English abstract

Invasive species are considered one of the most serious threats to the balance of aquatic ecosystems worldwide. The primary source for the spread of aquatic invasive species is transport and discharge of ballast water by cargo ships which represent around 90% of the global transportation of goods. Annual costs linked to aquatic invasive species are in the order of hundreds of billions of USD. To mitigate the spread, the United Nations' International Maritime Organization (IMO) adopted the Ballast Water Convention that entered into force in 2017. The convention requires all ships with ballast water to apply a type approved Ballast Water Treatment System (BWTS) to eliminate organisms. Results from the present thesis showed that the implementation of the ballast water convention caused a growing trend in the number of ships using BWTS and that future regulatory decisions regarding type approval process and enforcement protocols may influence the adoption of certain BWTS types. Furthermore, it was found that the two most prevailing treatment types were electrolysis and UV irradiation although 80% of ships in 2017 still used ballast water exchange to reduce presence of organisms in the ballast water.

The IMO discharge standard for phytoplankton (10-50 μm size class) in treated ballast water is <10 *viable* org. ml^{-1}. The United States Coast Guard (USCG) has implemented similar rules but with a critical difference in the discharge standard where the requirement is <10 *living* org. ml^{-1}. This means that living, but *non-viable* (i.e. unable to reproduce) organisms also must be destroyed. These are allowed under IMO regulations since they pose no invasion risk. After treatment, organism concentration can be determined using two quantification approaches: Vital Stain (VS) or Most Probable Number (MPN) method. The VS method quantifies both *viable* and *non-viable* living organisms as enzyme activity in the cells activates the fluorescent stain. The MPN method is based on organisms' regrowth potential and estimates only the number of *viable* organisms. The MPN method is not approved by the USCG which has implications for UV-based BWTSs. The UV treatment does not instantly kill organisms but causes DNA damages which result in *non-viable* living organisms with delayed mortality. Results from the present thesis showed that UV-based BWTS could satisfy the USCG discharge standard for *living* organisms but 10 times more UV energy was needed to instantly kill organisms. The implementation of extended dark-hold periods after UV treatments could however reduce the extra energy needed to comply with regulations. Although requirements for <10 *living* org. ml^{-1} could be met, the results showed that the VS method was less suitable for validation of UV-BWTS because costly overtreatment was required. Quantification of *viable* organisms with the MPN method is slow (weeks) but was demonstrated to be a more robust approach because *non-viable* living organisms with not invasion potential are not quantified.

The IMO/USCG-requirement for a natural algal concentration of >1000 org. ml^{-1} in the test water to properly challenge BWTS performance cannot always be fulfilled. Results from the present thesis showed that robustness towards UV treatments was at least as high for monocultures of *Tetraselmis suecica* and *Odontella* sp. as for natural algal compositions. The two species were therefore considered good candidates as standard test organisms to validate BWTS performance to ensure more robust testing conditions and that concentration requirements in test water are met.

Current manual counting procedures are challenged by several factors such as quantification accuracy. In the present thesis a novel automated approach was developed. It combined a filter technique to fixate mobile species, a High Content Screening Platform and image analysis which significantly improved quantification accuracy, possibility for verification of results, and reduced sample counting time.

The exploration of a novel indicative analysis approach in the present thesis, showed that integration of Raman spectroscopy in future viability assessment techniques could be a promising tool for the simultaneous identification of microplankton species and their viability state. When further developed, the technique could be applied to validate BWTS performance and for monitoring harmful algal bloom events.