

Exploring the diversity of DNA viruses in the North Sea using a metagenomic approach

Shaikh Hisham M.¹, Sheam Md Moinuddin², Piedade Gonçalo J.³, De Keulenaer Sarah⁴, Van Nieuwerburgh Filip⁴, Brussaard Corina³ and De Rijcke Maarten¹

¹ Research department, Flanders Marine Institute (VLIZ), InnovOcean Campus, Jacobsenstraat 1, 8400 Oostende, Belgium
E-mail: hisham.shaikh@vliz.be

² International Master of Science in Marine Biological Resources, Ghent University, Ghent, Belgium

³ Department of Marine Microbiology and Biogeochemistry, NIOZ—Royal Netherlands Institute for Sea Research, Den Burg, The Netherlands

⁴ NXTGNT, Faculty of Pharmaceutical Sciences, Ghent University, Ghent, Belgium

Infamous for causing diseases and wreaking havoc, viruses also play significant roles in keeping our planet functional. They are the most abundant biological entities in the ocean and represent the greatest reservoir of genetic diversity on Earth. Viruses play crucial roles in driving global biogeochemical cycles by (re-)shaping planktonic community structure and influencing microbial host metabolism and evolution through lysis and horizontal gene transfer. Some viruses (*i.e.* temperate viruses) may also enter a lysogenic cycle where, instead of killing their hosts, they incorporate themselves within the host genome and proliferate with the host cell. The choice between lytic and lysogenic cycle of temperate viruses is dependent on interactions with their hosts and environmental factors. Understanding the diversity of viruses and their interactions is imperative in understanding the functioning of an ecosystem.

The North Sea is a relatively shallow but highly productive sea that receives input from the Atlantic Ocean, and 5 major European rivers, causing considerable ecosystem heterogeneity. Despite its proximity to land, making it vulnerable to overexploitation, understanding is scarce on how viruses influence microbial diversity and ecosystem functioning in the North Sea. To date, only one study (4 stations) has attempted to delineate bacteriophage (viruses that infect bacteria) diversity in the North Sea. With our study, we aimed at increasing our understanding of the diversity of viruses in the North Sea and shining a light on the complexity of their role. We collected seawater samples at 7 sampling stations in the central North Sea. We then employed size filtration followed by iron chloride flocculation to enrich for viruses below 0.22 µm in size. As DNA virus genomes can be single (ssDNA) or double-stranded (dsDNA), we utilized a library preparation kit for Adaptase-Linker Amplification to reduce bias for or against ssDNA. Libraries were sequenced on Illumina NextSeq 500 and further analyzed using various bioinformatic tools to extract information on viral diversity, lifestyle, and putative hosts.

Overall, we identified more than 50,000 unique viral sequences, with 99 % sequences belonging to the viral realm *Duplodnaviria* (dsDNA viruses). We used PhaBOX to identify bacteriophage sequences, assign taxonomy, and predict viral lifestyle and host. Viruses representing 12 bacteriophage families were present in our samples, and over 30 % of our viruses exhibited a temperate lifestyle. We predicted 35 potential bacterial host species for these viruses. With this study, we have pushed the known diversity of viruses in the North Sea by employing state-of-the-art technologies, providing a baseline for future studies on marine viral ecology in the North Sea.

Keywords

North Sea; Marine Viruses; Bacteriophages; Metagenomics; Viromics; Ecogenomics; Lysogeny