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IN VITRO ANTI CANCER AND *IN VITRO* ANTIOXIDANT POTENCY OF ROOTS OF HYDRO ALCOHOLIC EXTRACT OF *PLECTRANTHUS VETTIVEROIDES*

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ABSTRACT

The main aim of the study was to screen the hydro alcoholic extract of *Plectranthus vettiveroides* roots for its *in vitro* anti-oxidant and anticancer activity and find its efficacy against various cell lines. Hydro alcoholic extract of *Plectranthus vettiveroides* were prepared and assayed for the presence of phytochemicals. *In vitro* antioxidant assay were performed Superoxide radical scavenging activity, Hydroxyl radical scavenging activity, DPPH radical scavenging activity. The effect of hydro alcoholic extract of *Plectranthus vettiveroides* on various cell lines were evaluated by MTT colorimetric assay. The preliminary phytochemical screening of hydro alcoholic extract of *Plectranthus vettiveroides* showed the presence of significant secondary metabolites. The efficacy of *Plectranthus vettiveroides* against various cancer cell lines showed that the incubation of cancer cells reduced the viability of all cancer cell lines and the dead cells were significantly increased with high extract concentration. Hence hydro alcoholic extract of *Plectranthus vettiveroides* exhibited high cytotoxicity. Also the extract showed potent antioxidant activity against all the three tested methods. Even at very low concentration *Plectranthus vettiveroides* showed high efficacy. In conclusion *Plectranthus vettiveroides* possess significant antioxidant activity and anticancer activity.

Key words: *Plectranthus vettiveroides*, Anti-oxidant, Anticancer, Cytotoxicity.

INTRODUCTION

Currently, one in four deaths in the United States is due to cancer. When ranked within age groups, cancer is one of the five leading causes of death amongst both male and female and the single largest cause of death worldwide (Jemal *et al*, 2008). Cancer is a group of diseases initiated by loss of cell cycle control. Cancer is associated with abnormal uncontrolled cell growth (Krishnamurthi, 2007).

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Cancer is caused by both external factors (tobacco, chemicals, radiation and infectious organisms) and internal factors (inherited mutations, hormones, immune conditions, and mutations that occur from metabolism). Indeed, the struggle to combat cancer is one of the greatest challenges of mankind (Divisi, 2006).This growing trend indicates efficiency in the present cancer therapies which include surgical operation, radiotherapy and chemotherapy. Since the average survival rates have remained essentially unchanged despite of such aggressive treatments, there is a critical need for anticancer agents with higher efficacy and less side effects that can be acquired at an affordable cost (Saudat, 2013).

Chemoprevention is recognized as an important approach to control malignancy and recent studies have focused on the search for desirable chemopreventive agents. Natural products, particularly dietary substances, have played an important role in creating new chemopreventive agents (Surh, 2003). According to Cragg and Newman over 50 % of the drugs in clinical trials for anticancer properties were isolated from natural sources (Cragg, GM and Newman DJ, 2000).

Several natural products of plant origin have potential value as chemotherapeutic agents. Some of the currently used anticancer agents derived from plants are podophyllotoxin, taxol, vincristine and camptothecin (Pezzuto, 1997). The areas of cancer and infectious diseases have a leading position in utilization of medicinal plants as a source of drug discovery. Among FDA approved anticancer and anti-infectious drugs, drugs from natural origin have a share of 60 % and 75 % respectively (Newman *et al*, 2003). Anticancer drugs discovered from herbal medicines have a long history and some of them have been used in clinical setting as a conventional anticancer drug (Yibin Feng *et al.*, 2011). Colorectal cancer, also known as colon cancer or bowel cancer is a cancer caused by uncontrolled cell growth in the colon or rectum, or in the appendix. Symptoms of colorectal cancer typically include rectal bleeding and anemia which are sometimes associated with weight loss and changes in bowel habits.

Colorectal cancer is the third most commonly diagnosed cancer in the world, but it is more common in developed countries. Around 60% of cases were diagnosed in the developed countries. It is estimated that worldwide, in 2008, 1.23 million new cases of colorectal cancer were clinically diagnosed and 608,000 people died of the disease (Ferlay, J *et al*., 2008). *Plectranthus vettiveroides* is also known as coleus vettiveroides, coleus zeylanicus, plectranthus zeynanicus (Lamiaceae). The main phytochemical constituents of the genus Plectranthus are diterpenoids, essential oils and phenolics. About 140 diterpenoids were identified from the coloured leaf –glands of plectranthus species. The main constituents of essential oils of plectranthus are mono and sesquiterpenes. Flavonoides seem to be rare in plectranthus, only two flavonoides were identified 4',7-dimethoxy -5,6-identified, vone in plectra thus ambigns and chrysosplevetin from p.marruboides. Traditionally it has been used as an antibacterial, deodorant, cooling agent and also used against eye burning head ache and fever. Therefore, in the present study we analyzed *Plectranthus vettiveroides* root for the presence of bioactive components and also evaluated *in vitro* anti-cancer activity various cell lines.

MATERIALS AND METHODS

Plant Identification and Collection

The plant was collected in January 2014 from Namakkal, Tamilnadu, India. A herbarium specimen of the plant was deposited in the Department of Pharmacognosy. The plant was identified by Dr.G.V.S.Murthy, Joint Director of the Botanical Survey of India, Southern circle, TNAU Campus, Coimbatore, who authenticated the plant from information available in the literature. The roots were dried in the shade for 10–12 days. After complete drying, the dried roots were pulverized to a coarse powder of 40 mesh size in a mechanical grinder. The powdered material was subjected to soxhlet extraction for 18 h at 50–55^{o}C using methanol and water. The extract was thereafter concentrated under vacuum and air-dried.

Qualitative Phytochemical Analyses

The phytochemical tests below were carried out on the hydro alcoholic extract of *Plectranthus vettiveroides* to determine the active constituents according to the procedures and methods outlined (Barnes, J. 1999; Harborne 1998). These phytochemical tests were done to detect the presence of secondary metabolites, such as alkaloids, tannins, saponins, resins, flavonoids, steroid, glycosides and terpenoids in the plant under investigation.

IN VITRO ANTI CANCER ACTIVITY:

Cell proliferation kit

MTT (Roche applied sciences, Cat. No. 11465 007 001)

Media

DMEM (Dulbecoo's Modified Eagels Medium, high glucose), DMEM (Dulbecco's Modified Eagels Medium, low glucose), FBS (Fetal Bovine Serum) (Bioclot, Lot No: 07310).

Glasswares and plastic wares

96-well micro titer plate, Tissue culture flasks, Falcon tubes, Reagent bottles

Equipments

Fluorescence inverted microscope (Leica DM IL), Biosafety cabinet classII (Esco), cytotoxic safety cabinet (Esco), CO_2 incubator Deep freezer, ELISA plate reader (Thermo), Micropipettes (Eppendorff), RO water system (Millipore)

Cell line used:

Rat skeletal muscle cell line (L6), *Ehrlich ascites carcinoma* EAC, *Breast cancer cell line* (MCF 7), *Hepato cellular carcinoma cell line* (HEP G2).

All the cell lines were brought from Amala Cancer Institute Thrissur, Kerala.

Principle

This Colorimetric assay is based on the capacity of Mitochondria succinate dehydrogenase enzyme in living cells to reduce the yellow water soluble substrate 3-(4, 5-dimethyl thiazol-2-yl)-2, 5-diphenyl tetrazolium bromide (MTT) into an insoluble, colored formazan product which is measured spectrophotometrically. Since reduction of MTT can only occur in metabolically active cells, the level of activity is a measure of the viability of the cells. (Thirumal. 2012)

Procedure

The monolayer cell culture was trypsinized and the cell count was adjusted to 3 lakhs cells/ml using medium containing 10% newborn calf serum. To each well of 96 well microtitre plates, 0.1ml of diluted cell suspension was added. After 24 hours, when the monolayer formed the supernatant was flicked off and 100 µl of different test compounds were added to the cells in microtitre plates and kept for incubation at 37ºC in 5 % CO_2 incubator for 72 hours and cells were periodically checked for granularity, shrinkage, swelling. After 72 hours, the sample solution in wells was flicked off and 50µl of MTT dye was added to each well. The plates were gently shaken and incubated for 4 hours at 37^{o}C in 5% CO_2 incubator. The supernatant was removed, 50 µl of Propanol was added, and the plates were gently shaken to solubilize the formed formazan. The absorbance was measured using a microplate reader at a wavelength of 490 nm. The percentage growth inhibition was calculated using the formula below:

The percentage growth inhibition was calculated using following formula,

% cell inhibition= 100-{(At-Ab)/(Ac- Ab)}x100

Where,

At= Absorbance of test compound,

Ab= Absorbance of blank,

Ac=Absorbance of control

Data interpretation

Absorbance values that are lower than the control cells indicate a reduction in the rate of cell proliferation. Conversely, a higher absorbance rate indicates an increase in cell proliferation. Rarely, an increase in proliferation may be offset by cell death; evidence of cell death may be inferred from morphological changes.

% cell survival= {(At-Ab)/ (Ac-Ab)} x100

% cell inhibition= 100-cell survival.

***IN VITRO* ANTIOXIDANT ACTIVITY**

Superoxide radical scavenging activity

Superoxide radical scavenging activity of plant extract was measured according to the method of McCord and Fridovich (McCord, JM. and Fridovich, I. 1969). This works based on light induced superoxide generation by riboflavin and the subsequent reduction of nitroblue tetrazolium. 0.1 ml of different concentrations of plant extract/ascorbic acid, 0.1 ml of 6 mM ethylenediamine tetraacetic acid containing 3µg NaCN, 0.05 ml of 2 Mm riboflavin and 0.1 ml of 50 µM nitroblue tetrazolium were transferred to a test tube and final volume was made up to 3 ml using 67 µM phosphate buffer pH 7.8. Then the assay tubes were uniformly illuminated with an incandescent light (40 Watts) for 15 minutes and thereafter the optical densities were measured at 560 nm. A control was prepared using 0.1 ml of respective vehicle in the place of plant extract/ascorbic acid. The percentage inhibition of superoxide production was evaluated by comparing the absorbance values of control and experimental tubes.

Hydroxyl radical scavenging activity

Hydroxyl radical scavenging activity was measured by studying the competition between deoxyribose and the extracts for hydroxyl radicals generated from the Fe2+/EDTA/H2O2 system (Fenton reaction). The hydroxyl radical attacks deoxyribose, which eventually results in the formation of thiobarbituric acid reacting substances (TBARS) (Elizabeth, S. and Rao, MNA. 1990). 200 ml of 10 mM ferrous sulphate (FeSO4. 7H2O), 200 ml of 10 mM EDTA and 200 ml of 10 mM 2-deoxyribose was mixed with 1.2 ml of 0.1 M phosphate buffer (pH 7.4) and 200 ml of plant extract/ascorbic acid. Thereafter, 200 ml of 10 mM H2O2 was added before the incubation at 37°C for 4 h. Then, 1 ml of this reaction mixture was treated with 0.2 ml of 8.1% sodium dodecyl sulphate, 1.5 ml of 0.8% thiobarbituric acid and 1.5 ml of 20 % acetic acid. The total volume was then made to 5 ml by adding distilled water and kept in an oil bath at 100° C for 1 hour. After the mixture had been cooled, 5 ml of 15:1 v/v butanolpyridine mixture was added. Following vigorous shaking, the tubes were centrifuged at 4000 rpm for 10 min and the absorbance of the organic layer containing the thiobarbituric acid reactive substances was measured at 532 nm. A control was prepared using 0.1 ml of vehicle in the place of plant extract/ascorbic acid.

DPPH radical scavenging activity

The scavenging activity for DPPH free radicals was measured according to the procedure described (Braca *et al.,* 2011). An aliquot of 3 ml of 0.004% DPPH solution in ethanol and 0.1 ml of plant extract/ascorbic acid at various concentrations were mixed. The mixture was shaken vigorously and allowed to reach a steady state

at room temperature for 30 min. Decolorization of DPPH was determined by measuring the absorbance at 517 nm. A control was prepared using 0.1 ml of respective vehicle in the place of plant extract/ascorbic acid. The percentage inhibition of DPPH radicals by the extract/compound was determined by comparing the absorbance values of the control and the experimental tubes.

Statistical analysis

The percentage inhibition was calculated using:

$$\% \text{ Inhibition} = \frac{(A0 - A1) \times 100}{A0}$$

Where,

A0 is absorbance of control;

A1 is the absorbance of extracts/ascorbic acid.

IC50 value (a concentration at 50% inhibition) was determined from the curve between percentage inhibition and concentration. All determinations were done in triplicate and the IC50 value was calculated by using the equation of line (Papuc *et al*., 2008).

RESULTS AND DISCUSSION

PRELIMINARY PHYTOCHEMICAL ANALYSIS

The preliminary phytochemical studies showed the presence of alkaloids, carbohydrates, Steroids, Terpenoides, Phenols, Tannins and Flavanoids. The results were showed in the table:2.

IN VITRO ANTI CANCER ACTIVITY:

Hydroalcoholic extract of *Plectranthus vettiveroides* at various doses were tested against various cell lines. The extract at various doses does not show any cytotoxicity against the normal L6 cell line. The IC_{50} Value was found to be 2000 µg/ml. The results were showed in the table 2, Figure 1 and 2.

Hydroalcoholic extract of *Plectranthus vettiveroides* at various doses were tested against various cell lines. The extract at various doses showed a potent cytotoxicity against the EAC cell line. The IC_{50} Value was found to be 45 µg/ml. The results were showed in the table 3, Figure 3 and 4.

The extract at various doses showed significant cytotoxicity against the MCF-7 cell line. The IC_{50} Value was found to be 30 µg/ml. The results were showed in the table 4, Figure 5 and 6.

The extract at various doses showed dose dependent cytotoxicity against the HEP-G2 cell line. The IC_{50} Value was found to be 120 µg/ml. The results were showed in the table 5, Figure 7 and 8.

IN VITRO ANTIOXIDANT ACTIVITY:

Superoxide radical scavenging activity

Superoxide anion plays an important role in the formation of more reactive species such as hydrogen peroxide, hydroxyl radical and singlet oxygen, which induce oxidative damage in lipids, proteins, and DNA (Pietta, PG. 2000). Therefore, studying the scavenging activity of plant extract on superoxide radical is one of the most important ways of clarifying the mechanism of antioxidant activity.

The hydroalcoholic extract of *Plectranthus vettiveroides* showed concentration dependent scavenging activity on superoxide generated by photoreduction of riboflavin. The mean IC50 values for superoxide radical of hydroalcoholic extract were found to be 191.40 µg. The mean IC50 value of ascorbic acid was found to be 128.53 µg. The results were given in Table 6.

Hydroxyl radical scavenging activity

Among the reactive oxygen species, the hydroxyl radicals are the most reactive and predominant radicals generated endogenously during aerobic metabolism (Harsh, M. 2010). A single hydroxyl radical results in the formation of many molecules of lipid hydroperoxides in the cell membrane which may severely, disrupts its function and leads to cell death.

The hydroalcoholic extract *Plectranthus vettiveroides* possesses concentration dependent scavenging activity on hydroxyl radicals. The mean IC50 values for hydroxyl radical of hydro alcoholic extract were found to be 290.12 µg. The mean IC50 value of ascorbic acid was found to be 212.48 µg. The results were given in Table 7.

DPPH radical scavenging activity

The 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical is a stable radical with a maximum absorption at 517 mm that can readily undergo reduction by an antioxidant. Because of the ease and convenience of this reaction it now has widespread use in the free radical-scavenging activity assessment (Eyob *et al*., 2008). The hydroalcoholic extract possess concentration dependent scavenging activity on DPPH radicals The mean IC50 values for DPPH radical of hydroalcoholic extract was found to be 122.20 µg. The mean IC50 value of ascorbic acid was found to be 71.56 µg. The results were given in Table 8. *In vitro* free radical scavenging efficacies of hydroalcoholic extract of Plectranthus vettiveroides were assessed by its ability to scavenge superoxide, hydroxyl and DPPH radicals. Concentration of sample at which the inhibition percentage reaches 50% is its IC50 value. IC50 values are negatively related to the antioxidant activity, as it express the amount of antioxidant needed to decrease its radical concentration by 50%. The lower IC50 value represents the higher antioxidant activity of the tested sample. The selected plant extract produced concentration dependent percentage inhibition of antioxidant activity. Hydroalcoholic extract of *Plectranthus vettiveroides* showed moderate effect on hydroxyl and DPPH radical; low activity on superoxide radical as compared to

standard ascorbic acid. The results of the present study suggest that the tested hydroalcoholic extract have antioxidant activity and/or free radical scavenging activity. Literature survey reveals that flavonoids (Lamson, DW and Brignall, MS. 2000; Torres *et al*., 2006), phenolic compounds (Visioli *et al*., 1998; Stratil *et al*., 2006) are responsible for antioxidant activity. Preliminary phytochemical studies of the extract showes the presence of flavonoids and phenolic compounds, therefore the "antioxidant activity might be due to the presence of flavonoids and phenolic compounds in the selected plant extract". However, we do not know what components in the plant extract show these activities. More detailed studies on chemical composition of the plant extract, as well as other *in-vitro* assays are essential to characterize them as biological antioxidants.

Table 1: Preliminary qualitative phytochemical study of Hydro alcoholic extract of *Plectranthus vettiveroides.*

S.No	Constituents	Inference &Intensity
1.	Alkaloides	+++
2.	carbohydrates	++
3.	Steroids & Terpenoides	+
4.	Proteins	-
5.	Phenols	+
6.	Tannins	++
7.	Flavanoides	+++
8.	Glycosides	-
9.	Saponin	-

Table: 2 Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on L6 cell lines

S.No	Conc. (µg/ml)	% Cell Viability	% Cytotoxicity	IC_{50} Value (µg/ml)
1.	Control	99.17±0.21	0.90±0.12	2000
2.	15	89.52±0.23	10.53±0.21	
3.	30	80.36±0.17	19.72±0.24	
4.	60	71.60±0.32	28.4±0.52	
5.	120	69.22±0.25	30.86±0.51	
6.	240	65.43±0.65	34.66±0.24	
7.	500	58.35±0.31	41.73±0.54	
8.	1000	55.63±0.24	44.43±0.51	
9.	1500	54.23±0.51	45.83±0.32	
10.	2000	43.65±0.32	56.43±0.15	

Table 3: Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on EAC cell lines.

S.No	Conc. (µg/ml)	% Cell Viability	% Cytotoxicity	IC_{50} Value (µg/ml)
1.	Control	99.10±0.10	0.93±0.23	45
2.	15	65.51±0.21	34.50±0.21	
3.	30	49.33±0.23	50.76±0.37	
4.	60	40.64±0.36	59.46±0.81	
5.	120	32.65±0.32	67.49±0.71	
6.	240	20.45±0.65	79.65±0.34	
7.	500	11.56±0.25	88.52±0.35	
8.	1000	1.64±0.35	98.65±0.45	
9.	1500	0.90±0.30	99.10±0.32	
10.	2000	0.05±0.33	99.05±0.21	

Table 4: Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on MCF-7 cell lines.

S.No	Conc. (µg/ml)	% Cell Viability	% Cytotoxicity	IC_{50} Value (µg/ml)
1.	Control	99.11±0.10	0.89±0.23	30
2.	15	63.51±0.21	36.50±0.21	
3.	30	45.33±0.23	54.76±0.37	
4.	60	38.64±0.36	61.46±0.81	
5.	120	30.65±0.32	69.49±0.71	
6.	240	21.45±0.65	78.65±0.34	
7.	500	14.56±0.25	85.52±0.35	
8.	1000	1.61±0.35	98.39±0.45	
9.	1500	0.90±0.30	99.10±0.32	
10.	2000	0.03±0.33	99.07±0.21	

Table 5: Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on HEP-G2 cell lines using

S.No	Conc. (µg/ml)	% Cell Viability	% Cytotoxicity	IC_{50} Value (µg/ml)
1.	Control	97.29±0.0	2.71±0.05	120
2.	15	80.44±0.36	19.56±0.21	
3.	30	75.75±0.29	24.25±0.43	
4.	60	64.64±0.32	35.46±0.42	
5.	120	42.45±0.26	57.63±0.21	
6.	240	31.46±0.54	68.64±0.26	
7.	500	22.43±0.48	77.63±0.44	
8.	1000	11.76±0.49	88.34±0.24	
9.	1500	3.20±0.31	96.85±0.66	
10.	2000	0.40±0.20	99.60±0.25	

Table 6: Percentage inhibition and IC50 values of superoxide radical scavenging activity *in vitro* by hydroalcoholic extract of *Plectranthus vettiveroides*

EXTRACT	Quantity in micrograms (µg), Mean±S.E.M					IC_{50} values
	25	50	100	200	400	
Ascorpic acid	53.4±1.27	40.50±1.06	53.59±1.51	72.98±1.56	83.15±1.56	128.53
HAEPV	20.10±1.42	31.38±0.76	53.26±1.71	60.02±1.05	69.51±1.05	191.40

Table 7: Percentage inhibition and IC50 values of Hydroxyl radical scavenging activity *in vitro* by hydroalcoholic extract of *Plectranthus vettiveroides.*

EXTRACT	Quantity in micrograms (µg), Mean±S.E.M					IC_{50} Values
	1.25	50	100	200	400	
Ascorpic acid	20.31±2.11	31.69±1.14	45.44±1.28	64.77±2.31	76.94±2.26	212.48
HAEPV	14.63±1.73	24.46±0.77	32.50±0.82	43.63±2.07	52.88±1.06	290.12

Table 8: Percentage inhibition and IC50 values of DPPH radical scavenging activity *in vitro* by hydroalcoholic extract of *Plectranthus vettiveroides*

EXTRACT	Quantity in micrograms (µg), Mean±S.E.M					IC_{50} values
	25	50	100	200	400	
Ascorpic acid	26.48±1.11	40.85±0.80	54.98±1.79	72.42±2.37	87.85±2.05	71.56
HAEPV	19.44±2.33	31.13±1.26	37.92±1.13	46.53±1.01	49.23±0.83	122.20

Figure 1. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on L6 cell lines

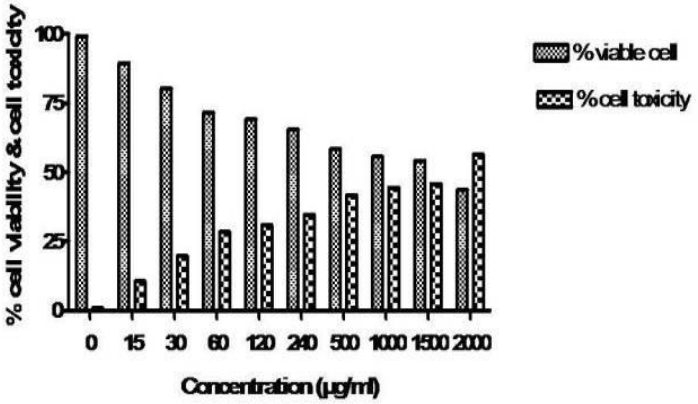
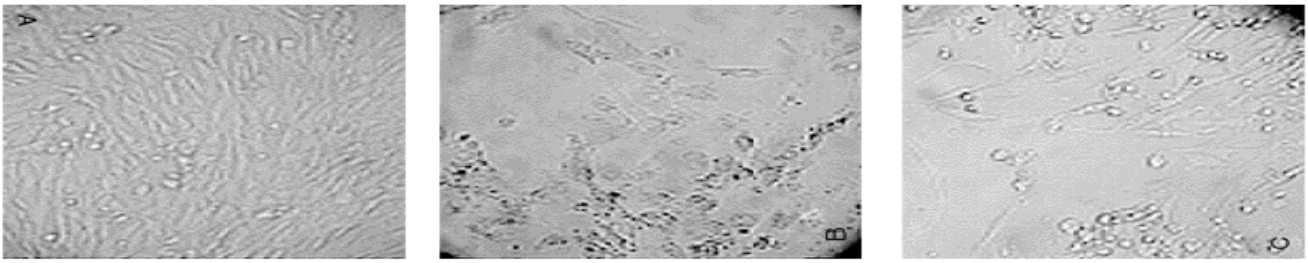


Figure 2. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on L6 cell lines



CONTROL **1000 µg/ml** **2000µg/ml**

Figure 3. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on EAC cell lines.

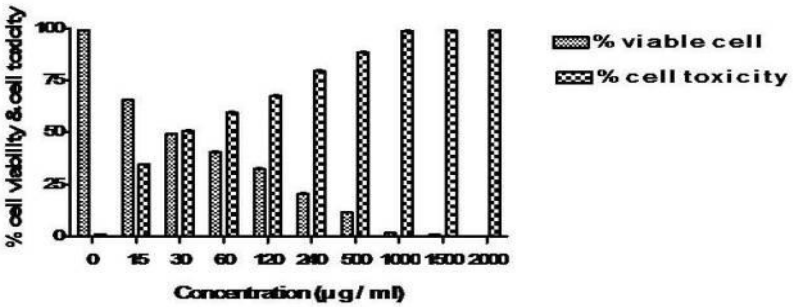
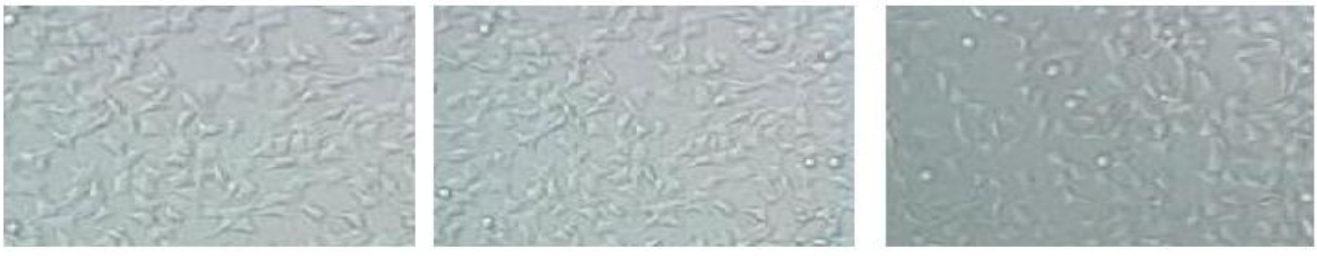


Figure 4. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on EAC cell lines.



CONTROL **1000 µg/ml** **2000 µg/ml**

Figure 5. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on MCF-7 cell lines.

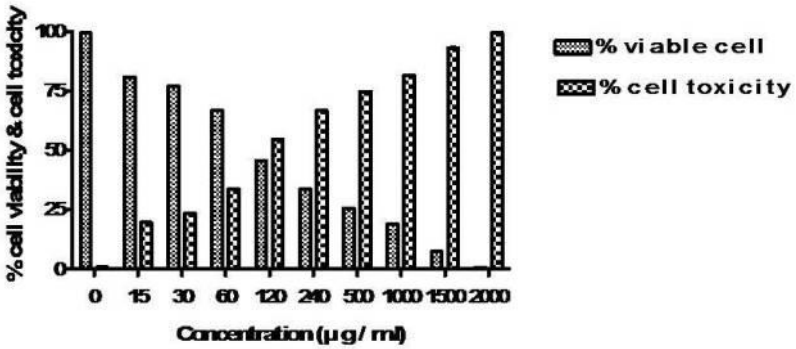


Figure 6. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on MCF-7 cell lines.

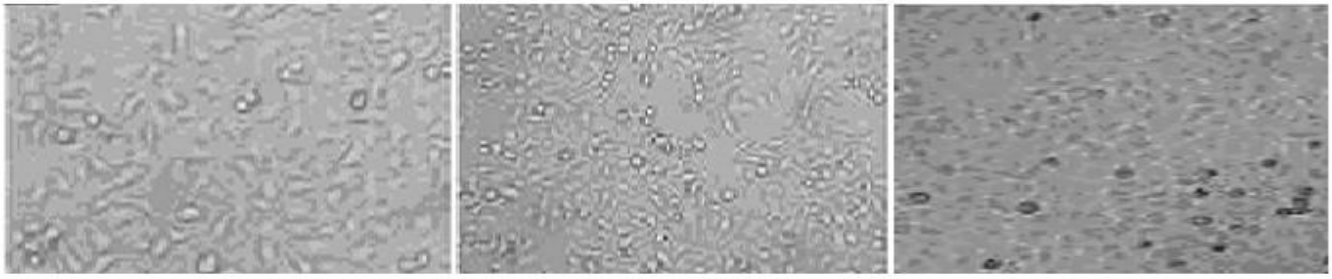


Figure 7. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthus vettiveroides* on HEP-G2 cell lines using.

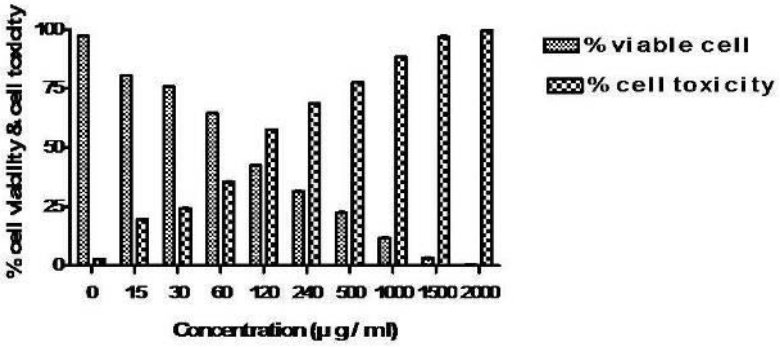


Figure 8. Evaluation of cytotoxicity and cell viability for hydroalcoholic extract of *Plectranthuss vettiveroides* on HEP-G2 cell lines using.

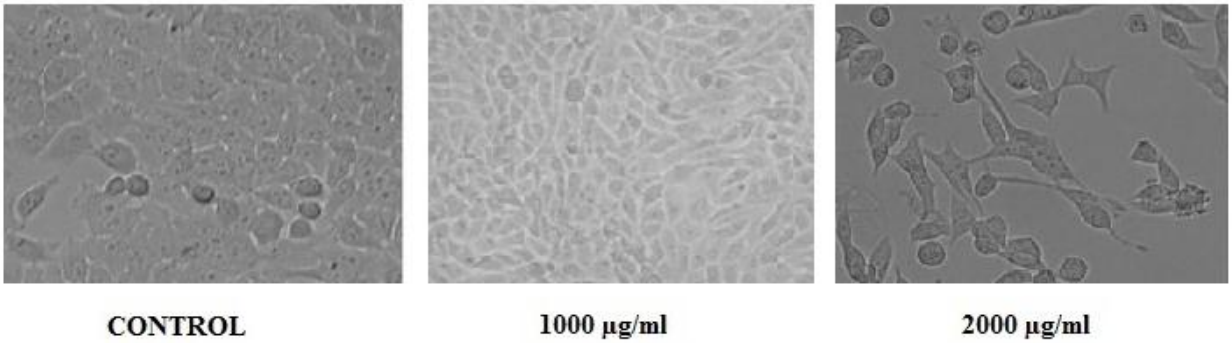


Figure 9. Percentage inhibition and IC50 values of superoxide radical scavenging activity *in vitro* by hydroalcoholic extract of *Plectranthus vettiveroides*

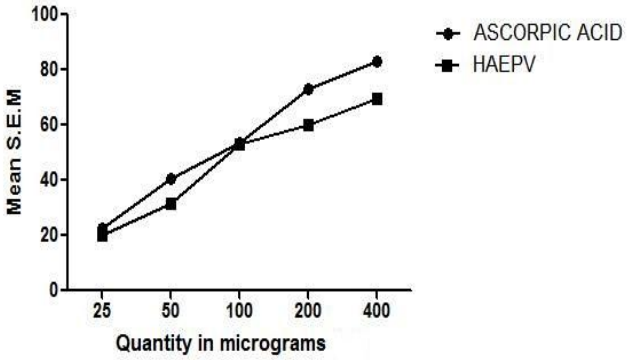


Figure 10. Percentage inhibition and IC50 values of Hydroxyl radical scavenging activity *invitro* by hydroalcoholic extract of *Plectranthus vettiveroides*

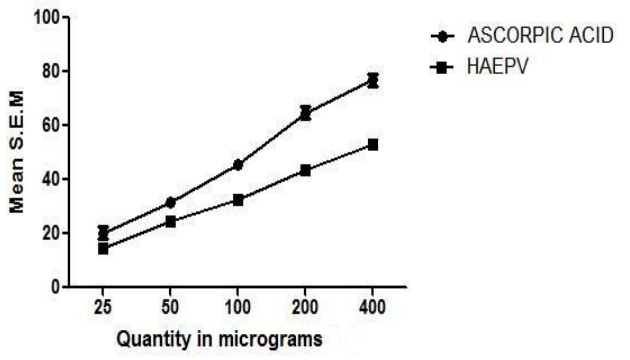
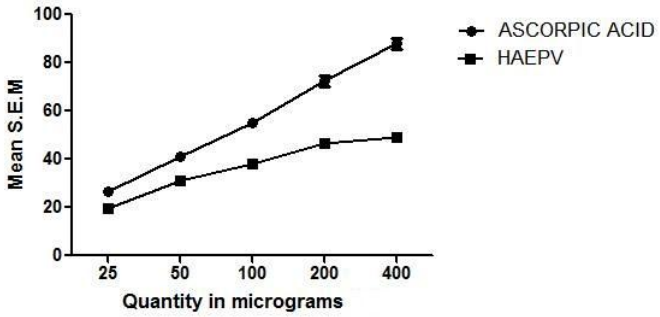


Figure 11. Percentage inhibition and IC50 values of DPPH radical scavenging activity *in vitro* by hydroalcoholic extract of *Plectranthus vettiveroides*



CONCLUSION

Hydro alcoholic extract of *Plectranthus vettiveroides* showed significant *in vitro* anti-cancer activity and *in vitro* anti-oxidant activity, The obtained data suggested that the phytoconstituents present in the extract is responsible for *in vitro* anticancer activity and *in vitro* antioxidant activity. If other related further studies and clinical trials are carried out, it will definitely open up a new vistas in modern medicine.

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CONFLICT OF INTEREST:

The authors declare that they have no conflict of interest.

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