

## CYTOTOXICITY OF NUTRACEUTICALS ON *ARTMIA SALINA*

SK. NARUL ISLAM\*, PARVEEN BEGUM AND MONIRA AHSAN<sup>1</sup>

*Institute of Nutrition and Food Science, University of Dhaka,  
Dhaka-1000, Bangladesh*

*Nigella sativa*, garlic and clove are being used as traditional medicine in the treatment of various ailments since time immortal. Very recently these products have been termed as Nutraceutical.<sup>(1)</sup> They have a versatile spectrum of medicinal properties,<sup>(2-3)</sup> immunomodulating responses,<sup>(4)</sup> and even reported to be potent in prevention of HIV/AIDS.<sup>(1,5)</sup> We report here the cytotoxicity of *Nigella sativa*, garlic and clove on *Artemia salina* (brine shrimp nauplii).

The nutraceuticals were prepared as described by Begum.<sup>(6)</sup> Volatile oil of *Nigella sativa* seeds (NSVO) was prepared by steam distillation of seed oil followed by hexane extraction and removal of n-hexane under reduced pressure. Allicin and eugenol were prepared from garlic and clove, respectively by chloroform extraction.

To test the cytotoxicity of nutraceuticals, brine shrimp lethality bioassay<sup>(7)</sup> was employed. Shrimp nauplii (*Artemia salina*) were produced by culturing the shrimp eggs (Ocean 90 USA) in brine solution (3.5 g NaCl in 100 ml H<sub>2</sub>O) with constant O<sub>2</sub> supply for 24 h. Different dilutions of each of the test products were prepared separately in the brine solution using dimethyl sulfoxide to dissolve the products. Shrimp nauplii were transferred to the test preparations and incubated for 24 h at room temperatures. Then the numbers of live shrimp nauplii for each concentration of each of the products were counted. LC<sub>50</sub> (Lethal cytotoxic dose that kills 50% of shrimp nauplii) were calculated from the per cent shrimp mortality. It was estimated by linear regression model.

Result of cytotoxicity of the nutraceuticals against shrimp nauplii is presented in the Table 1. LC<sub>50</sub> values for the NSVO, allicin and eugenol were found to be 330.04, 737.89, and 1340.38 µg/ml, respectively. Per cent cell mortality indicated that NSVO might have some promising cytotoxic activity. Allicin and eugenol showed mild or poor activity on the shrimp nauplii. Some investigators<sup>(8-9)</sup> have also reported the cytotoxicity of *Nigella sativa* and garlic. Mention is to be made here that the brine shrimp lethality bioassay is a non specific assay technique for testing cytotoxicity or lethality of natural products on biological system. It is, therefore, worthy to test the natural products against cancer cell lines along with some cytotoxic agents as control.

---

\*Author for correspondence. Email: snislam@bangla.net.sislam@smpt.udhaka.net

<sup>1</sup>Department of Pharmaceutical Chemistry, University of Dhaka, Dhaka-1000, Bangladesh.

**Table 1. Cytotoxicity of nutraceuticals on brine shrimp nauplii.**

Test material	Dose in µg/ml	Per cent mortality <sup>a</sup>	LC <sub>50</sub> in µg/ml <sup>*b</sup>
Volatile oil of <i>Nigella sativa</i>	50	18.33 ± 6.1	336.04
	100	29.00 ± 2.0	(298.98 - 373.10)
	200	47.33 ± 5.7	
	400	61.66 ± 5.0	
	800	82.66 ± 4.5	
Allicin of garlic	100	9.00 ± 2.0	737.89
	200	13.33 ± 3.2	(626.39 - 849.39)
	400	27.33 ± 3.5	
	800	55.66 ± 6.1	
	1200	78.00 ± 9.0	
Eugenol of clove	100	5.00 ± 2.0	1340.38
	200	9.00 ± 2.0	(1163.36 - 1517.4)
	400	16.66 ± 5.9	
	800	25.66 ± 5.0	
	1200	50.00 ± 7.9	

\*LC<sub>50</sub> value for the shrimp nauplii. It was calculated by linear regression model.

<sup>a</sup>value expressed in mean ± SD, <sup>b</sup>confidence limits 95% given in parentheses.

## References

1. Biro G 1995. The use of results of scientific nutrition research in clinical practice. *Orv. Hetil* **136**(19): 979-983.
2. Islam SN and M Ahsan 1999. Drug resistance in shigellosis and efficacy of natural products as an anti-shigella agents. *Pub. Health J.* **1, 2** : 20-30.
3. Guo NL, DP Lu, GL Woods, E Reed, GZ Zhou, LB Zhang and RH Waldman 1993. Demonstration of the antiviral activity of garlic extract against human cytomegalovirus in vitro. *Chines Med. J.* **106**(2): 93-96.
4. Islam SN, P Begum, T Ahsan, S Huque and M Ahsan 2004. Immunosuppressive and Cytotoxic Properties of *Nigella sativa*. *Phytotherapy Res.* **80**: (in press).
5. Abdullah TH, O Kandil, A Elkadi, J Carter 1988. Garlic revisited: therapeutic for the major disease of our times? *J. Natl. Med. Assoc.* **80**(4): 439-445.
6. Parveen B 2000. Cytotoxic and immunomodulating properties nutraceuticals and/or spices. M. Phil thesis, Institute of Nutrition and Food Science, University of Dhaka.
7. Meyer BN, JE Ferrigni, LB Putnam, DE Jacobsen, Nicols and JL McLaughlin 1982. Brine shrimp: a convinient general bioassay for active plant constituents. *Planta Med.* **45**:31-45.
8. Sundaram SG and JA Milner 1996. Diallyl disulfide inhibits the proliferation of human tumour cells in culture. *Biochim. Biophys. Acta.* **1315**(1): 15-20.
9. Salomi NJ, SC Nair, KK Jayawardhanan, CD Varghese and KR Panikkar 1992. Antitumour principles from *Nigella sativa* seeds. *Cancer Lett.* **63**(1): 41-46.

(Manuscript received on 12 April, 2004; revised on 13 March, 2005)