DETERMINING OPTIMAL SAMPLING STRATEGIES TO PRESERVE *IN SITU* MICROBIAL GENE EXPRESSION FOR ENVIRONMENTAL MONITORING IN THE BALTIC SEA.

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Bacterial communities react rapidly to changing environmental conditions in the ocean by modifying their functional activities at the genetic level. Due to this ability, bacterial gene expression patterns have the potential to be good indicators for environmental monitoring. However, the rapid turnover of messenger RNA molecules within cells can become a major obstacle to the study of gene expression among *in situ* bacterial communities, as sampling procedures tend to cause a transcriptomic response that masks the original environmental expression profiles. High-frequency long-term monitoring surveys of gene expression in bacterial communities would require a refined sampling tool, to instantaneously preserve water samples by injection of a fixative. With the objective of developing such an instrument, we empirically determined the minimal seawater volume necessary for adequate coverage of community gene expression and the suitability of fixatives for long-term preservation of metatranscriptomes.

An artificial bacterial community was aliquoted into different experimental volumes, injected with a fixative Stop Solution (5% phenol, 95% pure ethanol), and filtered at different time intervals (0-192h). The number of cells in these aliquots remained constant (~10^7cells mL^{-1}) in contrast to the unfixed aliquots of the artificial community, which exhibited increases in cell counts over time. The total RNA content of both fixed and unfixed aliquots of 1L showed a marked decrease over time (from 6-8 µg to <3 µg), and after 48h some aliquots had lost more than half their initial RNA content. As expected, at lower volumes, such as 250 mL, total RNA contents remained proportionally lower than in the 1L aliquots, with a maximum of 1 µg RNA. The quality of the total RNA, assessed by Fragment Analyzer, also showed a rapid decrease over the duration of the experiment, especially marked within the few 24 hours of incubation. All aliquots were affected by this trend, but the unfixed communities retained higher RNA quality numbers than those preserved with the fixative. These results imply that the Stop Solution accelerated degradation of the total RNA in artificial bacterial communities. Whether this apparent degradation of the total RNA is truly representative of a degradation of transcripts remains to be assessed by the comparative metatranscriptomic analyses of gene expressions among the different experimental treatments. In the meantime we must test other fixation methods, which would allow preservation the gene expression profiles for a longer period.