



## Proof of life using molecular tools



Molecular tools are an emerging technology for marine surveillance and monitoring. Scientists can use them to detect the presence of particular plants or animals in an environment by screening for specific segments of DNA and RNA within water, sediment or other types of samples. Fragments of DNA can, however, persist in the environment for some time after the organism itself has died or moved away. In many biosecurity applications it is important to know if unwanted organisms are currently present and living in an environment. This project investigated the utility of screening for both DNA and RNA to decipher which organisms are likely to be present and living.

Researchers from the Cawthron Institute used samples of bilge water (i.e., sea water that accumulates on-board a vessel during its operations) collected from 15 small recreational and commercial vessels in the Nelson and Marlborough regions for the study. The planktonic life stages of non-native marine species can potentially be transported in the bilge water of small vessels, but only represent a biosecurity threat if they are discharged alive into a new environment.

Environmental DNA and RNA molecules were extracted from the bilge water samples and sequenced to identify the organisms that they were associated with. Because RNA molecules degrade much more quickly in natural environments than DNA, they are likely to be better indicators of living ("viable") organisms in the samples. The study compared the numbers of organisms that were identified only from analysis of DNA, only from RNA, and those that were retrieved using both methods.

Just over 62% of the organisms identified from the bilge water were found using both methods. A further 19% were recorded only in the DNA analyses and 17% were unique to the RNA method. For biosecurity applications that require detection of living organisms, the study suggests using only organisms that were present in both eDNA and eRNA analyses.

Further research is needed to improve understanding of the persistence of RNA in the environment, and the underlying reasons for the presence of RNA-only organisms in environmental samples

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## Additional reading

- Pochon, X., Zaiko, A., Fletcher, L.M., Laroche, O., Wood, S.A. (2017) Wanted dead or alive? Using metabarcoding of environmental DNA and RNA to distinguish living assemblages for biosecurity applications. PLoS ONE, 12(11): e0187636.10.1371/journal.pone.0187636
- Can marine pests be transported and spread in bilge water?
- Fletcher, L., Zaiko, A., Atalah, J., Richter, I., Dufour, C., Pochon, X., Wood, S., Hopkins, G. (2017) Bilge water as a vector for the spread of marine pests: a morphological, metabarcoding and experimental assessment. Biological Invasions, 19: 2851–2867. doi:10.1007/s10530-017-1489-y

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