

8.4 CHRONIC KIDNEY DISEASE OF UNKNOWN ETIOLOGY (CKDu) : IS IT CYANOTOXINS

Along the novel risk factors, cyanotoxins are well recognized as a cause for number of livestock and human poisonings. Among the cyanotoxins, microcystins are the predominant toxins in freshwater lakes worldwide and have been implicated in several cases of animal and human intoxications. There are two aspects of chronic Microcystin, damage to the liver-progressive active liver injury and the potential for promotion of tumour growth. A number of human deaths have been reported in exposure to cyanobacterial toxin; microcystin through renal dialysis and also microcystin and cylindrospermopsin implicated in drinking-water. Cylindrospermopsin is a general cytotoxin that blocks protein synthesis, the first clinical symptoms being kidney and liver failure. In contrast to the pure toxin, crude extracts of the organism also cause injury to the lungs, adrenals and intestine, indicating further, unknown toxins in the organism.

Under these circumstances, investigations of cyanotoxins present in Sri Lanka water bodies' needs extensive attention. Therefore, this project aims to identify whether cyanotoxins is a risk factor for the CKDu prevail in North Central Province of Sri Lanka using morphological, molecular and bio assay methods. Targeting this objective, preliminarily, 25 water samples were collected from Kala wewa, Tissa wewa, Nuwara wewa and Jaya ganga in Anuradhapura. Collected water samples were cultured in cyanobacterial specific media and morphological observations were made for both environmental and cultured samples under the microscope. Under microscope, *Cylindrospermopsis* species were recorded as the dominant cyanobacterial species in the environmental water samples. Apart from that, *M. aeruginosa*, *Cylindrospermopsis*, *Anabaena*, *Chroococcus*, *Phormidium*, *Microcystis spp.*, *Oscillatoria*, *Limnothrix*, *Lyngbia*, *Calothrix*, *Anabaenopsis*, *Arthrospira*, *Nostoc*, *Synechocystis*, *Synechococcus*, *Chroococciopsis* and *Merismopedia* species were identified as toxin producers in cultured samples. Environmental and cultured samples were then subjected to PCR to detect cyanobacteria, *Microcystis*, microcystin, *Cylindrospermopsis* and cylindrospermopsin targeting the specific genes. All tested were positive for unicellular and five were positive for filamentous cyanobacteria. In all three reservoirs, positive amplification was observed for *mcyA / mcyE* genes indicating the presence of cyanobacterial strains with genetic potential to produce microcystins. A positive amplification was resulted in Nuwara wewa cultured sample for *rpoC1* gene confirming the presence of *C.raciborskii* in the reservoir. Further, an environmental sample from Nuwara wewa was positive for PS gene confirming the presence of toxic *Cylindrospermopsis* species and thereby cylindrospermopsin in that reservoir. Bioassay was performed using *A. salina* to test the lethality of water samples. Both filtrate and filter was assayed and higher toxicity was recorded from filters of both environmental and cultured samples. Among filters, highest toxicity was recorded in two Nuwara wewa environmental samples with a LD₅₀ of -694ppm. Among cultured samples, highest toxicity of -1128.28ppm was recorded from Kala wewa. According to molecular and bioassay findings, all three lakes has a vast distribution of cyanobacterial species with toxic generating ability and it might be a major risk factor for the CKDu in the dry zone.

Moreover, extending our studies towards Girandurukotte, we gained the Ethical Clearance for Kidney disease patients' information assessment from the Medical Faculty, University of Peradeniya and collected disease information from 204 kidney patients of unknown etiology attending to Kidney Dialysis Clinic at Giradurukotte hospital. 100 blood and urine samples and 80 water samples from different water sources (well water, streams, paddy etc) of kidney patients of unknown etiology and 37 water samples from major water reservoirs (Ulhitiya, Rathkinda, Henanigala, Minipe, Bathalayaya and Madagama) were also collected. In contrast, 40 well water samples from other different geographic areas were also collected as controls. Collected water samples were cultured in cyanobacterial specific media and morphological observations, molecular detections and bioassays for water, blood and urine samples are under progress.

- This Research is being carried out by Ms.H.M. Liyanage as part of for her M.Phil degree under the supervision of Myself and Dr.N.V.Chandrasekaran of Department of Chemistry, University of Colombo
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