



# Bangladesh Journal of Pharmacology

Volume: 11; Number 2; Year 2016



Cite this article as: Ravisankar N, Jerrine J, Radhakrishana M, Rajasekar T. *In vitro* cytotoxicity of methanol extracts of *Hypericum wightianum* and *Hypericum hookerianum* against 3T3L1 cell lines. Bangladesh J Pharmacol. 2016; 11: 328-29.

## Letter to the Editor

### *In vitro* cytotoxicity of methanol extracts of *Hypericum wightianum* and *Hypericum hookerianum* against 3T3L1 cell lines

Sir,

*Hypericum* (guttiferae or *Hypericaceae*) is a large genus of herb, which grows widely at temperate region of the earth and used as traditional medicinal plant across the globe (Yazaki and Okada, 1994). Different species of *Hypericum* have been used for the treatment of wounds, eczema and burns. During the last few years antimicrobial antifungal and anti-oxidant properties have been reported by many experimental studies (Rabanal et al., 2003). Nearly 27 species of *Hypericum* are reported to possess strong anticancer properties (Agostinis et al., 2002). The European *H. perforatum* have been studied in details for their potent anticancer properties and on its medicine value and presence of rice bioactive secondary metabolic. The present study was focused to investigate cytotoxicity properties of *H. wightianum* and *H. hookerianum*

The plant materials (*H. wightianum* and *H. hookerianum*) were collected from different locations of from Kodaikanal, Southern India during the month of April 2015. Plant materials was extracted with methanol: water (60:40) using soxhlet apparatus. Cytotoxicity studies were carried out using 3T3L1- mouse adipose cell line procured from National Centre for Cell Sciences, Pune, India. The cytotoxic effect of the crude extracts from the two *Hypericum* species were investigated using MTT assay (Shahneh et al., 2013). The percentage growth inhibition was calculated and concentration of test drug needed to inhibit cell growth by 50% (CTC<sub>50</sub>) values is generated from the dose-response curves for each cell line.

The cytotoxic effect of the three extracts from each *Hypericum* species was determined. All the extracts were tested against a panel of normal and cancer cell lines at a range of 1,000 to 62.5 µg/mL using MTT exclusion methods. The CTC<sub>50</sub> values were shown separately for normal and cancer cell lines as in Table I for MTT assay; the CTC<sub>50</sub> values for short term study are depicted in Figure 1. The cytotoxicity of three different extracts viz., methanolic, aqueous and hydro-methanolic from the studied species showed similar pattern with respect to their specificity towards toxicity. The toxicity of extracts are in increasing order, methanolic > hydro-methanolic > aqueous extract. The

|                       | Concentration (µg/mL) | % Cytotoxicity | CTC <sub>50</sub> (µg/mL) |
|-----------------------|-----------------------|----------------|---------------------------|
| <i>H. wightianum</i>  | 1000                  | 47.2 ± 3.3     | >1000                     |
|                       | 500                   | 34.8 ± 1.9     |                           |
|                       | 250                   | 26.3 ± 3.1     |                           |
|                       | 125                   | 22.4 ± 4.4     |                           |
|                       | 62.5                  | 19.3 ± 2.7     |                           |
| <i>H. hookerianum</i> | 1000                  | 26.3 ± 4.5     | >1000                     |
|                       | 500                   | 24.2 ± 1.0     |                           |
|                       | 250                   | 23.6 ± 3.0     |                           |
|                       | 125                   | 20.7 ± 1.7     |                           |
|                       | 62.5                  | 10.9 ± 7.7     |                           |

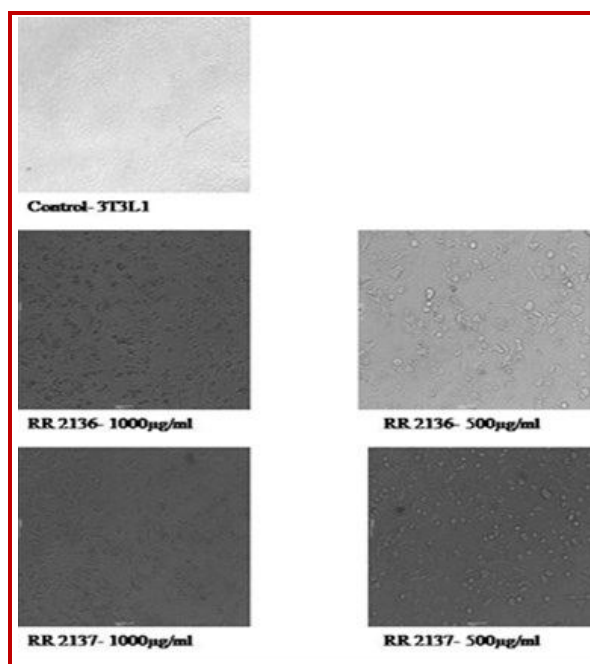


Figure 1: Cytotoxic effect of the *H. wightianum* (RR2136) and *H. hookerianum* (RR2137) on 3T3L1 cell line

extracts also exhibited moderate cytotoxicity against cancer cell lines, among the cancer cell lines. Overall the CTC<sub>50</sub> was found to be >1000 µg/mL.

The trypan blue assay based on the assumption that the dead cells will take the dye and viable cells won't (Unnikrishnan et al., 1988). From the study, it was observed that extracts showed moderate cytotoxic against both cancer and normal cell lines. The cytotoxicity of extracts found to be in dose dependent and non selective as reflected by uniform CTC<sub>50</sub> values independent of cell line origin. The species from are reported to possess various biological activities (Rahul and Thangaraj 2012) from the study it was observed that extracts are found to be moderately toxic. Hence, there is important need for further investigations on identification of phytoconstituents responsible for toxicity and also studying nature of toxicity using animal models.

The *in vitro* cytotoxicity assays offers quick, simple and cost-efficient way of testing the toxicity and forms an important tool for high throughput screening of plant extracts. From the present findings, it can be concluded that the studied extracts shows moderate toxicity against both cells irrespective of their origin. Hence the extracts need to be thoroughly studied using animal models.

**Narayanan Ravisankar<sup>1</sup>, Joseph Jerrine<sup>2</sup>, Manikkan Radhakrishana<sup>2</sup> and Thirunavukkarasu Rajasekar<sup>2</sup>**

<sup>1</sup>Department of Chemistry, Sathyabama University, Chennai, India;

<sup>2</sup>Centre for Drug Discovery and Development, Sathyabama University, Chennai, India.

Corresponding author:

email: ravisankarnarayanan@yahoo.co.in

## References

- Agostinis P, Vantieghem A, Merlevede W, de Witte PAM. Hypericin in cancer treatment: More light on the way. *Int J Biochem Cell Biol.* 2002; 34: 221-41.
- Rabanal RM, Arias A, Prado B, Hernandez-Perez M, Sanchez-Mateo CC. Antimicrobial studies on three species of *Hypericum* from the Canary Islands. *J Ethnopharmacol.* 2002; 81: 287-92.
- Rahul C, Thangaraj P. Total phenolic content and anti-radical property of *Caralluma diffusa* (Wight) N.E. Br. *Asian Pac J Trop Biomed.* 2012; 1: 1-4.
- Shahneh FZ, Valiyari S, Azadmehr A, Hajiaghaye R, Yaripour S, Bandehagh A, et al. Inhibition of growth and induction of apoptosis in fibrosarcoma cell Lines by *Echinophora platyloba* DC: *In vitro* analysis. *Adv Pharmacol Sci.* 2013; 5; 129-31.
- Unnikrishnan MC, Ramadasan K. Cytotoxicity of extracts of spices to cultured cells. *Nutr Cancer.* 1988; 11; 251-57.
- Yazaki K, Okada T. Medicinal and aromatic plants. VI. In: *Biotechnology in agriculture and forestry.* Bajaj YPS (ed.). Vol. 26. Berlin, Springer-Verlag, 1994, pp 167-78.