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Anticancer activity of *Lantana Camara* hexane extract against brain carcinoma

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Abstract
Cancer becomes a global problem now days. A natural derivative becomes a new hope for the researchers. Lantana *camara* is known for anti-inflammatory, anti-oxidant, insecticidal activities. It is also having good anticancer activity against breast cancer cells. The purpose of the study to evaluate the anticancer activity of Lantana *camara* extracts against brain carcinoma whereas Hexane extract showed potent anticancer activity compare to ethyl acetate extract.

Keywords: lantana *Camara,* brain carcinoma, hexane extract, ethyl acetate extract

Introduction

Lantana genus having 150 species of perennial floweing plants belongs to Verbenaceae family. Lantana camara linn a sherb found easily and having applications in folk medicine [1]. The antibacterial and antifungal activities of lantana camara essential oil were reported [2-10]. Furthermore, it showed several other activities such as insect repellent [11], insecticidal [12] anti-nematodes [13], larvicidal [14], anti-inflammatory [15] and antioxidant activities [16]. After the analysis of Lantana found presence of flavonoids, glycosides and alkaloids in the aerial part and roots for fixed compounds of genus lantana [15, 17-19]; while the volatile constituents from showed abundance of monoterpenes and sesquiterpenes [8, 9, 20].The emergency and spread of cancer disease justifies searching for save and effective solutions through natural ways. Therefore, the present study aimed to identify natural anticancer components from the yellow flower of L. camara against brain carcinoma.

Materials and Methods

Plant Collection and Extraction:

The L. camara wild plant was collected from the Roorkee and Bhagwanpur areas of Uttarakhand, India. The plant was identified in Forest Research Institute, Dehradun, Uttarakhand, India. The plant flowers were dried in shadow and homogenized. Since this study aimed at extraction of any bioactive compound from flowers per formed using different organic solvents with increasing polarity, not by hydro distillation that extract the volatile compounds only. To do that, ten gm of the dried flowers, the extraction were sequentially extracted by soaking in 100 ml of different solvents with increasing polarity (hexane, petroleum ether, ethyl acetate and methanol) in a conical flask on a rotary shaker for one day at room temperature. The supernatant was separated by filtration and evaporated by rotary evaporator at 40°C. The remaining residue after evaporation was dissolved in 1 ml of dimethyl sulfoxide (DMSO) for anticancer test.

Anticancer Test

MTT test [3-(4, 5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] was used. Cell lines (5.0 × 104) were plated in 96-well plates with serum-free RPMI-1640 media. Aliquots from each plant extracts at 0, 2, 4, 8, 16, 32, 64, and 128 µg/ml concentration in triplicates were added followed by incubation for 24 h at 37°C in a 5% CO_2 incubator. The media were then removed and 100 µl of 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) reagent was added to each well and was incubated again for 3-4 hr. Before adding 100 µl dimethyl sulfoxide to each, MTT reagent was removed and gently shaken. The untreated cells were compared to plant extract treated cells. The absorbance was measured at 570 nm using a microplate-reader.

Result and Discussion

Cytotoxic and growth inhibitory effects of the selected plant extracts on human cancer cells were studied by MTT assay. Whereas for hexane extract A549 (human lung cancer cells), A-172(Brain Cancer cells) and PC3 (Prostate cancer cells) showed IC50 µg/ml 42.39±3.08, 8.30±1.48, 66.40±2.68 showed respectively.it showed that hexane extracts is having potent constituents against cancer cells. The positive control, doxorubicin imparted cytotoxic and dose dependent inhibition of cell proliferation and the IC_{50} values of doxorubicin was found to be 1.25 ± 0.05 µM for A549 (human lung cancer cells), 4.10 ± 0.03 µM for A-172 (Brain Cancer cells) and 2.12 ± 0.14 µM for PC3 (Prostate cancer cells), respectively. This study can be carry forward for future scope.

Table 1: IC_{50} values of *Lantana Camera* extracts IC_{50} µg/ml

Groups	A549 (lung Cancer Cell line)	A-172 (Brain Cancer cell line)	PC3(Prostate Cancer cell line)
Lantana Camara Hexane Extract	42.39±3.08	8.30±1.48	66.40±2.68
Lantana Camara Ethyl acetate Extracts	430.90±13.44	589.80±24.56	830.70±38.54

Doxorubicin	1.25±0.05	4.10±0.03	2.12±0.14

IC_{50} (Concentration of the drug required to reduce the percentage cell viability to 50 percentage) were obtained from the graph by non-linear regression analysis as best curve- fit values. IC_{50} values of Lantana camara extracts and standard anticancer drug, doxorubicin were compared with human Lung cancer cells (A549); human brain cancer cells (A-172) and human prostate cancer cells. *The IC_{50} of doxorubicin is represented as µM. Numerical data are means ± S.D of three independent experiments (n=3). Statistically significant difference from IC_{50} values for the control cells exposed to vehicle control (blank) ($p < 0.001$) The yellow flowers of Lantana camara plant, only the hexane extract showed potent anticancer activity against brain cancer cells. TLC analysis of the hexane extract using hexane petroleum ether: hexane 1:1 and 1:2 mobile phases showed no separation. Therefore, other mobile phases with higher polarity index values (ethyl acetate (4.4) and Methanol (5.1)) were used. Sabinene was also found in the essential oil of Myristica fragrans (16.1%) that showed anticancer activity against colon cancer [23] and the essential oil of Tridax procumbens that showed anticancer activity against melanoma cell line [24]. Sabinene was also found in the essential oil of the aerial parts of Pituranthos tortuosus that caused apoptosis of B16F10 melanoma cells [25] and in the essential oil of Thymus vulgaris (0.8%) that showed anticancer activity against breast adeno carcinoma, Human alveolar basal epithelial and hepatocellular carcinoma cells [26]. The essential oil (EO) of Tanacetum annuum aerial parts, containing 22.3% sabinene showed anticancer activity against human rhabdomyosarcoma cancerous cell line [27]. The essential oil of Neolitsea variabillima showed anticancer activity against human oral, liver, lung, colon, melanoma and leukemic cancer cells. The GC/MS analysis of this oil showed presence of trans-beta-ocimene (13.4%), alpha-cadinol (10.5%), terpinen-4-ol (9.3%), tau-cadinol (9.2%), beta-caryophyllene (8.8%) and sabinene (6.7%). The anticancer activity of this oil was attributed to beta-caryophyllene, tau-cadinol, and alpha-cadinol [28]. L. camara essential oil from leaves was cytotoxic to V79 mammalian cells and also to Artemia salina, showing 50% lethal concentration (LC50) value from 0.23 µg/mL. The major components of the essential oil were varying with the season of sample collection. Generally, the basic components were β-caryophyllene (10.5%), sabinene (7.98%), limonene (7.68%) and spathulenol (11.64%) [29]. The Haplophyllum tuberculatum essential oil showed activity against lung and liver cancer cells. The GC/MS analysis of this oil showed presence of 3-Carene (3.8%) and Eucalyptol (1.6%) [30]. β-Caryophyllene was a major component of the Commiphora gileadensis essential oil that showed anticancer activity against lymphoma cell line. β-Caryophyllene was tested alone and showed the same effect [31]. The anticancer activity of β-Caryophyllene was also reported by Klaudyna *et al*. [32].

Conclusion

The volatile components of the L. camara yellow flower showed anticancer activity against brain cancer cells investigated by MTT test. The GC/MS analysis showed presence of monoterpenes (sabinene, cis- delta carene, eucalyptol (1,8-Cineole), geranyl isovalerate and menthol-derivative) and sesquiterpenes (7-epi-trans-sesquisabinene hydrate, trans-caryophyllene, tau.-cadinol and limonen-6-ol, pivalate). The literature showed presence of these components as major constitutes in the different essential oils that showed anticancer activities against variable cancer cells.

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