Using ultraviolet light for disinfection of finished water

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Abstract Ultraviolet light is now recognised to be very effective for inactivation of Cryptosporidium parvum oocysts; however, its application for disinfection of finished water necessitates validation of UV reactors prior to their installation. Although reactor performance will likely be assessed using non-pathogenic microorganisms as biodosimetry surrogates, it would be prudent for the water industry to simultaneously measure Cryptosporidium oocysts inactivation in controlled bench-scale studies using the water matrix intended for disinfection. The likelihood of that occurring is dependent upon the availability of infectivity measurement procedures that are more user-friendly than the mouse infectivity assays currently used. This study describes a modified cell culture procedure that would enable reliable measurement of changes in oocysts’ infectivity following their UV treatment. Also, a number of different biodosimetry surrogates were examined and one selected for comparing the UV doses delivered between bench-scale and full-scale biodosimetry studies. Impacts of UV disinfection on production of disinfection byproducts, effects of lamp ageing on effectiveness of disinfection and the costs associated with employing this technology were also examined.

Keywords Cryptosporidium; detection; infectivity; ultraviolet

Introduction

Recently the use of low levels of medium- and low-pressure ultraviolet light for successful inactivation of Cryptosporidium parvum oocysts has generated tremendous excitement in the water industry. While these findings have been corroborated by numerous independent studies at the bench-scale level, there is little experience in the US with full-scale disinfection of finished water using UV light. Concerns exist with respect to the reliability of UV technology and, in addition to biodosimetry experiments to validate reactor performance for organism inactivation, additional issues include (a) determination of the performance efficiency of the reactors, (b) effects of suspended metals or other water characteristics on lamp sleeve fouling, (c) effectiveness and reliability of lamp cleaning mechanisms, (d) stability of UV measurement sensors, (e) costs associated with retrofitting UV systems into existing water treatment plants, (f) operation and maintenance costs associated with employment of UV disinfection, (g) impact of lamp ageing on delivery of target UV doses and (h) impact of UV on disinfection byproduct (DBPs) formation or degeneration.

Although it is anticipated that experience with UV disinfection will provide information on some of these issues, certain parameters need to be investigated before water utilities can commit to using UV disinfection as one of the multiple barriers for protection of public health from waterborne disease-causing organisms. To address some of these operational issues, American Water, the largest investor-owned water utility in the US, has been actively involved in ascertaining the long-term feasibility of applying UV for treatment of finished water. A 12-inch (0.3 m) diameter 4 × 1 KW closed chamber UV reactor was installed after granular activated carbon filtration at the Pennsylvania American Water treatment plant at Hayes Mine and was operated continuously (finished water flow rate of 600 gpm). Over a 12-month period, various chemical (THM, HAA, UV254, DOC, TOC, metals, nitrate, nitrites) and physical measurements (lamp voltage, current, sensor measurements)
were monitored. Also, parameters such as power consumption and other operational issues associated with reactor function (i.e. frequency of power outages, failing components, etc.) were recorded. Reactor validation studies were performed using MS2 bacteriophage as a biodosimetry surrogate and utilised various lamp configurations as well as lamp age. These inactivation data in the pilot unit were compared with controlled bench-scale inactivation data to ascertain the effectiveness of full-scale disinfection using UV light.

**Materials and methods**

*Cryptosporidium parvum oocysts*

*C. parvum* oocysts (Iowa isolate) were used to examine inactivation levels at extremely low UV doses delivered by the collimated beam apparatus. The infectivity of UV-treated oocysts was determined with a cell culture–immunofluorescence (CC–IFA) based infectivity assay that was developed and optimised at American Water.

**Surrogate organisms**

*Bacillus subtilis* (ATCC 6633), MS2 bacteriophage (ATCC 15597-B1) and *Deinococcus radiodurans* (ATCC 13939) were propagated according to standard procedures and enumerated by using modified starch agar plates, double agar layer method (US-EPA Method 1602) and nutrient broth containing 1% glucose respectively.

**Bench-scale UV disinfection experiments**

A Rayox UV collimated beam unit, supplied by the Calgon Carbon Corporation and housing a medium-pressure UV lamp (1 kW), was used to conduct bench-scale inactivation experiments to develop dose-response curves.

**Full-scale biodosimetry trials**

A Sentinel™ UV reactor (12 inches, 0.3 m, diameter and 40 inches (1 m) in length) was supplied by Calgon Carbon Corporation and consisted of 4 × 1 kW medium pressure lamps. Accompanying the reactor was an electrical control cabinet (36 × 12 × 72 inches, 0.9 × 0.3 × 1.8 m), which controlled reactor function as well as monitoring various operational parameters. This UV reactor was capable of treating up to 700 gpm of water and contained four sensors (one for each lamp) for determining UV irradiance, as well as sensors for temperature, flow and pressure. Prior to installation of the UV reactor, a minor-permit of approval was obtained from the Pennsylvania Department of Environmental Protection. The UV reactor was installed on a dedicated filter at the Hays Mine water treatment plant of the Pennsylvania American Water Company.

**Results**

**Bench-scale dose-response curves for C. parvum oocysts**

Application of a UV dose of 1 mJ/cm² rendered 0.44-log inactivation of oocysts and delivery of UV doses between 1 mJ/cm² and 4 mJ/cm² yielded a linear decline in oocyst inactivation with 3 mJ/cm² rendering 2.79–2.84 logs inactivation and 4-log inactivation occurring at 4 mJ/cm². Between 4 mJ/cm² and 20 mJ/cm², measurements with the cell culture procedure continued to indicate that oocyst inactivation levels remained at 4-logs. It was likely that the actual levels of inactivation at UV doses between 4 mJ/cm² and 20 mJ/cm² were higher than 4-logs; however, as the level of oocyst inactivation measurable by the cell culture procedure was dependent upon the highest original inoculum of UV-treated oocysts (i.e. 1 × 10⁵ oocysts), the maximum levels of measurable inactivation with this cell culture procedure were limited to 4-logs or lower (Figure 1).
Bench-scale dose-response curves for surrogate organisms
The collimated beam apparatus was used to perform bench-scale dose-response curves for three surrogate organisms (MS2 bacteriophage, *Bacillus subtilis* and *Deinococcus radiodurans*). Five separate trials were performed for each surrogate organism at three different UV doses (10 mJ/cm², 20 mJ/cm² and 40 mJ/cm²). Additionally, for two surrogates (MS2 bacteriophage and *Bacillus subtilis*), triplicate and duplicate trials respectively were performed at 80 mJ/cm². For each disinfection trial, the inactivation data for these various UV doses have been presented in Figure 2.

The rate of inactivation with increasing UV doses was considerably higher for both *B. subtilis* spores and MS2 bacteriophage than *Deinococcus*. In replicate experiments at a given dose, higher inter-trial variability was noted for both *Deinococcus* and *B. subtilis* spores (data not shown).

Comparison of bench- and full-scale disinfection studies
For two surrogate organisms (*B. subtilis* spores and MS2 bacteriophage) an excellent agreement was observed between bench- and full-scale inactivation data at a target UV dose of 40 mJ/cm²; however, when the UV dose was reduced to either 20 mJ/cm² or 10 mJ/cm², an increasing disparity between the bench- and full-scale data was observed. Nonetheless, the inactivation levels with the full-scale reactor were always higher than those predicted from the bench-scale data, indicating that the level of protection provided by the reactor was greater than anticipated. The associated increase in power consumption costs with this increased delivery of UV light will be discussed.
Discussion

During these investigations, it was noted that UV treated *C. parvum* oocysts could undergo excystation and the sporozoites from inactivated oocysts were able to invade monolayers of HCT-8 cells to generate pin-points of invasion. Occurrence of this phenomenon was likely to be the primary reason that a cell culture quantitative PCR procedure, which was developed previously within our laboratory (DiGiovanni et al., 1999), could not be used to measure inactivation of UV-treated oocysts, as this assay did not have the capacity to discriminate between DNA originating from invasive sporozoites and that of infectious sporozoites. Optimisation of excystation triggers and cell culture incubation periods led to development of a cell culture–IFA procedure that enabled detection of as few as 10 infectious oocysts. Using this optimised cell culture–IFA procedure, our data indicated that extremely low UV doses (i.e. 2–5 mJ/cm²) were very effective for inactivation of *C. parvum* oocysts. For UV doses where our data could be compared with previously published data, using either tissue culture infectivity or mouse infectivity assays, an excellent agreement was also observed. Numerous studies (Clancy et al., 2000; Craik et al., 2000; Landis et al., 2000; Shin et al., 2001) have assessed levels of oocyst inactivation delivering UV doses ranging between 1 mJ/cm² and <40 mJ/cm². Not surprisingly, there was wide variability associated with the data generated from these various studies that may be associated with differences in oocyst preparation, disinfection conditions and methods for infectivity measurements. Nonetheless, using UV doses between 1 mJ/cm² and 3 mJ/cm², the oocysts’ inactivation data generated in our current study were within the range of inactivation data generated in the previous studies. Our data provide further evidence to support the fact that UV light was very effective for oocyst inactivation, with extremely low UV doses generating levels of inactivation that were greater than those anticipated from previously published studies.

Bench-scale UV dose-response data indicated that MS2 bacteriophage was the most suitable surrogate organism for validating UV doses in full-scale reactors. The factors favouring the choice of this surrogate included the ease of propagation/analysis of MS2 bacteriophage, the near-linear response curve to increasing UV doses and, perhaps most importantly, the excellent inter-trial reproducibility. Although *B. subtilis* demonstrated greater sensitivity than MS2 bacteriophage to increasing doses of UV light, the inter-trial variability was greater than for MS2 bacteriophage. In our experience, lower inter-trial reproducibility coupled with the time-consuming nature of growing sufficient quantities of *B. subtilis* spores for full-scale studies rendered *B. subtilis* spores a less desirable surrogate than MS2 bacteriophage.

An excellent agreement between the full-scale and bench-scale data was noted for 40 mJ/cm², and 20 mJ/cm² also revealed a good agreement between the full-scale and bench-scale data. Full-scale data using a target dose of 10 mJ/cm² indicated approximately twice the levels of inactivation expected from the bench-scale studies. There may be several explanations for why this may have occurred.

(a) Firstly, a working stock of MS2 bacteriophage used for one of the trials at 40mJ/cm², one of the trials at 20 mJ/cm² and all the trials at 10 mJ/cm² was derived from the same stock preparation. All of these trials demonstrated higher than anticipated levels of inactivation. During these trials, a higher concentration of organisms than usual was used for the spiking studies. As organism inactivation can be a function of the original spike dose, perhaps the variation in the original spike dose accounted for higher levels of inactivation than anticipated. While the possibility also existed that this propagation of MS2 bacteriophage had a higher susceptibility than the other isolates used in both the collimated beam and full-scale studies, the likelihood of this occurring was low. This was based on the fact that numerous propagations were used to generate the bench-scale and full-scale data, with very little variability in the observed levels of inactivation.
(b) Secondly, only a single lamp at full power was used for a target dose of 10 mJ/cm² and it was likely that the actual dose delivered to the organisms was considerably higher than the target UV dose. Based on the bench-scale inactivation data of MS2 bacteriophage, the levels of inactivation achieved at a full-scale target dose of 10 mJ/cm² indicated that a dose of approximately 20 mJ/cm² may have actually been delivered. When one accounts for the geometric distribution of the UV dose in a reactor, it seemed reasonable that organisms in close proximity to the lamps may have received a higher total dose than those further away from the lamps. If this was an influencing factor, then, in the absence of good mixing within the reactor, biodosimetry experiments would have led to over-estimation of the delivered dose. Immediately prior to each series of UV disinfection trials, process control samples were spiked into the reactor with all the lamps off. This procedure served a dual purpose in verifying the extent of the mixing during biodosimetry experiments, which was excellent throughout this study, and also helped to account for the impact of the experimental manipulations that could be responsible for organism inactivation. In this manner the impact of UV disinfection during biodosimetry experiments was differentiated from that due to either poor mixing or other experimental handling procedures.

During the course of this study, the impact of delivering a UV dose of approximately 40 mJ/cm² on total haloacetic acids (HAA-5) and total trihalomethanes (THM) was also examined. On all sampling occasions, the HAA-5 concentrations in the influent to the reactor were significantly below the total MCL of 60 µg/L. Peaks were noted between October 2001 and March 2002, with influent concentrations of 3.2–9.9 µg/L and, during these peak events, their respective post-UV effluent concentrations were equal to or lower than the levels detected in the influent samples. Overall, on 5/13 sampling events, HAA-5 concentrations were greater in the influent than the effluent post-UV treatment, whereas 4/13 sampling events indicated higher concentrations in the effluent samples. On two sampling occasions, both the influent and effluent samples showed the same total HAA-5 values. The total HAA-5 values for both the influents and effluents were low, which led to the possibility that the differences between influent and effluent samples may have been associated with sampling variability. Nonetheless, it was highly probable that employment of UV disinfection at a target dose of 40 mJ/cm² did not contribute further to the generation of HAA-5. Similarly for THMs, the levels in the influent and effluent only indicated marginal differences, indicating further the minimal impacts of UV disinfection on generations of these DBPs.

Although biodosimetry experiments confirmed that UV lamps aged up to 5,800 h of operation remained effective for organism inactivation, in consideration of their increased power consumption, perhaps it would be prudent to change lamps with greater frequency. Currently the lamp manufacturer advocates a lamp operation time of 3,000 h; however, according to the data accrued in this study, approximately $10 per MG increase in power consumption occurred with lamps aged between 2,200 h and 3,600 h. Thus, operating lamps for the additional 800 h (~33 d) above 2,200 h (i.e. lamp age up to 3,000 h) would likely yield an increase in daily power consumption costs ($10/MG × 33 d) to $330. Clearly, factors such as the costs associated with purchase of replacement lamps and labour for their installation have to be accounted for in order to determine the optimal usage hours at which the lamps are replaced.

Conclusions

The following conclusions resulted from this study: (a) bench-scale studies using three surrogate organisms (MS2 bacteriophage, Bacillus subtilis and Deinococcus radiodurans), and various UV doses indicated that MS2 bacteriophage yielded highly reproducible inactivation levels compared to the other two surrogate organisms; (b) MS2 bacteriophage was
identified as the most suitable surrogate for measuring UV inactivation due to highly reproducible inactivation data, ease of propagation of large numbers of organisms and simple procedures for organism enumeration; (c) development of an optimised cell culture–immunofluorescence procedure led to more reliable measurement of Cryptosporidium oocyst inactivation following UV disinfection; (d) using a 12-inch (0.3 m) diameter UV reactor, biodosimetry experiments using the three surrogate organisms confirmed that the reactor accurately delivered a target dose of 40 mJ/cm²; and (e) where UV doses <40 mJ/cm² were targeted, there was significant disparity between the targeted UV doses and those predicted by biodosimetry – although there may be cost implications associated with this; nonetheless, the doses delivered by the reactor always exceeded the anticipated UV doses, indicating that the reactor provided greater than projected levels of protection when targeting UV doses <40 mJ/cm².

References


