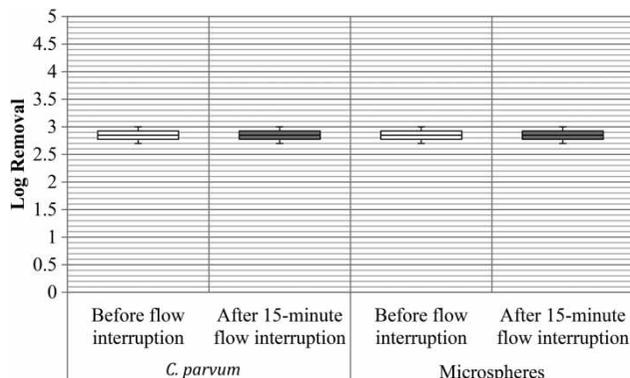
**Figure 7** | Microsphere removals by perlite-sand filter (1.8 #/mL microspheres, 30 cm sand, 37 m/h filtration rate; duplicate pilot-scale experiments with triplicate samples).

The initial pressure loss was 20 kPa for the sand-only filter prior to perlite addition, but the pressure increased as the amount of perlite increased. Initial filter influent pressures for the 0.24 kg-perlite/m<sup>2</sup>, 0.37 kg-perlite/m<sup>2</sup>, 0.49 kg-perlite/m<sup>2</sup>, and 0.61 kg-perlite/m<sup>2</sup> conditions at a filtration rate of 37 m/h, were 55 kPa, 55 kPa, 62 kPa, and 69 kPa, respectively.

## Full-scale results

### DE

Figure 8 displays the full-scale microsphere and *C. parvum* oocyst and removals by DE filtration (0.7 kg DE/m<sup>2</sup>, and filtration rate of 3.6 m/h) before flow interruption and after



**Figure 8** | *C. parvum* and microsphere removals through DE filtration during 5 days, 0.7 kg DE/m<sup>2</sup>, 3.6 m/h filtration rate (white boxes for removals by DE before flow interruption; gray boxes – removals after flow interruption with a 15-minute filter stop), full-scale study.

flow interruption times. When the DE filter flow was not interrupted or interrupted with a 15-minute stop, *C. parvum* and microspheres were removed by an average of more than 2.7 log (99.8%), similar to the pilot-scale results which averaged 2.5 log (99.7%) for the same amount of pre-coat and filtration rate of 3.6 m/h.

Removals after flow interruption were not significantly decreased by 15-minute stop of the DE filter at a filtration rate of 3.6 m/h (t-test  $p=3.9$ ). Removals obtained from full-scale filters after flow interruption were slightly higher than those at the pilot-scale.

### Perlite-sand

Microsphere removals by the full-scale perlite-sand filter (having a thin perlite layer overlying the sand) were 2.7 log (99.8%) (data not shown), compared with 2.4 log (99.7%) in pilot-scale trials at 0.5 kg-perlite/m<sup>2</sup>. Replicate samples were lost due to an unplanned backwash. Data for *C. parvum* oocyst removal could not be collected because oocysts were mistakenly seeded at a level that was too low (resulting in bulk water concentration <0.001 oocysts/mL). The full-scale pool could not be studied further, so only microsphere data were collected for the perlite-sand filter.

## CONCLUSION

The results indicate that sand filters are relatively ineffective for *C. parvum* oocyst and microsphere removal from swimming pools, with removals in the range of 0.1–0.4 log (20% to 63%). The DE precoat filter and perlite-sand filter improved removals significantly. At least 0.7 kg DE/m<sup>2</sup> was required to achieve 2 log (99%) of microsphere removals at 5 m/h indicated by pilot-scale studies. When DE filtration was performed with 0.7 kg DE/m<sup>2</sup> and a filtration rate of 3.6 m/h, greater than 2.7 log (99.8%) of *C. parvum* oocysts and microspheres were removed from full-scale swimming pool water. Pilot-scale results for DE filters showed filter restarts with flow interruption times of 15 minutes led to decreased removals of *C. parvum* and microspheres at the same filtration rate, but this effect was not observed for the full-scale pool filter.

Adding a layer of perlite on the top of the sand filter increased microsphere removal compared with a sand filter only. The pilot-scale results showed average microsphere removals in the pilot study were 2.3 log (99.4%), 2.4 log (99.7%), and 2.5 log (99.8%) for 0.37 kg-perlite/m<sup>2</sup>, 0.49 kg-perlite/m<sup>2</sup>, and 0.61 kg-perlite/m<sup>2</sup>, respectively. Filter influent pressure increased since the perlite was captured at the surface of sand in the filter. Perlite-sand filtration (0.5 kg-perlite/m<sup>2</sup>) in the full-scale study removed a similar level of microspheres [2.7 log (99.8%)].

The data from this study demonstrate that under the tested conditions the removal of *Cryptosporidium*-sized microspheres and *C. parvum* oocysts from swimming pool water can be significantly increased above typical sand filter levels through the use of a DE precoat filter or by applying a perlite layer of at least 0.37 kg-perlite/m<sup>2</sup> to a sand filter. Pool operators and pool water treatment engineers may want to consider using DE or perlite to improve the performance of pool filters for removing *Cryptosporidium* oocysts and other particulates.

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## REFERENCES

- Amburgey, J. E. 2011 Removal of *Cryptosporidium*-sized polystyrene microspheres from swimming pool water with a sand filter with and without added perlite media. *Journal of Environmental Engineering* **137** (2), 1205–1208.
- Amburgey, J. E., Walsh, K. J., Fielding, R. R. & Arrowood, M. J. 2012 Removal of *Cryptosporidium* and polystyrene microspheres from swimming pool water with sand, cartridge, and precoat filters. *Journal of Water and Health* **10** (1), 31–42.
- APHA 2005 *Standard Methods for the Examination of Water and Wastewater*, 21st edn. American Public Health Association/American Water Works Association/Water Environment Federation, Washington, DC, USA.
- Cantey, P. T., Kurian, A. K., Jefferson, D., Moerbe, M. M., Marshall, K., Blankenship, W. R., Rothbarth, G. R., Hwang, J., Hall, R., Yoder, J., Brunkard, J., Johnston, S., Xiao, L., Hill, V. R., Sarisky, J., Zarate, M. A., Otto, C. & Hlavsa, M. C. 2012 Outbreak of *Cryptosporidiosis* associated with a man-made chlorinated lake – Tarrant County, Texas, 2008. *Journal of Environmental Health* **74** (4), 14–19.
- CDC 2012 *Promotion of Health Swimming after a Statewide Outbreak of Cryptosporidiosis Associated with Recreational Water Venues – Utah, 2008–2009*. Centers for Disease Control and Prevention, Atlanta, GA, USA. Available at: <http://www.cdc.gov/mmwr/preview/mmwrhtml/mm6119a5.htm>.
- Hlavsa, M. C., Roberts, V. A., Anderson, A. R., Hill, V. R., Kahler, A. M., Orr, M., Garrison, L. E., Hicks, L. A., Newton, A., Hilborn, E. D., Wade, T. J., Beach, M. J. & Yoder, J. S. 2011 Surveillance for waterborne disease outbreaks and other health events associated with recreational water – United States, 2007–2008. *Morbidity and Mortality Weekly Report* **60** (SS-12), 1–37.
- Joce, R. E., Bruce, J., Kiely, D., Noah, N. D., Dempster, W. B., Stalker, R., Gumsley, P., Chapman, P. A., Norman, P., Watkins, J., Smith, H. V., Price, T. J. & Watts, D. 1991 An outbreak of cryptosporidiosis associated with a swimming pool. *Epidemiology and Infection* **107** (3), 497–508.
- Karanis, P., Sotiriadou, I., Kartashev, V., Kourenti, C., Tsvetkova, N. & Stojanova, K. 2006 Occurrence of *Giardia* and *Cryptosporidium* in water supplies of Russia and Bulgaria. *Environmental Research* **102** (3), 260–271.
- Letterman, R. D. & Yiacoymi, S. 2010 *Coagulation and Flocculation. Coagulation and Flocculation in Water Quality and Treatment: A Handbook on Drinking Water*. McGraw-Hill Professional, New York, USA.
- Lisle, J. T. & Rose, J. B. 1995 *Cryptosporidium* contamination of water in the USA and UK: a mini review. *Journal of Water Supply: Research and Technology* **44** (3), 103–117.
- Lu, P. & Amburgey, J. E. 2016 A pilot-scale study of *Cryptosporidium*-sized microsphere removals from swimming pools via sand filtration. *Journal of Water and Health* **14** (1), 109–120.
- Lu, P., Yuan, T., Feng, Q., Xu, A. & Li, J. 2013 Review of swimming-associated *Cryptosporidiosis* and *Cryptosporidium* oocysts removals from swimming pools. *Water Quality Research Journal of Canada* **48** (1), 30–39.
- McAnulty, J. M., Fleming, D. W. & Gonzalez, A. H. 1994 A community-wide outbreak of cryptosporidiosis associated with swimming at a wave pool. *Journal of the American Medical Association* **272** (20), 1597–1600.

- Puech, M. C., McAnulty, J. M., Lesjak, M., Shaw, N., Heron, L. & Watson, J. M. 2001 A statewide outbreak of cryptosporidiosis in New South Wales associated with swimming at public pools. *Epidemiology and Infection* **126** (2), 389–396.
- Rolfs, R. T., Beach, M. J., Hlavsa, M. C. & Calanan, R. M. 2008 Communitywide cryptosporidiosis outbreak – Utah, 2007. *Journal of the American Medical Association* **300** (15), 1754–1756.
- Shields, J. M., Hill, V. R., Arrowood, M. J. & Beach, M. J. 2008 Inactivation of *Cryptosporidium parvum* under chlorinated recreational water conditions. *Journal of Water and Health* **6** (4), 513–520.
- USEPA 1999 *Method 1623: Cryptosporidium and Giardia in Water by Filtration/IMS/FA*. EPA-821-R-99-006. Office of Water, US Environmental Protection Agency, Washington, DC, USA.

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