Status of EPA Development of New Ambient Water Quality Criteria for Bacteriophage

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Background

Human pathogenic enteric viruses, such as adenovirus, enterovirus and norovirus, are found in human wastewater and have been implicated as important causative agents of gastroenteritis (GE) in humans from exposure to contaminated recreational waters and consumption of contaminated shellfish. Human pathogenic enteric viruses have very low infectious doses as low as one to 10 virus particles, are highly transmissible and have biochemical characteristics that permit them to persist in bathing waters and in shellfish (EPA 2015). There is some previous work that suggests the current US Environmental Protection Agency (EPA) Ambient Water Quality Criteria, based on fecal indicator bacteria, such as *E. coli* and *enterococci*, do not adequately predict the presence of human viral pathogens in receiving waters. However, there is also no clear-cut epidemiological evidence linking viral GE outbreaks from exposure to bathing waters that do meet criteria based on fecal indicator bacteria (Dorevitch 2016). Thus, while FIB may not predict viral pathogen concentrations, it is difficult to conclude that FIB are entirely inadequate at their intended purpose – protecting public health.

Though it would be ideal to monitor concentrations of specific viral pathogen concentrations that cause GE, the primary reason that viral pathogens of concern are not used is because there are methodological limitations that make monitoring viral pathogens challenging. Human viral pathogens are not easily quantified in wastewater effluent, storm water or coastal receiving waters. Thus, similar to using bacterial indicators, viral indicators are being explored as indicators of the actual viral pathogens. Coliphage (viruses that infect *E. coli* bacteria, but not humans) have potential to be used as fecal indicator viruses as a surrogate for human pathogenic viruses. Coliphage benefit from being quantifiable in a range of water types, and specific subgroups of coliphage (e.g., somatic or F+ coliphage) have been proposed to show relationships with human health outcomes in recent epidemiological studies. As a result, a number of federal agencies and groups including US EPA, US Food and Drug Administration (FDA), and the Interstate Shellfish Sanitation Conference (ISSC) have been investigating the possibility of using coliphage in water quality and shellfish harvesting water quality management plans. EPA is considering developing new ambient water criteria under the Clean Water Act (CWA) based around viral indicators rather than the existing recommended criteria for *E. coli* and *enterococci* (EPA 2015).

EPA bacteriophage criteria development

In the US, water quality standards are the foundation of the water quality-based pollution control program mandated by the CWA. As such, water quality standards define goals for a waterbody by designating its uses, setting criteria to protect those uses and establishing provisions such as anti-degradation. Section 304(a)(1) of the CWA also requires EPA to develop criteria for water quality that accurately reflects the latest scientific knowledge. These criteria are based solely on data and scientific judgments of pollutant concentrations and environmental or human health effects; no considerations are made for cost or other implementation requirements. And, interestingly, unlike drinking water, where standards are developed by first establishing an acceptable human health risks, for ambient water quality, EPA develops a dose-response relationship and then makes a policy decision to establish acceptable risk to set the criteria.

As a first step in this criteria development process, the EPA conducted a literature review of the scientific information that will be evaluated to develop coliphage-based ambient water quality criteria for the protection of swimmers (EPA 2015). This literature review establishes that coliphages are equally good indicators of fecal contamination as EPA's currently recommended criteria for *E. coli* and *enterococci* (EPA 2015). The review also indicates that coliphages may be better indicators of viruses in some treated wastewater than bacteria, although there are a limited number of published studies, and many of these studies show that conclusions are site-specific. This is probably one of the most important limitations in development of such a criteria. A secondary limitation is that while it is anticipated that the literature review would establish that there is a public health issue associated with viruses in surface water, CDC data indicate that the relative issue associated with viruses appears to be of lesser concern than agents such as algal toxins with respect to human health (CDC 2014).

With respect to the additional activities in criteria developing, a recent presentation by EPA staff indicated that that several activities have been conducted and the EPA has proposed a schedule for the criteria development, as outlined in Table 1.

Table 1. Proposed schedule for bacteriophage criteria (Nappier 2016)

Date	Milestone
4/17/15	Review of coliphages as possible viral indicators of fecal contamination for ambient water quality
10/15/15	EPA webinar for stakeholders
3/1/16	Coliphage expert workshop. Fact sheet anticipated in summer 2016; proceedings anticipated in winter 2017.
2016	Listening sessions/webinars • Conferences (New Orleans and Chapel Hill) • States • Other stakeholders (industry/environmental groups)
Summer 2016	Analytical method multilaboratory validation
Late 2017	Draft criteria released for public view

The Coliphage Expert Workshop, held in March 2016, had the purpose of having internationally recognized experts who could engage on the topic of how best to protect public health from viral contamination of water, given currently available information. The specific goals of the workshop included obtaining input on science questions from experts in the fields of environmental microbiology, microbial risk assessment and environmental epidemiology. Additionally, the experts were to support EPA in gathering scientific insight into determination of the best coliphage type (male specific and/or somatic) for use on CWA 304(a) criteria. This included a discussion on identifying situations where these coliphage types may be most useful for preventing illness and identifying impaired waters.

Moving forward, the EPA proposes to conduct additional meta-analysis of National Epidemiological and Environmental Assessment of Recreational Water (NEEAR) and Southern California Coastal Water Research Project (SCCWRP) data. The NEEAR study data was derived from an investigation of human health effects associated with recreational water use. It was a collaborative research study between two laboratories of the EPA and the Centers for Disease Control and Prevention to investigate human health effects and rapid water quality methods associated with recreational water use. This study provided near real-time water quality measurements to better define the link between water pollution, swimming at the beach and public health. A main goal of the NEEAR study is to determine how new ways of measuring water pollution can be used effectively to protect swimmers' health. The SCCWRP data was derived from several epidemiology studies at beaches with varying characteristics between 2007 and 2014 (SCCWRP 2016).

Concurrent with the ongoing criteria derivation process, the EPA continues to work on validation of two culturable methods for bacteriophage that were used in the four Great Lakes beaches study that was conducted during summer 2015. With this background information, EPA anticipates that a draft 304(a) AWQC for viruses (coliphage) will be published for peer-review and public comment in late 2017.

Implications of bacteriophage criteria on design of municipal UV disinfection systems

Disinfection is at the heart of the sanitary and public health aspects of wastewater treatment and even secondary treated wastewater contains large numbers of pathogenic (disease causing) organisms. The purpose of wastewater disinfection is to inactivate pathogens that have not been removed in the upstream treatment process to the extent necessary to protect the public health, at some acceptable risk. This should be clearly distinguished from sterilization, which is the elimination of all microbial life from the water – which is not an objective of wastewater disinfection.

To achieve the end goal of protecting human health, UV irradiation often is used for wastewater disinfection. In order to understand how wastewater treatment plants (WWTPs) would be impacted considering a new bacteriophage criteria, it is also important to evaluate the treatment performance of UV disinfection with respect to both indicators and pathogens. This has huge potential impacts on the economics of UV disinfection and while the development of EPA criteria do not need to consider economic factors, the implementation of such a criteria could significantly impact current practices that have been demonstrated to be protective of human health for decades.

page 14 ▶

◀ page 13

UV irradiation impacts on indicators and pathogens

The germicidal action of UV irradiation is a result of photochemical reactions. When UV photons (polychromatic) are absorbed by a microbe, in bacteria, viruses or protozoans, most of the germicidal action of UV light is due to nucleic acid absorption. This is because nucleic acids absorb in the range of 240-280 nm, 10-20 times higher per weight compared to protein; although proteins, can also be involved in inactivation of microorganisms by UV (Jagger 1967). Various proteins and enzymes have been found to absorb UVB and UVC, resulting in further damage to the organisms (Harm 1980; Oguma et al. 2002; Sinha and Häder 2002).

Because most disinfection using UV irradiation is a result of disruption of nucleic acids, it is of note that although the absorption spectra of different nucleic acids are similar, nucleotide bases of DNA are adenine, guanine, thymine and cytosine, whereas RNA contains uracil instead of thymine. Nucleic acids are heterocyclic aromatic compounds that show significant absorption of UV photons; in DNA, UV absorption results in dimerization of adjacent thymine molecules, inhibiting transcription of the microbe's genetic code and reproduction. Dimers in DNA that can be formed from thymine (T) and cytosine (C) include T<>T, C<>T and C<>C, and in RNA dimers can be formed from uracil and cytosine. Cytosine dimers absorb less than thymine in the germicidal range (Harm 1980) and the quantum yield of T<>T formation is greater than for the other dimers C<>C and C<>T (Patrick and Rahn 1976). Thus, organisms rich in thymine (found only in DNA) tend to be more sensitive to UV irradiation; conversely, microbes such as MS2 bacteriophage that is a single stranded RNA virus is less sensitive to UV radiation; although, adenovirus is a DNA virus that requires very high doses of UV to achieve inactivation.

Because the nucleotide composition of genetic material varies from one organism to another, so does the sensitivity to UV disinfection. A graphical summary of low-pressure UV doses required to achieve 4-log inactivation of various bacteria, protozoans and viruses are shown in Figure 1. It is of note that there are additional confounding issues associated with interpretation of virus inactivation using medium pressure UV, which has been at the center of a significant body of recent research; and that information is not presented here.

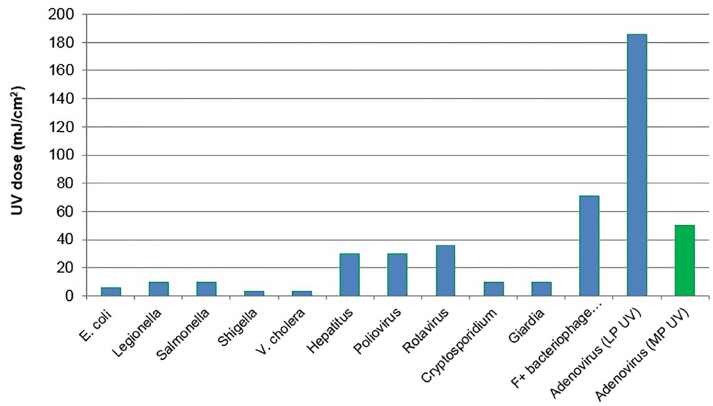

Figure 1. Low-pressure UV dose requirements for meeting 4-log inactivation of various microorganisms (EPA 2006)

Implications for NPDES permitting at wastewater treatment facilities

In the US, limits for microbial indicators are typically enforced at the "end-of-pipe," meaning that the ambient water quality criteria must be met at the end of the treatment process, before it is discharged to the receiving water body. This issue is somewhat murky in the US wastewater community because, while the EPA, in documents such as the Ephraim King Letter (EPA 2008), has indicated that there is a prohibition on the use of mixing zones for bacteria in primary contact recreation waters, individual primacy states may in fact, use mixing zones to calculate the effluent limits for bacteria. While the mixing zone calculation should be allowable, most states typically implement bacteria criteria at the end-of-pipe and utilize the criteria directly in National Pollutant Discharge Elimination System (NPDES) permits as discharge limits. However, if the ambient water quality criteria are updated to reflect coliphages, many utilities may initiate more site specific investigations to leverage the benefits of mixing zones to provide dilution factors that could be used in permitting because methods of wastewater disinfection that are most commonly employed, are not adequate to provide high levels of coliphage inactivation, although these practices already provide protection of human health (Dorevitch 2016).

Summary

The EPA develops criteria for determining when water has become unsafe for people and wildlife, using the latest scientific knowledge. Ambient water quality criteria for human health are intended to establish guidance for how much of a specific pollutant can be present in surface water before it is likely to cause harm. The EPA's commitment to develop new bacteriophage criteria by 2017 for public review should strengthen public health protection compared to the existing 2012 criteria and provide a mechanism for the various Clean Water Act needs to be met. While the EPA has conducted work toward a bacteriophage criteria, there are additional policy decisions that will establish numeric criteria that support derivation of effluent limits in NPDES permits. It is this information that is critical in understanding how new criteria could impact the design of UV systems for WWTPs. As a result, it will be important for the UV community to participate in EPA stakeholder events and provide new information to the EPA during this process if it emerges during the ongoing criteria development process. ■

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