

Report for 2003DE25B: Undergraduate Internship: Enumeration of Aquatic RNA Viruses from a Mixed Viral Sample

- Water Resources Research Institute Reports:
 - Simon, Matthew, K. Eric Wommack, Kurt E. Williamson, 2003, Enumeration of Aquatic RNA Viruses from a Mixed Viral Sample, Delaware Water Resources Center, University of Delaware, Newark, Delaware, 11 pages.

Report Follows

Undergraduate Internship Project #6 of 10 for FY03

The project is co-sponsored by the *University of Delaware College of Marine Studies (CMS) and the DWRC*. Mr. Simon hopes to better characterize viruses, which are now known to be the most abundant life-form in natural waters and also to be efficient transformers of bacteria and plankton into organic matter.

“I have been using epifluorescence microscopy to detect and quantify viruses. Ultimately, I hope to extract viruses from soils in a number of ways, discovering which are most efficient at helping us understand the biological diversity of viral communities in soils.”

– Matt Simon, University of Delaware undergraduate junior, Biology major.

Abstract:

Many studies have demonstrated a high abundance of viruses in aquatic ecosystem and there seems to be a growing interest in viruses. However, methods used to date have focused on dsDNA viruses and little effort has been made to determine the overall abundance of RNA viruses. In this study, we attempted to determine the efficacy of quantifying RNA viruses from a mixed sample using epifluorescence microscopy (EFM). Prior to enumeration of RNA viruses, it was necessary to determine if unencapsidated viral nucleic acids could be visualized using EFM. This was accomplished by using dsDNA bacteriophage T4 as a model system. Viral capsids were lysed using physical (heat) and enzymatic (Proteinase K) treatments. Additionally, attempts were made to visualize naked DNA of λ -HindIII (~23 kb) under EFM. Phage T4 seemed to be resistant to protease treatment since the number of viruses in protease-treated samples did not differ significantly from untreated controls. When phage capsids were denatured by heating, discrete virus particles could not be detected. The λ -HindIII preparations provided further evidence that naked DNA cannot be enumerated using EFM. The inability to resolve un-encapsidated DNA as countable units demonstrated that discrimination of RNA viruses from a mixed sample is not possible using EFM techniques.