

MOLECULAR IDENTIFICATION OF CYLINDROSPERMOPSIS PRODUCING *CYLINDROSPERMOPSIS RACIBORSKII* FROM ANURADHAPURA WATER RESERVOIRS

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Most of the world's population relies on surface freshwaters as its primary source for drinking. The drinking water industry is therefore constantly challenged with surface water contaminants that must be removed to protect human health. Most chemicals in drinking water are of health concern only after exposure of several years, rather than months. A number of chemical contaminants have been shown to cause adverse health effects in humans. Among chemical contaminants, cyanotoxins are well recognised as a cause for number of livestock and human poisonings. Therefore, the presence of cyanobacteria and their toxins in surface waters used for drinking and recreational activities are now readily acknowledged as a serious human health risk. Chronic Kidney Disease of unknown aetiology (CKDu) prevailing in Sri Lanka also focuses on a probable cause arising from drinking water. The disease is a major health problem in Sri Lanka. CKDu is likely to be triggered by an environmental factor and therefore, we focused our study on cyanotoxin due to its potentiality to cause adverse health effects.

Among cyanotoxins, cylindrospermopsin (CYN) is one of the potent cyanotoxin which effects to the kidney and liver function. Therefore, this study was performed to identify cyanotoxin; CYN in Anuradhapura water reservoirs using molecular, biochemical and bioassay methods. Water samples were collected from Kala wewa, Nuwara wewa, Tissa wewa and Jaya ganga. Under microscope, *Cylindrospermopsis* species were recorded as the dominant cyanobacterial species along with *Microcystis*, *Anabaena*, *Chroococcus*, *Phormidium*, *Oscillatoria*, which were comparatively moderate to low. In molecular detection, the presence of cyanobacteria, the presence of *Cylindrospermopsis* and *Cylindrospermopsis* strains that have the genetic potential to produce CYN were detected using specific PCR primers targeting 16S rRNA gene, *C. raciborskii* specific cylindrospermopsin synthetase gene and cylindrospermopsin specific peptide synthase (PS) gene respectively. The presence of toxin producing *Cylindrospermopsis raciborskii* in those water samples was reconfirmed by a nested PCR using *C. raciborskii* specific primers cyl2, cyl4 and cyl-int and by direct sequencing the PCR products at commercial facility and NCBI data bases.

All water samples were subjected to cylindrospermopsin ELISA detection kit to confirm the presence of CYN and to quantify the toxin. All were positive for CYN with a mean concentration of 0.137ng/ml. Further, water samples collected from the water purification centre in Anuradhapura showed 0.245 and 0.154 ng/ml of CYN before purification and after addition of chlorine respectively. However, a sample treated with chlorine and alum collected from the same purification centre was negative for CYN. Therefore, water purification processes have been playing a major role in reducing CYN toxin in drinking water. Further, bioassay results also confirmed the presence of CYN in collected water samples.

Therefore, this molecular, biochemical and bioassay findings may be an answer to the prevalence of CKDu in North Central Province and lack of the disease among people in the peripheral areas who consume purified drinking water. However, there is

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people in the peripheral areas who consume purified drinking water. However, there is a need of extensive studies to identify the types of cyanobacteria and cyanotoxins present in those water reservoirs using molecular, biochemical and bioassays methods to assess the present issue of CKDu.

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