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**TOXICITY AND BIOLOGICAL ACTIVITY STUDIES
OF GREEN LEAFY VEGETABLES CONSUMED
IN SRI LANKA**

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BY

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Toxicity and biological activity studies of green leafy vegetables consumed in Sri Lanka

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Green leafy vegetables (greens) play a major role in the human diet, due to the low cost, easy accessibility and the influence of traditional herbal medicine. Due to their dietary importance, many scientific studies have been carried out on the nutritive values of greens. However, there is a lack of scientific literature on their toxic effects on consumers. Hence, there is a strongly felt need to conduct systematic scientific evaluation of their potential toxic effects, and screen them for possible biological activities to have a better understanding and educate the consumers on their choice of greens. Consequently, the present research project was initiated. The aim of the present investigation was to determine the toxicity and biological effects of thirty green leafy vegetables consumed in Sri Lanka.

Water extracts of 30 vegetable greens and culinary herbs consumed in Sri Lanka were tested for their cytotoxic properties using brine shrimp (*Artemia salina*) lethality bioassay. As anticipated most of the greens were found to have insignificant cytotoxicity. However, the water extracts of *Alternanthera sessilis*, *passiflora edulis*, *Aerva lanata* and *Baccopa monierri* showed significantly higher ($p < 0.001$) cytotoxicity, when compared with the positive control. Of them *Alternanthera sessilis* (Mukunuwenna/Ponnakkani) is the most popular leafy vegetable among Sri Lankans. As large quantities of *A. sessilis* are often consumed in a single meal, there is an urgent need for more investigations on toxicological aspects. Therefore experiment was conducted using Swiss mice. Results indicated that oral administration of water extract of *A. sessilis* in high doses (0.96 g/kg and 1.92 g/kg of freeze dried extract) for 15 days leads to histopathological changes in the liver and kidney tissues of Swiss mice. Further, histopathological, serum biochemical and hematological changes caused after 21 days and 42 days of oral administration of water extract of *A. sessilis* to Male Wistar

Rats in 3 different doses were examined. Concentrations of AST (Aspartate aminotransferase) and ALT (Alanine aminotransferase) were significantly elevated in rats that received the highest dose for 21 days. Significant changes were observed in the AST, ALT, ALP (Alkaline phosphate), urea, creatinine, magnesium and albumin levels in all treatment groups, after 42 days. Mild histopathological changes were observed after 21 days in liver and kidneys of rats that received the higher doses. Mild to moderate histological lesions and/or necrosis in liver and kidney tissues were found in all treatment groups after 42 days, while both control groups were devoid of significant histopathological changes. Present findings indicated that, the oral administration of fresh *A. sessilis* extract leads to mild to moderate hepatic and renal toxicities in male Wistar rats. Therefore, frequent consumption of larger quantities of *A. sessilis* should not be recommended, especially for patients with chronic hepatic and renal diseases. However, further investigations are necessary in order to understand the long-term effects of consuming of cooked *A. sessilis*.

The 30 green extracts were screened for antioxidant capacities using DPPH (2,2'-diphenyl-1-picrylhydrazyl) radical scavenging assay. The results revealed the presence of radical scavenging activities of all the extracts. When the antioxidant activities of commonly used 10 extracts were quantified, none had a comparable EC_{50} to that of the positive control, vitamin C (EC_{50} 82.3 ppm). However, *Sesbania grandiflora* (EC_{50} 219.9 ppm) and *Brassica sativa* (EC_{50} 249.4 ppm) had comparatively high antioxidant activities.

Antibacterial activity and antifungal activity of the 30 green extracts were conducted using disk diffusion method against, human pathogenic bacteria such as *Staphylococcus aureus*, *Enterococci faecalis*, *Pseudomonas aeruginosa*, *Klebsiella P.W.*, *Escherichia coli*, 10 MRSA strains and human pathogenic fungi, 18 strains of *Candida*. Water extract (at a concentration of 200 μ g/disk) of *Dregea volubili*, *Cardiospermum halicacabum*, *B. sativa*, *Carum petriselinum* and hexane extract of *Alternanthera sessilis* showed activity against *Staphylococcus aureus*. *D. volubili*, *C. halicacabum*, *B. sativa* and *C. petriselinum* had activity against MRSA (Methicillin resistant *Staphylococcus aureus*) strains. None of the extracts had antifungal activity against *Candida* strains. The leafy vegetables can be further explored to isolate promising antibacterial compounds.