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First report of a microcystin-producing cyanobacterium isolated from sponge

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The occurrence and ecotoxicology of cyanobacteria are well documented in freshwater habitats, however studies in marine environments have not been carried out to the same extent. To date, toxicity has been reported in marine cyanobacteria belonging to the genera *Oscillatoria, Synechococcus, Synechocystis,* and there are scarce records for *Leptolyngbya* and *Geitlerinema* [1,2]. The present study is aiming to investigate the presence of cyanotoxins in cyanobacteria associated with marine sponges, on which no information exists. Sponge samples were collected by Scuba diving from North Aegean Sea. Nine cyanobacteria strains were isolated from the sponges *Acanthella acuta, Aplysina aerophoba, Axinella damicornis, Chorndrilla nucula, Dysidea avara, Ircinia variabilis,* and *Petrosia ficiformis*. The nine stains were characterized based on morphological and molecular (16S rRNA gene sequences) characters. In order to detect the presence of genes implicated in the production of cyanotoxins, we used PCR using the specific primers *mcyA*, *mcyB*, and *mcyE* for microcystins, *mcyE/ndaF* gene for microcystin/nodularin, and PKS for cylidrospermopsin, respectively. Moreover, the strains were extracted and analysed for a wide range of cyanotoxins (Cylindrospermopsin, Anatoxin-a, Nodularin and 12 Microcystins: [D-Asp3]MC-RR, MC-RR, MC-YR, MC-HtyR, [D-Asp3]MC-LR, MC-LR, MC-HilR, MC-WR, MC-LA, MC-LY, MC-LW and MC-LF) using ESI-LC-MS/MS in MRM mode. The strains were assigned to the following taxa: *Leptolyngbya* sp., *Pseudanabaena* cf. *persicina, Schizotricaceae* sp., *Synechococcus* sp. and *Xenococcus* sp.. No amplification of any of the toxins genes was obtained, with the exception of *mcyE/ndaF* in *Pseudanabaena* cf. *persicina* AUTH 1415 strain. No cyanotoxin, however, was detected by LC-MS/MS in this strain. The strain *Leptolyngbya* sp. AUTH 1115 produced microcystin-RR (0.14 μg g^{-1}) as determined by LC-MS/MS analysis. Microcystins have been reported previously in cyanobacteria of the genus *Leptolynbgya,* only in a marine strain which was isolated from Black Band Disease on the coral *Montastraea annularis* [2]. This strain is phylogenetically close to *Leptolyngbya* sp. AUTH 1115 of the present study. This is the first report of microcystins in cyanobacteria associated with sponges. The results of the present study indicate that microcystin production among marine cyanobacteria could be more widespread than previously demonstrated and further investigation is required.

References

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