Guidance for Implementing Action Spectra Correction With Medium Pressure UV Disinfection

Web Report #4376

Subject Area: Water Quality
Guidance for Implementing Action Spectra Correction With Medium Pressure UV Disinfection
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Guidance for Implementing Action Spectra Correction With Medium Pressure UV Disinfection

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6666 West Quincy Avenue, Denver, CO 80235-3098

U.S. Environmental Protection Agency
Washington, D.C.

and

American Water Works Association
Denver, CO

Published by:
Water Research Foundation
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This study was funded by the Water Research Foundation (WRF) and the U.S. Environmental Protection Agency (EPA) under Cooperative Agreement #EPA-EM-83484801-0, with additional funding from the American Water Works Association (AWWA). WRF, EPA, and AWWA assume no responsibility for the content of the research study reported in this publication or for the opinions or statements of fact expressed in the report. The mention of trade names for commercial products does not represent or imply the approval or endorsement of WRF, EPA, or AWWA. This report is presented solely for informational purposes.

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ISBN 978-1-60573-222-0

Printed in the U.S.A.
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FOREWORD

The Water Research Foundation (WRF) is a nonprofit corporation dedicated to the development and implementation of scientifically sound research designed to help drinking water utilities respond to regulatory requirements and address high-priority concerns. WRF’s research agenda is developed through a process of consultation with WRF subscribers and other drinking water professionals. WRF’s Board of Trustees and other professional volunteers help prioritize and select research projects for funding based upon current and future industry needs, applicability, and past work. WRF sponsors research projects through the Focus Area, Emerging Opportunities, and Tailored Collaboration programs, as well as various joint research efforts with organizations such as the U.S. Environmental Protection Agency and the U.S. Bureau of Reclamation.

This publication is a result of a research project fully funded or funded in part by WRF subscribers. WRF’s subscription program provides a cost-effective and collaborative method for funding research in the public interest. The research investment that underpins this report will intrinsically increase in value as the findings are applied in communities throughout the world. WRF research projects are managed closely from their inception to the final report by the staff and a large cadre of volunteers who willingly contribute their time and expertise. WRF provides planning, management, and technical oversight and awards contracts to other institutions such as water utilities, universities, and engineering firms to conduct the research.

A broad spectrum of water supply issues is addressed by WRF’s research agenda, including resources, treatment and operations, distribution and storage, water quality and analysis, toxicology, economics, and management. The ultimate purpose of the coordinated effort is to assist water suppliers to provide a reliable supply of safe and affordable drinking water to consumers. The true benefits of WRF’s research are realized when the results are implemented at the utility level. WRF's staff and Board of Trustees are pleased to offer this publication as a contribution toward that end.

Denise L. Kruger
Chair, Board of Trustees
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Robert C. Renner, P.E.
Executive Director
Water Research Foundation
ACKNOWLEDGMENTS

Authors would like to acknowledge the advice and guidance of the Project Advisory Committee (PAC) members that included Yann Le Gouellec (Greater Cincinnati Water Works), Mark Angles (Sydney Water), and Sam Hayes (EPA). Authors would also like to acknowledge the support of the Water Research Foundation project manager Hsiao-wen Chen. The action spectra research in this report would not have been possible without the support and expertise of Tom Hargy and Randi McCuin (Tetra Tech/Clancy Environmental) and Tom Larason (National Institute of Standards and Technology). Lastly, the authors would like to acknowledge the input from the American Water Works Association (AWWA) Action Spectra Working Group, made up of industry stakeholders of UV system manufacturers, utilities, regulators, academics, and consultants, including:

- Steve Via (AWWA)
- Brian Petri (Trojan Technologies Inc.)
- Keith Bircher (Calgon Carbon Corporation)
- Christian Bokermann (Xylem-WEDECO)
- Phyllis Posy (Atlantium Technologies Ltd)
- Oliver Lawal and Paul Ropic (Aquionics)
- Jon McClean and Patrick Bollman (ETS)
- Joel Ducoste (North Carolina State University)
- Jim Malley (University of New Hampshire)
- Chris Schulz and Kati Bell (CDM-Smith)
- Paul Swaim (CH2M Hill)
- Bryan Townsend (Black & Veatch)
- Tom Hargy (Tetra Tech)
- Chengyue Shen (HDR Inc.)
- Jeff Adams (EPA)
- Brian Bernados (California DPH)
- Paul Rochelle (MWDSC)
- Eva Nieminski (Utah DEQ)
- Sam Perry (Washington DOH)
- William Sullivan (MWRA)
- Kurt Wells (LADWP)
- Enio Sebastiani (San Francisco PUC)
EXECUTIVE SUMMARY

BACKGROUND

Recent research shows significant differences between the wavelength responses or action spectra of validation test microbes and regulated pathogens, namely Cryptosporidium, Giardia, and adenovirus. These differences impact the interpretation of UV validation data used to define UV dose monitoring algorithms and disinfection credit with UV systems using polychromatic MP UV lamps. While the U.S. Environmental Protection Agency’s (EPA) UV Disinfection Guidance Manual (UVDGM) (EPA 2006a) accounts for these differences, it does not account for the UV transmittance spectrum of the quartz sleeve housing the MP lamp, the UV absorbance spectra of the water passing through the reactor, or the configuration of the lamps within the UV reactor. As such, the UVDGM approach can be overly conservative with many commercial UV systems. The approach is also limited by the quality of the action spectra data in the peer-reviewed literature, which show a high degree of variability due to the methods used, and do not extend over the full germicidal range (from 200 to 300 nm). Understanding the action spectra over the full germicidal range is important for the calculation with many MP UV reactors.

OBJECTIVES

The objectives of this research were to:

1. Develop data on the UV dose-response and action spectra of adenovirus, Cryptosporidium, Giardia, and commonly used validation microbes using a tunable laser, thereby addressing issues with the methods used by previous studies.

2. Develop recommendations for calculating action spectra correction factors (ASCF) that account for differences in the action spectra of common validation test microbes and regulated pathogens, UV output from the MP lamp, UV transmittance of the quartz sleeve, UV absorbance spectra of the water, and the reactor’s lamp configuration.

3. Develop recommendations for applying ASCF for regulatory credit.

APPROACH

A tunable laser from the National Institute of Standards and Technology (NIST) was used to measure the action spectra from 200 to 290 nm in 10 nm increments of the regulated pathogens adenovirus, Cryptosporidium parvum, Giardia lamblia, and validation microbes MS2, T1UV, Q Beta, T7, and T7m Coliphage. The action spectra of Bacillus pumilus spores was measured using bandpass filters.

UV dose models based on computational fluid dynamics and UV intensity field (CFD-I) models, developed for three commercial UV reactors equipped with MP UV lamps, were used to predict reduction equivalent doses (REDs) for the target pathogens and validation microbes over the validated range of flows, UV transmittance at 254 nm (UVTs), and relative lamp output for those reactors. While the validation microbe REDs were calculated using the full action spectra, the target pathogen REDs were calculated assuming no action below 240 nm, because current UV sensor technology does not provide proper monitoring below 240 nm. With each reactor, the
calculations were repeated with three sleeve types (Type 219, 214, and synthetic quartz) and five validation water types, thereby varying the relative contribution of low wavelength UV dose delivery below 240 nm to the calculated RED. ASCF values were calculated as the RED predicted, using the validation microbe action spectrum divided by the RED predicted using the pathogen action spectrum. For a given reactor sleeve and validation water combination, ASCF values for a given validation microbe and regulated pathogen pair were modeled as a function of flow, UVT and lamp output. The models were used to predict ASCF values required for various levels of Cryptosporidium and adenovirus disinfection credit.

RESULTS/CONCLUSIONS

Figures ES.1 and ES.2 present the action spectra of the validation microbes and the pathogens Cryptosporidium and adenovirus. These data illustrate the differences between the action spectra at wavelengths above 254 nm and below 240 nm. Above 240 nm, all organisms tested exhibited a relative peak sensitivity between 260 and 270 nm. Of the coliphage, MS2 exhibited the highest relative sensitivity below 240 nm, relative to its sensitivity at 254 nm, followed by Q Beta, T1UV, T7m, and T7 coliphage. The pathogen adenovirus exhibited the highest UV sensitivity at wavelengths below 240 nm, higher than all the phages, but similar to B. pumilus. Giardia action spectrum is plotted in Figure 2.21 in comparison to Cryptosporidium. Above 240 nm, there is no statistical difference in the two action spectra.


Figure ES.1 Relative spectral sensitivity of MS2, T1UV, Q Beta, T7m, and T7 Coliphage and Cryptosporidium (C. parvum) to UV light from the tunable laser

ASCF Tables

ASCF tables for Cryptosporidium and adenovirus inactivation credit were developed for 0.5 to 4.0 log inactivation credit in 0.5 log increments using the reactor configuration that provided the more conservative values. The ASCF values are presented as a function of UVT from 70 to 98
percent in 5 percent increments for each combination of sleeve and validation water type modeled. Tables were also developed for adenovirus credit assuming that adenovirus was used during validation and for UV reactors used to deliver a specific MS2 RED. With both of these cases, the ASCF was calculated as the RED calculated using the full action spectrum divided by the RED calculated using the action spectrum set to zero below 240 nm.


Figure ES.2 Relative spectral sensitivity of *B. pumilus* spores and adenovirus to UV light

**ASCF Values for Cryptosporidium and Giardia Credit**

With sleeve and validation water types that maximize UV dose delivery below 240 nm, ASCF values for *Cryptosporidium* calculated using the full *Cryptosporidium* action spectra were notably less than ASCF values calculated with the *Cryptosporidium* action spectrum set to zero below 240 nm. Because UV sensors used with today’s commercial MP UV reactors do not monitor UV intensity at wavelengths below 240 nm, and because UV dose delivery below 240 nm realized during validation may not occur at the WTP because of lamp aging, sleeve fouling, or UV absorbance at wavelengths below 240 nm, this guidance recommends calculating ASCF values using the pathogen action spectrum set to zero at wavelengths below 240 nm, at least until low wavelength UV sensors are developed and proven. Because the action spectra of *Giardia* and *Cryptosporidium* were statistically similar, the ASCFs for *Cryptosporidium* can be directly used for *Giardia*.

ASCF values decreased with lower UVT and greater log inactivation credit, varied with reactor configuration, and increased with more operating lamps within a given configuration.
ASCF Values for Adenovirus Credit

With the validation microbes MS2, T1UV, T7, QB, and T7m, ASCF values for adenovirus credit calculated using the full adenovirus action spectra were notably less than 1.0. But values calculated using the adenovirus action spectra set to zero below 240 nm ranged from about 0.85 to 2.15. Values less than 1.0 were obtained with sleeve and validation water combinations that minimize low wavelength UV dose delivery below 240 nm. Values much greater than 1.0 were obtained with sleeve and validation water combinations that maximize low wavelength UV dose delivery. If the reactor was validated with adenovirus, the ASCF values calculated relative to an adenovirus action spectrum set to zero below 240 nm ranged from 1.1 to 5.5. Low values were associated with sleeves and validation water combinations that minimize low wavelength dose delivery, and high values associated with combinations that maximize low wavelength dose delivery.

Like the ASCF values for Cryptosporidium, adenovirus ASCF values decreased with lower UVT and higher log inactivation credit, varied with the reactor configuration, and increased with more operating lamps within a given reactor configuration.

APPLICATIONS/RECOMMENDATIONS

The ASCF calculation methods and implementation approaches presented in this report are designed to assist existing and future UV disinfection facilities to address the disinfection benefits and biases inherent in MP UV disinfection for regulatory compliance. Choosing the best approach to implement and monitor the ASCF is a site-specific determination. Implementation guidance was grouped into three broad categories 1) Generic ASCF Tables, 2) CFD-I UV reactor-specific modeling, and 3) new validations. Table ES.1 provides a general overview of each option.
Table ES.1
Low wavelength disinfection implementation approaches

<table>
<thead>
<tr>
<th>Approach</th>
<th>Target Pathogen</th>
<th>Include Wavelengths &lt;240 nm in Disinfection Calculation</th>
<th>Site-Specific Water Quality</th>
<th>Monitoring&lt;sup&gt;1&lt;/sup&gt;</th>
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<tbody>
<tr>
<td>Generic ASCF Tables</td>
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<td>No</td>
<td>No</td>
<td>No additional monitoring</td>
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<td>CFD-I UV reactor-specific modeling</td>
<td>Cryptosporidium and Giardia</td>
<td>No</td>
<td>No</td>
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<td></td>
<td>Cryptosporidium, Giardia and virus</td>
<td>Yes w/ aging and fouling&lt;sup&gt;2&lt;/sup&gt;</td>
<td>No</td>
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<td></td>
<td>Cryptosporidium, Giardia and virus</td>
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<tr>
<td>New Validations</td>
<td>Cryptosporidium and Giardia</td>
<td>Eliminated/minimized with sleeve, water quality, and UV absorber selection</td>
<td>No</td>
<td>No additional monitoring</td>
</tr>
<tr>
<td></td>
<td>Cryptosporidium, Giardia and virus</td>
<td>Yes</td>
<td>Yes</td>
<td>Additional monitoring</td>
</tr>
</tbody>
</table>

<sup>1</sup> Monitoring should include measurement of lamp output and water absorbance at wavelengths below 240 nm if either wavelengths below 240 nm or site-specific water quality is included in CFD-I modeling or new validations.

<sup>2</sup> CFD-I modeling should account for changes in lamp emission spectra due to aging and fouling if action below 240 nm is included.

Use of the generic ASCF tables provides the option to select an ASCF value with no additional modeling, testing, or monitoring required. However, this approach will result in a conservative ASCF value and may result in higher capital and annual operations and maintenance costs associated with higher power consumption. CFD-I modeling or new validation techniques may require additional upfront costs, but could result in annual operations and maintenance cost savings due to a more site and UV reactor-specific ASCF calculation. Potential cost savings of lowering the ASCF through CFD-I modeling or new validation work may be offset by the cost of reactor-specific modeling, new validations, and any potential additional monitoring requirements, which are unknown at this point (e.g., cost of UV sensors, calibration frequency, and maintenance needs).

If a validation is conducted using multiple validation microbes (e.g., MS2 and T1UV), it is recommended that a weighted average be calculated based on the relative percent of validation testing done with each microbe. Similarly, if the validation was conducted using different validation waters, it is recommended that a weighted average be calculated based on the relative percent of each water type used.

If reactor specific ASCF values are determined using CFD-based UV dose models, it is recommended that the models meet the recommendations provided in the checklist for CFD-based UV dose models provided in the appendices of this report. The appendices are available on the #4376 project page (under Project Resources/Project Papers) on the WRF website.

Once UV sensor technologies improve to allow for monitoring of lower wavelengths, new validations could be completed to better account for the influence of the low wavelength on disinfection performance. At this time, the UV sensor technology does not allow for development
of new validation protocols or recommendations, and should be evaluated once the technology becomes available.

Depending on the implementation approach selected, monitoring of the low wavelengths emitted by MP UV lamps may be required. To understand and account for the low wavelengths, both the lamp output and water absorbance must be monitored. Due to the lack of proven technologies for real-time monitoring of UV absorbance or lamp output, specific guidance for monitoring cannot be developed at this time. Once the technologies are available and proven, utilities should work with state regulators to develop accepted monitoring requirements and frequencies.

Once the ASCF value or equation is selected based on the above approach, the ASCF should be implemented as part of the overall Validation Factor (VF) calculation used to determine inactivation credit of the target pathogen. The ASCF should be implemented within the polychromatic bias factor as outlined in the UVDGM (Equation ES.1).

\[
VF = B_{RED} \times B_{poly} \times \left(1 + \frac{U_{Val}}{100}\right)
\]

where

- \(VF\) = Validation Factor
- \(B_{RED}\) = RED Bias
- \(B_{poly}\) = Polychromatic Bias Factor
- \(U_{Val}\) = Uncertainty of validation expressed as a percentage

VF inputs are defined in the UVDGM and should be documented in the UV reactor validation report. It is recommended that for all MP UV disinfection systems, ASCF calculations should be based on the approaches outlined in this report in place of the recommendations provided in the UVDGM. As discussed in Chapter 1, the UVDGM approach may overcorrect for the influence of action spectra differences on disinfection performance by not accounting for the water and sleeve absorbance.

If the target pathogen is Cryptosporidium or Giardia, an ASCF value less than 1.0 should not be used and any value less than 1.0 should default to 1.0. The UVDGM states that an ASCF is not required if the value is less than 1.05 or 1.06. However, it is recommended that an ASCF should be calculated and applied to all MP UV reactors, even if the calculated value is less than 1.05 or 1.06. If the target pathogen is adenovirus, an ASCF value less than 1.0 can be used to account for the disinfection benefits provided by low wavelength disinfection.

RESEARCH PARTNERS

- American Water Works Association
- U.S. Environmental Protection Agency

PARTICIPANTS:

This research was made possible through technical and financial support from numerous organizations:

- Tetra Tech/Clancy Environmental
- National Institute of Standards and Technology

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• LA Department of Water and Power
• Metropolitan Water District of Southern California.
• Aquionics (Erlanger, KY; USA)
• Atlantium Technologies (Bet Shemesh, Israel)
• Calgon Carbon (Pittsburgh, PA; USA)
• ITT Wedeco (Charlotte, NC; USA)
• ETS (Beaverton, Wisconsin; USA)
• Trojan Technologies (London, Ontario; Canada)
• Hydroqual/HDR (NJ, USA)
• CDM (Denver, CO; USA)
• Black & Veatch (Overland Park, KS; USA)
• CH2M Hill (Denver, CO; USA)
CHAPTER 1
INTRODUCTION

UV disinfection is an available technology for virus inactivation, but the high doses required for inactivation dictated in the Long Term 2 Enhanced Surface Water Treatment Rule (LT2) cause some challenges for validation of UV systems. In fact, the groundwater rule notes that there are no adequate methods (surrogates or procedures) for assessing the capabilities of a UV system to achieve 4-log virus inactivation. Recent research published by Linden and colleagues (Linden et al. 2007; Eischeid et al. 2009, Linden et al. 2009) has demonstrated that polychromatic UV irradiation, such as from medium pressure (MP) mercury vapor lamps have significant advantages for inactivation of adenoviruses compared to monochromatic UV 254 nm emitted from low pressure (LP) mercury vapor lamps. UV doses for 4-log inactivation of adenovirus on the bench scale from MP UV systems were up to 4 times lower than the LT2 rule. Linden, Scheible, and colleagues also developed and executed the first validation of a MP UV system for 4-log inactivation of viruses under LT2, using a live adenovirus challenge procedure. This validation was accepted by the NY State Department of Health for use of MP UV systems in groundwater disinfection of viruses. Since then a number of surrogates have been identified that may be key in validating MP UV systems at high UV doses (Rochelle et al. 2010). Because of these advances, there is now a need to develop sound guidance on the use of surrogates and MP UV for validating 4-log adenovirus inactivation.

Development of such guidance requires studies on the wavelength response (action spectra) of pathogens and their surrogates such as adenovirus, *Cryptosporidium*, and validation test microbes. Action spectra measurements of pathogens and test microbes help to resolve questions that impact the general application of MP UV systems for pathogen credit.

Action spectra data on pathogens and surrogates typically do not extend below 220 nm. Yet measured validation data show that wavelengths below 220 nm can have a significant impact on reduction equivalent dose (RED) measured using MS2, T1 and T7 phage. Furthermore, action spectra data is typically measured using bandpass optical filters, which introduces a significant error (up to 50 percent) that impacts the interpretation of that data.

While the United States Environmental Protection Agency (EPA) UV Disinfection Guidance Manual (UVDGM) (EPA 2006a) states that MS2 is a good surrogate for *Cryptosporidium*, that statement is only true for wavelengths above 240 nm. Below 240nm, published data indicates that validation using MS2 phage is overstating *Cryptosporidium* dose by as much as 50 percent, raising an important public health concern. Hence, the issue with MP systems is much bigger than just adenovirus and any MP-specific validation protocol should be applicable to all target pathogens.

Two issues are evident with the testing of MP UV systems. First, the LT2 specifies UV dose requirements for virus credit based on adenovirus [40 CFR 141.720(d)(1)]. Adenovirus is very resistant to UV light, requiring a relatively high UV dose of 186 mJ/cm², based on LP UV light at 254 nm, for 4-log inactivation credit. Many of the test microbes currently used for UV reactor testing, such as MS2 phage, *B. subtilis* spores, and T1 phage, are too sensitive to UV light to demonstrate such high UV dose values (Figure 1.1). However, Water Research Foundation’s Project 3105 has recently identified UV-resistant microbes, such as *B. pumilis* spores and *D. aquaticus*, as viable candidates for demonstrating adenovirus inactivation. Guidelines developed through this project address the application of current and new test microbes for adenovirus credit.
Secondly, the wavelength response of adenovirus differs significantly from the wavelength response of currently used test microbes like MS2 phage (Figure 1.2), especially at wavelengths below 240 nm. If a UV system using MP UV lamps has a significant UV output below 240 nm, UV testing using MS2 will under-predict the UV dose delivered to adenovirus by a factor of two or more (Linden et al. 2007). Hence, the ideal test microbes should have a wavelength response similar to adenovirus. Alternatively, approaches need to be developed for applying action spectra correction factors (ASCF) to validation data measured using MS2 phage or other surrogates to account for these differences. Unfortunately, for the purposes of this report, the analysis presented only uses ASCFs calculated based on action spectra set to zero below 240 nm because of the lack of current sensor technology that allows effective monitoring below 240 nm. With such a sensor, these analyses could be re-worked to include those wavelengths down to 200nm.

This research collected and analyzed published data on the UV dose-response of adenovirus and other UV-resistant pathogens, and candidate validation test microbes with the objective of defining UV dose monitoring and validation strategies for adenovirus credit, consistent with approaches recommended by EPA within the UVDGM. The research filled in some important gaps in the current published data by conducting further measurement and analysis on the wavelength response of pathogens and test microbes. That data was used as inputs to proven CFD-based models for commercial MP UV reactors to define UV dose monitoring and validation strategies for virus credit. These strategies provided the foundation for this guidance document, which was developed with input and consensus from UV academics, regulators, consultants, utilities, and UV vendors.

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1 UV validation testing typically is limited to demonstrating 5 to 6 log inactivation of the test microbe.
Figure 1.2  Wavelength response of adenovirus and MS2 phage shows adenovirus is more sensitive to UV light below 240 nm

CRYPTOSPORIDIUM RESPONSE AT LOW WAVELENGTHS

Over 300 drinking water utilities in North America are implementing ultraviolet (UV) disinfection at flowrates up to 2,200 mgd, primarily for the inactivation of Cryptosporidium and Giardia (Wright et al. 2012). Approximately 75 percent of those systems use polychromatic medium-pressure (MP) UV lamps. MP lamps generate germicidal UV light at wavelengths from 200 to 320 nm.

Through the course of this research, it was recognized that the challenge in testing MP systems is not only relevant to adenovirus. Figure 1.3 compares the action spectra or wavelength response of Cryptosporidium and MS2 phage, taken from published data. The action spectra is normalized to a value of 1.0 at 254 nm, since UV dose requirements for UV disinfection are based on UV dose-response curves measured at 254 nm using a collimated beam apparatus. Figure 1.3 also shows the UV output of a MP UV lamp.

Source: Adapted from Linden et al. 2001.

Figure 1.3 Comparison of the wavelength response of MS2 phage (Rauth 1965) and Cryptosporidium
While the action spectra of MS2 and Cryptosporidium at wavelengths above 240 nm are similar, the action spectrum of MS2 phage is greater than that of Cryptosporidium at wavelengths below 240 nm. If wavelengths below 240 nm significantly contribute to UV dose delivery measured during validation, validation conducted using MS2 phage will overstate the inactivation expected with Cryptosporidium. As we discovered, a similar issue with action spectra differences exists with T1 and T7 phage, currently used as alternates to MS2 phage.

The difference between the action spectra of MS2 and Cryptosporidium at wavelengths below 240 nm represents a potential risk to public health protection that is not addressed by the UVDGM. Instead, the UVDGM states that the action spectra of Cryptosporidium and MS2 phage are similar enough that no action spectra correction factor is required. However, this conclusion is based on an analysis of the germicidal UV output of MP lamps as provided in the 2003 draft UVDGM. That analysis was based on a MP lamp that had negligible UV output (below 240 nm) and is not applicable with today’s MP lamps.

**INTERPRETING PATHOGEN AND SURROGATE ACTION SPECTRA**

The action spectrum of a microbe is obtained by measuring the microbe’s UV dose response at different wavelengths and plotting the UV sensitivity as a function of wavelength. The UVDGM states that the impact of the microbe’s action spectra on dose delivery by a UV reactor may be estimated by calculating the lamp’s germicidal output, $P_g$, using:

$$P_g = \sum_{\lambda=200nm}^{320nm} P(\lambda) \times G(\lambda) \times \Delta\lambda$$  \hspace{1cm} (1.1)

where
- $P(\lambda)$ = spectral UV output of the lamp at wavelength $\lambda$
- $G(\lambda)$ = action spectra of the microbe at wavelength $\lambda$
- $\Delta\lambda$ = 1 nm increment

The UVDGM states the ratio of the germicidal output defined using the action spectra of the validation test microbe to that defined using the action spectra of the pathogen (such as adenovirus or Cryptosporidium) may be used to define an action spectra correction factor (ASCF) that is applied to the UV dose algorithms developed through validation.

At the time the UVDGM was developed, correction factors based on MS2 validation were 1.06 because the lamp considered in the calculation had little output below 240 nm (EPA 2006a). However, using today’s lamps, the correction factor with typical MP lamps in some cases ranges from 1.7 to 2.0 using the UVDGM approach. Applying the correction factor as defined by the UVDGM to MP UV systems could increase UV system capital and O&M costs for these cases by a factor of 70 to 100 percent.

Because the UVDGM approach for determining the correction factor does not account for the impact of the sleeve UV transmittance (UVT) and the UV absorbance of the water during validation, it can be very conservative. For example, including the UV transmittance of Type 219 sleeves in the definition of the germicidal output lowers the correction factor to values of approximately 1.05. While UV systems in Europe are validated using Type 219 sleeves, UV systems in North America are typically validated using Type 214 and synthetic quartz sleeves.

The impact of UV absorbance on the correction factor depends on the placement of lamps within the reactor and can be determined using UV dose models based on Computational Fluid
Dynamics (CFD). Using CFD, the correction factor is defined as the ratio of UV dose predicted using the action spectrum of the validation test microbe to that predicted using the action spectrum of the pathogen, where both are calculated at a given flow, UVT and UV sensor reading.

The impact of sleeve type and water absorbance on the UV dose measured during validation was first determined during Fall 2010 with the validation of a MP system manufactured by ITT-WEDECO (Wright et al. 2011). For a given UVT at 254 nm, the sleeve type and UV absorbance of the water below 240 nm affected the measured UV dose by as much as 60 percent. The validation data demonstrated the importance of UV light below 240 nm on UV system sizing and technology selection.

One issue with action spectra data is the use of bandpass filters. Most of the action spectra data previously published for pathogens (adenovirus and Cryptosporidium) and validation microbes (MS2, T1, T7, B. pumilis) were measured using bandpass filters to restrict the UV light from a MP lamp to a given wavelength (Rochelle et al. 2010; Wright et al. 2007; etc.). These bandpass filters have a bandwidth on the order of 10 nm at half the maximum peak transmittance and bandpass of 20 nm or more at 10 percent of peak. In contrast, UV photobiologists measure action spectra using monochromatic light obtained with either narrow bandpass monochromators or a tunable laser (Rauth 1965; Sutherland 2002). Use of bandpass filters may cause significant errors during the action spectra measurement because the effective wavelength of the bandpass filtered light differs from the nominal wavelength of the bandpass filter (See Figure 1.4).

Source: Adapted from Rauth 1965, Linden et al. 2000, Mamane-Gravetz et al. 2005.2

Figure 1.4 Reported action spectra of MS2 phage show significant differences above and below 254 nm

Some of the differences between action spectra may also be related to the interpretation of UV dose response data used to define the action spectra. Some researchers define the action spectra using the reciprocal of the UV dose at a given wavelength required to obtain a fixed level of microbe inactivation (e.g. 2 log inactivation). This approach is valid if the microbe has first order inactivation kinetics. However, the UV dose response of many microbes, including adenovirus, MS2 phage, and B. pumilis, shows curvature, tailing, and shoulder affects. In those cases, the action spectra could be defined using the kinetic constants that define the shoulder, exponential, and

---

2 These differences have a large impact on action spectra correction factors applied to validation data.
tailing affects. For example, Cabaj et al. (2001) report that the action spectra for the kinetic constant that defines the shoulder with *B. subtilis* spores differs from the action spectra for the kinetic constant that defines the exponential reduction. Furthermore, the integrated impact of MP light on *B. subtilis* inactivation depended on the shape of the inactivation kinetics (Cabaj et al. 2001), confirming what has been reported by UV photobiologists (Sutherland 2002).

Clearly, to develop guidelines for the validation of MP UV systems for adenovirus, *Giardia* or *Cryptosporidium* credit, one needs to understand how the action spectra of the target pathogen and validation microbes impacts the inactivation of those microbes. This is important for defining what is an appropriate test microbe, defining action spectra correction factors if needed, and defining what wavelengths matter for UV dose monitoring. To address these issues, this project generated resources to experimentally measure the action spectra of adenovirus, *Giardia* and *Cryptosporidium*, as well as a number of surrogates used in validation testing, from 210 to 320 nm using a tunable UV laser obtained from NIST. Collecting the data down to 210 nm filled in an important gap with current datasets. Using a UV laser to produce monochromatic light (bandwidth $\ll 1$ nm) eliminated the errors with previous work obtained using bandpass filters.

**UV DOSE MONITORING**

A major challenge for developing guidance for testing MP UV systems for adenovirus or *Cryptosporidium/Giardia* credit is UV dose monitoring. UV dose monitoring is critical for public health protection with UV disinfection and is a major focus of the UVDGM. With UV dose monitoring, the programmable logic controller (PLC) of the UV reactor calculates the delivered UV dose from measurements of flow rate through the reactor, UVT of the water, and UV sensor readings using an algorithm that is developed using UV validation test data. Currently, UV systems use the UVT measured at 254 nm and UV sensor readings that have a peak response near 260 nm and negligible response below 220 nm (see Figure 1.5). As such, UV dose monitoring will not indicate whether the enhanced inactivation of adenovirus due to wavelengths below 240 nm is occurring with the application of the UV reactor at the water treatment plant (WTP). The enhanced inactivation will not occur if lamp aging and sleeve fouling or changes in the spectral UVT of the water cause a reduction in UV intensity at wavelengths below 240 nm (see Figure 1.6). For example, Petri (2009) reports that the benefits of MP light with adenovirus fall away as the UV absorbance of the waters at low wavelengths increases. Alternatively, UV dose monitoring will not indicate whether the dose algorithm is over-predicting disinfection performance for *Cryptosporidium* or *Giardia*.

To address this issue, UV systems using MP lamps need UV dose monitoring algorithms that account for UVT measured at 220 to 240 nm and UV intensity measured using a UV sensor with a peak response below 240 nm. Furthermore, the validation test plan needs to be designed to account for the impact of the UVT between 220 and 230 nm and the UV intensity at those wavelengths. Factors that impact the UVT at low wavelengths include the spectral UVT of the water and the UV absorber used during validation, such as Super Hume™ and lignin sulfonic acid (LSA). Factors that impact the UV intensity at low wavelengths include the UV output of the lamp and the UV transmittance spectra of the quartz sleeves used by the reactor. While MP lamps considered with the development of the UVDGM emitted minimal UV light below 240 nm, MP lamps currently being used by UV vendors have a significant output at wavelengths as low as 200 nm, especially those systems used for advanced oxidation process (AOP) applications (Figure 1.7). There has also been a trend with UV vendors moving from type 214 quartz sleeves with a UV
transmittance of 51 percent at 200 nm to synthetic quartz sleeves with a UV transmittance of 89 percent at 200 nm (Figure 1.7).

Figure 1.5 UV sensors used by MP UV systems have negligible response below 240 nm

Figure 1.6 Waters with similar UV absorbance at 254 nm may have very different UV absorbance below 240 nm, impacting the inactivation of adenovirus

This project used proven CFD-based UV dose models for commercial UV reactors to understand and quantify how each of these factors impacts UV dose delivery and monitoring with commercial MP UV reactors. The project also leveraged a recently developed CFD model for the WEDECO KM2300 MP UV system. This reactor was validated during the fall of 2010 with three different sleeve types and two different water types (see Figures 1.6 and 1.7). The measured MS2, T1 and T7 REDs were significantly impacted by both sleeve and water UV transmittance below 240 nm, making this dataset valuable for providing data on the very issues that impact this project.
Figure 1.7. Differences in lamp output and sleeve UVT below 240 nm will also significantly impact adenovirus inactivation

The action spectra data for adenovirus, *Cryptosporidium*, and test microbes like *B. pumilis* and MS2 phage developed through this project were used as inputs to MP UV system CFD models. The models quantified how UV lamp output, quartz sleeve UV transmittance, water UV absorbance spectra and UV sensor spectral response impacts dose delivery and monitoring with each microbe. Results were analyzed to define ASCFs and provide updated guidance for UV dose monitoring algorithms and validation test plans.
CHAPTER 2
ACTION SPECTRA OF SURROGATES AND PATHOGENS

INTRODUCTION

This chapter reports on the methods used and data generated for action spectra used in development of the ASCFs. An extensive literature review on action spectra state of the art prior to this research is provided in Appendix K. In this project, the action spectra for MS2, T1UV, T7, T7m, Q beta, adenovirus, Cryptosporidium parvum, and Giardia lamblia were determined through a series of collimated beam tests. UV irradiations were conducted using a tunable laser from the National Institute of Standards and Technology (NIST).

Surrogates Tested

MS2

MS2 (Male-specific 2) coliphage is a single-stranded RNA virus that infects the F pilus of male-specific strains of *E. coli* and serves as an indicator of fecal contamination. Frequently used for reactor validation in North America, MS2 is similar in size and shape to other viruses, yet it is often more resistant, making it an ideal surrogate for testing. The MS2 action spectrum was first determined in 1965 using a large diffraction grating monochromator that dispersed monochromatic UV light approximately 1.2 nm in bandwidth. The spectrum was measured again in 2005 with a monochromator with a maximum bandwidth of 10 nm (Mamane-Gravetz et al. 2005). UV inactivation of MS2 is very straightforward with 65-70 mJ/cm² of LP UV light required for 4-log inactivation (Meng and Gerba 1996; Park, et al. 2011).

T1UV

T1UV, an organism with a genome similar to coliphage T1, was isolated from swine manure by GAP Microbial Services. Like T1, T1UV is also a double-stranded DNA virus with a non-contractile tail (Stefan et al. 2007). T1UV has similar sensitivity to low-pressure UV light as Cryptosporidium and Giardia and has been suggested as a surrogate microorganism for reactor validation. An action spectrum was developed for T1UV using a medium-pressure mercury arc-lamp with six bandpass filters between 9-11 nm in bandwidth. An LP UV dose of ~18 mJ/cm² is capable of 4-log reduction of T1UV.

T7 and T7m

T7 and T7m coliphage are double-stranded DNA viruses that infects *E. coli*. It is commonly applied as a biological dosimeter for solar UV radiation (Ronto et al. 1994) and also used for calibration of chemical actinometry (Kuhn et al. 2004). Like T1UV, T7m coliphage exhibits inactivation kinetics similar to *Cryptosporidium* and *Giardia* and has been considered for reactor validation. For 4-log inactivation of T7, 16.6 mJ/cm² of LP UV is required (EPA 2006a). An action spectrum was developed for T7 coliphage from 240 nm to 340 nm using a xenon lamp and a monochromator with ~4 nm dispersion (Ronto et al. 1992). In this project, a strain of T7, called T7m, was also tested.
**Qβ**

Qβ (Q beta) coliphage is a virus similar to MS2 that infects E. coli through the F pilus of male-specific strains. The coliphage is sometimes used as a biological indicator to detect the state of host E. coli cells and determine if they are active viable but non-culturable (VBNC) bacteria (Ben Said et al. 2010). Q beta has been used in the past for UV reactor validation (EPA 2006a). It is more sensitive to UV light than MS2. The accepted dose required for 4-log inactivation of Q beta, is 48 mJ/cm² (EPA 2006a). An action spectrum was developed for Q beta in the same study as for T1UV (Stefan et al. 2007).

**Bacillus pumilus**

*Bacillus pumilus* is a non-pathogenic spore-forming bacterium, highly resistant to monochromatic UV light. Given its UV resistance, *B. pumilus*, in its spore form, is being considered as a surrogate microorganism for adenovirus (Rochelle et al. 2010). The UV dose-response of spores varies by strain and cultivation method.

**Pathogens Tested**

**Adenovirus**

*Adenovirus* is responsible for respiratory and gastrointestinal illness in humans, primarily affecting immunocompromised individuals including infants and the elderly. As a known public health risk subject to future regulation, adenovirus is one of few microbial contaminants listed on the EPA’s Contaminant Candidate List (CCL 3). Due to its strong resistance to UV light relative to other pathogens, adenovirus governs the current regulations for virus inactivation in the Long Term 2 Enhanced Surface Water Treatment Rule and the Groundwater Rule. The EPA Surface Water Treatment Rule requires 4-log inactivation of viruses (EPA 1984). Based on empirical results of adenovirus inactivation by low-pressure UV light, the agency established a required dose of 186 mJ/cm² for 4-log inactivation of viruses (EPA 2006a) for surface water treatment. An action spectrum was developed for adenovirus type 2 using medium-pressure UV light with bandpass filters (Linden et al. 2007).

**Cryptosporidium parvum**

*Cryptosporidium parvum* is a protozoan parasite that infects the cell lining of the digestive tract, causing the diarrheal disease, cryptosporidiosis. The pathogen’s resistant oocysts, 4-6 μm in size, can infect humans through multiple transmission pathways and have been associated with numerous waterborne disease outbreaks (Fayer et al. 2000, Mackenzie 1994). *Cryptosporidium* is important to the water treatment industry due to its resistance to chemical disinfection with chlorine. The accepted dose for 4-log inactivation of *Cryptosporidium parvum* is 22 mJ/cm² (EPA 2006a). An action spectrum was previously developed for *Cryptosporidium parvum* in 2001 using bandpass filters with a medium-pressure mercury vapor lamp (Linden et al. 2001).

**Giardia lamblia**
*Giardia lamblia* (also called *G. duodenalis*) is a flagellate protozoan that infects the small intestines, causing giardiasis. *Giardia*’s highly resistant oocysts are prevalent in surface water sources (Marshall et al. 1997) and have been responsible for several waterborne outbreaks (Gibson et al. 1998). The accepted dose for 4-log inactivation of *Giardia* is 22 mJ/cm² (EPA 2006a). No references are available with an action spectrum for *Giardia lamblia*.

**METHODS**

**UV Light Source – NIST Laser**

UV irradiations were conducted using a NT242 series Ekspla laser provided by the National Institute of Standards and Technology (NIST, Gaithersburg, MD). The Ekspla incorporates a pulsed (1 kHz) Nd:YAG pump laser and OPO (optical parametric oscillator), which provide the capability to tune the laser output between 210 to 2600 nm. For action spectra work, only the wavelengths between 200 and 300 nm were necessary. Outputting these UV wavelengths involved doubling the frequency of wavelengths in the visible range. Visible light from the laser that was co-aligned with the UV wavelengths was either filtered by dielectric mirrors or diverted from aluminum mirrors and a prism from the optical path. A NIST compact array spectrometer (Instrument Systems CAS 140 CT) was used to verify that no visible light reached the samples. The full width at half maximum (FWHM) bandwidth of the laser radiation was calculated at 0.04 nm at 300 nm emission and 0.07 nm at 210 nm. When measured with a Maya 2000 Pro spectrometer (Ocean Optics, Dunedin, Fl), the FWHM varied between 1.1 nm at 210 nm and 2.7 nm at 300 nm, as shown in Figure 2.1. The wider FWHM was caused by the bandwidth of the Maya 2000 Pro.


**Figure 2.1 Germicidal emission spectra for a NT242 series Ekspla laser from NIST for wavelengths used to generate action spectra**

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UV Light Source – Low Pressure and Medium Pressure Mercury Vapor Lamps

Collimated beam exposures were also conducted with a low-pressure UV mercury vapor lamp (G12T6L, Atlantic Ultraviolet, Hauppauge, NY) for all surrogates and pathogens tested and in some cases a medium-pressure UV mercury vapor lamp (RAYOX 1 kW, Calgon Carbon Corporation, Pittsburgh, PA) for MS2, adenovirus, and *Giardia*. Low-pressure (LP) UV lamps emit relatively monochromatic light, primarily at 253.7 nm whereas medium-pressure (MP) UV lamps emit polychromatic light. The emission spectra for both lamps used in this study are given in Figure 2.2. The LP exposures were performed to compare to the data obtained to that generated with the NIST tunable laser at 253.7 nm. The data for MP were generated to evaluate if the action spectra generated in this study could be used to predict the performance of MP UV lamp inactivation using the microorganism action spectra as the germicidal weighting factor. This analysis was performed for MS2 and adenovirus and is presented below.

![Figure 2.2. Relative emission of the medium-pressure and low-pressure UV mercury vapor lamp used for testing](image)

**Collimated Beam Testing**

At wavelengths between 210 and 290 nm, at approximate 10 nm intervals, four collimated beam exposures were conducted to generate a dose response curve. Irradiance was measured at the water surface by a photodiode detector (IRD SXUV 1000, Opto Diode Corporation, Thousand Oaks, CA) and precision aperture (SK#030483-1073, Buckbee Mears, Cortland, NY), both supplied by NIST. Average UV doses for the collimated beam tests were determined as described in Bolton and Linden (2003), adjusting for reflection off the water surface, UV absorption (measured by a Spectronic Genesys 10uv™ spectrophotometer, Thermo Electron Scientific Instruments Corp, Madison, WI), depth of the water sample, as well as the non-uniform distribution of light across the sample surface. Beam divergence was assumed to be negligible from the laser diffuser, but measured and accounted for in the mercury lamp irradiations. Quiescently stirred samples of 5 mL (0.6 sample depth) were irradiated in 3.5 cm diameter petri dishes located at a distance of ~26.5 cm from the light source. Petri factors were calculated by
measuring irradiance in an X-Y axis pattern in 0.5 cm increments. At these distances from the light source, the petri factors for the laser varied from 0.98 to 1.04.

Depending on the wavelength and pathogen or surrogate tested, UV doses ranged from 8 mJ/cm² to 160 mJ/cm². The laser irradiance varied across wavelengths between 10 μW/cm² and 300 μW/cm². Immediately after exposure and prior to starting the microbial assays, the irradiated samples were refrigerated at 4 °C. Single replicate dose-response analysis was conducted for QB, T7m, T7 and adenovirus while several replicates were conducted at each wavelength for MS2, T1UV, Cryptosporidium, and Giardia.

Collimated beam exposures were also conducted to compare the adenovirus dose response to LP UV and MP UV light. UV doses with the low-pressure mercury vapor lamp were determined as described above. For the MP UV mercury vapor lamp, the UV dose accounted for the relative lamp emission of the light source (as measured by the Maya 2000 Pro spectrometer), the sensitivity of the radiometer used to measure the irradiance (from radiometer calibration data), the absorbance and sample depth of the water, as well as a germicidal weighting. In this study, average irradiance was weighted germicidally with DNA absorption as is the common practice (Linden and Darby 1997), as well as with the pathogen or surrogate action spectrum, determined below, for comparison.

**Microbial Assays**

This section describes the assays used to enumerate the inactivation of the surrogates and pathogens studied for action spectra in the project.

**Surrogates**

Tested surrogates consisted of phages and a spore used in validation testing of UV disinfection systems.

**Coliphage.** Stock solutions of coliphage were prepared by inoculating the propagation media listed in Table 2.1 with the log-phase host bacteria and an aliquot of the phage stock also listed.

<table>
<thead>
<tr>
<th>Coliphage (Source/ID)</th>
<th>Host (Source/ID)</th>
<th>Propagation Media</th>
<th>Total Time to Propagate</th>
</tr>
</thead>
<tbody>
<tr>
<td>MS2 (ATCC/15597-B1)</td>
<td><em>E. coli</em> HS(pFamp)R (ATCC/700891)</td>
<td>Tryptic Soy Broth supplemented with Ampicillin/Streptomycin</td>
<td>5 hours</td>
</tr>
<tr>
<td>T1UV (Laval University/HER#468)</td>
<td><em>E. coli</em> CN13 (ATCC/700609)</td>
<td>Modified TYGB</td>
<td>21-27 hours</td>
</tr>
<tr>
<td>Qβ (ATCC/23631-B1)</td>
<td><em>E. coli</em> (ATCC/23631)</td>
<td>Tryptone Yeast Glucose Broth (TYGB)</td>
<td>5 hours</td>
</tr>
<tr>
<td>T7m (ATCC/11303-B38)</td>
<td><em>E. coli</em> B (ATCC/11303)</td>
<td>Nutrient Broth supplemented with 0.5% Sodium Chloride</td>
<td>7 hours</td>
</tr>
<tr>
<td>T7 (ATCC/BAA-1025-B2)</td>
<td><em>E. coli</em> CN13 (ATCC/700609)</td>
<td>Tryptic Soy Broth</td>
<td>24 hours</td>
</tr>
</tbody>
</table>
The broth was incubated at 35 °C ± 0.5 °C with constant shaking for the time specified in Table 2.1. Bacterial debris was removed by centrifugation. The clarified supernatant containing the phage was decanted to sterile containers and stored at 4 °C. For some exposures (i.e., MS2, T1UV, and T7), coliphage stocks were received from other laboratories involved in UV reactor validations.

Phage concentrations were determined using the double agar layer method as described previously (Fallon et al. 2007). Briefly, log phase host bacteria were added to “soft agar” with an aliquot of the sample containing an estimated 20 – 200 plaque forming units (pfu). The inoculated soft agar was poured over an agar plate and allowed to harden. Agar plates were incubated inverted for 16 to 24 hours at 35°C ± 0.5 °C and then examined for plaque formation. Viral plaques were counted to determine the original concentration of coliphage in the original broth culture and the concentrations after exposure to UV light. Each sample was analyzed at multiple dilutions with three replicates plated at each dilution. Controls were analyzed each day samples were analyzed. Negative controls were analyzed at the beginning of each analytical batch and periodically during the plating procedure to ensure the sterility of plating area. A negative control was also analyzed at the end of the plating. A positive control sample was plated each day to ensure the assay was working correctly.

**Spore.** The ASFUVRC strain of *B. pumilus* was isolated from native aerobic spore forming (ASF) bacteria after exposure to high doses of LP UV light. Spore preparation was conducted as described previously (Rochelle et al. 2010). Briefly, spores were cultured for 6 days by incubating at 37 °C in a modified sporulation medium supplemented with 0.1 mM MnSO₄. Spores were harvested by centrifuging at 4000 g at 4 °C for 15 min. The resulting cell pellets were washed three times by resuspending in sterile phosphate buffered water (PBW) and centrifuging at 4000 g at 4 °C for 15 min. The final cell pellet was resuspended in 10 mL of PBW, pasteurized at 80 °C for 20 min to kill vegetative cells, and then stored at 4 °C prior to UV exposure.

*B. pumilus* spore assays were conducted as described previously (Rochelle et al. 2010). The stock suspension was serially diluted and then aliquots were filtered using 0.45 µm pore-sized mixed cellulose esters membranes. Membranes were applied to plates of tryptic soy agar (TSA) and incubated inverted for 22 to 26 hours at 35 °C. Colonies were enumerated to determine the initial spore concentrations. For exposed samples, sample volumes or dilutions containing an estimated 20 to 60 colony-forming units (cfu) per membrane were analyzed to determine log₁₀ inactivation. Each volume analyzed was plated in triplicate.

Quality control samples were analyzed with each batch of samples and include a negative control of the rinse water, a negative control of vegetative bacteria, *E. coli* stock (ATCC #25922) after pasteurization, and a positive control of a known concentration of *B. pumilus*.

**Pathogens**

**Adenovirus.** Virus and cell lines used for inactivation studies were obtained from American Type Culture Collection (ATCC). A549 human lung carcinoma cells (ATCC #CCL-185) were cultured in DMEM high glucose media with 1X penicillin streptomycin (100X solution, Gibco Invitrogen) and 10% fetal bovine serum and incubated at 37 °C in 5% CO₂ and 95% humidity. Confluent flasks of A549 cells were infected with adenovirus type 2 (ATCC #VR-846), incubated, and checked daily for cytopathic effects (CPE). When the monolayer of A549 cells was 90% destroyed, flasks were freeze-thawed three times and the adenovirus was purified from the flask by centrifugation and polyethylene glycol precipitation, and titered.
For UV-irradiated samples containing adenovirus, a Most Probable Number (MPN) assay (EPA 1996 and Cromeans et al. 2008) was performed. After exposures, all samples were frozen at -80 °C until cell monolayers were ready for infection. Briefly, 24-well plates were seeded with A549 cells (5 x 10^5 cells/1 mL), DMEM supplemented with antibiotics, antimitotic, and 10% fetal bovine serum (FBS). The plates were incubated for 1 d to 2 d at 37 °C ± 1 °C and 5% CO₂. Once the monolayers were 90% to 100% confluent, adenovirus samples were removed from the freezer, thawed, and serially diluted. Four sequential dilutions were analyzed, 10 wells per dilution, so that the most dilute applied to the monolayers resulted in no cytopathic effects (CPE). In each 24-well plate, the first well was a negative control. The media was aseptically removed from the well. A fixed volume (100 μL) of each sample dilution was applied to the monolayer and incubated at 37 °C ± 1 °C for 1 h to 2 h to enhance virus absorption. After the required incubation, 900 μL of DMEM supplemented with antibiotics, antimitotic, and 2% fetal bovine serum (FBS) was added to each well. All plates were incubated at 37 °C ± 1 °C and 5% CO₂ for 15 d. The media was changed on wells showing no CPE after 5 d to 7 d incubation. An MPN result was determined using a spreadsheet based on the calculation methods given in Standard Methods (APHA 2005).

**Cryptosporidium parvum.** Cryptosporidium parvum oocysts (Iowa isolate) were purchased from Waterborne, Inc. (New Orleans, LA). These had been produced by experimentally infected mice. Feces were collected and purified using sucrose and Percoll density gradient centrifugation. Oocysts were < 1 month old post shedding and had ≥ 5% infectivity rate for inactivation trials at time of testing.

Infectious Cryptosporidium concentrations in samples and controls were determined using HCT-8 (human ileocaecal adenocarcinoma) cells (American Type Culture Collection, ATCC # CCL-244). The HCT-8 cell stock solutions were prepared by placing the cells in a volume of maintenance media (RPMI), supplemented with antibiotics, an antifungal agent and 5% fetal bovine serum. The culture flask was incubated at 35 °C +/- 1.0 for 2 – 5 days under 5% CO₂ atmospheric conditions. When the cells formed a confluent monolayer, they were removed from culture flask by trypsinization, concentrated by slow speed centrifugation at 400 g for 2 min, and re-suspended in maintenance media. A small portion of the re-suspended cells was placed in a new culture flask containing fresh media for continued growth. The Cryptosporidium infectivity assay was conducted by placing 10^5 HCT-8 cells in each chamber of a well slide containing 8 chambers and incubating at 35 °C +/- 1.0 °C and 5% CO₂ for 2 to 3 days. When the monolayers in the well chambers were 80% to 90% confluent, they were ready to support Cryptosporidium infections. HCT-8 cells were discarded after 24 passages.

**Cryptosporidium** infectivity was measured within 24 hours of UV exposure using a protocol described previously (Johnson et al. 2012). Briefly, oocysts were concentrated using immunomagnetic separation (IMS) as described in US EPA Method 1623 within 2 hours of exposure. Each sample was transferred to an individual Leighton tube containing 1 mL each of 10 X SL-A and 10 X SL-B buffers. Sample tubes were rinsed with approximately 1 mL to 2 mL of phosphate buffered saline (PBS) to remove residuals and the rinse was transferred to its respective Leighton tube. One hundred microliters of Cryptosporidium IMS beads were added to each tube and were incubated for 1 h at room temperature under constant rotation. Each Leighton tube was then placed in a magnetic particle concentrator (MPC-1) and gently rocked for 2 min, through a 90° angle. The supernatant in each tube was decanted to waste. The tube was removed from the MPC-1 and the bead-oocyst complex was gently re-suspended in 1 mL PBS. The tube was replaced in the MPC-1, and the bead isolation procedure was repeated and the supernatant...
discarded. The beads in each tube were re-suspended in 100 µL Hank’s Balanced Salts Solution (HBSS) and refrigerated overnight at 4°C.

The following morning, samples and controls (method blanks, heat-inactivated oocysts and unexposed oocysts) were incubated at 35 °C for one hour in acidified HBSS containing a concentration of 1 % trypsin to stimulate excystation of the oocysts. Sample aliquots were rinsed to remove the acidified HBSS and known concentrations of Cryptosporidium oocysts were applied to confluent monolayers of HCT-8 cells and then incubated for 68 to 72 hours under 5 % CO₂. After incubation, inoculated monolayers were washed with phosphate buffered saline and stained with fluorescein isothiocyanate (FITC) conjugated polyclonal antibodies. Detection and enumeration of the infectious foci were performed using epifluorescence and differential interference contrast (DIC) microscopy. An infection focus was defined as a cluster of intracellular reproductive stages of Cryptosporidium oocysts as previously described (Rochelle et al. 2001)

Quality assurance/quality control samples were analyzed with each analytical batch. Method blanks, positive controls, a negative control consisting of heat-inactivated oocysts, and inoculation process controls were analyzed. Initial concentrations of oocysts used in each experiment were determined by hemocytometer counts. Initial oocyst infection rates were used to calculate the number of oocysts needed for each exposure to accurately measure the log₁₀ inactivation. Six subsamples of the oocyst stock used for all tests on a given day were assessed for percent infectivity. This value was determined from the number of foci detected against the number of oocysts applied, with each focus representing one infectious oocyst. For each UV-treated sample, triplicate applications of three subsamples of oocysts were applied in triplicate to the cell layers across a range of concentrations sufficient to achieve detectable numbers of foci. Percent infective UV-treated oocysts were then calculated for each concentration, and the log inactivation achieved was determined by averaging the log₁₀ of the percent infectious untreated oocysts divided by that of the irradiated oocysts. Variability between replicates was consistently less than 0.1 log₁₀.

Giardia duodenalis. G. duodenalis (H3 strain; Assemblage B) cysts were propagated at the Cincinnati US EPA National Exposure Research Laboratory in Mongolian gerbils (Meriones unguiculatus) and purified by sucrose flotation followed by Percoll sedimentation (GE Healthcare Life Sciences, Piscataway, NJ, USA) (Belosevic et al. 1983; Sauch 1984). The cysts were stored at 4º C in 0.01% Tween-20 amended with Pen-Strep, and used within 14 days of preparation.

For each study, 27-73 age-matched, specific-pathogen free, 30-40 g, female Mongolian gerbils (Meriones unguiculatus) were purchased from Charles Rivers Laboratories (Wilmington, MA) for the experimental studies. One study used male gerbils. Gerbils were group caged until cyst dosage, and then individually caged. Animals were housed in a ventilated caging system, with sterile bedding, food, and water. Animal studies were approved and overseen by the Cincinnati US EPA Institutional Animal Care and Use Committee.

Animals were dosed by oral gavage with flow cytometry enumerated cysts. The infection status of the animals was determined by microscopy as previously described (Hayes et al. 2003). Briefly, fecal samples were examined for cysts after flotation. Cyst free gerbils were euthanized and the small intestines were examined for trophozoites. An animal was considered free of infection if no cysts or trophozoites were detected.

The ID₅₀ was established using a dose-response curve of 25 gerbils divided into five cohorts of 5 gerbils. Each cohort was exposed to cysts increasing in 0.5 log increments. UV exposed cysts studies were controlled with either 10 gerbils exposed to the ID₅₀, or two cohorts of five each exposed to the ID₅₀ and ID₅₀+1log. Five animals were exposed the ID₅₀+1log cysts and
5 animals were exposed to the ID$_{50}+2$ log cysts, and two studies exposed five gerbils to ID$_{50} +3$ logs. Up to three UV exposure levels were evaluated at one time and shared the same controls. The results were examined by comparing dose response curves and an MPN approach described by Shin et al. (2009).

**Statistical Analysis**

**Coliphage, Spore, and Cryptosporidium**

Sutherland (2002) describes an action spectrum as a spectrum of constants that map the UV dose response of an organism at different monochromatic wavelengths to its UV dose-response at a reference wavelength, such as 253.7 nm. In this study, a second-order polynomial equation for the coliphage and *Cryptosporidium* fit well the UV dose-response at each wavelength. Hence, mapping constants were defined using:

\[
\log I_\lambda = A \times (\alpha_\lambda \times D_\lambda) + B \times (\alpha_\lambda \times D_\lambda)^2
\]  

[2.1]

where 

- $\log I_\lambda$ = predicted log inactivation
- $D_\lambda$ = UV dose
- $\lambda$ = wavelength associated with $\log I_\lambda$ and $D_\lambda$
- $\alpha_\lambda$ = action spectrum constant at wavelength $\lambda$

A and B are constants defining the UV dose-response at 253.7 nm.

The values of the action spectra constants were identified as the constants that maximized the R-squared value of the relation between measured $\log I_\lambda$ and $D_\lambda$. The action spectra constants from repeated measurements of the UV dose-response at a given wavelength were averaged.

The full action spectra were generated by fitting the averaged values at the wavelength intervals tested with a cubic spline. Error bars represent one standard deviation.

The dose response of MS2 to MP UV light was determined given two different MP UV dose calculations. The doses were calculated as described in Linden and Darby (1997) with the spectral weighting being the standard measure of germicidal effectiveness (EPA 2006a), the DNA absorption curve, in one calculation and the MS2 action spectrum from this study in the other. The two dose response curves were compared with the dose response of MS2 to LP UV light using a dummy variable analysis.

With *Bacillus pumilus*, the UV dose response data obtained using the MP UV lamp with bandpass filters was mapped to the UV dose-response data from a LP UV lamp emitting at 253.7 nm. With all other microbes, the UV dose response data obtained from the tunable laser emitting at various wavelengths described above was mapped to the UV dose response data obtained using the tunable laser emitting at 253.7 nm.

**Adenovirus**

The relationship between the log concentration of adenovirus and UV dose was linear, showing no statistical curvature. Therefore, adenovirus data was analyzed with linear approximations. The kinetic constant for the relationship between log (N) and UV dose was obtained from regression analysis. The log reduction of adenovirus was calculated as:
Log reduction = \log (N_0) - \log (N) \quad \text{[2.2]}

Where \log (N_0) = \log \text{concentration of the unexposed sample obtained from the regression analysis (i.e. the intercept)}

\log (N) = \text{measured log concentration}

Inactivation rate constants were taken relative to their values at 253.7 nm to illustrate the action spectrum or spectral sensitivity of adenovirus to UV light emission from a LP UV lamp. ANCOVA was used to compare the linear regression for the dose response of adenovirus 2 to LP UV with its dose response to MP UV light given two different MP UV fluence calculations.

**Giardia**

The action spectrum for Giardia was developed by analyzing the relationship between \log \text{inactivation (similar to Equation 2.2) and UV dose using a linear fit of the data and determining the dose required for 2-log inactivation. The inverse of this dose was normalized to the dose required at 253.7 nm and plotted as a function of UV exposure wavelength.}

**RESULTS AND DISCUSSION**

**Surrogates**

**Coliphage**

Results from the four collimated beam trials with MS2 Coliphage are shown in Figure 2.3. At the tunable laser emission of 253.7 nm, 2-log inactivation was attained at a UV dose between 29.4 mJ/cm² and 41.8 mJ/cm². This is consistent with the literature in which 2-log inactivation was reported at LP UV doses of 32.8 mJ/cm² and 35.1 mJ/cm² (Meng and Gerba 1996; Park et al. 2011). Four of the six trials lay outside the National Water Research Institute (NWRI) bounds of 34 mJ/cm² to 54.5 mJ/cm² because the bounds assume a linear dose response where, in fact, the dose-response has curvature. The bounds were also developed using dose response data where the UV dose calculations did not account for divergence, reflection, or the non-uniformity of the light source across the Petri dish.
Figure 2.3 Dose response of MS2 Coliphage to wavelength-specific UV light from the tunable laser

The dose response data was used to determine the spectral sensitivity or action spectrum of MS2 Coliphage, which is shown in Figure 2.4, compared with two action spectra from the literature. The MS2 Coliphage action spectrum was first determined in 1965 using a large diffraction grating monochromator, which dispersed monochromatic UV light approximately 1.2 nm in bandwidth (Rauth 1965). In 2005, the action spectrum was measured with a monochromator with a maximum bandwidth of 10 nm (Mamane-Gravetz et al. 2005). The MS2 action spectrum from the present study agrees with the other two, with a relative maximum near 260 nm and an increased sensitivity below 240 nm.

Note different x-axis values. Symbols represent data from four separate trials: R1 (●) and R2 (▲) from one laboratory and R3 (+) and R4 (×) from two additional laboratories. Average standard deviation between replicate platings = 6.3 pfu/mL.
Results from the collimated beam trials with T1UV Coliphage are given in Figure 2.5, with its corresponding action spectrum in Figure 2.6. The T1UV action spectrum agreed well with that developed previously using a medium-pressure mercury arc-lamp and bandpass filters (Stefan et al. 2007) at wavelengths above 240 nm, but differed significantly below 240 nm. In both current and past studies, T1UV exhibited a relative peak sensitivity at 265 nm. Because T1UV Coliphage has similar inactivation kinetics at 253.7 nm as the UV dose-requirements for *Cryptosporidium parvum* and *Giardia lamblia*, specified by the LT2ESWTR, it is used as a surrogate organism for those pathogens for UV reactor validation. However, differences in the action spectra of T1UV and *Cryptosporidium* at wavelengths below 240 nm and above 253.7 nm may require application of an action spectra correction factor.

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4 Error bars represent 1 standard deviation from the mean sensitivity value. n = 4 for 240 nm, 253.7 nm, 260 nm, and 270 nm and n = 3 for all other wavelengths tested.

Figure 2.5 Dose response of T1UV Coliphage to wavelength-specific UV light from the tunable laser\(^5\)

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\(^5\) Note different x- and y-axis values. Symbols represent data from four separate trials: R1 (●) and R2 (▲) from one laboratory and R3 (+) and R4 (×) from two additional laboratories. Average standard deviation from replicate platings = 5.5 pfu/mL.
Results from the collimated beam trials with Q Beta Coliphage are given in Figure 2.7, with its corresponding action spectrum in Figure 2.8. The Q Beta action spectrum also agreed well with the spectrum developed previously using an MP UV lamp and bandpass filters (Stefan et al. 2007). In both studies, Q Beta exhibited a peak relative sensitivity between 260 and 265 nm. Note that the Q Beta action spectrum was very similar to that of MS2.

Source: Adapted from Stefan et al. 2007, Beck et al. 2015.

Figure 2.6 Relative spectral sensitivity of TIUV Coliphage to UV light compared to a past study

Error bars represent 1 standard deviation from the mean sensitivity value. n = 4 for 240 nm, 253.7 nm, 260 nm, and 270 nm and n = 3 for all other wavelengths tested.

Figure 2.7 Dose response of Q Beta Coliphage to wavelength-specific UV light from the tunable laser\(^7\)

Source: Adapted from Stefan et al. 2007, Beck et al. 2015.

Figure 2.8 Relative spectral sensitivity of Q Beta Coliphage to UV light compared to a past study \(n = 1\)

\(^7\) Average standard deviation from triplicate platings = 8.9 pfu/mL.
Results from the collimated beam trials with T7m and T7 Coliphage are given in Figures 2.9 and 2.10 respectively. The action spectra for T7m and T7 Coliphage are shown in Figure 2.11 as compared with the action spectra for MS2 Coliphage, T1UV, Q Beta, and Cryptosporidium. The action spectrum for T7 Coliphage matches that developed previously at 260 nm, where both exhibited a sensitivity of 1.3 (relative to 253.7 nm). However, the spectrum in the literature, obtained with a xenon lamp and a monochromator with 4 nm dispersion, decreased above 260 nm, whereas the T7 Coliphage action spectrum developed in this study increases to a relative peak at 270 nm (Ronto et al. 1992). T7m Coliphage was slightly less sensitive than T7 with a relative peak of 1.1 at 260 nm and a decreasing sensitivity at higher wavelengths.


Figure 2.9 Dose response of T7m Coliphage to wavelength-specific UV light from the tunable laser

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8 Average standard deviation from triplicate platings = 8.4 pfu/mL.
Figure 2.10 Dose response of T7 Coliphage to wavelength-specific UV light from the tunable laser

Figure 2.11 Relative spectral sensitivity of MS2, T1UV, Q Beta, T7m, and T7 Coliphage and Cryptosporidium (C. parvum) to UV light from the tunable laser

9 Average standard deviation from triplicateplatings = 6.6 pfu/mL.

10 Note data points at 200 and 300 nm are extrapolated.
Table 2.2 contains the required UV doses for 1- to 4-log inactivation of each phage tested using the tunable laser emission of 253.7 nm along with results for 2-log inactivation with low-pressure mercury vapor lamps for comparison. Inactivation rate constants were not determined because the dose response data exhibited curvature and was not fit linearly; therefore, the dose for 2-log inactivation was used as a point of comparison between phage.

Table 2.2  UV dose response results for coliphage irradiated with the tunable laser at 253.7 nm and LPUV as compared with previous studies.

<table>
<thead>
<tr>
<th>Coliphage</th>
<th>Tunable laser at 253.7 nm (± 95 % CI)</th>
<th>LPUV (± 95 % CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n 1-log  2-log  3-log  4-log</td>
<td>2-log this study</td>
</tr>
<tr>
<td>MS2</td>
<td>ssRNA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>6 16.0 (± 2.0) 33.6 (± 3.8) 53.4 (± 5.4) 77.6 (± 6.4)</td>
<td>4 32.8a (± 2.9) 32.8a</td>
</tr>
<tr>
<td>T1UV</td>
<td>dsDNA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4 4.3 (± 0.4) 8.5 (± 0.9) 12.8 (± 1.3) 17.0 (± 1.8)</td>
<td>2 8.3 (± 0.1) 35.1b</td>
</tr>
<tr>
<td>Q Beta</td>
<td>ssRNA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 10.8 22.0 33.8 46.2</td>
<td>1 19.8 23.9c</td>
</tr>
<tr>
<td>T7</td>
<td>dsDNA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1 1.6 3.6 6.6 --</td>
<td>1 3.8 --</td>
</tr>
<tr>
<td>T7m</td>
<td>dsDNA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2 1.7 (± 0.2) 3.8 (± 0.4) 6.3 (± 0.5) 10.6 (± 0.2)</td>
<td>2 3.4 (± 0.6) --</td>
</tr>
</tbody>
</table>

a(Meng and Gerba 1996); b(Park et al. 2011); c(Stefan et al. 2007)

Figure 2.12 shows a comparison of the dose response of MS2 to LP UV and MP UV light with the MP UV dose calculated two different ways. When the MP UV emission spectrum was weighted by the DNA absorption, the dose response of MS2 was significantly different from its response to LP UV light (p = 0.0036). However, when the MP UV dose was weighted by the action spectrum measured in this paper, the dose response of MS2 was similar to its response to LP UV light (p = 0.375). This theoretical comparison confirms the accuracy of the MS2 action spectrum, which was developed with respect to 253.7 nm. It also suggests that the low wavelengths, which are less prominent in a DNA weighted UV dose, play a role in MS2 coliphage inactivation.
Results from the collimated beam dose-response with Bacillus pumilus spores across the germicidal UV range are given in Figure 2.13. The action spectrum for Bacillus pumilus spore ASFUVRRC developed from these dose-response data are presented in Figure 2.14 and is consistent with that developed previously (Rochelle et al. 2010) for spores cultivated with a slightly different method (1 mmole/L MnSO₄ for 5 days). B. pumilus spores exhibited a relative peak in the 260 to 265 nm range. At 220 nm, the spores were 8 to 9 times more sensitive than at 253.7 nm. The reason behind increased effectiveness of this low wavelength light was not investigated but is postulated in more detail in Beck et al. (2014). As a point of reference, when irradiated with LP UV light (253.7 nm), 2-log inactivation resulted from a UV dose of 174.7 mJ/cm² (data not shown).

From 220 to 290 nm, the Bacillus pumilus spore action spectrum is very similar to the action spectrum for adenovirus 2 (Figure 2.14), which also was developed for this study and published previously (Beck et al. 2014). This similarity between B. pumilus spore and adenovirus is consistent with other research (Rochelle et al. 2010); however, the relative spectral sensitivity of B. pumilus spore may decrease sharply below 220 nm given the absorbance of the spore homogenate (Rochelle et al. 2010). Adenovirus 2 is more sensitive than B. pumilus spore between 200 and 220 nm.
Figure 2.13  Dose response of *B. pumilus* to wavelength-specific UV light from MP UV output filtered by bandpass filters

Source: Adapted from Rochelle et al. 2010, Beck et al. 2014, Beck et al. 2015.

Figure 2.14 Relative spectral sensitivity of *B. pumilus* (this study and previous research), MS2, and adenovirus to UV light across the germicidal wavelength range

Source: Adapted from Rochelle et al. 2010, Beck et al. 2014, Beck et al. 2015.
Pathogens

Adenovirus

Dose response data of adenovirus 2 inactivation by the NIST laser at 210 nm, 220 nm, 230 nm, 240 nm, 254 nm, 260 nm, 270 nm, 280 nm, and 290 nm is shown in Figure 2.15 as a function of UV dose.

![Dose response data of adenovirus 2 inactivation by the NIST laser at various wavelengths](image)


**Figure 2.15 Dose response of adenovirus 2 to UV irradiation from the NIST tunable laser**

From the figure, it is evident that adenovirus 2 was significantly more sensitive at the lower wavelengths of 210 - 230 nm and least sensitive at 290 nm. The inactivation rate coefficients at each wavelength were taken relative to the inactivation rate at 253.7 to determine the spectral sensitivity of adenovirus 2. The resulting spectrum is shown in Figure 2.16 with two different scales to give more definition at the higher wavelengths. The spectrum shown is a linear fit between the points. A cubic spline fit and 5th order polynomial fit both showed a possibly false minimum between 240 and 254 nm, with a spectral sensitivity as low as 0.5 relative to 254 nm. Otherwise, dose response data of adenovirus 2 demonstrated one peak at an apparent maximum of 270 nm and a valley at an apparent minimum between 240 and 260 nm. The spectrum agrees relatively well with a spectrum in the literature (Linden et al. 2007), which was obtained using a medium-pressure mercury vapor lamp and bandpass filters with a full width at half maximum (FWHM) of 10-12 nm.
The action spectrum of adenovirus 2 was used to calculate UV dose delivered by a collimated beam apparatus equipped with a medium pressure lamp. Because polychromatic UV sources such as the medium pressure mercury vapor lamp emit UV irradiation that varies in its germicidal effectiveness, (e.g., 260 nm and 300 nm are both UV emissions but 260 is much more germicidally effective than 300 nm) it is common in disinfection applications to weight the polychromatic emissions by some germicidal effectiveness weighting. A default weighting for germicidal effectiveness is the absorption spectrum of DNA (Linden and Darby 1997), however when an action spectrum is known, this is typically the preferred weighting for a specific microorganism. Figure 2.17 shows the dose response of adenovirus 2 to LP UV and MP UV light. When the MP UV dose was calculated by weighting the MP UV emission spectrum by adenoviral DNA absorption, the LP UV and MP UV results differed considerably (ANCOVA $p = 1.5 \times 10^{-4}$) and MP UV was significantly more effective than LP UV. However, when the MP UV dose was determined by weighting the MP UV emission by the adenovirus action spectrum, the dose response of adenovirus to MP UV light was similar to its response to LP UV light (ANCOVA $p = 0.592$). This theoretical comparison confirms that the adenovirus action spectrum, which was developed relative to 254 nm, is accurate. It also highlights the importance of the low wavelengths for adenovirus disinfection, which are not accounted for as strongly in dosimetry using DNA absorption weighting.

Figure 2.17 Dose response of adenovirus 2 to LP UV light and MP UV light with the MP UV dose determined by weighting the lamp emission with DNA absorption or the adenovirus action spectrum as measured by cell culture infectivity

Cryptosporidium parvum

Results from the three collimated beam trials with Cryptosporidium parvum are shown in Figure 2.18. At the tunable laser emission of 253.7 nm, 2-log inactivation was attained at an average UV dose of 1.9 mJ/cm². This is consistent with the literature in which 1.7-log inactivation was reported at a LP UV dose of 2 mJ/cm² (Shin, Linden et al. 2001). The required dose for 2-log inactivation of Cryptosporidium, stipulated in the Federal Register is a conservative 5.8 mJ/cm² (EPA 2006b). Cryptosporidium dose response exhibited a slight shoulder at each wavelength tested.
The dose response data was used to determine the spectral sensitivity or action spectrum of *Cryptosporidium parvum* relative to its response at 253.7 nm (Figure 2.19). This spectrum was compared to an action spectrum developed previously from a medium-pressure UV mercury vapor lamp and bandpass filters providing half-peak bandwidths of 9 nm to 11 nm (Linden et al. 2001). Above 240 nm, the two spectra were comparable. Below 240 nm, the spectrum developed in this study displayed a greater relative sensitivity of *Cryptosporidium parvum* to the lower UV wavelengths. This difference could be due to differences between the narrow-band light emitted from the monochromatic tunable laser used in this study and the broader output from the medium-pressure UV lamp with bandpass filters used previously. It could also be due to differences in the *Cryptosporidium* assays. Nevertheless, unlike for some of the phage presented earlier, the lower wavelengths from polychromatic lamps do not provide increased inactivation of *Cryptosporidium* relative to inactivation from LP UV light, which is consistent with the previous study (Linden et al. 2001).

11 Symbols represent data from three independent trials: R1 (●), R2 (▲), and R3 (■). Average standard deviation between triplicate platings = 8.3 oocysts.

*Source: Copied from Beck et al. 2015.*
**Giardia duodenalis**

The *Giardia* testing included development of an ID$_{50}$ and calculated MPN for UV exposures to LP, MP, and UV wavelengths of 212, 220, 240, 254, 260, 270, and 280 nm using the NIST tunable laser. UV disinfection dose-response curves were generated for each wavelength plotting log inactivation as a function of UV dose. A compilation of these plots is presented in Figure 2.20. From these data, the UV dose for 2 log inactivation was calculated for each wavelength data set. Similar to *Cryptosporidium*, the dose response of the *Giardia* illustrated its high sensitivity to UV irradiation such that any dose over 3 mJ/cm$^2$ resulted in “greater than” log inactivation, typically around 3 log inactivation (the assay limit).

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12 Error bars represent 1 standard deviation from the mean sensitivity value. n = 3 for all wavelengths except 230 nm and 290 nm, where n = 2.
Figure 2.20  UV dose response of *Giardia* cysts across the wavelengths tested\(^{13}\)

The relationship between the UV wavelength and relative effectiveness (dose required to achieve 2 log inactivation) compared to UV 254 emitted by the NIST tunable laser system is presented in Figure 2.21 along with the action spectrum of *Cryptosporidium*. The *Giardia* action spectrum was calculated based on the dose required for 2-log inactivation.

![Figure 2.21 Action spectrum for *Giardia duodenalis*, compared to *Cryptosporidium*, relative to the 254 nm dose-response](image)

\(^{13}\) Black symbols are from testing performed in August/September 2012 and red symbols are from testing in December 2012.
Based on the action spectra data, the low wavelengths did not result in any increased log inactivation over the base case of 254 nm. If there is any trend, it is for lower effectiveness at wavelengths less than 240 nm. The only data that indicated enhanced inactivation was at 260 nm and that was only about 1.5 times greater than at 254 nm. A statistical comparison was performed to compare the action spectra of *Giardia* and *Cryptosporidium* and based on this analysis there was no statistical difference between the action spectra at wavelengths above 230 nm. The comparative analysis of the *Giardia* and *Cryptosporidium* action spectra data is the subject of a forthcoming publication.
CHAPTER 3
CFD-BASED UV DOSE MODELING

In this work, UV dose models based on Computational Fluid Dynamics (CFD) were used to understand the impact of UV lamp output, sleeve UV transmittance, and water UV absorbance spectra on the inactivation of the pathogens and the validation test microbes by three commercial UV reactors equipped with MP UV lamps. Table 3.1 describes the three UV reactors. The models were developed by the Project Team through previous work conducted for the WRF project 4107 (Ho et al. 2011) and UV vendor validation by WEDECO.

Figure 3.1 provides a flowchart describing CFD-based UV dose models. The use of CFD to model UV dose delivery by UV reactors has been described in several WRF Projects (Ducoste and Linden 2006; Wright et al. 2007; Wright et al. 2009; Ho et al. 2011). This chapter describes details on how the CFD-based UV dose models were used for this work.

Table 3.1
Three commercial UV reactors modeled for this work

<table>
<thead>
<tr>
<th>Manufacturer</th>
<th>Reactor</th>
<th>Lamp operation</th>
<th>Project</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calgon</td>
<td>12” Chevron</td>
<td>1, 2 and 3 lamps</td>
<td>WRF 4107</td>
</tr>
<tr>
<td>Trojan</td>
<td>30” TrojanUVSwift™</td>
<td>10 lamps</td>
<td>WRF 4107</td>
</tr>
<tr>
<td>Xylem-WEDECO</td>
<td>Quadron 3000</td>
<td>5 lamps</td>
<td>Validation</td>
</tr>
</tbody>
</table>

Figure 3.1 Flow chart describing CFD-based UV dose models
GEOMETRIC MODEL

The geometric model for each UV reactor and the upstream and downstream piping used during validation was developed using the software Gambit (ANSYS Inc., Canonsburg, PA). The geometric model captured the significant features that promote flow jetting, short-circuiting or flow recirculation, and thus impact UV dose delivered by a reactor, such as valves, lamps, baffles, UV sensors, and piping elements. Dimensions used to define the geometric model were obtained from the validation reports and vendor drawings.

The piping was meshed using hexahedral elements using a Cooper scheme, which includes boundary layer elements. The reactor was typically meshed using a Cooper scheme through the center along the lamp axis, and a tetrahedral/hybrid mesh with a hexahedral core to transition to the outer walls. A finer grid was used in regions where flow is influenced by the reactor components. Figures 3.2, 3.3 and 3.4, respectively, show the physical models developed for the Calgon, Trojan, and Xylem-WEDECO reactors.

HYDRAULIC MODEL

The CFD software FLUENT (ANSYS Inc., Canonsburg, PA) was used to simulate the hydraulic conditions within the UV reactors. The inlet velocity to the model system was a uniform velocity field based on the modeled flow rate. The inlet velocity was applied at the entrance to the inlet piping, which was sufficiently upstream of the reactor to allow the flow to fully develop prior to entering the reactor. The outlet from the system, located downstream of the reactor, was treated as an outflow boundary. All other surfaces were treated as wall boundaries.

The k-ε realizable turbulence model was used in all cases with the standard wall function. The term “realizable” refers to the fact that the model constrains the Reynolds stresses to positive values. Neither the standard k-ε model nor the RNG k-ε model are realizable, leading to non-positive Reynolds stress terms. The realizable k-ε model is expected to provide more accurate predictions of flows that show rotation, separation, recirculation, and boundary layers with strong adverse pressure gradients.

The models were run to steady state in double precision. A converged solution was defined by an absolute residual of less than 0.001 for all solved variables. After the flow solution converged for each of the reactor models, approximately 2,000 virtual particles were released at the inlet and tracked in time as they travelled through the system. The particle paths were outputted to a text file and were used in conjunction with the UV intensity model to determine the UV dose distributions delivered by the reactor.
Figure 3.2 GAMBIT/FLUENT model of the Calgon 12” Sentinel® reactor and piping
Figure 3.3  GAMBIT/FLUENT model of the TrojanUVSwift™10L30 reactor and piping

Figure 3.4  GAMBIT/FLUENT model of the Xylem WEDECO Quadrom 3000 reactor and piping
UV INTENSITY MODEL

The UV intensity and RED analyses was performed using UVXPT software (Ho et al. 2011). UVXPT software was developed by Carollo Engineers to model UV intensity fields, UV sensor readings, UV dose delivery, and microbial inactivation with UV reactors. UVXPT uses Microsoft Excel as a user interface. The software is programmed in Visual Basic and is accessed using command buttons located on the Excel user interface.

The algorithm treats UV lamps as a line source modeled using 100 discrete points. The algorithm calculates the UV intensity field about each point source using an approach that accounts for reflection and refraction of UV light through the quartz sleeve, absorption of UV light by the water, and divergence of light due to geometry (Bolton, 2000). The UV intensity throughout the UV reactor is then calculated by adding the contributions by each point that defines each lamp within the reactor.

UV Lamp Output

Spectral outputs for new lamps were collected from two Water Research Foundation Projects – Development of a UV Knowledgebase (Wright et al. 2012) and Optimization of UV Disinfection (Wright et al. 2007). With these projects, UV intensity from new medium pressure lamps was measured using a spectral radiometer in air from 200 to 400 nm in 1 nm increments at various locations along length and about circumference of lamps. All measurements at a given wavelength with a given lamp were averaged and then normalized to give an integrated UV intensity from 250 to 275 nm of 5.63 W/m². The value of 5.63 W/m² is an arbitrary value obtained by integrating the measured results with the new lamp used with the 24” Trojan UVSWIFT™. Figure 3.5 compares the normalized data with the new lamps. The average of the normalized spectral output was used to define a standard MP lamp spectrum for this project, which is provided in Appendix A.1. When modeling a particular UV reactor, the standard spectrum was adjusted to provide a one-to-one relation between measured and predicted UV sensor readings.

![Figure 3.5 Normalized UV spectral output with new MP lamps](image-url)
As stated, the UV lamps were modeled as line sources consisting of 100 discrete points. Each point was treated as either a point or a Lambertian source. As shown in Figure 3.6, a point source radiates evenly in all directions in three-dimensional space in accordance with:

\[ I = \frac{P_n}{4\pi} \]  

(3.1)

where \( I \) = UV intensity (W/steridian) emitted by the lamp  
\( P_n \) = total UV output from the point source (W)

In contrast, a Lambertian (or cosine) source radiates in accordance with (Sasges 2006):

\[ I = \frac{P_n}{\pi^2} \times \cos(\theta) \]  

(3.2)

where \( \theta \) = angle between the emitted light and a normal to the line source

Phillips (1983) states that the emission from UV lamps depends on the optical density of the arc at the emitted wavelength, with higher optical densities behaving more like Lambertian sources. Which emission model that is most appropriate for medium pressure lamps will vary with wavelength and the lamp design. For this work, the model that gave the best correlation between predicted and measured RED was used to model the lamp.

The UV output from new medium pressure lamps will vary about the circumference due to arc deflection (Phillips 1983; Wright et al. 2007). With a horizontally aligned lamp, the emission

Figure 3.6 UV Lamps were modeled as point or cosine sources
is greater in the upwards direction and least in the downwards direction. Mayor-Smith and Templeton (2014) reported that the non-uniform output about the circumference occurred with the resonant radiation about 254 nm and the broad peak below 240 nm but did not occur with other line wavelengths. Validation data also shows the extent of the non-uniformity depends on the operating power of the lamps. For this work, the non-uniform output about the circumference was modeled using a cosine function:

\[ I = I_0 + I' \cos(\alpha) \]  

(3.3)

where \( I_0 \) = emission assuming a uniform emission about the circumference
\( \alpha \) = angular position about the circumference where 0 degrees indicates the upwards direction
\( I' \) = constant defining the amplitude of the non-uniformity from the value \( I_0 \)

With the Trojan and Calgon reactors, the value for the amplitude \( I' \) was obtained by analyzing the UV sensor data provided in the reactor’s validation report. With the WEDECO reactor, the UV sensors all viewed the lamps from the same orientation and the amplitude could not be defined. Lacking data to define the amplitude, the value was set to zero. Furthermore, with all reactors, the non-uniform output about the lamp was assumed to apply to all wavelengths, as opposed to the resonant wavelengths and wavelengths below 240 nm. Future model work should only apply the non-uniform output to resonant wavelengths around 254 nm and wavelengths below 240 nm.

**Quartz Sleeve**

UV lamps are typically housed within quartz sleeves. As shown in Figure 3.7, UV light emitted from the lamp is reflected and refracted at the air-quartz and quartz-water interfaces of the sleeves. The transmittance of UV light as it propagates through the quartz sleeve housing the lamp is calculated using:

\[ T_q = (1 - R_{aq}) \times (1 - R_{qw}) \times 10^{-\alpha_q \times d} \]  

(3.4)

where \( T_q \) = UV transmittance
\( R_{aq} \) = reflectance at the air-quartz interface
\( R_{qw} \) = reflectance at the quartz-water interface
\( \alpha_q \) = UV absorbance coefficient of the quartz material
\( d \) = distance the UV light propagates through the quartz

The reflectance at the air-quartz and quartz-water interfaces is calculated using Fresnel’s Law, which accounts for the index of refraction of the water and quartz and the angles of incidence and transmission of the UV light as it propagates through the interfaces:

\[ R_{12} = \frac{1}{2} \times \left\{ \left( \frac{n_1 \cos \theta_1 - n_2 \cos \theta_2}{n_1 \cos \theta_1 + n_2 \cos \theta_2} \right)^2 + \left( \frac{n_1 \cos \theta_2 - n_2 \cos \theta_1}{n_1 \cos \theta_2 + n_2 \cos \theta_1} \right)^2 \right\} \]  

(3.5)

where \( n_1 \) and \( n_2 \) = indices of refraction for the first and second medium
\( \theta_1 \) and \( \theta_2 \) = the incident and transmitted angles relative to the normal at the interfaces

The refraction at the interface is defined by Snell’s Law:

\[
\frac{n_1 \sin \theta_1}{n_2 \sin \theta_2} = \frac{n_1}{n_2}
\]

(3.6)

Figure 3.8 gives the spectral index of refraction for quartz and water. Figure 3.9 shows the spectral UVA of type 214, type 219, and synthetic quartz sleeves used by the model. Figure 3.10 shows an example of the UV transmittance calculated using Equation 3.4 for a 2 mm thick quartz sleeve. Data on the index of refraction and the UV absorbance of the sleeve material is tabulated in Appendices A.2 and A.3, respectively.

Refraction and reflection at air-quartz and quartz water interfaces

Figure 3.7 Transmittance of UV light from the lamp through the quartz sleeve
Figure 3.8 Index of refraction for quartz and water

Figure 3.9 UV absorbance coefficient for Type 214, Type 219, and synthetic quartz
Figure 3.10 UV transmittance of 2 mm thick quartz sleeve material for a zero degree incidence angle

Water UV Absorbance

As UV light travels through the water, it is attenuated in accordance to Beer’s Law:

\[ I = I_0 \times 10^{-axd} \]  

(3.7)

where

- \( d \) = distance through the water
- \( a \) = UV absorbance coefficient of the water (m\(^{-1}\))
- \( I_0 \) = UV intensity at the starting point
- \( I \) = attenuated UV intensity

Validation Water UV Absorbance Spectra

The spectral UV absorbance of validation waters were defined using data obtained from validation reports for the following cases:

1. Portland test facility water, Blue Lake Aquifer plus Super Hume (BLA-SH)
2. Portland test facility water, Blue Lake Aquifer plus Lignin Sulphonate (BLA-LSA)
3. Portland test facility water, Sand and Gravel Aquifer plus Super Hume (SGA-SH)
4. Portland test facility water, Sand and Gravel Aquifer plus Lignin Sulphonate (SGA-LSA)
5. New York test facility water plus Lignin Sulphonate (NY-LSA)

Data was used to define UV absorbance coefficients for UVTs at 254 ranging from 60 to 98 percent in 2 percent increments, as shown in Figures 3.11 to 3.15. Tabulated data is provided in Appendices A.4 to A.8. UVTs values in the legend are defined at 254 nm over a 1 cm pathlength.
Figure 3.11 UV absorbance spectra of Portland test facility water - Blue Lake Aquifer plus Super Hume

Figure 3.12 UV absorbance spectra of Portland test facility water - Blue Lake Aquifer plus LSA
Figure 3.13 UV absorbance spectra of Portland test facility water - Sand and Gravel Aquifer plus Super Hume

Figure 3.14 UV absorbance spectra of Portland test facility water - Sand and Gravel Aquifer plus LSA
Figure 3.15 UV absorbance spectra of New York test facility water with LSA

Divergence

As shown in Figure 3.16, as UV light propagates from the lamp into the water, it diverges. The divergence reduces the UV intensity in accordance with:

\[
I_2 = I_1 \times \frac{A_1}{A_2}
\]  \hspace{1cm} (3.9)

UVXPT calculates the areas A1 and A2 using ray tracing using a small differential angle between the two emitted rays.
Figure 3.16 Divergence of UV light

Shadowing

UV light emitted from one lamp will be reflected, refracted, and absorbed by quartz sleeve and lamp of an adjacent lamp. Resonant radiation from one lamp may be absorbed and re-emitted by the mercury atoms within an adjacent lamp. Given the complexity of these effects, UVXPT models adjacent lamps as either being transparent to UV light from an adjacent lamp or fully absorbing that light. The premise of this approach is that the true effects would lie between these two assumptions. The approach that gives the best correlation between measured and predicted UV sensor readings and REDs was used for this work.

Action Spectra

UVXPT uses the action spectrum of the microbe to calculate a germicidal UV intensity, defined as the integrated UV intensity from 200 to 320 nm weighted by the action spectrum normalized to 1.0 at 254 nm. With UVXPT, the germicidal UV intensity can be integrated in 1 or 5 nm increments, the latter shortening the time required to run the model. For this work, the action spectra presented in Chapter 2 was used within UVXPT to calculate the germicidal UV intensity.

UV SENSOR MODEL

UVXPT calculates UV sensor readings accounting for the spectral and angular response of the UV sensor. The spectral response is defined from 200 to 400 nm in 1.0 nm increments. The angular response is defined from -90 to 90 degrees in 0.1 nm increments.
Typically, the UV sensor angular response is measured in air. Yet, the UV sensor is inserted into a UV sensor port and monitors the UV light through a water-quartz interface. As such, the angular response of the UV sensor measured in air needs to be adjusted to account for the differences in refraction with an air-quartz and a water-quartz-interface (Ho et al. 2011).

The UV intensity model was calibrated by adjusting the lamp UV output to provide a one-to-one relation between measured and predicted UV sensor readings. If the model is correct, the calibrated model should predict the log inactivation and RED observed during validation to within the percentage uncertainty of the UV sensor readings and the UV validation. The measurement uncertainty of UV sensors is typically on the order of 5 to 15 percent.

UV DOSE DISTRIBUTION MODEL

UV dose delivered to each particle track was calculated by integrating the germicidal UV intensity along each particle track as a function of time. The results are used to define the UV dose distribution delivered by the reactor. Since approximately 2,000 particle tracks were generated by the hydraulic model, the UV dose distribution was defined by approximately 2,000 particle tracks.

MICROBE UV DOSE RESPONSE KINETICS

The log inactivation and RED delivered by a given UV dose distribution was determined using the microbe’s UV dose-response curve measured at 254 nm. UV dose-response curves measured during validation are typically modeled using:

\[ D = A \times \log I + B \times \log I^2 \]  

(3.10)

where \( D \) = UV dose in mJ/cm\(^2\)  
A and B = fitting coefficients provided in the validation reports  
\( \log I \) = log inactivation of the microbe

In many cases, the UV dose distribution predicted using CFD-based UV dose models extends to UV dose values much greater than the measured range used to define UV dose response relation. To provide reasonable extrapolation of the UV dose-response of bacteriophage (Ho et al. 2011), the measured UV dose-response was modeled using:

\[ \log I = a \times (1 - \exp(-b \times D)) \]  

(3.11)

To account for shoulders and tailing, the UV dose response of \( B. pumilus \) spores were modeled using:

\[ \log I = \frac{a \times b + c \times D^d}{b + D^d} \]  

(3.12)

Both equations are empirical where a through b are constants obtained by fitting the equation to UV dose-response data.
MICROBE LOG INACTIVATION AND RED

The log inactivation was determined from a UV dose distribution using:

\[
\log I = -\log \left[ \sum_{i=1}^{P} \frac{10^{-f(D_i)}}{P} \right]
\]  

(3.13)

where \( \log I \) = log inactivation of the microbe
\( D_i \) = UV dose delivered to the ith particle track
\( f() \) = microbe’s UV dose-response function expressing log inactivation as a function of UV dose
\( P \) = total number of particle tracks

The RED was then back calculated using the UV dose-response function (e.g. equation 3.11).
CHAPTER 4
ACTION SPECTRA CORRECTION FACTORS

All results presented in this chapter are based on UV dose models conducted on three commercial UV reactors with assumed UV lamp outputs, sleeve UV transmittance, and water UV absorbance spectra as described in chapter 3. The UV lamp output, sleeve UV transmittance, and water UV absorbance spectra given in Chapter 3 do not match what is given in each of those reactor’s validation reports. The results of this report were used to understand the impact of sleeve and validation water type on ASCF values and provide guidance for applying ASCF values for broad application with UV systems. Values for a particular reactor should be determined using lamp, sleeve, and water absorbance data specific to the validation of that reactor.

Because the results presented in this Chapter are based on generic data for UV lamp output, sleeve UV transmittance, and water UV absorbance spectra, the UV reactors are referred to as reactors 1, 2 and 3 as opposed to their commercial names given in Chapter 3.

BACKGROUND ON ANALYSIS

WRF Project 4421 entitled “Development of Action Spectra Correction Factors for UV Inactivation of Cryptosporidium” used CFD-based UV dose models for seven commercial UV reactors equipped with MP UV lamps to determine ASCF values for Cryptosporidium inactivation credit based on validation using MS2 phage. The analysis used interim action spectra for MS2 phage and Cryptosporidium determined through this project (WRF Project 4376). The interim action spectra for MS2 phage and Cryptosporidium were developed before the completion of the action spectra measurements for this project, and hence differ somewhat from the final MS2 and Cryptosporidium action spectra given in Chapter 2 of this report.

Project 4421 evaluated the impact of lamp aging, sleeve fouling/aging, and WTP water UVA spectra on RED delivered to MS2 and Cryptosporidium. Lamp aging and sleeve fouling/aging has a greater impact reducing UV output at wavelengths below 240 nm compared to wavelengths above 254nm. For a given UVT at 254 nm, WTP waters can have a lower or greater UV absorbance at wavelengths below 240 nm than validation waters. For this reason, benefits of UV dose delivery at wavelengths below 240 nm realized during validation may not occur at the WTP if the lamps age, the sleeve age or foul, or the water UV absorbance at wavelengths below 240 nm is greater than occurred during UV validation.

Project 4421 also evaluated the ability of current commercial UV sensors to monitor low wavelength UV dose delivery. Current commercial UV sensors have a peak spectral response around 260 to 270 nm and little if any response below 240 nm (Wright et al. 2009). The analysis showed that current commercial UV sensors do not provide adequate monitoring of UV dose delivery when the benefits of dose delivery at 240 nm are reduced due to lamp aging, sleeve fouling/aging, and changing water UV spectra.

Given that lamp aging, sleeve fouling, and changing water absorbance will occur at the WTP but current UV sensor systems will not respond to those changes, to provide public health protection, Project 4421 concluded that ASCF values should be determined where the pathogen RED is calculated without the contributions of UV light below 240 nm. The analysis on ASCFs provided in this chapter builds on this conclusion.
MODELED CONDITIONS

Table 4.1 lists the range of flow, UVT, and relative lamp output modeled with each reactor configuration. The reactors modeled included low flow reactors (0.3 to 4.9 mgd) and high flow reactors (2.9 to 42.4 mgd). The UVTs ranged from 65 to 98 percent and the range of relative lamp output ranged from 21 to 104 percent.

The models were conducted with the reactor equipped with Type 214, Type 219 or synthetic sleeves, as described in Figures 3.9 to 3.11. The models were also conducted with five validation water UVA spectra as described in Figures 3.12 to 3.16. The models were used to predict the log inactivation and RED of the validation microbes MS2, QB, T1UV, T7 and T7m bacteriophage. B. pumilus spores REDs were only predicted with Reactor 1-3 (because analysis with B. pumilus was done at the end of this project after selection of Reactor 1-3 for defining generic ASCF values). The predictions were done using the UV dose-response kinetics at 254 nm measured using the collimated beam during UV validation and the action spectra of the validation test microbes determined through this project and presented in Chapter 2. The predictions were repeated using the full action spectra of Cryptosporidium and adenovirus determined through this project and presented in Chapter 2, as well as the action spectra of Cryptosporidium and adenovirus with the values set to zero at wavelengths less than 240 nm. The log inactivation and RED of MS2 phage was also predicted with the MS2 action spectrum set to zero at wavelengths below 240 nm.

<table>
<thead>
<tr>
<th>Reactor</th>
<th>Lamp operation</th>
<th>Flow (mgd)</th>
<th>UVT (%)</th>
<th>Relative lamp output (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-1</td>
<td>1</td>
<td>0.3 – 4.8</td>
<td>70 – 98.7</td>
<td>21 – 84</td>
</tr>
<tr>
<td>1-2</td>
<td>2</td>
<td>0.3 – 4.8</td>
<td>70 – 98.6</td>
<td>26 – 95</td>
</tr>
<tr>
<td>1-3</td>
<td>3</td>
<td>0.3 – 4.9</td>
<td>70 – 98.6</td>
<td>27 – 104</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
<td>2.9 – 42.4</td>
<td>79.2 – 98.5</td>
<td>21 – 104</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>1 – 26.3</td>
<td>65 – 98</td>
<td>24 – 104</td>
</tr>
</tbody>
</table>

ASCF CALCULATION

With each modeled test condition of flow, UVT, and relative lamp output, the ASCF values were calculated using:

$$ASCF = \frac{RED_{val}}{RED_{target}}$$

(4.1)

where $RED_{val}$ = RED calculated using the UV dose-response at 254 nm and action spectra of the validation test microbe

$RED_{target}$ = the RED calculated using the UV dose-response at 254 nm of the validation test microbe but the action spectra of the target pathogen or microbe for which disinfection is required with the applications of the UV system (e.g. Cryptosporidium or adenovirus).

The ASCF values with a given reactor, sleeve type, and validation water were fitted using:
\[ ASCF = 10^a \times UVA^{b+cUVA} \times \left( \frac{RLO}{D_L \times Q} \right)^{c+dUVA+eUVA^2} \]  

(4.2)

where  
UVA = UV absorbance of the water at 254 nm  
RLO = relative lamp output  
D_L = UV sensitivity of the validation microbe  
Q = flowrate through the reactor  

\( a \) through \( e \) are constants

The UV sensitivity of the microbe is defined as the RED divided by the log inactivation of the microbe, as defined by the microbe’s UV dose response curve at 254 nm. The constants \( a \) through \( e \) were obtained by fitting Equation 4.2 using non-linear multi-variate linear regression.

Equations 4.2 was used to predict the ASCF values under operating conditions of UVT, flow, and relative lamp output that would provide 0.5 to 4.0 log inactivation credit for Cryptosporidium and adenovirus. To define those operating conditions, validation microbe REDs required for 0.5 to 4.0 log inactivation of Cryptosporidium and adenovirus were defined as a function of UVT using:

\[ RED = VF \times D_{Req} \]  

(4.3)

where  
\( D_{Req} \) = UV dose requirement specified by the LT2ESWTR  
VF = validation factor

The validation factor was calculated per the UVDGM as:

\[ VF = B_{RED} \times B_{Poly} \times \left( \frac{1 + U_{val}}{RED} \right) \]  

(4.4)

where  
\( B_{RED} \) = RED bias factor  
\( B_{Poly} \) = polychromatic bias factor  
\( U_{val} \) = uncertainty of validation in units of mJ/cm\(^2\)

The RED bias factors were taken from Appendix G of the UVDGM. The uncertainty of validation was calculated as 0.42 log multiplied by the UV sensitivity of the validation microbe, \( D_L \). The polychromatic bias was assumed to have a value of 1.0. The calculations also assumed that the uncertainty of the UV dose-response and UV sensors used during validation was set to zero. Tables 4.2 to 4.7 summarize the target MS2, QB, T1UV, T7, T7m and \( B. pumilus \) REDs for Cryptosporidium credit obtained using these calculations. Tables 4.8 to 4.10 summarize the target MS2, \( B. pumilus \), and adenovirus REDs for adenovirus credit obtained using these calculations. Target adenovirus REDs were defined because validation for adenovirus credit has been conducted using adenovirus as a test microbe. The target T7 and T7m REDs for a given level of Cryptosporidium credit and the target MS2 and adenovirus REDs for a given level of adenovirus credit do not vary with UV because the RED bias is set to a value of 1.0.
### Table 4.2
**MS2 REDs for Cryptosporidium inactivation credit as a function of UVT**

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required MS2 REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>11.2</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>17.1</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>22.4</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>27.6</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>33.0</td>
</tr>
<tr>
<td>3.0</td>
<td>12</td>
<td>38.4</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>42.9</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>50.7</td>
</tr>
</tbody>
</table>

### Table 4.3
**QB REDs for Cryptosporidium inactivation credit as a function of UVT**

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required QB REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>7.1</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>10.8</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>14.4</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>17.9</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>21.8</td>
</tr>
<tr>
<td>3.0</td>
<td>12</td>
<td>25.9</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>29.6</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>35.9</td>
</tr>
</tbody>
</table>

### Table 4.4
**T1UV REDs for Cryptosporidium inactivation credit as a function of UVT**

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required T1UV REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>4.0</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>6.1</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>8.4</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>10.7</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>13.5</td>
</tr>
<tr>
<td>3.0</td>
<td>12</td>
<td>16.6</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>19.4</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>24.8</td>
</tr>
</tbody>
</table>
Table 4.5
T7 REDs for Cryptosporidium inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required T7 REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>2.8</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>3.9</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>5.4</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>7.4</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>10.2</td>
</tr>
<tr>
<td>3.0</td>
<td>12</td>
<td>13.8</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>16.9</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>23.9</td>
</tr>
</tbody>
</table>

Table 4.6
T7m REDs for Cryptosporidium inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required T7m REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>3.5</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>4.7</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>6.4</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>8.6</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>11.6</td>
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<tr>
<td>3.0</td>
<td>12</td>
<td>15.3</td>
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<tr>
<td>3.5</td>
<td>15</td>
<td>18.4</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>25.6</td>
</tr>
</tbody>
</table>

Table 4.7
B. pumilus REDs for Cryptosporidium inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Cryptosporidium log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required T7m REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>14.4</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>24.0</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>34.9</td>
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<td>2.0</td>
<td>5.8</td>
<td>43.4</td>
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<td>60.0</td>
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<td>3.5</td>
<td>15</td>
<td>66.5</td>
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<tr>
<td>4.0</td>
<td>22</td>
<td>76.8</td>
</tr>
</tbody>
</table>
Table 4.8
MS2 REDs for adenovirus inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Adenovirus log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required MS2 REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>45.8</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>65.1</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>86.3</td>
</tr>
<tr>
<td>2.0</td>
<td>5.8</td>
<td>107.4</td>
</tr>
<tr>
<td>2.5</td>
<td>8.5</td>
<td>128.5</td>
</tr>
<tr>
<td>3.0</td>
<td>12</td>
<td>150.6</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>170.6</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>193.7</td>
</tr>
</tbody>
</table>

Table 4.9
B. pumilus REDs for adenovirus inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Adenovirus log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required Adenovirus REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>54.2</td>
</tr>
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<td>1.0</td>
<td>2.5</td>
<td>74.4</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
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<tr>
<td>4.0</td>
<td>22</td>
<td>205.1</td>
</tr>
</tbody>
</table>

Table 4.10
Adenovirus REDs for adenovirus inactivation credit as a function of UVT

<table>
<thead>
<tr>
<th>Adenovirus log Credit</th>
<th>Required UV Dose (mJ/cm²)</th>
<th>Required Adenovirus REDs (mJ/cm²) for a UVT of:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>65 %</td>
</tr>
<tr>
<td>0.5</td>
<td>1.6</td>
<td>45.8</td>
</tr>
<tr>
<td>1.0</td>
<td>2.5</td>
<td>65.1</td>
</tr>
<tr>
<td>1.5</td>
<td>3.9</td>
<td>86.3</td>
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<tr>
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<td>5.8</td>
<td>107.4</td>
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<tr>
<td>2.5</td>
<td>8.5</td>
<td>128.5</td>
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<tr>
<td>3.0</td>
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<td>150.6</td>
</tr>
<tr>
<td>3.5</td>
<td>15</td>
<td>170.6</td>
</tr>
<tr>
<td>4.0</td>
<td>22</td>
<td>193.7</td>
</tr>
</tbody>
</table>

The log inactivation values of MS2, QB, T1UV, T7 and T7m predicted by the CFD-based UV dose models with a given reactor, sleeve type, and validation water were fitted using:
\[ \log I = 10^{a'} \times UVA^{b\times UVA} \times \left( \frac{RLO}{Q \times D_L} \right)^{c\times d\times UVA + e\times UVA^2} \]  

(4.3)

where \( a' \) to \( e' \) are constants. The RED is predicted as:

\[ RED = D_L \times 10^{a'} \times UVA^{b\times UVA} \times \left( \frac{RLO}{Q \times D_L} \right)^{c\times d\times UVA + e\times UVA^2} \]  

(4.4)

Equation 4.4 was used to calculate the RLO/Q values required at a given UVT to deliver the validation microbe REDs required for *Cryptosporidium* or adenovirus credit given in Tables 4.2 to 4.10. The value of RLO/Q was then substituted into Equation 4.2 to predict the ASCF value associated with a given level of *Cryptosporidium* or adenovirus inactivation credit at a given UVT.

Equation 4.2 was used to predict ASCF values with the value of \( D_L \) set to that of the pathogen, such as *Cryptosporidium*, as opposed to that of the validation test microbe, such as MS2 phage. The log inactivation and RED delivered by a UV reactor will depend on the action spectra of the microbes, the UV dose-response kinetics at 254 nm, and the reactor’s UV dose distribution. The difference in the RED of MS2 phage and *Cryptosporidium* with a given UV dose distribution delivered by a MP UV reactor is caused by RED bias affects related to differences in the UV dose-response at 254 nm and polychromatic bias affects due to differences in the action spectra. The RED bias factor specified by the UVDGM accounts for the difference in the UV dose-response at 254 nm of the validation test microbe and the target pathogen but not the differences in action spectra (Figure 4.1). Since the RED bias accounts for the differences in the UV dose-response at 254 nm of the validation microbe and the target pathogen, the ASCF need only account for differences in the action spectra, and hence should be calculated using the \( D_L \) of the target pathogen.
Figure 4.1 The RED bias and ASCF act to transform the validation RED to a pathogen RED accounting for differences in the UV dose response at 254 nm and the action spectra.

MICROBIAL UV DOSE RESPONSE AND UV SENSITIVITY

Figure 4.2 presents the UV dose-response of the validation test microbes used in the analysis with the WEDECO UV reactor. The UV dose-response of T1UV measured using the collimated beam during validation of the reactor showed first order kinetics and was modeled using:

$$\log I = \frac{D}{a}$$  \hspace{1cm} (4.5)

where a is a constant. In contrast, the UV dose-response of MS2, QB, T7 and T7m measured with the collimated beam showed curvature and was modeled using:

$$\log I = a \times (1 - \exp(-b \times D))$$  \hspace{1cm} (4.6)
where $a$ and $b$ are constants. The UV dose-response of *B. pumilus* spores was fitted using:

$$\log I = \frac{a \times b + c \times D^d}{b + D^d}$$  \hspace{1cm} (4.7)$$

where $a$ through $d$ are constants.

Figure 4.2 UV dose response at 254 nm of validation test microbes modeled with reactor 3

The UV sensitivity of the validation microbes was defined as RED/log$I$. Because the UV dose-response of MS2, QB, T7, and T7m shows curvature, the UV sensitivity depends on the log$I$, increasing in value as the log inactivation increases. This dependence was accounted for when
fitting Equation 4.3 to the CFD-predicted log inactivation values using non-linear multi-variate regression.

The UV sensitivity of Cryptosporidium and adenovirus was defined as the UV dose required for disinfection credit, specified by the LT2ESWTR, divided by the log inactivation credit. For example, the UV sensitivity of Cryptosporidium for 3 log inactivation credit was defined as 4 mJ/cm² per log inactivation (calculated as 12 mJ/cm² / 3 log).

LOG INACTIVATION MODEL

Figure 4.3 shows the relation between log inactivation predicted by the CFD-based UV dose model and that predicted by Equation 4.3. The data is presented for the reactor 3 equipped with Type 214, Type 219 and synthetic quartz sleeves and validated with the BLA-SH water type. Equation 4.3 was fitted to all data predicted using the CFD-based model with a given sleeve type. As shown, Equation 4.3 does a good job predicting the log inactivation of the validation tests microbes as a function of UVT, flow, relative lamp output, and microbial UV sensitivity. The relations have a slope within a few percent of 1.00 and an R-squared value greater than 0.98. Similar observations were made with Equation 4.3 fitted to the predicted log inactivation with all three reactors and water types modeled. The quality of the relations show that Equation 4.4 will do a good job predicting the value of RLO/Q associated with a target RED for Cryptosporidium or adenovirus inactivation credit, and used as an input to Equation 4.2.

ASCF MODEL

Figure 4.4 shows the relations between ASCF values for Cryptosporidium credit calculated using Equation 4.1 and the values predicted using Equation 4.2 where Equation 4.2 was fitted to ASCF values for all validation microbes. The data is presented for reactor 3 equipped with Type 214, Type 219 and synthetic quartz sleeves and validated with the BLA-SH water type. As shown, Equation 4.2 fitted to all ASCF values for all microbes with a given sleeve and water type does not do a good job predicting the ASCF values. The R-squared values with reactor 3 validated with the BLA-SH water are 0.57 with Type 214 sleeves, 0.026 with the Type 219 sleeves, and 0.63 with the synthetic sleeves.

Figure 4.5 shows the relations between ASCF values for Cryptosporidium credit calculated using Equation 4.1 and the values predicted using Equation 4.2 where Equation 4.2 was fitted individually to ASCF values for a given validation microbe. The data is presented for reactor 3 equipped with Type 214, Type 219 and synthetic quartz sleeves and validated with the BLA-SH water type. In contrast to Figure 4.4, Equation 4.2 fitted to data for individual validation microbes does a good job predicting the ASCF values calculated using Equation 4.1. The overall relations have a slope of 1.0 and an R-squared that ranges from 0.95 to 0.99.

Equation 4.2 does a better job predicting ASCF values when fitted to data for individual validation microbes because each validation microbe has a unique action spectrum that differs from other validation microbes. As shown in Chapter 2, the action spectra of validation microbes differ at wavelengths below 240 nm and wavelengths above 254 nm. Because those differences have a significant impact on the ASCF values, Equation 4.2 should be fitted individually to ASCF values calculated with a given combination validation microbe and target pathogen.
Figure 4.3 Relation between CFD-predicted log inactivation of validation microbes with reactor 3 and log inactivation predicted using Equation 4.3

\[
y = 1.0014x \\
R^2 = 0.9925
\]

\[
y = 1.0019x \\
R^2 = 0.9919
\]

\[
y = 1.0134x \\
R^2 = 0.9846
\]
Figure 4.4 Relation between ASCF values for *Cryptosporidium* credit with reactor 3 calculated using Equation 4.1 and ASCF values predicted using Equation 4.2 fitted to all ASCF data.
Figure 4.5 Relation between ASCF values for Cryptosporidium credit with reactor 3 calculated using Equation 4.1 and ASCF values predicted using Equation 4.2 fitted to ASCF values for individual validation microbes.
ANALYSIS OF ASCF VALUES FOR CRYPTOSPORIDIUM CREDIT

The following slides show relations between ASCF values for Cryptosporidium credit given in the appendices and the table parameters UVT, log inactivation credit, validation water type, sleeve type, reactor type, validation test microbe, and whether or not the action spectrum of Cryptosporidium is set to zero below 240 nm.

Figures 4.6 and 4.7 compare ASCF values for 3.0 log Cryptosporidium credit based on MS2 validation calculated using the full Cryptosporidium action spectrum and the Cryptosporidium action spectrum set to zero at wavelengths below 240 nm. The comparison is provided as a function of UVT and validation water type for Reactor 3 equipped with Type 219 and synthetic quartz sleeves, respectively. With both sleeve and all validation water types, the ASCF values increase with higher UVT. With both sleeve types, the ASCF values with and without the Cryptosporidium action spectra below 240 nm set to zero are very similar with the BLA-LSA validation water. This is expected since that water type has a high UV absorbance below 240 nm, which minimizes UV dose delivery below 240 nm. With the SGA-LSA and NY-LSA water types, the ASCF values are similar at low UVTs but the values with the Cryptosporidium action spectrum set to zero below 240 nm are greater than those calculated using the full action spectrum at high UVTs. Those differences increase with higher UVT, and are most pronounced with the synthetic quartz sleeve and least pronounced with the Type 219 quartz sleeves. The similar values at low UVT occur because LSA has a high absorbance below 240 nm. Hence, as more LSA is added to lower the UVT during validation, the contributions of low wavelengths to UV dose delivery are reduced. With the BLA-SH and SGA-SH validation waters, the ASCF values calculated using the Cryptosporidium action spectrum set to zero are higher at all UVTs than the values with the full action spectra. The differences are relative small with the BLA-SH water and large with the SGA-SH water. Because SuperHume has a relatively flat UV absorbance spectra, Super Hume has a relatively uniform impact lowering UV across all wavelengths, and hence does not have as great an impact lowering UV dose delivery at wavelengths below 240 nm as does LSA. At low UVTs, the ASCF values follow the order:

BLA-LSA ~ SGA-LSA ~ NY-LSA < BLA-SH < SGA-SH

The order reflects the differences in the UV absorbance of LSA and Super Hume at wavelengths below 240 nm. At 98 % UVTs, the ASCF values follow the order:

BLA-LSA ~ BLA-SH < NY-LSA < SGA-LSA ~ SGA-SH

Because there is minimal UV absorber addition at 98 percent UVT, this order reflects the differences in the UV absorbance spectra of the source water used for validation.
Figure 4.6 Comparison of ASCF values for 3 log Cryptosporidium credit based on MS2 validation calculated using the full Cryptosporidium action spectrum (closed symbols) and the Cryptosporidium action spectrum set to zero below 240 nm (open symbols) – Reactor 3 equipped with Type 219 sleeves.
Figure 4.7 Comparison of ASCF values for 3 log Cryptosporidium credit based on MS2 validation calculated using the full Cryptosporidium action spectrum (closed symbols) and the Cryptosporidium action spectrum set to zero below 240 nm (open symbols) – Reactor 3 equipped with synthetic quartz sleeves

Figures 4.8 and 4.9 show the impact of the validation microbe action spectrum on ASCF values for 3.0 log Cryptosporidium credit with Reactor 3 equipped with Type 219 and synthetic quartz sleeves, respectively. ASCF values were calculated using the Cryptosporidium action spectrum set to zero below 240 nm. Each figure includes two charts. The top chart shows ASCF values for the BLA-LSA validation water that blocks low wavelength light, and the bottom chart shows the ASCF values for the SGA-SH validation water type that transmits low wavelength light. With Type 219 sleeves, the ASCF values are slightly greater with the SGA-SH validation water compared to the BLA-LSA water, and follow the order:
Figure 4.8 Impact of validation microbe action spectra on ASCF values for 3 log *Cryptosporidium* credit – Reactor 3 equipped with Type 219 sleeves and validated with BLA-LSA (Top chart) and SGA-SH (Bottom chart) waters

T7 > T1UV > QB > MS2 > T7m

Because the Type 219 sleeves block the low wavelength light, this order reflects the relative differences in the action spectra of these microbes at wavelengths above 240 nm. With the synthetic sleeves, the ASCF values with the SGA-SH validation water are much greater than with the BLA-LSA water because the synthetic sleeve with the SGA-SH water transmits low wavelength UV light. Because the BLA-LSA validation water has relatively high UV absorbance below 240 nm, the ASCF values follow a similar order with validation microbe type as mentioned above for the Type 219 sleeves. However, with the synthetic sleeves and the SGA-SH validation water, the ASCF values follow the order:

MS2 ~ QB > T1UV > T7 > T7m
at high UVTs and the order:

\[
T1UV \sim QB \sim T7 > MS2 \sim T7m
\]

at low UVTs. Because the combination of synthetic sleeves with SGA-SH water has the greatest effect transmitting low wavelength UV light at high UVT, the order at high UVT primarily reflects differences in the action spectra at wavelengths below 240 nm. At lower UVTs, the order is more affected by the action spectra above 240 nm.

Figure 4.9 Impact of validation microbe action spectra on ASCF values for 3 log Cryptosporidium credit – Reactor 3 equipped with synthetic quartz sleeves and validated with BLA-LSA (Top chart) and SGA-SH (Bottom chart) waters

Figure 4.10 shows the impact of the validation microbe action spectrum on ASCF values for 3.0 log Cryptosporidium credit with Reactor 1-3 equipped with Type 219 and validated with
BLA-LSA and synthetic quartz sleeves validated with SGA-SH. ASCF values were calculated using the *Cryptosporidium* action spectrum set to zero below 240 nm. Unlike Figures 4.8 and 4.9, Figure 4.10 also includes ASCF values for *B. pumilus*. The ASCF values with *B. pumilus* are notably greater than those with MS2, T1UV, T7, T7m and QB, which follow an order similar to that observed with Reactor 3. As shown in Figure 2.14, the action spectrum of *B. pumilus* spores is notably greater than that of MS2 both at wavelengths above 254 nm and below 240nm, hence giving rise to greater ASCF values.

![Graph showing impact of validation microbe action spectra on ASCF values for 3 log *Cryptosporidium* credit – Reactor 1-3 equipped with Type 219 quartz sleeves and validated with BLA-LSA (Top chart) and synthetic quartz sleeves and validated with SGA-SH (Bottom chart).](image)

*Figure 4.10 Impact of validation microbe action spectra on ASCF values for 3 log *Cryptosporidium* credit – Reactor 1-3 equipped with Type 219 quartz sleeves and validated with BLA-LSA (Top chart) and synthetic quartz sleeves and validated with SGA-SH (Bottom chart).*
Figure 4.11 shows the impact of Cryptosporidium log inactivation credit on the ASCF values with reactor 3 equipped with Type 219 sleeves and validated with MS2. Figure 4.12 shows similar data for Reactor 3 equipped with Type 214 sleeves. ASCF values were calculated using the Cryptosporidium action spectrum set to zero below 240 nm. ASCF values in all cases decreased with increasing log inactivation credit.

Figure 4.11 Impact of Cryptosporidium log inactivation credit on ASCF values at 75 and 98 percent UVT – Reactor 3 equipped with Type 219 sleeves and validated using MS2
Figure 4.12 Impact of Cryptosporidium log inactivation credit on ASCF values at 75 and 98 percent UVT – Reactor 3 equipped with synthetic quartz sleeves and validated using MS2

Figure 4.13 compares the ASCF values for 3 log Cryptosporidium inactivation credit with Reactor 3 equipped with Type 214 and synthetic sleeves. ASCF values are based on MS2 validation and calculated using the Cryptosporidium action spectrum set to zero below 240 nm. The ASCF values with Type 214 and synthetic sleeves are very similar with validation waters that absorb the low wavelength light, such as the BLA-LSA and BLA-SH validation waters across all UVTs and the SGA-LSA and NY-LSA validation water at low UVTs. The ASCF values with synthetic sleeves are greater than those with Type 214 sleeves with validation waters where low wavelength UV dose delivery becomes significant, such as the SGA-SH validation water at all UVTs and the SGA-LSA and NY-LSA validation water at high UVTs
Figure 4.13 Comparison of ASCF Values for 3 log *Cryptosporidium* credit with Reactor 3 equipped with Type 214 and synthetic sleeves and validated using MS2.

Figure 4.14 shows ASCF values for 3 log *Cryptosporidium* credit with Reactor 3 equipped with Type 219 sleeves and validated using MS2, QB and T7m. Figure 4.15 shows similar ASCF values for validation using T1UV and T7. Values were calculated using the *Cryptosporidium* action spectrum set to zero below 240 nm. Both figures show how the ASCF values compare to the UVDGM ASCF criteria of 1.06, below which the UVDGM states an ASCF does not need to be applied when defining disinfection credit. ASCF values with MS2, QB and T7m are all below the UVDGM criteria of 1.06. ASCF values with T1UV are above the criteria for the SGA-SH validation water at all UVTs and the NY-LSA and SGA-LSA waters at high UVTs, but are below those criteria for all other validation waters. ASCF values with T7 are above the criteria for all validation waters. Since the Type 219 sleeves block the low wavelength light, the values reflect the differences in the action spectra of the validation microbes relative to *Cryptosporidium* at high wavelengths above 240 nm.
With Reactor 1-3, ASCF values for Cryptosporidium credit based on *B. pumilus* validation were greater than the UVDGM 1.06 criteria with all sleeve and validation water types.

Figure 4.14 ASCF values for 3 log *Cryptosporidium* Credit with Reactor 3 equipped with Type 219 sleeves and validated using MS2 (Top), QB (middle) and T7 (bottom)
Figure 4.15 ASCF values for 3 log *Cryptosporidium* Credit with Reactor 3 equipped with Type 219 sleeves and validated using T1UV (Top) and T7 (bottom)

Figures 4.16 and 4.17 show the ASCF values for 3.0 log *Cryptosporidium* credit based on MS2 validation as a function of reactor type for Type 219 and synthetic sleeves, respectively. Each figure shows two charts, one for the BLA-LSA validation water and one for the SGA-SH validation water. Values were calculated using the *Cryptosporidium* action spectrum set to zero below 240 nm. As shown, the ASCF values vary significantly with reactor type with the highest values observed with Reactor 1-3. With Reactor 1, the values increase with the number of operating lamps. With a given reactor shell, increasing the number of lamps reduces the average water distance between lamps. Because the total UV absorbance is less over a short distance, low wavelengths better propagate and have a greater impact on UV dose delivery at lower wavelengths, hence increasing the ASCF.
Figure 4.16 ASCF values for 3 log *Cryptosporidium* credit with different reactors equipped with Type 219 sleeves and validated using MS2
Figure 4.17 ASCF values for 3 log Cryptosporidium credit with different reactors equipped with synthetic quartz sleeves and validated using MS2

ANALYSIS OF ASCF VALUES FOR ADENOVIRUS CREDIT

Table 4.11 compares the range of ASCF values for adenovirus credit based on MS2 validation calculated using Equation 4.1 using the full adenovirus action spectra and the adenovirus action spectrum set to zero at wavelengths below 240 nm. With the full action spectrum, the range to ASCF values are much less than 1.0, reflecting the observation that the action spectrum of adenovirus is greater than that of MS2 at wavelengths below 240 nm and at wavelengths above 254 nm. With the adenovirus action spectrum set to zero below 240 nm, the ASCF values are less than 1.0 with the Type 219 sleeves. They are also less than 1.0 with Type 214 and synthetic quartz sleeves validated with BLA-LSA and BLA-SH validation waters. However, with Type 214 and synthetic quartz sleeves, ASCF values range from less than 1.00 to much greater than 1.0 with the NY-LSA and SGA-LSA validation waters, and are much greater
than 1.0 with the SGA-SH validation water. ASCF values greater than 1.0 occur because the action spectrum of adenovirus is set to zero at wavelengths below 240 nm.

### Table 4.11
Range of ASCF Values for adenovirus credit based on MS2 validation with Reactor 2 calculated using Equation 4.1

<table>
<thead>
<tr>
<th>Sleeve Type</th>
<th>Validation Water</th>
<th>ASCF Range</th>
</tr>
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<tr>
<td></td>
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<td>Full Adenovirus Action</td>
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<td></td>
<td>Spectrum</td>
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<td>219</td>
<td>BLA-LSA</td>
<td>0.79 – 0.87</td>
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<td>BLA-SH</td>
<td>0.76 – 0.81</td>
</tr>
<tr>
<td></td>
<td>NY-LSA</td>
<td>0.72 – 0.86</td>
</tr>
<tr>
<td></td>
<td>SGA-LSA</td>
<td>0.71 – 0.86</td>
</tr>
<tr>
<td></td>
<td>SGA-SH</td>
<td>0.71 – 0.77</td>
</tr>
<tr>
<td>214</td>
<td>BLA-LSA</td>
<td>0.70 – 0.85</td>
</tr>
<tr>
<td></td>
<td>BLA-SH</td>
<td>0.62 – 0.75</td>
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<tr>
<td></td>
<td>NY-LSA</td>
<td>0.49 – 0.81</td>
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<tr>
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<td>SGA-SH</td>
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<tr>
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<td>BLA-SH</td>
<td>0.60 – 0.74</td>
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<td>NY-LSA</td>
<td>0.46 – 0.81</td>
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<td></td>
<td>SGA-SH</td>
<td>0.34 – 0.49</td>
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Figure 4.18 shows the ASCF values for 4 log adenovirus credit with Reactor 1-3 based on MS2 validation. ASCF values are calculated using the adenovirus action spectrum set to zero below 240 nm. With Type 219 sleeves, the ASCF values with the five validation water types range from 0.89 to 1.01 with values typically below 1.0. The values are also range from about 0.90 to 1.0 with synthetic sleeves and the BLA-LSA and BLA-SH validation waters. In contrast, ASCF values with synthetic sleeves increase from about 0.83 at 75 percent UVT to values much greater than 1.0 at 98 percent UVT with the NY-LSA and SGA-LSA validation waters. With the SGA-SH validation waters, the ASCF values with synthetic sleeves are much greater than 1.0 over all UVTs.

The order of the ASCF values for adenovirus credit with validation water type follows similar patterns as observed with Cryptosporidium. At low UVTs, the order reflects differences in the UV absorbance of LSA and Super Hume below 240 nm, and follows:

BLA-LSA ~ SGA-LSA ~ NY-LSA < BLA-SH < SGA-SH

At high UVTs, the order reflects differences in the UV absorbance of the source waters below 240 nm, and follows:

BLA-LSA ~ BLA-SH < NY-LSA < SGA-LSA ~ SGA-SH
Figure 4.18 ASCF values for 4 log adenovirus credit with Reactor 1-3 based on validation using MS2

Figures 4.19 and 4.20 shows the impact of validation microbe action spectra on ASCF values for 4 log adenovirus credit with Reactor 1-3 equipped with Type 219 and synthetic sleeves, respectively. Each figure includes two charts. The top chart showing ASCF values for the BLA-LSA validation water that blocks low wavelength light, and the bottom chart shows the ASCF values for the SGA-SH validation water type that transmits low wavelength light. ASCF values are calculated using the adenovirus action spectrum set to zero below 240 nm. With the Type 219 sleeves and both validation waters and with synthetic sleeves with the BLA-LSA validation water, the ASCF values follow the order:

*B. pum.* >> T7 ~ T1UV > QB > MS2 > T7m
In contrast, with synthetic sleeves and the SGA-SH validation waters, the ASCF values follow the order:

\[ B. \text{pum} \gg \text{MS2} > \text{QB} > \text{T1UV} > \text{T7} > \text{T7m}. \]

The order of the ASCF values for adenovirus credit with validation microbe type is similar to that observed with Cryptosporidium.

![Figure 4.19 Impact of validation microbe action spectra on ASCF values for 4 log adenovirus credit – Reactor 1-3 equipped with Type 219 sleeves and validated with BLA-LSA (Top chart) and SGA-SH (Bottom chart) waters](image-url)
Figure 4.20 Impact of validation microbe action spectra on ASCF values for 4 log adenovirus credit – Reactor 1-3 equipped with synthetic quartz sleeves and validated with BLA-LSA (Top chart) and SGA-SH (Bottom chart) waters

Figure 4.21 shows the impact of adenovirus log inactivation credit on the ASCF values with Reactor 1-3 equipped with Type 219 sleeves and validated with MS2. Figure 4.22 shows similar data for Reactor 1-3 equipped with synthetic sleeves. ASCF values were calculated using the adenovirus action spectrum set to zero below 240 nm. The ASCF values tended not to change significantly with log inactivation, with some values increasing slightly with UVT and some values decreasing.
Figure 4.21 Impact of adenovirus log inactivation credit on ASCF values at 75 and 98 percent UVT – Reactor 1-3 equipped with Type 219 sleeves and validated using MS2
Figure 4.22 Impact of adenovirus log inactivation credit on ASCF values at 75 and 98 percent UVT – Reactor 1-3 equipped with synthetic quartz sleeves and validated using MS2.

Figure 4.23 compares the ASCF values for 4 log adenovirus credit with Reactor 1-3 equipped with Type 219, Type 214, and synthetic sleeves and validated using MS2 and BLA-LSA and SGA-SH validation waters. ASCF values are calculated using the adenovirus action spectrum set to zero below 240 nm. ASCF values with synthetic sleeves somewhat greater than those with Type 214 sleeves, and both are much greater than ASCF values with Type 219 sleeves.
Figure 4.24 compares ASCF values for 4 log adenovirus credit with different UV reactors validated with Type 219 sleeves and BLA-LSA waters and validated with synthetic sleeves and SGA-SH waters. ASCF values are calculated using the adenovirus action spectrum set to zero below 240 nm. Similar to ASCF values for Cryptosporidium credit, the ASCF values for adenovirus credit vary significantly with reactor type with the highest values observed with Reactor 1-3. ASCF values also increase with the number of operating lamps.
Figure 4.24 ASCF values for 4 log adenovirus credit with different reactors validated using MS2

Figure 4.25 shows ASCF values for 4 log adenovirus credit with Reactor 1-3 based on validation conducted using adenovirus. ASCF values are calculated using Equation 4.1 with the adenovirus RED used in the numerator calculated using the full adenovirus action spectrum and the adenovirus RED used in the denominator calculated using the adenovirus action spectrum set to zero at wavelengths below 240 nm. The ASCF values for adenovirus credit based on validation conducted using adenovirus are notably greater than values based on validation conducted using MS2, T1UV, T7, QB or T7m. The higher values occur because the action spectrum of adenovirus at lower wavelengths is notably greater than the action spectrum of those five validation microbes, and hence has a greater relative impact increasing the RED used in the numerator of Equation 4.1.
Figure 4.25 ASCF values for 4 log adenovirus credit with Reactor 1-3 equipped with Type 219 (top) and synthetic (bottom) quartz sleeves and validated using adenovirus

ANALYSIS OF ASCF VALUES FOR MS2 REDS

Figure 4.26 gives ASCF values for an MS2 RED of 40 mJ/cm² with Reactor 1-3 equipped with Type 219 and synthetic quartz sleeves. ASCF values are calculated using Equation 4.1 with the MS2 RED used in the numerator calculated using the full MS2 action spectrum and the MS2 RED used in the denominator calculated using the MS2 action spectrum set to zero at wavelengths below 240 nm. ASCF values follow the patterns similar to those observed with ASCF values for Cryptosporidium and adenovirus inactivation credit.
Figure 4.26 ASCF values for an MS2 RED of 40 mJ/cm² with Reactor 1-3 equipped with Type 219 (top) and synthetic (bottom) quartz sleeves and validated using MS2.
ASCF MODEL

For a given validation water, sleeve, and validation microbe, the ASCF values for a given UVT can be modeled as a function of inactivation credit using:

\[ ASCF = 10^a \times \log b \times c^{\log I} \] \hspace{1cm} (4.8)

Coefficients \( a \), \( b \) and \( c \) of Equation 4.8 can be modeled using a third order polynomial as a function of UVT using:

\[ a = A + B \times UVT + C \times UVT^2 + D \times UVT^3 \]
\[ b = E + F \times UVT + G \times UVT^2 + H \times UVT^3 \]
\[ c = I + J \times UVT + K \times UVT^2 + L \times UVT^3 \] \hspace{1cm} (4.9)

Equation 4.9 substituted into Equation 4.8 was fitted to the ASCF values predicted using Equation 2 with Reactor 1-3 with a given sleeve and validation water type. Appendices F, G, and H give model coefficients (A through L) obtained from those fits.

Figure 4.27 compares ASCF values for Cryptosporidium credit for Reactor 1-3 determined Equation 2 to those predicted by Equations 4.8 and 4.9. The comparison is given for the reactor equipped with Type 219 sleeves, validated with BLA-LSA waters, equipped with synthetic sleeves, and validated with SGA-SH waters. Figure 4.28 provides similar comparisons for the ASCF values for adenovirus credit. As shown, Equations 4.8 and 4.9 provide accurate predictions of the ASCF values for Cryptosporidium and adenovirus credit (i.e. R-squared values > 0.99).

The ASCF values for MS2 REDs were calculated using Equation 4.1, where the MS2 RED in the numerator was calculated using the full MS2 action spectrum, while the MS2 RED used in the denominator was calculated with the MS2 action spectrum set to zero below 240 nm can be modeled as a function of RED using:

\[ ASCF = 10^a \times RED^b \times c^{RED} \] \hspace{1cm} (4.10)

The coefficients of Equation 4.10 can also be modeled as a function of UVT using Equation 4.9. Appendix I gives the model coefficients (A through L) for the ASCF values for Reactor 1-3 operating to deliver a target MS2 RED.
Figure 4.27 Comparison of ASCF values for Cryptosporidium credit given in Appendix B to values predicted using Equations 4.7 and 4.8
Figure 4.28 Comparison of ASCF values for adenovirus credit given in Appendix C to values predicted using Equations 4.7 and 4.8

GENERIC ASCF TABLES

Appendices B through E provide tables of ASCF values that can be broadly applied to MP UV systems for disinfection credit. The tables are based on the ASCF values determined with Reactor 1-3, since that configuration resulted in the most conservative values compared to the other reactor configurations modeled in this work. The ASCF values with Appendices B, C and D are predicted using Equations 4.8 and 4.9 using the coefficients in Appendices F, G, and H, respectively. The ASCF values with Appendix E are predicted using Equations 4.10 and 4.9 using the coefficients in Appendix I.
Appendix B presents ASCF values for Cryptosporidium credit calculated using the action spectrum of Cryptosporidium set to zero below 240 nm. Figure 4.29 shows a snapshot of a section of the Appendix B tables. The tables are structured with Cryptosporidium log inactivation credit and UVT in the first two columns. The remaining columns present the ASCF values for the reactor equipped with Type 214, Type 219 and synthetic quartz sleeves. With each sleeve type, ASCF values are provided for six validation microbe types.

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</table>

Figure 4.29 Structure of the ASCF tables provided in Appendix B

Appendix C provides ASCF tables for adenovirus credit calculated where the action spectrum of adenovirus is set to zero at wavelengths below 240 nm. The tables have a similar structure to the tables provided for Cryptosporidium credit.

Appendix D provides ASCF values for adenovirus credit if adenovirus is used as the validation microbe. In this case, the adenovirus RED used in the numerator of Equation 4.1 was calculated using the full adenovirus action spectrum and the adenovirus RED used in the denominator was calculated with the action spectrum of adenovirus set to zero at wavelengths below 240 nm.

Appendix E provides ASCF tables for MS2 REDs. The ASCF values are provided as a function of MS2 RED as opposed to pathogen inactivation credit.
CHAPTER 5
IMPLEMENTATION AND MONITORING

This chapter discusses the strategies developed to effectively implement and monitor action spectra correction for MP UV disinfection. The best implementation approach will be a site-specific decision, depending on the UV reactor and water system preferences. It is important the user understands the factors influencing the action spectra correction and evaluate the options to ultimately determine the best implementation approach for a particular system.

The methods for ASCF implementation depend on whether low wavelengths can be monitored by the UV reactor. Currently, there are few proven and reliable technologies to monitor low wavelengths to either utilize the benefits of low wavelength disinfection (e.g., virus inactivation) or precisely adjust for biases between the target pathogen and the validation microbe. The implementation approaches discussed in this chapter were developed to account for currently available technologies with adequate flexibility to allow for technology advancements. The following sections outline general implementation approaches and monitoring recommendations.

GOALS AND OBJECTIVES

The implementation and monitoring strategies outlined in this section are based on two primary goals and objectives 1) protect public health and 2) provide adaptable approaches to account for advances in technology.

Protect Public Health

The overall goal for UV disinfection is to provide a robust disinfection barrier for the protection of public health. In recent years it has been discovered that UV systems emitting low wavelength UV light (e.g., MP lamps) may be advantageous to water systems requiring virus inactivation due to the increased UV sensitivity of adenovirus to low wavelengths. The low wavelength UV sensitivity of adenovirus results in lower required doses compared to those presented in the Long Term 2 Enhanced Surface Water Treatment Rule, which are based on low pressure UV lamp. For Cryptosporidium or Giardia inactivation, current validation techniques may over estimate disinfection performance due to the low sensitivity of Cryptosporidium and Giardia to wavelengths below 240 nm and higher sensitivity for validation microbes (e.g., MS2). Validation microbes can also exhibit lower sensitivities than the target pathogens at wavelengths above 240 nm as well. Thus, the action spectra correction should account for all difference at both low and higher wavelengths. The implementation of the action spectra correction is aimed at accounting for the uncertainty associated with dose calculations for both virus and Cryptosporidium/Giardia inactivation. The correction for action spectra differences is reactor, site, and validation specific. UV disinfection provides many public health benefits but the disinfection performance should be accurately calculated to maintain protection of public health.
Provide Adaptable Approaches

The implementation and monitoring approaches were developed to provide a high degree of adaptability for possible technology advancements and flexibility for both current and future installations.

Allow for Future Advancements in Monitoring Technologies

Currently, UV sensor technologies are being developed to reliably measure UV intensity at lower wavelengths. The implementation approaches provide flexibility for current installations to account for low wavelength disinfection as well as provide flexibility for improved monitoring capabilities once the technology is available.

Flexibility for Current and Future Installations

The implementation approaches were developed to provide flexibility for currently validated UV reactors given low wavelength monitoring limitations as well as future validations that may be able to directly account for the presence of low wavelengths through validation modifications.

Inclusive of Both Virus and Cryptosporidium/Giardia Inactivation

The methods developed take into consideration both installations that are targeting virus or Cryptosporidium/Giardia inactivation. Cryptosporidium and Giardia are less sensitive to low wavelength UV disinfection compared to validation microbes, so it is important to ensure the ASCF calculations effectively corrects for the validation microbe’s higher UV sensitivity. However, since adenovirus has a higher UV sensitivity to low wavelengths compared to validation microbes, it may be advantageous to fully utilize the low wavelengths for disinfection credit once monitoring becomes available.

IMPLEMENTATION CONSIDERATIONS

ASCF is influenced by site-specific characteristics such as water quality, target pathogen, UV reactor configuration, and lamp aging and fouling. Implementation may also be affected by the user’s preferences for monitoring requirements. The following discusses factors that should be considered when selecting an implementation approach.

Target Pathogen

As discussed earlier, the ASCF was developed to account for the different sensitivities of target pathogens and validation microbes to UV light. The target pathogen may determine the preferred implementation approach. Cryptosporidium and Giardia are less sensitive to low wavelengths than the validation microbes typically used in the validation process. This creates the possibility that the validation process could over-predict disinfection performance. Cryptosporidium and Giardia are also more sensitive to higher wavelengths than some validation microbes but these wavelengths are monitored with current UV sensor technology. For systems targeting Cryptosporidium or Giardia, the inclusion of low wavelengths in calculating disinfection
performance provides minimal benefit but would increase monitoring requirements. Thus, an implementation approach that does not include the influence of the low wavelength may be preferred (i.e., action below 240 nm set to zero).

Virus inactivation credit is based off of adenovirus, which has a higher UV sensitivity to low wavelengths than typical validation microbes. Therefore, if targeting virus inactivation, it may be advantageous to the water system to include low wavelengths in the disinfection performance calculations and an implementation approach that includes low wavelengths could provide capital and annual operations and maintenance cost savings.

Inclusion of Low Wavelengths

Current UV sensor technologies do not allow for monitoring of low wavelengths in a UV reactor; however, the technology is under development at the time of this publication. It is recommended that disinfection credit for low wavelengths only be provided if monitoring of the low wavelengths is available. Additional monitoring recommendations are provided in the following sections. Once the monitoring technologies become available, the decision to include low wavelengths in the implementation approach will depend on the target pathogens and utility preferences. The monitoring requirements will vary depending on the developed UV sensor technology. The UV sensor technology may provide easy online monitoring or may require more operator intensive spot monitoring, similar to current UV reference sensor calibration checks. The benefits of including low wavelengths in the disinfection performance calculations should be considered with respect to any additional monitoring requirements.

Site-Specific Water Quality

The propagation of UV light through the water is highly dependent on water quality. Water quality can be highly variable and largely site-specific. Site-specific water quality may be beneficial or detrimental for low wavelength disinfection depending on the UV absorbance and the target pathogens (e.g., water with a low UV absorbance would be beneficial when targeting adenovirus inactivation). The selected implementation approach should consider the benefit of including site-specific water quality and any associated increased monitoring requirements.

Lamp Aging and Fouling

Lamp aging can affect the spectral output from lamps. Lamp fouling can also affect wavelength emission by altering the UV absorbance of the lamp sleeves. If the selected implementation approach includes low wavelengths in the disinfection performance calculations, the potential effects of lamp aging and fouling should be accounted for in the modeling or monitoring efforts. Changes to spectral output of the lamp due to aging should be verified through independent third party evaluation of the lamps and may be included in lamp aging reports. The influence of fouling on the UV transmittance of the UV lamps sleeves will be site specific and may require a fouling study to determine the rate of fouling and influence on sleeve UV transmittance.
Monitoring

As previously described, if the contribution of low wavelengths is accounted for in the disinfection performance calculations, the low wavelengths should be monitored. Monitoring will result in additional labor costs to conduct monitoring. The potential energy savings of including the low wavelengths should be evaluated against the additional costs and effort needed for monitoring. The extent of the monitoring requirements will be a function of the developed technologies.

IMPLEMENTATION OPTIONS

Choosing the best approach to implement and monitor the ASCF is a site-specific determination. Implementation guidance was grouped into three broad categories 1) Generic ASCF Tables, 2) CFD-I UV reactor-specific modeling, and 3) new validations. Table 5.1 provides a general overview of each option and additional details are provided in the sections below.

Table 5.1
Low wavelength disinfection implementation approaches

<table>
<thead>
<tr>
<th>Approach</th>
<th>Target Pathogen</th>
<th>Include Wavelengths &lt;240 nm in Disinfection Calculation</th>
<th>Site-Specific Water Quality</th>
<th>Monitoring¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Generic ASCF Tables</td>
<td>Cryptosporidium, Giardia and virus</td>
<td>No</td>
<td>No</td>
<td>No additional monitoring</td>
</tr>
<tr>
<td>CFD-I UV reactor-specific modeling</td>
<td>Cryptosporidium and Giardia</td>
<td>No</td>
<td>No</td>
<td>No additional monitoring</td>
</tr>
<tr>
<td></td>
<td>Cryptosporidium, Giardia and virus</td>
<td>Yes w/aging and fouling²</td>
<td>No</td>
<td>Additional monitoring</td>
</tr>
<tr>
<td></td>
<td>Cryptosporidium, Giardia and virus</td>
<td>Yes w/ aging and fouling²</td>
<td>Yes</td>
<td>Additional monitoring</td>
</tr>
<tr>
<td>New Validations</td>
<td>Cryptosporidium and Giardia</td>
<td>Eliminated/minimized with sleeve, water quality, and UV absorber selection</td>
<td>No</td>
<td>No additional monitoring</td>
</tr>
<tr>
<td></td>
<td>Cryptosporidium, Giardia and virus</td>
<td>Yes</td>
<td>Yes</td>
<td>Additional monitoring</td>
</tr>
</tbody>
</table>

¹ Monitoring should include measurement of lamp output and water absorbance at wavelengths below 240 nm if either wavelengths below 240 nm or site-specific water quality is included in CFD-I modeling or new validations.
² CFD-I modeling should account for changes in lamp emission spectra due to aging and fouling if action below 240 nm is included.

Use of the generic ASCF tables provides the option to select an ASCF value with no additional modeling, testing, or monitoring required. However, this approach will result in a conservative ASCF value and may result in higher capital and annual operations and maintenance costs associated with higher power consumption. CFD-I modeling or new validation techniques may require additional upfront costs but could result in annual operations and maintenance cost savings due to a more site and UV reactor specific ASCF calculation. Potential costs savings of
lowering the ASCF through CFD-I modeling or new validation work may be offset by the cost of reactor-specific modeling, cost of new validations, and any potential additional monitoring requirements, which are unknown at this point (e.g., cost of UV sensors, calibration frequency, maintenance needs).

Inclusion of wavelengths below 240 nm or site-specific water quality would be most beneficial for systems targeting virus inactivation as the inclusion of the low wavelengths has the potential to reduce the required dose for virus inactivation. A lower required dose could decrease capital as well as annual operations and maintenance costs. For systems targeting Cryptosporidium or Giardia inactivation, inclusion of low wavelength or site-specific water would provide minimal benefits while increasing monitoring requirements.

The following sections outline the recommended ASCF implementation approaches.

**Generic ASCF Tables**

The first option for utilities to calculate an ASCF value is to use the generic ASCF tables included in this report. These tables were developed by modeling a range of water quality, log inactivation, and sleeve types on five different UV reactor configurations as documented in Chapter 4. The goal for developing the generic tables was to develop ASCF values that would be conservative and applicable to a range of UV reactors. Using the tables, the selected ASCF is a function of target pathogen, validation microbe(s), UVT, log inactivation, validation water UV absorbance, and sleeve type. The generic ASCF tables are designed to be conservative and do not account for variations in lamp output or lamp spacing, which may result in lower ASCF values. Additional research is required to adjust the ASCF tables as a function of lamp spacing.

The generic ASCF tables can be implemented with no additional testing or additional modeling as may be required with other implementation approaches. However, in order to use the tables, a review of the UV reactor validation report is required. The validation water UV absorbance, sleeve type, validation microbe(s), and operating conditions must be determined.

**Target Pathogen**

ASCF calculation is a function of the target pathogen’s action spectra and UV sensitivity as compared to the validation microbe. The generic tables are first organized based on the target microorganism (i.e., Cryptosporidium, virus, MS2). The generic tables assume no action below 240 nm for all of the target pathogens, which eliminates the need for additional monitoring during operation. As discussed in Chapter 2, Giardia and Cryptosporidium have no statistically significant differences in action spectra above 240 nm. Thus, ASCF calculations in the generic tables for Cryptosporidium and Giardia are assumed identical with the action below 240 nm set to zero. Cryptosporidium does have a higher required dose between 0.5 and 3-log inactivation. The generic ASCF tables are based on Cryptosporidium log inactivation and would be conservative for Giardia.

For utilities operating based on a required MS2 RED (e.g., 40 mJ/cm²), the target pathogen would be MS2, and the ASCF calculation should be based on the UV sensitivity of MS2. Separate ASCF tables are provided for targeting MS2 (Appendix E)
Validation Water UV Absorbance

Validation water UV absorbance affects the amount of distribution of UV wavelengths and the resulting influence on the validation microbe REDs. The generic ASCF tables are organized by validation water source and UV absorber used during validation.

- Portland test facility water
  - Blue Lake Aquifer (BLA)
  - Sand and Gravel Aquifer (SGA)
- New York test facility water (NY)
- UV absorbers
  - SuperHume™ (SH)
  - Lignin Sulfonic Acid (LSA)

Water UV absorbance curves were assumed for each source water to complete the modeling for the generic ASCF tables. The UV absorbance of an individual validation can vary from the assumed curves. It is recommended that the validation water absorbance be compared to the assumed water types in the generic ASCF tables and a representative water type be selected regardless of the water source identified in validation report. The selection of a representative water source should be based on best engineering judgment given the variability of the validation data. To compare the UV absorbance of the validation water to the generic ASCF table assumptions, the calculation of the ratio of UVA_{230} and UVA_{254} over the range of validated UVTs at 254 nm is recommended (Equation 5.1). The validation water UVA at 230 nm and 254 nm should be documented in the validation report. The UVA ratios should be compared to the data provided in Table 5.2.

\[
\frac{UVA_{230}}{UVA_{254}}
\]

(5.1)

### Table 5.2

<table>
<thead>
<tr>
<th>UVT_{254}</th>
<th>BLA-LSA</th>
<th>SGA-LSA</th>
<th>BLA-SH</th>
<th>SGA-SH</th>
<th>NY-LSA</th>
</tr>
</thead>
<tbody>
<tr>
<td>98</td>
<td>7.44</td>
<td>1.74</td>
<td>7.44</td>
<td>1.74</td>
<td>2.02</td>
</tr>
<tr>
<td>96</td>
<td>5.32</td>
<td>2.42</td>
<td>4.36</td>
<td>1.46</td>
<td>2.38</td>
</tr>
<tr>
<td>94</td>
<td>4.58</td>
<td>2.67</td>
<td>3.28</td>
<td>1.37</td>
<td>2.54</td>
</tr>
<tr>
<td>92</td>
<td>4.20</td>
<td>2.79</td>
<td>2.74</td>
<td>1.32</td>
<td>2.63</td>
</tr>
<tr>
<td>90</td>
<td>3.98</td>
<td>2.86</td>
<td>2.41</td>
<td>1.29</td>
<td>2.67</td>
</tr>
<tr>
<td>88</td>
<td>3.83</td>
<td>2.91</td>
<td>2.19</td>
<td>1.27</td>
<td>2.71</td>
</tr>
<tr>
<td>86</td>
<td>3.73</td>
<td>2.94</td>
<td>2.04</td>
<td>1.26</td>
<td>2.73</td>
</tr>
<tr>
<td>84</td>
<td>3.65</td>
<td>2.97</td>
<td>1.92</td>
<td>1.25</td>
<td>2.75</td>
</tr>
<tr>
<td>82</td>
<td>3.58</td>
<td>2.99</td>
<td>1.83</td>
<td>1.24</td>
<td>2.76</td>
</tr>
<tr>
<td>80</td>
<td>3.53</td>
<td>3.00</td>
<td>1.76</td>
<td>1.23</td>
<td>2.77</td>
</tr>
</tbody>
</table>

(continued)
Table 5.2 (Continued)

<table>
<thead>
<tr>
<th>UVT254</th>
<th>BLA-LSA</th>
<th>SGA-LSA</th>
<th>BLA-SH</th>
<th>SGA-SH</th>
<th>NY-LSA</th>
</tr>
</thead>
<tbody>
<tr>
<td>78</td>
<td>3.49</td>
<td>3.02</td>
<td>1.70</td>
<td>1.23</td>
<td>2.78</td>
</tr>
<tr>
<td>76</td>
<td>3.46</td>
<td>3.03</td>
<td>1.65</td>
<td>1.22</td>
<td>2.79</td>
</tr>
<tr>
<td>74</td>
<td>3.43</td>
<td>3.04</td>
<td>1.61</td>
<td>1.22</td>
<td>2.79</td>
</tr>
<tr>
<td>72</td>
<td>3.41</td>
<td>3.05</td>
<td>1.57</td>
<td>1.22</td>
<td>2.80</td>
</tr>
<tr>
<td>70</td>
<td>3.38</td>
<td>3.05</td>
<td>1.54</td>
<td>1.21</td>
<td>2.80</td>
</tr>
<tr>
<td>68</td>
<td>3.37</td>
<td>3.06</td>
<td>1.52</td>
<td>1.21</td>
<td>2.81</td>
</tr>
<tr>
<td>66</td>
<td>3.35</td>
<td>3.06</td>
<td>1.49</td>
<td>1.21</td>
<td>2.81</td>
</tr>
<tr>
<td>64</td>
<td>3.33</td>
<td>3.07</td>
<td>1.47</td>
<td>1.21</td>
<td>2.82</td>
</tr>
<tr>
<td>62</td>
<td>3.32</td>
<td>3.07</td>
<td>1.45</td>
<td>1.20</td>
<td>2.82</td>
</tr>
<tr>
<td>60</td>
<td>3.31</td>
<td>3.08</td>
<td>1.43</td>
<td>1.20</td>
<td>2.82</td>
</tr>
</tbody>
</table>

If the validation included multiple water sources (e.g., BLA and SGA), it is recommended that the ASCF be calculated as a weighted average of the ASCF tables for each water source. The weighted average should be calculated based on the percentage of test conditions conducted with each water source.

**Sleeve Type**

The sleeve type used in validation has an effect on the ASCF due to variations in the UV transmittance of each sleeve type (Figure 3.11). The generic ASCF tables are based around three commonly used sleeve types with assumed UV transmittances for each sleeve type.

- Type 214
- Type 219
- Synthetic quartz

The transmittance of a sleeve can vary based on the manufacturer and sleeve thickness. It is recommended that the UV transmittance of the sleeves used in validation be compared to Table 5.3 to select the appropriate sleeve category in the generic tables. The validation report should document the UV transmittance (or UV absorbance) of the validated lamp sleeve over a range of wavelengths. It is recommended that the UV transmittance be evaluated at 230 nm and compared to the values in Table 5.2 to determine the appropriate sleeve type in the generic ASCF tables.

Table 5.3

<table>
<thead>
<tr>
<th>Validated Lamp Sleeve UVT at 230 nm</th>
<th>Generic ASCF Table Sleeve Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤53%</td>
<td>Type 219</td>
</tr>
<tr>
<td>&gt;53% and ≤84%</td>
<td>Type 214</td>
</tr>
<tr>
<td>&gt;84%</td>
<td>Synthetic</td>
</tr>
</tbody>
</table>
Validation Microbe

Reactors can be validated with a single or multiple validation microbes. For example, one reactor may be validated with only MS2, while another reactor may be validated with a combination of MS2, T1UV, and T7 to bracket the UV sensitivity of Cryptosporidium and Giardia. As previously discussed, ASCF is a function of the validation microbe’s action spectra. The validation microbes each have unique action spectra and could not be combined into one correction factor within the generic ASCF tables.

The generic ASCF tables provide ASCF values for the following validation organisms and target pathogens.

- MS2, T1UV, T7, Qβ, T7m and Bacillus pumilus relative to Cryptosporidium
- MS2, T1UV, T7, Qβ, T7m, and B. pumilus relative to adenovirus
- Adenovirus relative to adenovirus
- MS2 relative to MS2

Validation microbes in addition to the ones included in the generic tables can be evaluated but each microbe should be evaluated and compared to the provided validation microbes on a case-by-case basis. Alternatively, one of the other ASCF implementation approaches can be utilized for alternative validation microbes.

Each validation microbe will yield a different ASCF value for a given set of conditions; therefore, the final ASCF needs to reflect a combined ASCF value relative to each microbe used in the UV reactor’s validation testing. For validations completed with only one validation microbe or that developed independent RED equations for each validation microbe, the ASCF can be selected directly from the generic ASCF tables with no weighting. It is recommended that for validation equations that are developed using multiple validation microbes, a weighted ASCF calculation based on the percentage of testing done with each microbe should be completed (Equation 5.2).

\[
ACSF_{\text{Final}} = ASCF_{\text{Microbe 1}}(%Microbe 1) + ASCF_{\text{Microbe N}}(%Microbe N) \tag{5.2}
\]

where: ASCF_{\text{Final}} = Final ASCF value
ACSF_{Microbe 1, N} = ASCF specific to the validation microbe
% Microbe 1, N = Percent of validation test runs using each validation microbe expressed as a decimal

Final ASCF Selection

The generic ASCF tables can be used to select a single conservative ASCF value or can be used to develop an ASCF equation that is a function of log inactivation and UVT. The selection of a single ASCF value can reduce UV reactor programming modifications but could result in higher power costs.

To following steps (see also Figure 5.1) should be used to select a site-specific ASCF value based on the generic ASCF tables.

1. Select the table for the target pathogen
   a. Cryptosporidium/Giardia (Appendix B)
   b. Virus (Appendix C or D)
c. MS2 (Appendix E)

2. Compare validation water UV absorbance ratio (UVA\textsubscript{230}/UVA\textsubscript{254}) to the values in Table 5.2. Select the generic ASCF table corresponding to the water source that is most appropriate to the validation water ratios.

3. Compare the lamp sleeve UV transmittance at 230 nm to Table 5.3. Select the appropriate UV sleeve type and select the columns in the generic ASCF table that correspond to the sleeve type.

4. Determine the validation microbes used to develop the dose monitoring equations.
   a. If the dose monitoring equation is specific to one validation microbe, select the column corresponding to the validation microbe and select an ASCF value conservative for all operating conditions at the target log inactivation or utilize an ASCF prediction equation.
   b. If the dose monitoring equation is based on multiple validation microbes, calculate the percentage of test conditions utilizing each validation microbe. Calculate a weighted average ASCF value using Equation 5.2. The selected ASCF should be conservative for all operating conditions at the target log inactivation or a new equation based on the weighting can be developed.

5. If multiple water sources were used during validation. Calculate a weighted average of the selected value from step 4 based on the percentage of test conditions using each water source.
Figure 5.1. Generic ASCF Table Implementation

If a single ASCF value is desired, the highest ASCF over the potential operating range should be selected to cover all potential operating UVTs. The ASCF values in the table may be interpolated for intermediate UVT values (e.g., UVT between 90 and 95 percent). Alternatively, the ASCF can be incorporated as an equation with log inactivation and UVT as input variables to adjust the ASCF based on actual operating conditions. Appendices F, G, H, and I provide equations and coefficients that can be programmed to calculate the generic ASCF table values as a function of log inactivation and UVT. If multiple validation microbes or water sources were used during validation, the equations for each microbe and water type should be programmed and
the results of the equations should be used to calculate a weighted average based on the test conditions with each microbe or water source. Alternatively, a new equation can be developed based on the weighted averages of the ASCF tables.

The following examples show how to calculate an ASCF value based on multiple validation microbes or multiple validation water sources when targeting Cryptosporidium or virus inactivation.

**Example 5.1: Using Generic ASCF Tables for Cryptosporidium Credit**

A UV system is validated for 3-log Cryptosporidium inactivation credit with 60% of validation testing performed using MS2 and the remaining 40% performed with T1UV. The validation testing was completed using type 214 UV lamps sleeves with a UV transmittance of 82% at 230 nm. The source water was the Blue Lake Aquifer with SuperHume™ as the UV absorber.

1.) Selecting a Single ASCF
A single ASCF is selected using the steps outlined in this section and Figure 5.1.

1. Appendix B is selected based on *Cryptosporidium* being the target pathogen.

2. UV scans from the validation report were used to calculate the ratio of UVA$_{230}$/UVA$_{254}$ over the range of validated UVTs. The ratios were compared to the values presented in Table 5.2. The calculated ratio fit closest to the assumed BLA-SH ratios. The table in Appendix C for BLA-SH is selected.

3. The sleeve UV transmittance at 230 nm is 82% and is compared to Table 5.3. 82% transmittance corresponded to the assumed transmittance for type 214 sleeves.

4. The validation included MS2 and T1UV. The most conservative ASCF values from the selected table at 3-log inactivation for MS2 and T1UV are:

   ASCF$_{MS2} = 1.11$
   ASCF$_{T1UV} = 1.18$

   A weighted average ASCF value is calculated based on the dose monitoring equation being based on 60% MS2 and 40% T1UV.

   \[
   ASCF_{\text{Final}} = (0.60 \times 1.11) + (0.40 \times 1.18)
   \]

   \[
   ASCF_{\text{Final}} = 1.14
   \]

5. The validation was only completed with one water sources and does not require weighting based on water source. The final ASCF is 1.14.
Example 5.1: Using Generic ASCF Tables for Cryptosporidium Credit (continued)

2.) Equation
Equations 4.7 and 4.8 can also be used to calculate the ASCF value as a function of UVT and log inactivation. Equations and coefficients are found in Appendix F. The coefficients are selected based on the validation water, UV absorber, and sleeve type. The coefficients for this example would be as follows:

<table>
<thead>
<tr>
<th></th>
<th>MS2</th>
<th>T1UV</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>-8.48342E-02</td>
<td>6.19479E-02</td>
</tr>
<tr>
<td>B</td>
<td>-1.03052E-03</td>
<td>-1.72705E-03</td>
</tr>
<tr>
<td>C</td>
<td>7.28845E-05</td>
<td>4.91814E-05</td>
</tr>
<tr>
<td>D</td>
<td>-5.01017E-07</td>
<td>-3.17850E-07</td>
</tr>
<tr>
<td>E</td>
<td>1.32051E+00</td>
<td>3.51970E-01</td>
</tr>
<tr>
<td>F</td>
<td>-5.25590E-02</td>
<td>-1.39750E-02</td>
</tr>
<tr>
<td>G</td>
<td>6.46424E-04</td>
<td>1.60826E-04</td>
</tr>
<tr>
<td>H</td>
<td>-2.52321E-06</td>
<td>-5.63028E-07</td>
</tr>
<tr>
<td>I</td>
<td>5.32367E-01</td>
<td>7.71244E-01</td>
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<td>J</td>
<td>1.95324E-02</td>
<td>8.42765E-03</td>
</tr>
<tr>
<td>K</td>
<td>-2.53119E-04</td>
<td>-9.86096E-05</td>
</tr>
<tr>
<td>L</td>
<td>1.04561E-06</td>
<td>3.71867E-07</td>
</tr>
</tbody>
</table>

Equation 4.7 and 4.8:

\[
ASCF = 10^a \times \log^b \times e^{c \log I} \\
\begin{align*}
a & = A + B \times UVT + C \times UVT^2 + D \times UVT^3 \\
b & = E + F \times UVT + G \times UVT^2 + H \times UVT^3 \\
c & = I + J \times UVT + K \times UVT^2 + L \times UVT^3
\end{align*}
\]

The selected equation and coefficients would be programmed into the PLC to calculate the ASCF for MS2 and T1UV based on real-time monitoring of UVT and log inactivation. The final ASCF would then be weighted accordingly to the amount of testing performed with each microbe (i.e., 60% MS2, 40% T1UV).

At an operating UVT of 90% and 3-log Cryptosporidium inactivation, the calculated ASCF values would be as follow:

\[
\begin{align*}
ASCF_{MS2} & = 1.11 \\
ASCF_{T1UV} & = 1.17 \\
\end{align*}
\]

\[
ASCF_{final} = (0.60 \times 1.11) + (0.40 \times 1.17) \\
ASCF_{final} = 1.13
\]
**Example 5.2: Using Generic ASCF Tables for Adenovirus Credit**

A UV system is validated for 2-log virus inactivation credit with 100% of validation testing performed using MS2. The validation testing was completed using synthetic UV lamps sleeves with a UV transmittance of 90% at 230 nm. Validation testing was completed using Blue Lake Aquifer water for 90% of the test conditions and Sand and Gravel Aquifer water for 10% of the test conditions. The UV absorber used was SuperHume™.

1.) Single ASCF

A single ASCF is selected using the steps outlined in this section and Figure 5.1.

1. Appendix C is selected based on adenovirus being the target pathogen.

2. The validation was completed with two water sources. UV scans from the validation report were used to calculate the ratio of UVA\textsubscript{230}/UVA\textsubscript{254} over the range of validated UVTs for each water source. The ratios were compared to the values presented in Table 5.2. The calculated ratio fit closest to the assumed BLA-SH and SGA-SH ratios. First, the table in Appendix C for BLA-SH is selected.

3. The sleeve UV transmittance at 230 nm is 90% and is compared to Table 5.3. 90% transmittance corresponded to the assumed transmittance for synthetic sleeves.

4. A weighted average ASCF value based on the validation microbe is not required due to the validation being completed using only MS2. The most conservative ASCF values from the selected table at 2-log inactivation for MS2 is:

   
   \[
   \text{ASCF}_{\text{MS2,BLA-SH}} = 1.04
   \]

5. The validation was only completed with two water sources and it was decided to weight the ASCF based on each water source. Steps 1-4 were repeated for SGA-SH and the most conservative ASCF values from the selected table at 2-log inactivation for MS2 is:

   \[
   \text{ASCF}_{\text{MS2,SGA-SH}} = 1.77
   \]

A weighted average ASCF value is calculated based on the number of test conditions with each water source. The final ASCF is calculated based on the following equation.

\[
\text{ASCF}_{\text{Final}} = (0.90 \times 1.04) + (0.10 \times 1.77)
\]

\[
\text{ASCF}_{\text{Final}} = 1.11
\]
Example 5.2: Generic Tables for Adenovirus Credit (continued)

2.) Equation

Equation 4.7 and 4.8 can also be used to calculate the ASCF value as a function of UVT and log inactivation. Equations and coefficients are found in Appendix G. The coefficients are selected based on the water and sleeve types. Two equations are needed due to the validation testing being completed with two water sources. The coefficients for this example would be as follows:

<table>
<thead>
<tr>
<th></th>
<th>MS2 based on BLA and SuperHume™</th>
<th>MS2 based on SGA and SuperHume™</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>-1.83254E+00</td>
<td>-3.41853E+00</td>
</tr>
<tr>
<td>B</td>
<td>5.82260E-02</td>
<td>1.15149E-01</td>
</tr>
<tr>
<td>C</td>
<td>-6.02993E-04</td>
<td>-1.24773E-03</td>
</tr>
<tr>
<td>D</td>
<td>2.03400E-06</td>
<td>4.65406E-06</td>
</tr>
<tr>
<td>E</td>
<td>-3.61787E-01</td>
<td>-1.04000E+00</td>
</tr>
<tr>
<td>F</td>
<td>2.64245E-03</td>
<td>2.63659E-02</td>
</tr>
<tr>
<td>G</td>
<td>7.53549E-06</td>
<td>-2.57180E-04</td>
</tr>
<tr>
<td>H</td>
<td>-1.76498E-08</td>
<td>9.43094E-07</td>
</tr>
<tr>
<td>I</td>
<td>1.12700E+00</td>
<td>1.85043E+00</td>
</tr>
<tr>
<td>J</td>
<td>-4.60775E-03</td>
<td>-2.77523E-02</td>
</tr>
<tr>
<td>K</td>
<td>5.32388E-05</td>
<td>2.97287E-04</td>
</tr>
<tr>
<td>L</td>
<td>-1.99338E-07</td>
<td>-1.04648E-06</td>
</tr>
</tbody>
</table>

Equation 4.7 and 4.8:

\[ ASCF = 10^a \times \log I^b \times e^{c\log I} \]
\[ a = A + B \times UVT + C \times UVT^2 + D \times UVT^3 \]
\[ b = E + F \times UVT + G \times UVT^2 + H \times UVT^3 \]
\[ c = I + J \times UVT + K \times UVT^2 + L \times UVT^3 \]

The selected equation and coefficients would be programmed into the PLC to calculate the ASCF based on real-time monitoring of UVT and log inactivation. Since the validation was only completed with MS2 but with two water sources, it is necessary to develop a weighted ASCF value. The final ASCF would then be weighted accordingly to the amount of testing performed with each water source (i.e., 90% BLA, 10% SGA).

At an operating UVT of 90% and 2-log virus inactivation, the calculated ASCF values and weighted average would be as follow:

ASCF MS2_{BLA} = 1.03
ASCF MS2_{SGA} = 1.65

\[ ASCF_{Final} = (0.90 \times 1.03) + (0.10 \times 1.65) \]
\[ ASCF_{Final} = 1.09 \]
Computational Fluid Dynamics and Irradiance Modeling

CFD-I modeling is an approach that can be utilized to calculate a UV reactor specific ASCF based on the reactor geometry and lamp configurations as well as site-specific water quality. By accounting for reactor and site-specific conditions, CFD-I modeling allows for a more accurate calculation of an ASCF value compared to the generic ASCF tables. ASCF is heavily influenced by the distribution of UV light within a UV reactor, which can be accounted for with CFD-I modeling. CFD-I modeling may result in additional costs to complete the modeling. However, the additional costs may be recovered through lowering power costs based on a more accurate ASCF calculation.

CFD-I modeling should be completed by qualified personnel with experience modeling UV reactors. Qualified personnel may include UV manufactures, academics, or consultants with extensive CFD-I modeling experience. If modeling is completed by the UV manufacturer, independent review is not required but is recommended.

CFD-I modeling can be completed using multiple approaches that can yield similar results. To standardize CFD-I modeling for ASCF calculations, any modeling efforts should follow the procedures outlined in Chapter 3. Appendix J provided a CFD-I checklists that should be completed by the modeler to verify conformance with generally accepted CFD-I principles for ASCF calculations.

The outputs of the CFD-I modeling can include an ASCF prediction equation that models ASCF as a function of operating parameters, such as flow, UV intensity, UVT, and target pathogen UV sensitivity. This allows the ASCF calculation to be based on real-time operating conditions. Alternatively, a single conservative ASCF can be selected from the CFD-I modeling to represent all potential operating conditions.

CFD-I modeling can be used to calculate the ASCF based on the validation testing conditions. If CFD-I modeling is based on the validation testing (UV absorbance of the validation water) with no target pathogen action below 240 nm, no additional site-specific monitoring is recommended. CFD-I also provides the ability to include low wavelengths and/or site-specific water quality to more accurately calculate the ASCF for a specific application. Inclusion of the low wavelengths and site-specific water quality would be most beneficial for facilities targeting virus inactivation, which is aided by the presence of the low wavelengths. If the low wavelengths or site-specific water quality is included in the modeling, monitoring is recommended to verify operating conditions are within the evaluated range of the CFD-I modeling parameters. If operating conditions fall outside of the modeled range, the ASCF calculations would not be applicable.

New Validations

As an alternative to using the generic ASCF tables or developing complex CFD-I models, new validations could be used to calculate or reduce/eliminate the ASCF. New validation testing could fall into three categories 1) eliminating or minimizing the influence of low wavelengths; 2) measuring the influence of low wavelengths through testing of various UV absorbers and sleeve types; or 3) validations using new sensor technologies that can directly account for the presence of low wavelength in the dose monitoring equations.

If low wavelength disinfection is not beneficial (e.g., targeting Cryptosporidium or Giardia inactivation), testing could be completed to reduce the presence of low wavelengths as much as
practically possible through a combination of sleeve material, water source, and UVT absorber selection. The validation testing results could then be used as a method to empirically calculate the ASCF value by evaluating the validation microbe response with and without low wavelengths present or as a method to eliminate the ASCF if the system is validated without low wavelengths present and is operated with the same equipment (i.e., sleeve material). Depending on the ability of the testing conditions to reduce the influence of low wavelengths, new validations can be supported by CFD-I analyses to demonstrate that the validation approach either eliminated or reduced the presence of the low wavelengths.

UV sensor technologies are currently under development for monitoring low wavelengths but are currently not commercially available. As such, the design, operation, and reliability of the emerging technologies are not known. Once the UV sensor technology is available, validations could be done using the new sensor technology. This would allow dose-monitoring equations to include the low wavelengths in the RED calculations. Validation methods to effectively account for the low wavelength and dose monitoring equation development will have to be developed once the technology becomes available. If the presence of low wavelengths were an input variable to a dose monitoring equation, validation techniques would need to be modified to test a range of low wavelength contributions similar to current procedures validating a range of UVT, flow, and UV intensity. If the dose monitoring equations were adjusted to include the contribution of the lower wavelengths, the ASCF equations would also have to be adjusted to be a function of the contribution of the low wavelengths.

APPLICATIONS OF ASCF IN THE VALIDATION FACTOR

The ASCF is intended to correct the calculated validation microbe RED for biases resulting from action spectra differences. ASCF is one of several biases and uncertainty factors associated with current validation techniques. The EPA UVDGM corrects for the validation biases and uncertainties with the calculation of a Validation Factor (VF). ASCF should be implemented as a component of the polychromatic bias factor in the VF calculation as shown in Equation 5.3 below. For UV reactors with germicidal UV sensors, the polychromatic bias is typically 1 when the ASCF is not included.

\[
VF = B_{RED} \times B_{poly} \times \left(1 + \frac{U_{Val}}{100}\right)
\] (5.3)

where: 
- \(VF\) = Validation Factor
- \(B_{RED}\) = RED Bias factor
- \(B_{poly}\) = Polychromatic Bias Factor, includes correction for non-germicidal sensors and action spectra
- \(U_{Val}\) = Uncertainty of validation expressed as a percentage

VF inputs are defined in the UVDGM and should be documented in the UV reactor validation report. It is recommended that ASCF calculations be based on the approaches outlined in this report in place of the recommendations provided in the UVDGM. As discussed in chapter 1, the UVDGM approach may overcorrect for the influence of action spectra differences on disinfection performance by not accounting for the water and sleeve absorbance. Validation reports completed prior to the identification of the ASCF issue may not include ASCF calculations.
unless they are updated or amended. ASCF calculations could be included in future validation reports or revisions to existing reports.

As an input in the VF calculation, ASCF can be implemented as a single number or as a variable based on operating conditions. If a constant ASCF is selected, the ASCF should be selected to be conservative for all potential operating conditions. The ASCF can also be defined as a function of operating conditions, such as log inactivation, UVT, flow, UV sensor readings, and target pathogen UV sensitivity. Example 5.3 shows how to implement a single ASCF value within the VF.

**Example 5.3: VF Calculation using ASCF**

A UV system is targeting 2-log *Cryptosporidium* inactivation and was validated using only MS2. The UV reactor was validated with type 214 sleeves and was tested with BLA water and LSA. The design UVT is 90% UVT. ASCF values range from 0.96 to 1.10 based on Appendix B for the validation sleeve and water type. The highest ASCF value (i.e., 1.10) is selected to be conservative for all operating conditions. The following VF inputs were provided in the existing validation report for the design condition (not including ASCF):

\[
B_{\text{RED}} = 1.82 \\
B_{\text{poly}} = 1.00 \\
U_{\text{Val}} = 24\% \\
\]

\[
VF = 1.82 \times 1.00 \times \left(1 + \frac{24\%}{100}\right) \\
VF = 2.26 \\
\]

The required dose for 2-log *Cryptosporidium* inactivation is 5.8 mJ/cm² and the system was operating with a required MS2 RED of 13.1 mJ/cm².

The new VF calculation incorporating ASCF would be as follows:

\[
VF = 1.82 \times 1.10 \times \left(1 + \frac{24\%}{100}\right) \\
VF = 2.48 \\
\]

The new required MS2 RED including the ASCF would be 14.4 mJ/cm².

If the target pathogen is *Cryptosporidium* or *Giardia*, an ASCF value less than 1.0 should not be used and any value less than 1.0 should default to 1.0. The UVDGM states that an ASCF is not required if the value is less than 1.05 or 1.06. However, it is recommended that an ASCF should be calculated and applied to all MP UV reactors, even if the calculated value is less than 1.05 or 1.06.

If the target pathogen is adenovirus, an ASCF value less than 1.0 can be used to account for the disinfection benefits provided by low wavelength disinfection.

For many older installations, operation of the system may be based on a required RED and not a validated dose or log inactivation (e.g., MS2 RED of 40 mJ/cm²). These systems do not calculate a VF and the ASCF should be applied directly to the required RED. For example, if the
calculated ASCF were 1.10, the new MS2 RED target would be 44 mJ/cm² (i.e., 40 mJ/cm² x 1.10). ASCF values specific for MS2 REDs are shown in Appendix E. The water system should choose the ASCF value corresponding to the operating conditions and UV system criteria.

For existing installations, application of the ASCF value will require updating the existing programmable logic control (PLC) programming for the VF calculations to maintain the target level of disinfection. Reprogramming PLC equations may be cost prohibitive for some smaller systems. If PLC reprogramming cannot be accomplished, the ASCF could alternatively be applied to the required validated dose (i.e., validated dose x ASCF) or log inactivation depending on the operating setpoint. If log inactivation is the target setpoint, the required dose should be multiplied by the ASCF and then the new required dose should be converted to log inactivation. The ASCF should not be directly applied to the target log inactivation, as this will result in an overcorrection for the ASCF.

MONITORING GUIDANCE

The Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) states that UV reactors must be monitored in order to receive disinfection credit (40 CFR 141.720(d)(3)(i)). The UVDGM provides recommendations on how to perform the required monitoring as well as recommended monitoring frequencies. The LT2ESWTR requires that UV reactors be monitored to verify that the reactors are operating within the validated conditions. Required monitoring includes UV intensity, flow rate, lamp status, and UVT. These parameters are intended to be monitored continuously. In addition, the UVDGM also identified monitoring frequencies for evaluating the calibration of the UV sensors and UVT analyzer. Parameters critical to dose monitoring are intended to be monitored on a continuous basis and only the calibration of these instruments are monitored periodically. The goals of monitoring the low wavelengths should be selected to be consistent with the approach outlined in the UVDGM.

The influence of the low wavelength on dose calculations is a function of the lamp output as well as the UV absorbance of the water. To understand and account for the low wavelengths the lamp output and water absorbance should be monitored. The technology currently exists for monitoring UV absorbance below 240 nm using grab samples with a bench-top UV spectrophotometer. However, the ability to measure low wavelength UV absorbance in real time is limited and the reliability of the available systems for low wavelength monitoring is not well documented. Current UV sensor technology does not allow for monitoring of low wavelengths. Due to the lack of proven technologies for real-time monitoring of UV absorbance or lamp output, discrete guidance for monitoring cannot be developed at this time. Once the technologies are available and proven, utilities should work with State regulators to develop acceptable monitoring requirements and frequencies. The following sections outline general monitoring recommendations that should be considered.

Lamp Output

Current UV sensor technologies monitor a range of wavelengths, typically from approximately 240 nm to 300 nm. In order to properly monitor and account for low wavelength disinfection, a UV sensor should be used that isolates the contribution of the lower wavelengths. If a sensor measures a broad spectrum of UV light, it will not be possible to isolate the presence or absence of low wavelengths. If a UV sensor measures from 200 to 300 nm, identical UV sensor
reading could result in substantially different levels of virus inactivation. For example, waters with high nitrate concentrations experience high absorbance of low wavelengths. For a given sensor reading, the amount of low wavelength present and benefiting virus inactivation would vary based on the presence or absence of nitrate at a given sensor reading. The UV sensor and corresponding dose calculations would be incapable of isolating changes in the contribution of low wavelengths.

Low wavelength monitoring could be accomplished either by an online UV sensor or through periodic spot monitoring with a low wavelength reference sensor. Online monitoring would provide the inherent benefit of being able to monitor changes due to lamp aging or fouling in real-time but water systems with stable water quality and proven lamp aging and fouling could consider spot monitoring depending on the UV sensors under development. Lamp aging can be documented in a laboratory setting. However, fouling can be unpredictable and rapid depending on water quality. Fouling studies could be performed to define site-specific fouling rates. If spot monitoring is utilized, lamp aging and fouling characteristics should be taken into account when developing the monitoring frequency.

**UV Absorbance**

UV absorbance is typically monitored at 254 nm for UV reactor monitoring and control. Since absorbance curves below 240 nm can vary and may not track relative to 254 nm, monitoring should also include measuring the UV absorbance at the lower wavelengths unless a low wavelength UV sensor is located where it accounts for both changes in lamp output and water quality. As with lamp output, UV absorbance could be monitored via online analyzers or spot checks (e.g., grab samples). Online monitoring provides a better representation of actual operating conditions and more accurate dose calculations compared to spot checks. If spot monitoring is utilized, water quality characteristics and variability should be taken into account when developing the monitoring frequency. If spot checks are being considered, an upfront evaluation of UV absorbance variability should be conducted.
REFERENCES


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Petri, B. 2009. A Comparison of Adenovirus and MS2 Inactivation With Low-Pressure and Medium-Pressure UV. *In Proc. of the AWWA Water Quality Technology Conference (WQTC)*.


ABBREVIATIONS

A549  human lung carcinoma cells
AOP  Advanced Oxidation Process
ASCF  Action Spectra Correction Factor
ATCC  American Type Culture Collection
AWWA  American Water Works Association

BLA  Blue Lake Aquifer (Portland test facility water)

°C  degrees Celsius
California DPH  California Department of Public Health
CFD  Computational Fluid Dynamics
CFD-I  Computational Fluid Dynamics – light intensity
cm  centimeter
CO₂  carbon dioxide
CPE  cytopathic effect
d  day
DIC  differential interference contrast
DMEM  Dulbecco's Modified Eagle Medium
DNA  Deoxyribonucleic acid
ETS  Engineered Treatment Systems

FBS  fetal bovine serum
FWHM  full width at half maximum

g  gram

h  hour
HBSS  Hank’s Balanced Salts Solution
HCT-8  human ileocaecal adenocarcinoma cells

IMS  immunomagnetic separation

kHz  kilohertz

LADWP  Los Angeles Department of Water and Power
LP  low pressure
LSA  lignin sulphonate (UV absorber)
LT2  Long Term 2 Enhanced Surface Water Treatment Rule

mgd  millions of gallons per day
min  minute
mJ  millijoule
mL  milliliter
MP  medium pressure
MPC-1 magnetic particle concentrator
MPN Most Probable Number
MS2 Male-specific 2 coliphage
MWDSC Metropolitan Water District of Southern California
MWRA Massachusetts Water Resources Authority
μL microliter
μW microwatt
Nd:YAG neodymium-doped yttrium aluminum garnet
NIST National Institute of Standards and Technology
nm nanometer
np total number of particle tracks
NY New York validation test facility water
O&M Operation and Maintenance
OPO optical parametric oscillator
PBS phosphate buffered saline
pfu plaque forming units
PLC programmable logic control
Qβ Q beta coliphage
RED reduction equivalent dose
RLO relative lamp output
RPMI Roswell Park Memorial Institute
San Francisco PUC San Francisco Public Utilities Commission
SGA Sand and Gravel Aquifer (Portland test facility water)
SH Super Hume™ (UV absorber)
T1UV double-stranded DNA virus with a non-contractile tail
T7 double-stranded DNA viruses that infect *E. coli*
T7m double-stranded DNA viruses that infect *E. coli*
TYGB Tryptone Yeast Glucose Broth
EPA United States Environmental Protection Agency
Utah DEQ Utah Department of Environmental Quality
UV ultraviolet
UVDGM UV Disinfection Guidance Manual
UVT Ultraviolet Transmittance over a 1 cm pathlength
UVXPT software developed by Carollo Engineers
Washington DOH Washington Department of Health
WRF  Water Research Foundation
WTP  Water Treatment Plant