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# TEMPH 2014

TRENDS IN ENVIRONMENTAL MICROBIOLOGY FOR PUBLIC HEALTH

18 - 21 SEPTEMBER 2014

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TRENDS IN ENVIRONMENTAL MICROBIOLOGY FOR PUBLIC HEALTH

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Methanol preservation of filamentous cyanobacteria for molecular and morphological studies.

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Molecular studies on cyanobacteria often involve filtering and freezing of samples leading to loss of cell morphological features. Methanol is a coagulating fixative often used in cell preservation in association with other fixatives. This study intends to evaluate the application of methanol fixation in the preservation of viable DNA for PCR reactions and also preserve cell morphology for microscopic studies in filamentous cyanobacteria. Several types of samples – cultured isolates, mixed cultures and environmental bloom samples - were fixed using a cold methanol dehydration series (50, 70 and 100%) and stored at -20°C up to one year. Samples were analyzed at the time of fixation and after 6 and 12 months preservation in cold methanol. The DNA yield and DNA purity extracted from fixed samples was determined spectrophotometrically, while electrophoretic analysis of genomic DNA was carried out to investigate DNA integrity. The DNA template amplification was tested in both conventional and real-time PCR. Base pair alteration was analyzed by comparing the sequences obtained from preserved and unpreserved samples. Microscopic observations and cell measurements were performed in fixed samples. The DNA extracted from samples preserved up to one year was successfully amplified and quantified using conventional and real-time and PCR. The DNA sequence and cell morphology were also maintained during the preservation time. The applicability of methanol preservation in molecular studies is discussed.