Pond walls: inclined planes to improve pathogen removal in pond systems for wastewater treatment?
A. L. Hawley and H. J. Fallowfield

ABSTRACT
Attenuation of sunlight in wastewater treatment ponds reduces the depth of the water exposed to disinfecting irradiances. Shallow pond depth with paddlewheel rotation increases exposure of pathogens to sunlight in high rate algal ponds. Generation of thin films, using pond walls as inclined planes, may increase inactivation of pathogens by increasing sunlight exposure. The performance of a laboratory based model system incorporating an inclined plane (IP) was evaluated. F-RNA bacteriophage, in tap water or wastewater, was exposed to sunlight only on the IP with the bulk water incubated in the dark. MS2 inactivation was significantly higher when the IP was present (P < 0.05) with a 63% increase observed. Prolonged exposure increased MS2 die-off irrespective of IP presence. Versatility of the IP was also demonstrated with faster inactivation observed in both optically clear tap water and wastewaters. IPs of different surface areas produced similar inactivation rates when operated at similar hydraulic loading rates regardless of slope length.

Key words | dark inactivation, high rate algal ponds, inclined plane, pathogen removal, solar exposure

INTRODUCTION
Wastewater treatment ponds utilise the germicidal properties of sunlight for the disinfection of pathogens (Clancy et al. 2000). Ultraviolet (UV) and visible (Vis) light when absorbed, directly (photo-inactivation) or indirectly (photo-oxidation), cause the genetic material or membranes of the organism to be damaged (Muela et al. 2002). In wastewater, however, attenuation reduces the depth of light penetration through the water column, particularly of the more germicidal, shorter wavelength UVB spectrum, (Caslake et al. 2004). This light decay is more prominent in turbid waters, and increases with pond depth (Kirk 1994; Fallowfield et al. 1996). In turbid water, the majority of the light involved in inactivation is absorbed in the first 1 m of water (Haag & Hoigne 1986) and UVB in the first 0.03 m (Kohn & Nelson 2007). More specifically, Bolton (2012) identified the extinction depths of 0.03 m (UVB; 280–315 nm), 0.07 m (UVA; 315–400 nm) and 0.14 m (Vis; 400–700 nm), in waste stabilisation pond (WSP) effluent, respectively. For improved disinfection, it is essential that the availability of light within the water column is increased and the effects of attenuation reduced.

High rate algal ponds (HRAPs) are intentionally mixed, shallow treatment ponds (0.2–0.5 m) arranged in a raceway configuration (Park et al. 2011). The hydraulic retention time of these ponds is between 2 and 8 days (Shilton 2005). Increased exposure to sunlight is achieved in these ponds through large surface area to volume ratios and continual mixing, most commonly by paddlewheel (Fallowfield & Garrett 1985). Elevated removal rates are achieved in these ponds with disinfection up to six times faster than other WSP systems (Buchanan et al. 2011). The removal of helminth (El Hamouri et al. 1994), protozoa (Araki et al. 2001) and bacteria (El Hamouri et al. 1994; Fallowfield et al. 1996) have been reported with a focus on the reduction of faecal coliforms and Escherichia coli. The removal of enteric viruses is a priority to protect human health; however, limited information exists regarding the removal of these viruses and their bacteriophage surrogates. To help bridge the gap this study will focus on the removal of MS2, an F-RNA bacteriophage.

The existing pond walls surrounding both HRAPs and WSPs provide a natural inclined plane (IP) (45°), formed during construction. At present, these embankments serve no purpose other than to contain the pond water. However, they may provide an opportunity for a cost-effective means of increasing solar exposure, and consequently virus inactivation within the systems. Generation of a thin film of wastewater flowing down the pond wall will increase...
exposure to disinfecting wavelengths of light. The solar exposure (UV energy) experienced by an IP has been characterised. An increase of 3–4% was reported when the incline was 37° compared to a horizontal surface (Navntoft et al. 2012). A 10% increase was reported when the plane was in the direction of the sun and the incline equal to the degree of latitude (Iqbal 1985; Duffie & Beckman 2003).

The objective of this research was to establish if increasing the area available for solar exposure, through the addition of an IP and the generation of a thin film, improved the inactivation of the F-RNA coliphage MS2. To achieve this, the study examined and compared removal rates achieved in model HRAPs in the presence and absence of an IP.

**METHODS**

**Inclined planes (IP)**

IPs were constructed from black Perspex sheet (width 0.67 m) fixed to a steel frame base. Two sizes were used, a small IP (SIP; length 0.55 m, 0.37 m²) and a large IP (LIP; length 1.10 m, 0.75 m²). A valved manifold was attached to the top of the plane through which water was pumped (Aqua PRO AP950) and the flow rates controlled to generate the thin film on the plane. Figure 1 provides a schematic diagram of the IPs used.

**Model HRAP systems**

Model HRAPs were constructed from 100 L plastic vessels, filled to a depth of 0.30 m (87.0 L) with either optically clear (tap) water or wastewater collected from a treatment plant comprising an aerated lagoon and maturation pond (Mt Barker, South Australia). The bulk water in the plastic vessels was continuously mixed using aquarium pumps (Aqua One 102). In order to determine the inactivation potential of the IP the bulk water in the vessels was covered so that only the IP was exposed to sunlight. A similarly mixed and covered HRAP without an IP (HRAPd) was used for comparison (Figure 1). Operating conditions are presented in Table 1.

**Sample collection**

Triplicate 10 mL samples were collected in 1.5 h intervals over a 24 h period. For water analysis, 120 mL samples were collected every 3 hours with an additional 1 L collected prior to and at completion of each experimental run.

**Environmental conditions**

*In situ* water measurements were recorded at each collection interval. Parameters monitored included water temperature (YSI model 55, Xylem), pH (370 pH meter,
Inactivation rate constants (K) were determined using GlnaFiT; a Microsoft Excel add-in (Geeraerd et al. 2005). Statistical analyses were carried out using statistical software packages: R version 3.1.2 (Vienna, Austria) and SPSS version 20.0 (Armonk, NY). Normal distribution of data was determined from quantile-comparison (Q-Q) plots and Shapiro–Wilk normality tests. Statistical analyses included linear regression (multiple with stepwise regression), independent samples t-tests, one-way analysis of variance with Tukey’s post hoc comparison and Pearson’s product correlation. Statistical significance was inferred at $P < 0.05$.

**RESULTS AND DISCUSSION**

**MS2 inactivation with IP inclusion**

The global solar irradiation received during this experiment ranged from 26.0 to 28.2 MJ m$^{-2}$ with a mean of 26.9 ± 1.1 MJ m$^{-2}$. Water temperatures ranged from 17.4 to 32.6 $^\circ$C in the model systems with means of 26.0 ± 3.7 $^\circ$C (HRAP$_d$) and 25.9 ± 5.2 $^\circ$C (HRAP$_d$ + SIP). Elevated MS2 inactivation was achieved by the incorporation of the SIP (Table 2). The inclusion of the SIP resulted in an inactivation 1.6 times higher than the HRAP$_d$ operated in the absence of the IP. The difference between the mean LRV was statistically significant ($P < 0.05$).

Table 2 shows the inactivation rates obtained after 24 h incubation. The log$_{10}$ linear + tail model described by Geeraerd et al. (2000) was identified as the best representation of the data. Tailing of the data suggests a lag in die-off for the presence of a mixed or sub culture where one of the populations exhibits a greater resistance to disinfection (Bevilacqua et al. 2015). In Table 2, the $K_{\text{max}}$ values are derived from the log$_{10}$ linear + tail inactivation curves.

Since the bulk water of both HRAPs was in the dark, the elevated MS2 inactivation seen in the HRAP$_d$ + SIP can be attributed to the exposure of water to sunlight received...
whilst on the slope. The inclusion of the IP increased the LRV24 by 63% when compared with the HRAP$_d$.

A direct comparison could not be made with the current literature due to the novel nature of this research. Comparison was made with studies examining F-RNA phage inactivation in other pond systems, for example WSPs, under both dark and solar exposures. The values reported here were higher than the inactivation rates reported by Sinton et al. (2002), who examined the inactivation of F-RNA phage in WSP effluent exposed to sunlight (summer; 0.070 h/$C_0$, winter; 0.050 h/$C_0$) and in the absence of light (0.014 h/$C_0$).

The role of hydraulic loading rate

Further experiments comparing the performance of SIP and LIP at different flow rates which maintained different but constant hydraulic loading rate (HLR) on the two IPs within each experiment resulted in MS2 LRV$_{49.5}$ that were statistically similar ($P > 0.05$), an observation consistent across the four HLRS examined (Figure 2). The similar inactivation rates for slopes of different lengths and surface areas operated over a range of HLRS suggest that HLR is the factor which influences inactivation by the IP operated under the same climatic conditions. The higher inactivation exhibited for HLR 1 (Figure 2) was attributed to the higher solar irradiance received throughout the experimental period. Corresponding solar irradiances for the HLR experiments were 18.5 ± 4.3 (HLR 1), 8.9 ± 10.7 (HLR 2), 4.4 ± 3.5 (HLR 3) and 5.5 ± 6.2 MJ m$^{-2}$ (HLR 4).

Incubation time

Solar and dark disinfection are affected by the duration of incubation and exposure to sunlight (Kirk 1994). Figure 3 shows that inactivation was related to incubation time both in the dark-incubated HRAP$_d$ and in the HRAP$_d$ + IP exposed to sunlight. The LRV$_{5d}$ were 1.882 ± 0.282, 2.852 ± 0.627 and 3.046 ± 0.322 for the HRAP, HRAP$_d$ + SIP and HRAP$_d$ + LIP, respectively.

In situ water conditions

Inactivation can vary depending on different environmental parameters. Higher pH, DO and water temperature were identified in the HRAP$_d$ + LIP (pH 7.73 ± 0.42, 6.08 ± 2.01 mg DO L$^{-1}$, 21.9 ± 6.0 °C) and HRAP$_d$ + SIP (pH

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**Table 2** | MS2 inactivation in model systems: log$_{10}$ reduction values (LRV) and inactivation rate constant ($K_{\text{max}}$ mean ± standard deviation for HRAP$_d$ and HRAP$_d$ + SIP, after 24.0 h incubation

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>System</th>
<th>n</th>
<th>LRV</th>
<th>$K_{\text{max}}$</th>
<th>$R^2$</th>
<th>$P$-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>24.0</td>
<td>HRAP$_d$</td>
<td>21</td>
<td>1.445</td>
<td>0.240 ± 0.067</td>
<td>0.7795</td>
<td>2.2 × 10$^{-12}$</td>
</tr>
<tr>
<td></td>
<td>HRAP$_d$ + SIP</td>
<td>21</td>
<td>2.354</td>
<td>0.297 ± 0.078</td>
<td>0.9110</td>
<td>2.0 × 10$^{-12}$</td>
</tr>
</tbody>
</table>

$R^2$ and $P$-values relate to the strength of the statistics associated with the determination of $K_{\text{max}}$. 

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**Figure 2** | MS2 log$_{10}$ reduction values (mean ± standard deviation) obtained after an incubation time of 49.5 h for small (♦) and large (▴) IP operated at different HLRS. Mean HLRS were HLR 1, 350.3 L m$^{-2}$ h$^{-1}$; HLR 2, 171.1 L m$^{-2}$ h$^{-1}$; HLR 3, 232.75 L m$^{-2}$ h$^{-1}$ and HLR 4, 300 L m$^{-2}$ h$^{-1}$.

**Figure 3** | The relationship between LRV (mean ± 1 standard deviation) and the incubation time for the dark-incubated HRAP$_d$ (♦), HRAP$_d$ + SIP (▴) and HRAP$_d$ + LIP (▴). The inclined planes were operated at an HLR of 300.0 L m$^{-2}$ h$^{-1}$.
7.64 ± 0.41, 6.58 ± 2.11 mg DO L⁻¹, 20.7 ± 5.9 °C) compared to the HRAPd (pH 7.51 ± 0.43, 5.51 ± 2.26 mg DO L⁻¹, 20.4 ± 6.2 °C). However, pH was the only parameter identified as being significantly higher in the HRAPd + IP. This is probably a response to the elevated chlorophyll $a$ levels identified in the systems incorporating IPs. The chl $a$ levels were 0.76 ± 0.53 (HRAPd + LIP), 0.71 ± 0.38 (HRAPd + SIP) and 0.49 ± 0.21 mg L⁻¹ (HRAPd), respectively.

Summer incubations exhibited greater MS2 inactivation (Figure 4) indicative of the effect of higher solar irradiances received throughout summer. During the experimental period the mean summer irradiance, 27.9 ± 1.1 MJ m⁻², was 2.6 and 5.5 times higher than the mean irradiance received during autumn (11.2 ± 8.1 MJ m⁻²) and winter (9.9 ± 0.0 MJ m⁻²), respectively.

These results support those of Sinton et al. (1999) who identified slower inactivation of F-RNA phage throughout winter (both in sunlight and the dark).

Effect of water type

Figure 5 shows the variation of MS2 inactivation in clear (tap) waters and turbid wastewater. Inactivation was found to be 0.358 log₁₀ h⁻¹ slower in wastewater (0.186 ± 0.019 log₁₀ h⁻¹) than in tap water (0.544 ± 0.214 log₁₀ h⁻¹), suggesting inactivation efficiency was affected by water type. High turbidity and the presence of algae and particulate matter were probably responsible for the slower inactivation rates, with all capable of affecting light dispersion in the water column (Curtis et al. 1994). Davies-Colley et al. (1999) and Kohn & Nelson (2007) also reported lower F-RNA phage inactivation in WSP effluent than in reverse osmosis water. Inclusion of the IP continued to produce elevated inactivation in wastewater and tap water, with the HRAPd + SIP yielding an inactivation 1.5 and 1.2 times higher than the tap water and wastewater HRAPd, respectively.

CONCLUSION

It is clear from the results that pathogen inactivation can be improved by the inclusion of an IP to a model HRAP. Inactivation increased by 63% following inclusion of the IP compared to the dark-incubated pond. HLR was identified as an important factor influencing inactivation rates. The improvement in inactivation rates following incorporation of an IP was less for turbid wastewaters compared to operation with optically clear tap waters. Additional work is still required with the aim of transferring the concept to a fully functioning field system. It would be beneficial to gain an understanding of the inactivation efficiency when both pond and IP are solar exposed and exposed to different seasonal variations. Multiple disciplines within the water industry would benefit from this research with higher quality effluent produced, safer for reuse via the management of the risk associated with exposure to pathogens.

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REFERENCES


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