

Introduction: Sri Lankan ayurvedic treatments for colorectal cancer are primarily based on herbal decoctions. However, the anticancer effect of most of these decoctions has not been scientifically validated. Current study was targeted to prove the cytotoxicity of a herbal decoction of *Withania somnifera* (WS) which is used to treat colorectal cancer in Sri Lanka. The cytotoxicity of WS was compared with the cytotoxic activity of poly-herbal decoction (PH) composed of *Adenanthera pavonina*, *Thespesia populnea*, *Withania somnifera* and *Tinospora cordifolia* which is used to treat cancer.

Methods: The fresh plant materials were washed with deionized water and air dried for 1.5 hours. Dried materials were freeze dried and decoctions were prepared according to a recipe in Ayurveda. Cytotoxicity of WS and PH on Human Colorectal cancer (HCT-116) and Human Kidney (HEK-293) cell lines was determined by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. Morphological changes of the treated cells were observed by Ethidium Bromide and Acridine Orange (EB/AO) staining.

Results: WS showed IC₅₀ values of 83.09± 2.40 µg/mL for 24 hr, 17.42± 1.20 µg/mL for 48 hr and 3.10± 3.20 µg/mL for 72 hr. WS exhibited less cytotoxicity (IC₅₀= 534.45± 3.20 µg/mL for 24 hr) on normal cells compared to cancer cells. PH exhibited IC₅₀ values of 383.07± 2.51 µg/mL and 1159.16± 3.40 µg/mL for 24 hr against HCT-116 and HEK-293 respectively. EB/AO staining indicated characteristic apoptotic changes such as shrunken cells, fragmented nuclei and apoptotic bodies.

Conclusions: These results indicate that WS possess strong cytotoxic activity compared to PH with less cytotoxicity for normal kidney cells. Current study provides preliminary scientific evidence for the claimed anti-colorectal cancer activity of WS in Sri Lankan ayurvedic medicine.