Diversity of DNA viruses in Membrane Bioreactor Effluents in France and the United States

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Introduction

Metagenomic analysis has been applied to investigate viral diversity in wastewater, but previous studies have focused primarily on raw untreated wastewater and activated sludge, with little research directed towards studying viral diversity in treated wastewater effluent. Still less research has been performed to study the impact on viral diversity of the wastewater treatment process performed (e.g., conventional activated sludge vs. membrane bioreactor) and the disinfection method utilized, if any.

Objectives:
1) Investigate what human DNA viruses are detected in wastewater effluents
2) Assess the diversity of DNA bacteriophages in wastewater effluents
3) Describe and compare the diversity of DNA viruses in conventional and MBR wastewater treatment plant effluents
4) Investigate the impact of disinfection on DNA virus diversity in wastewater effluent.

Methods

Sample Collection: Treated effluent samples were collected from La Grande-Motte (LGM) in 2015 and East Lansing (EL) WWTP and Traverse City (TC) WWTP in 2013. Samples after disinfection were also taken for the two Michigan utilities. Argonide (electrosporative cartridge) filters were used in the filtration and concentration of the samples for viral isolation. Approximately 400 liters of effluent samples were passed through the sampler at a rate of about 11.2 L/min.

Sample Processing: All samples collected were eluted 12-24 hours after initial sampling. Further concentration of the eluate was performed by placing 500 mL into a bottle and centrifuged for 15 min at 2500-g at 4 °C. The supernatant was loaded into a 60 mL syringe and passed through a 0.22 mm sterilized filter for removal of bacteria, fungi and other contaminating agents. Virus DNA was extracted using a MagNA Pure Compact DNA extractor.

Sequencing: Virus-enriched DNA isolated from the samples was sequenced on an Illumina Hiseq platform. Sequences were assembled into contigs and diversity analysis was conducted using the MetaVir analysis platform. Principal component analysis was then performed using the Real Statistics Resource Pack software for Microsoft Excel.

Results

Viral diversity and sequence affiliation consistent across all three wastewater effluents
Disinfection introduces differentiation in viral diversity
Released effluent, after disinfection samples show significant differences in diversity to receiving surface waters according to PCA
Herpesviridae most commonly detected human viral order across all samples; much more common than the frequently studied Adenoviridae

References


Acknowledgments

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Table 1: Wastewater treatment plant characteristics.

<table>
<thead>
<tr>
<th>Sampling Locations</th>
<th>EAST LANSING</th>
<th>TRAVERSE CITY</th>
<th>LA GRANDE-MOTTE</th>
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</thead>
<tbody>
<tr>
<td>Wastewater Treatment Plants</td>
<td>Biological Reactor</td>
<td>Biological Reactor</td>
<td>Biological Reactor</td>
</tr>
<tr>
<td>Amount (MGD)</td>
<td>15.4</td>
<td>17.5</td>
<td>17.0</td>
</tr>
<tr>
<td>Average Flow</td>
<td>12.4 MGD</td>
<td>14.9 MGD</td>
<td>15.3 MGD</td>
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<tr>
<td>Capacity (MGD)</td>
<td>18.8 MGD</td>
<td>20.8 MGD</td>
<td>20.0 MGD</td>
</tr>
<tr>
<td>Disinfection</td>
<td>UV</td>
<td>UV</td>
<td>UV</td>
</tr>
</tbody>
</table>

Conclusions

Method of disinfection is more significant determinant of viral diversity than wastewater treatment process utilized
Released disinfectant could have impact on receiving surface waters whose viral diversity is dissimilar to that of the treated effluent
Viral order Herpesviridae could be a more useful indicator of human pollution due to higher levels of detection
Large majority of sequences unaffiliated with any known genome; more robust genomic data still required to fully assess biological diversity of a sample