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Original Article

In-vitro anticancer activity of different extracts of *Sesbania Grandiflora* against HEP2 cell lines

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ABSTRACT

Herbal medicine is established on the plants contains natural chemical substances that can promote health and have curative properties for illness and diseases. Medicinal herbs play an important role in the treatment of cancer. In this study we reported that potential anticancer activity of *Sesbania grandiflora* leaf extracts and compared with standard commercial anticancer drug. Water, ethanol and acetone extract of *S. grandiflora* leaves showed invitro anticancer activity against different human cancer cell lines like HEp2 (Human larynx carcinoma cell line). The potential of anticancer property of *S. grandiflora* leaf extract was assayed by MTT method. The activity was done at different concentration like 50-300 µg/ml of the extract. From the analysis the 50% inhibitory concentration (IC_{50}) is 200 µg/ml against HEp2 (Human larynx carcinoma cell line) cell lines for all the extracts. While increasing the concentration of extracts showed decrease in cell viability. Extracts of *S. grandiflora* showed dose dependent reduction of cell viability and induction of apoptosis in the HEp2 (Human larynx carcinoma cell line) cell lines. This in vitro outcomes suggest a significant clinical effects of *S. grandiflora* against human HEp2 (Human larynx carcinoma cell line) cell lines.

Keywords: *Sesbania grandiflora*, Anticancer, HEp2

INTRODUCTION

Cancer is a worldwide public health problem and one of the leading cause of death. Cancer is defined as an irregular growth of cells exhibited uncontrolled division unconventionally resulting a gradually increase in the number of cell dividing [1]. The development of therapies for rapidly spreading cancer is not been successful and increasing demands [2-3]. So it is a challenge to develop a drugs for the various types of diseases. HT29 cells are human epithelial cells which produce the secretory component of Immunoglobulin A (IgA), and carcinoembryonic antigen (CEA) [4]. Neuroblastoma is a common type of cancer in infants that affects infants and young children formed by neuro-blasts nerve cells. These immature cells grow and mature into functioning the nerve cells. But in they become cancer cells instead. Chemotherapy and radiation therapy is available for treatment and control of cancer cells but still it is exhibits low specificity and restricted by dose limiting toxicity.

Medicines from plants have played an important role in maintaining human health and improving the quality of human [5].In recent years, an increasing number of natural products have been reported to display anti-tumor compounds have been isolated from herbal plants used in various traditional medicinal systems. Herbal medicines are expected hopefully to revolutionize the cancer diagnosis and therapy. Most number of plants and their isolated constituents have been shown to potential anticancer activity [6].Several plants have pharmacological properties shown to have potential to cure human cancers without causing side effects due to they have anti-tumor substances [7].

Sesbania grandiflora is an Indian medicinal plant which is extensively used in Ayurveda and other alternative system of medicine. It is commonly known as "Sesbania" and "agathi," is widely used in Indian traditional medicine for the treatment of a wide-ranging of diseases like rheumatism, cancer and liver disorders. The plant *Sesbania grandiflora*is belongs to the family Fabaceae [8]. The plant leaves serves as a natural antioxidant

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activity and the juice of leaves used to treat worms, biliousness, fever, gout, and leprosy [9]. Leaves of this plant have medicinal effect due to its astringency property; hence it is used against inflammation, venom and other poisons, bacterial infections and tumors [10]. Recently, *Sesbania grandiflora* (antianxiety) anxiolytic, hepatoprotective, cardio, antiurolithiatic and antioxidant activities were reported. Extract of *S. grandiflora* leaves shows significant hepatoprotective [11], anti-microbial [12], analgesic and antipyretic activity [13]. The main objective of this study is to investigate the anticancer activity of Aqueous, Ethanol and acetone extract of *Sesbania grandiflora* leaf against HEp2 (Human larynx carcinoma cell line) cell lines at dose dependent method.

MATERIAL AND METHODS

Chemicals: Analytical graded3-(4,5-dimethyl thiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), dimethylsulfoxide (DMSO), doxorubicin and other chemicals were purchased from Himedia laboratories private limited, Mumbai.

Preparation of extraction of *Sesbania grandiflora* leaves: Plant leaves were collected from Vandhavasi, TN, India.. The collected plant leaves were shade dried and powdered. The powdered materials will be packed and extracted with 80% ethanol and acetone in two Soxhlet apparatus for 24 h at 55ºC. Water extract was prepared by immersing 100 g dried leaf powder into 200 ml double distilled for 24 hours. The extracts will be concentrated using rotary flash evaporator and the extract will be used to evaluate anticancer activity.

Anticancer activity against HEp2 (Human larynx carcinoma cell line) cancer cell line: Cells viability test was done by the MT(3-(4,5-dimethylthiazol-2-yl)- 2,5-diphenyl tetrazolium bromide) assay is s colorimetric analysis based on the measuring the activity of cellular enzymes that living cells were reduce the yellow MTT dye into insoluble purple color formazan. Cells were plated and grown with different concentration of plant extracts and incubated for 24 hours in CO2 atmosphere. After 24 hours treatment, MTT was added in each well and incubated at 37C for 4 hours in 5% CO2 chamber. Then the medium was removed and washed with Phosphate buffer solution. Then, DMSO was added to each well which dissolve the insoluble formazan crystals into colored solution. The intensity of the colored solution was measured using ELISA microplate reader at 570 nm. The results were expressed as the percent optical density of treated cells to that of the control cells. The 50 % of inhibitory concentration value (IC_{50}) of the extracts was identified for normal untreated cell line. Commercial anticancer drug Doxorubicin was used as a control. The assay was performed in triplicate for each extracts.

Statistical analysis: The quantitatively obtain data were analyzed using one way Analysis of Variance (ANOVA) and expressed as mean± S.E.M. value of $p< 0.05$ is considered as Statistical significant. The experiment data plots of the cell viability against drug and extract concentration.

RESULTS AND DISCUSSION

The medicinal herb *S. garandiflora* leaves showed anticancer activity against HEp2 (Human larynx carcinoma cell line) cell lines (Table 1). In the present study, the treatment with water, ethanol and acetone extracts suppressed the cell viability up to 50% at 200µg/ml against both cell lines (Figure1-3).Plant extracts shows more significant activity as compared to the positive control. The extract showed significant inhibition in the cell viability in a dose dependent manner. The treatment with ethanol extract against HEp2 (Human larynx carcinoma cell line) cell lines significantly decrease the viability of cells at 200g/ml when compared other extracts. The cells were contact with 50µg/ml, 100µg/ml, 150µg/ml, 200µg/ml, 250µg/ml, and 300µg/ml of extract showed decreased number of cell viability. The results indicate that extracts of *S. grandiflora* has an anticancer activity in HEp2 cell lines. The maximum cytotoxic effect was observe in ethanol extract. This variation in activity occurs due to presence of different phyto-constituents like flavonoids, alkaloids and steroids. Alkaloids [14] Flavonoids, [15], phenols, polyphenols and other derivatives have been associated with anticancer property [14]. Ethanol extract has high amount of alkaloids and flavonoids which actively involved in the cancer cell death. Cell death occur by apoptosis and necrosis caused by the drug. The phychemicals alkaloids, flavonoids and polyphenols were actively inhibit the cells in the protein synthesis either by damaging DNA or by blocking at transnational level which may be determine the mortality of cells [16].

Conclusion

The synthetic or semisynthetic medicines can cure the diseases but at the same time they are highly toxic in nature, whereas the herbal drug are minimize the adverse side effects. In this present report we concluded that anticancer activity of different solvent derived extracts of *S. grandiflora* like water, ethanol, and acetone determined by

colorimetric method of MTT assay. Live cells reduce the MTT yellow dye into insoluble purple color formazan at 50% in 200µg/ml concentrations. The IC_{50} concentration is 200 µg/ml for all the extracts. Hence, our report suggested that the herbal medicine from *S. grandiflora* leaves actively in the growth of cancer cells better than standard drug would be replace chemotherapy treatment. Further study needed to identify the exact active compound present in the *S. garandiflora* leaves underlying this high anticancer activity.

Table 1: Anticancer activity of extracts *S. grandiflora* leaves against HEp2 (Human larynx carcinoma cell line)

concentration (µg/ml)	Cell Viability %			
	Standard Drug	Aqueous extract	Ethanol extract	Acetone extract
50	92.53±0.99*	98.12±0.9*	94.21±1.14**	96.55±1.25**
100	76.44±1.27**	92.56±1.09*	84.87±1.65**	91.82±1.64*
150	51.33±1.14*	79.78±1.25**	66.65±1.25**	65.65±1.45**
200	29.65±0.64*	57.19±1.16**	52.15±0.85***	54.25±0.82**
250	19.23±0.81**	40.54±1.15**	42.15±1.09**	45.77±1.9**
300	10.89±0.47*	26.38±0.95**	21.64±0.95***	23.61±0.95**

* **p< 0.05,** ** **p < 0.01,*****p < 0.001** value are considered statistically significant (BMRT)

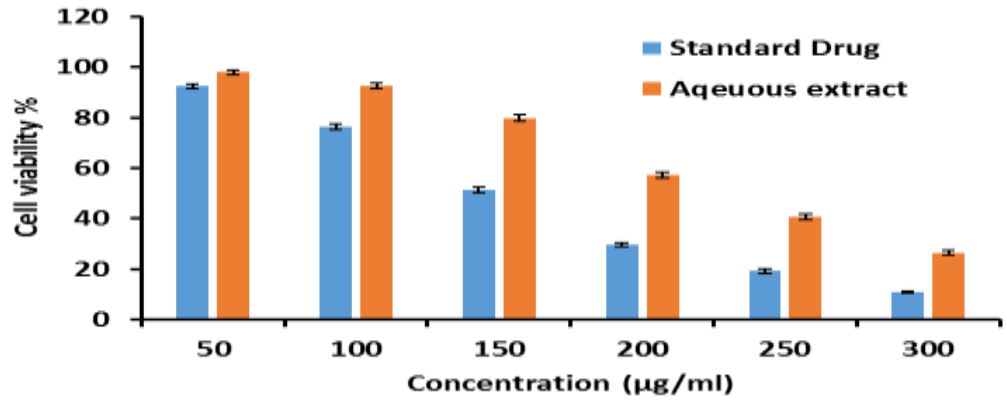

Figure 1: Anticancer activity of water extracts *S. grandiflora* leaves against HEp2 (Human larynx carcinoma cell line)

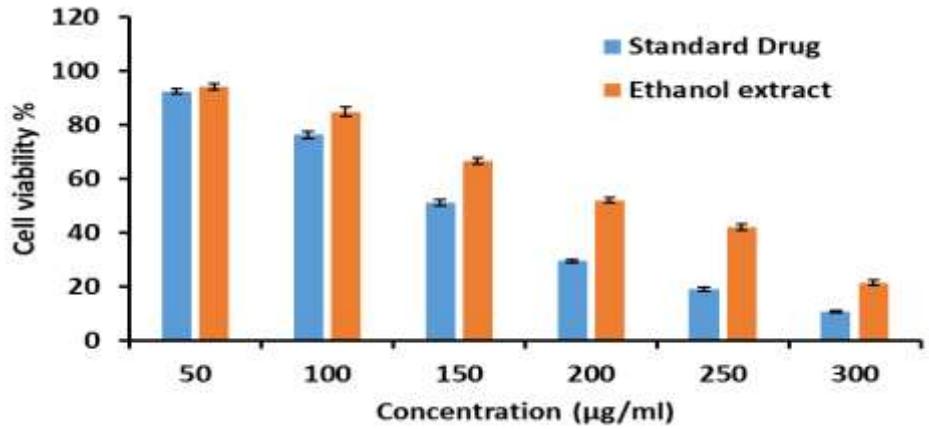

Figure 2:Anticancer activity of ethanol extracts *S. grandiflora* leaves against HEp2 (Human larynx carcinoma cell line)

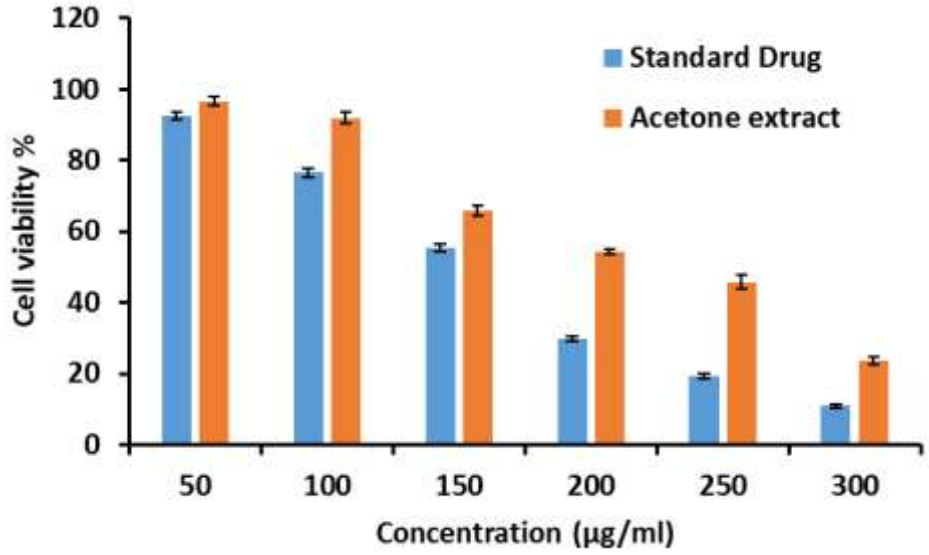

Figure 3: Anticancer activity of acetone extracts *S. grandiflora* leaves against HEp2 (Human larynx carcinoma cell line) cell lines

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