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ANALYSIS OF RESPONSIBLE GENES IN THE BIOSYNTHESIS OF GEOSMIN AND 2-MIB IN LABORATORY CULTURED CYANOBACTERIA

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Abstract

Geosmin and 2-MIB are common, earthy and musty-smelling sesquiterpenes, produced by Actinomycetes and cyanobacteria. These compounds result in objectionable and unpleasant tastes and odours (T & O) in drinking water. This is a common and recurrent problem in global municipal water supply. Hence, the current study was focused on the determination of genes responsible for biosynthesis of Geosmin and 2-MIB in cyanobacteria. Cyanobacteria were isolated from drinking water reservoirs and were cultured in the laboratory. Exponentially growing cultures were subjected to extraction of Geosmin and 2-MIB following Solid Phase Micro Extraction (SPME) and quantification was done using the Gas Chromatography Mass Spectrometry (GC/MS). Polymerase chain reaction (PCR) screening was carried out to confirm the presence of Geosmin and 2-MIB producing three major gene clusters; Geosmin synthase gene (geo A), SAM dependent methyl transferase gene (mtf) and monoterpene cyclase gene (mic). Out of four cyanobacteria species, Pseudanabaena sp., Nostoc sp., Lyngbya sp. and Oscillatoria sp., the highest concentrations of 2-MIB (532.5 \pm 2.1 ng/L) was recorded by *Pseudanabaena* sp. after four weeks of cultivation whereas *Nostoc* sp. $(162.6 \pm 1.8 \text{ ng/L})$ recorded the highest concentration of Geosmin. Both *Pseudanabaena* sp. and *Nostoc* sp. cultured showed significantly high Geosmin and 2-MIB production compared to all other cyanobacteria species tested and the lowest Geosmin (10.8± 2.8 ng/L) and 2-MIB (50.8± 2.2 ng/L) production was recorded in Lyngbya sp. culture. All four species harbored 'geo A' gene at 905 bp reconfirming Geosmin synthesizing ability. However, only Pseudanabaena sp. and Nostoc sp. showed presence of 'mtf' gene at 870 bp confirming 2-MIB synthesizing capability. None of four species showed the presence of 'mic' gene at 726 bp. This may open path to understand a new synthesizing pathway to 2-MIB and further research are on progress.

Keywords: Biosynthesis, Geosmin, 2-MIB, Pseudanabaena sp., Nostoc sp., PCR analysis