Development of Concentration Method for Adenovirus and Quantification of Its Removal and Inactivation Efficacies in Soil Aquifer Treatment

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1. Background

Many parts of the world are facing serious water shortage due to the uneven distribution of water resources and the concentration of the population in large cities. Also, extreme weather conditions caused by global climate change may further complicate the situation. Water reuse has become an important option to amend the limited access to water sources. However, membrane-based systems are not always cost- and energy- effective. In this study, the performance of soil aquifer treatment (SAT) was evaluated with respect to pathogens especially viruses.

2. Objectives

This research aims to establish an appropriate method for collecting adenovirus (AdV) removal and inactivation efficacies applicable to quantitative microbial risk assessment. Specific goals are as follows: (1) to establish a AdV concentration procedure for water samples at very low level, (2) to estimate removal efficacy of AdV in a pilot-scale SAT reactor, and (3) to evaluate inactivation efficacy of AdV in SAT.

3. Methods

One liter of SAT influent sample (treated wastewater) and 50 L SAT effluent sample were collected periodically to establish concentration methods for adenovirus. After concentrating the SAT effluent down to 1 L by ultrafiltration with a membrane of MWCO 50,000, both SAT influent and the preconcentrated SAT effluent samples were further concentrated by polyethylene glycol (PEG) precipitation. For the ultrafiltration, NaPP and Tween 80 were added for blocking ultrafilter cartridges prior to use and for eluting viruses, respectively. The recovery of AdV during each concentration step was determined by spike tests with AdV40. AdV concentrations were determined using a quantitative PCR (qPCR) method. Infectious AdV inactivation by concentrations was estimated from a virus infectivity assay consisting of viral culture using Caco-2 cell lines and the following PCR detection of intracellular virus genome. The removal and inactivation efficiencies of AdV in SAT were estimated from the infectious AdV concentrations in SAT influent and effluent samples concentrated by the established methods.

4. Results and Discussion

The recovery of AdV was greatly improved by optimizing the centrifuge separation, blocking by NaPP, and adding the elution step with Tween 80 (hereinafter called Tween 80 method). During the Tween 80 method, the infectivity of AdV in SAT effluent was not lost. This result shows the effectiveness of this method for the concentration of AdV in SAT effluent.

The established methods revealed the AdV removal and inactivation efficacies to be 0.7-1.2 $\log_{10}$ and 1.6 $\log_{10}$, respectively. The inactivation efficacy was slightly higher than the removal efficacy, indicating that both physico-chemical and biochemical mechanisms are involved in the reduction of AdV infectivity in SAT.

Table 1 The recovery of AdV in SAT effluent during ultrafiltration and PEG precipitation

	1st (%)	2nd (%)	3rd (%)	4th (%)	Avg (%)	*CV* (%)
Old method	0.0007	-	0.0588	0.159	0.0728	89.8
NaPP method	0.558	21.3	17.9	9.31	12.3	65.4
Tween 80 method	28.7	33.0	15.3	21.9	24.7	27.2