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In vitro anticancer activity of *Launaea procumbens* (Roxb.) against different cancer cell lines

Deepa K.N. and D. Kotresha*
Department of Studies in Botany,
Davangere University, Shivagangothri, Tholahunase, Davangere-577007 (Karnataka), India.

(Corresponding author: D. Kotresha)*
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ABSTRACT: The present study focuses on *Launaea procumbens*, a wild plant from the Asteraceae family, commonly found as a weed in maize and onion fields. It is utilized for treating different health conditions such as liver and kidney disorders, antifungal issues, antioxidant benefits, and cardio-protective properties. Additionally, in certain regions, it is consumed as a food item in the form of a salad. The plant samples were collected from maize fields in and around Davangere district, Karnataka. The primary aim is to conduct a phytochemical analysis, GCMS profiling, and assess anticancer activity against four different cancer cell lines. Phytochemical analysis involved three different solvents for extracting compounds derived from the aerial parts of the plant. The identification of chemical compounds was carried out using GCMS analysis from chloroform and methanol extracts. Cytotoxicity against four cancer cell lines (NCIH-460, MDAMB-231, K562, and HePG2) was evaluated using MTT assay with three different solvents of plant extracts. Phytochemical analysis revealed the presence of various components, indicating the existence of phytoconstituents like alkaloids, saponins, tannins, proteins, phenols, and terpenoids, glycosides and carbohydrates. GCMS analysis of plant extracts of chloroform and methanol revel the presence some active compounds and these are responsible for anticancer activity against selected cancer cell lines. Out if four cancer cell lines, MDAMB-231 and NCIH-460 shows good cytotoxic activity with IC_{50} values are 45.23 µg/ml and 35.23 µg/ml, respectively. *Launaea procumbens* was possess some significant secondary metabolites and exhibited potential as an anticancer agent against four different cancer cell lines. However, additional research is necessary to find out particular compounds responsible for its anticancer properties, and additional investigations are needed to elucidate its molecular mechanisms.

Keywords: *Launaea procumbens,* Aerial part, phytochemical analysis, GCMS, MTT assay.

INTRODUCTION

Nature has been an abundant provider of medicinal resources in the form of plants for thousands of years. Indeed, many traditional medicines used in healthcare originate from plants, highlighting the vital role of folk medicines in herbal remedies. Plants containing compounds are classified as medicinal plants, capable of therapeutic uses or serving as derivatives for chemical drug synthesis. Plants are traditionally used worldwide for various ailments and against many infectious agents (Shapiro, 2006). As per the World Health Organization (WHO) assessment, 80% of the populace in developing nation have faith in traditional medicine and use plant-derived drugs for their primary healthcare (Yirga, 2010). Despite several progressions in the field of chemical drugs and antibiotics, plants remain the principal materials for drugs treating many human diseases (Jain *et al.*, 2019). The second most significant cause of global mortality is cancer and in developing countries it is responsible for 9.6 million deaths. Seventy percent of people lose their lives due to cancer. Chemotherapeutic drugs for cancer, as indicated by references produce more effects such as pulmonary, renal toxicity, cardiac, and neurological problems. Therefore, plant-derived compounds may be a better remedy for cancer treatment with the least side effects (Alonso-Castro *et al.*, 2011; Shin *et al.*, 2019). Lung cancer holds the top position as the most prevalent form of cancer globally (Teixeira *et al.*, 2013). It occurs especially in males aged between 65-70 years, primarily in chronic tobacco users, accounting for in 2015, lung cancer accounted for 1.69 million death rates (Ballestreri *et al.*, 2018; Karczmarek *et al.*, 2014; Ginsburg, 2018). A serious note is that nearly 85-90% of patients suffer from lung cancer (Audrey Mouche & Rémy Pedeux 2020). This cancer forms an aggressive tumor with a poor diagnosis, resulting in an aggregate mean total survival of over five years. Recent developments show that the death rate of lung cancer is high because the disease is detected in late stage. Among women, breast cancer stands as second most prevalent form of cancer, affecting 30% of women with approximately 2.3 million new cases reported in 2020. Women, especially from middle age to older age, suffer from this type of cancer (Rudrappa *et al.*, 2023). The incidence of female breast cancer patients have been dramatically increased, with a simultaneous 6.9% rise in the mortality rate (Bray *et al.*, 2018). Leukaemia is most prevalent among a diverse hematologic

malignancies characterized by the abnormal proliferation of immature lymphoid cell (Redaelli *et al.*, 2003). In 2020, approximately 65,530 new leukaemia cases were recorded. Among those, there were 19,940 newly diagnosed cases, and 23,100 people lost their lives due to leukaemia cancer (Ginsburg, 2018). Worldwide, liver cancer holds the position of the third most prominent cause of cancer and approximately 75% of liver cancer patients are from Asia (Arzumanian *et al.*, 2021). According to the National Cancer Registry, in 2012, 92 females and 229 males in Asian countries were suffering from liver cancer. In 2020, 34,743 new cases were recorded, with nearly 33,793 death rates occurring due to liver cancer.

A member of the Asteraceae family, *Launaea procumbens*, is a wild annual edible plant that grows as a weed in crop fields and waste lands, particularly in rich black soil. It consists of small yellow flowers and simple leaves. This family, comprising 1000 genera and 20000 species (Rawat *et al.*, 2021). The plant holds significant medicinal value and is used in herbal and Ayurvedic preparations for longevity, wound healing, and as a washing agent and food supplement (Wazir *et al.*, 2007). It addresses various health issues such as reproductive disorders, painful urination**,** (Ahmad, 2006) hormonal imbalance (Qureshi and Bhatti 2008) and liver dysfunction (Khan *et al.*, 2013).

Additionally, the plant exhibits biological activities including insecticidal and antifungal properties, antipyretic effects (Baquar, 2014) anticancer, antioxidant, cardio-protective, and neuroprotective effects, (Rathod, 2014**)** antibacterial properties, (Reddy & Mishra 2012) antitumor effects (Khan *et al.*, 2016), antiurolithatic effects (Makasana *et al.*, 2014) hepato-protective effects (Khan *et al.*, 2014) and cytotoxic activity (Rawat *et al.*, 2016). In folk medicine, the leaves of *L. procumbens* are used for treating fever, swelling, toxiemia, and kidney stones. It is particularly effective for jaundice (Shaukat *et al.*, 2003). The current investigation adopted a novel approach by concentrating on the plant's aerial parts to assess anticancer activity against various cancer cell lines, marking the first-time reporting of such an investigation. Previous research primarily utilized the leaf part for similar purposes. The research study aimed to conduct preliminary phytochemical analysis and characterize the GCMS analysis to identify novel compounds present in the extract from the aerial parts. The results indicated significant cytotoxic activity against selected cell lines. This Pioneering work suggests the potentiality for developing a promising herbal medicine treatment for cancer.

MATERIALS AND METHOD

Taxonomic description:

Launaea procumbens (Roxb.) is a herbaceous plant leaves are entire, glabrous, radical lower leaves belong obovate along with obtuse segments leaf margin having spinulose with cartilaginous teeth leaves are distinct in position cauline sessile arranged in rosette manner. Flowers are yellow solitary heads are cylindrical with 1.2 -1.9 cm in length with short pedicle involucre of bract outer once are short acute ovate. Stamens are five basified are arranged in syngenious condition single ovule unilocular bicarpellary basal placentation style is terminal with bifid stigma. Fruits are achene.

Fig. 1. Habit of *Launaea procumbens.*

Plant samples collection: Plant samples were collected from local region around Davangere district, Karnataka, India in the maize field after harvesting in the month of July-August. The collected plant was identified by department of Central Ayurveda Research Institute; Bangalore with voucher specimen number (RRCBI18671) afterwards, the plant sample underwent a meticulous rinse with flowing tap water, followed by drying in the shade. Subsequently, it was ground into a powdered sample and deposited into an airtight container. The fine powder was used for Soxhelt extraction by using three different solvents.

Preparation of plant extract: The aerial parts of *Launaea procumbens* were collected, dried in the shade, ground into a fine powder with a blender, and subsequently stored at room temperature for a few days before further extraction. The fine powder was used for Soxhelt extraction by employing three different solvents they are methanol, chloroform and aqueous and maintain the temperature with respective solvent and the filtrate are reduced to get the desire consistency of the extracts and evaporate in a desiccator, storage the extract for further usage.

Phytochemical analysis: *Launaea procumbens* aerial part extracts of Methanol, Chloroform and Aqueous were examined the presence of alkaloids, flavonoids, Saponins, glycosides, tannins, proteins and carbohydrates. Standard methods with slight

modification are used for preliminary phytochemical analysis (Harborne, 1984).

GC-MS. Chemical compound identification in the sample was carried out using GCMS analysis. The aerial part of *Launaea procumbens* was analysed employing a Perkin Elmer instrument (GCMS QP2010 SE, Shimadzu instrument, Columbia, M.D., USA). The aerial part extract of *Launaea procumbens* was mixed with 100 ml of derivatization reagent (80µl BFSTA +20µl TMCS) for 1 hr and subjected to incubation at 65°C. Then, each extract was injected into an Rtx5 MS 30M of column with 0.25mm ID & 0.25 µM df. The interference temperature at 300ºC an ion source of temperature maintained at 25ºC & the injection temperature was held at 300°C, A carrier gas of helium was utilized at a flow rate of 1 ml/min. The analysis was conducted through isothermal heating at 100°C for 1 min. Afterward, a heating phase at 300°C for 20 minutes ensued, with mass spectra subsequently recorded at a rate of 2 scans per second, covering a varying scanning range of 40 to 850m/z by using turbo mass software quantified the component based on peck area & normalization as per internal standard. The chemical composition of aerial part extract of *L. procumbens* comparing the identified spectra with those of a NIST/Wiley library and with obtained compounds.

GC-MS (Gas chromatography mass spectrometry) Profiling: The identification of chemical compounds present in the sample was conducted through GCMS analysis. The aerial part of *Launaea procumbens* using a perkin Elmer instrument (GCMS QP2010 SE, shimudzu instrument, Columbia M.D USA). The prepared aerial part *Launaea procumbens* aerial part extract were added with 100 ml of derivatization reagent (80µl BFSTA +20 µl TMCS) for 1hr & subjected to incubated at 65ºC. Then each extract was injected in a Rtx5 MS 30M of column with 0.25mm ID & 0.25 µM df. The interference temperature at 300ºC an ion source of temperature maintained at 25ºC &the injection temperature was held at 300°C, and a flow rate of 1 ml/min of helium gas was employed as the carrier gas. The analysis was conducted through isothermal heating at 100°C for 1 min, followed by a subsequent heating phase at 300°C for 20 min. Mass spectra were then recorded at a rate of 2 scans per second, covering a varying scanning range of 40 to 850m/z by using turbo mass software quantified the component based on peck area & normalization as per internal standard. The chemical composition of aerial part extract of *Launaea procumbens* comparing the identified spectra with those of a NIST/Wiley library and with obtained compounds.

Cell culture method: The HePG2, NCIH-460, MDAMB-231 and K562 cancer cell lines were purchased from NCCS, Pune, India. The medium used for culture the cancer lines is DMEM (high-glucose Dulbecco's Modified Eagle Medium](#AL111 Himidia for NCIH-460 and MABMD-231 and low glucose medium (at AL149 HIGH MEDIA) for HePG2 and K562 supplemented with, 10% fetal bovin serum (FBS #RM00432 high media) humidified incubator 5% CO_2 at 37º C till confluence was produced.

***In vitro* cytotoxic assay:** A 200 ml cell suspension was distributed into 69 cell plates to achieve the desired cell density (20000 cells per plate) and incubated at 37°C with 5% CO_2 for 24 hrs. Resulting in the partial formation of a monolayer. Subsequently, the cells were subjected to treatment with methanol aerial part extract at various concentrations (12.5 µg/ml, 25 µg/ml, 50 µg/ml, 100 µg/ml, and 200µg/ml) were applied to different wells. The control wells, containing untreated cells, received only maintenance medium, and the positive control received Camptothecin. (at C9911sigma) for MDAMB-231, HepG2 cell lines and Doxorubin (at D515 sigma) for NCIH-460, K562 cell lines.

Afterward, the plates were placed in an incubator set at 37°C with 5% CO_2 and 75% relative humidity for 24 hours (Heal Force, China). Subsequent to this, A labelling compound of 0.5 mg/ml MTT was introduced, and the cells were subjected to incubation for an additional 3 hours. Following that, each well received 100 ml of DMSO to dissolve the formazan formation in the final step. Measurements of sample absorbance were conducted using a microplate (ELISA) reader at 570 nm and 630 nm. The IC50 value was calculated through a linear regression equation, with Y=50, and the M and C values were obtained from the viability graph. The influence of extract *Procumbens* the determination of viability was carried out using the as given below

$$\% \text{ viability} = \frac{\text{Mean abosrbance of treated cell}}{\text{Mean absorbance of untreated cell}} \times 100$$

The formula to calculate percentage of cytotoxicity is expressed as:

% cytotoxicity =100 - % cell viability

RESULTS

Table 1: Preliminary qualitative phytochemical analysis.

Phytochemical test	Methanol	Chloroform	Aqueous
Alkaloids	+	+	-
Flavonoids	+	+	-
Saponins	+	-	-
Glycosides	+	+	+
Tannins	+	-	-
Protein	+	+	-
Phenols	+	-	+
Saponins	-	-	+
Terpenoids	+	+	-
Carbohydrates	+	+	-
Positive (+) and Negative(-)			

The qualitative analysis of *Launaea procumbens* shows the presence in Phytochemicals in three different solvents such as methanol, chloroform and aqueous.

GCMS analysis: The analysis of mass spectra and identification of components based on their retention indices were conducted utilizing the National Institute of Standards and Technology (NIST) database. The database encompasses over 62,000 patterns of known chemical. For identification purposes, the mass spectra of unidentified components were cross-referenced with the standard mass spectra of known compounds archived in the NIST collections.

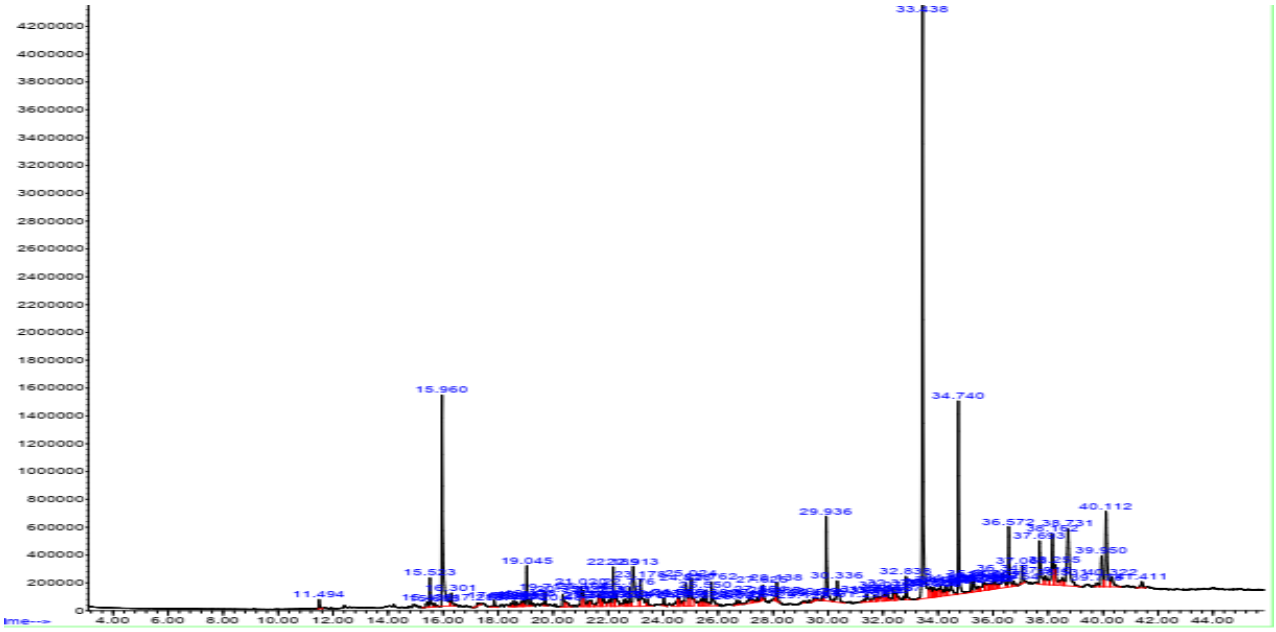


Fig. 2. GCMS graph showing chromatogram with different retention time.

Table 2: List of compounds obtained from GCMS from chloroform extract.

Sr. No.	Identified Compound Names	Molecular Weight	Molecular Formula	RT	Area%
1.	Heptadecane, 2,6,10,15-tetramethyl	296.5741	$C_{21}H_{44}$	15.43	0.27
2.	Heptadecane	240.4677	$C_{17}H_{36}$	15.52	0.89
3.	Hexadecane	226.4412	$C_{16}H_{34}$	16.3	0.51
4.	Octacosane	394.7601	$C_{28}H_{58}$	18.68	0.2
5.	9-methylheptadecane	268.5209	$C_{19}H_{40}$	18.91	0.21
6.	Pentacosane	352.6804	$C_{25}H_{52}$	19.17	0.25
7.	1-Octadecene	252.5253	$C_{18}H_{36}$	20.35	0.66
8.	1-Octadecanesulphonyl chloride	353.0254	$C_{18}H_{37}ClO_2S$	20.487	0.39
9.	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-,[1S-(1.alpha.,2.beta.,5.alpha.)]-	138.2524	$C_{10}H_{18}$	21.02	0.77
10.	Oxirane, tetradecyl	268.4778	$C_{18}H_{36}O$	21.38	0.39
11.	Heneicosane, 11-(1-ethylpropyl)	366.707	$C_{26}H_{54}$	21.66	0.7
12.	Hentriacontane	436.8399	$C_{31}H_{64}$	22.68	0.2
13.	1,2-Benzenedicarboxylic acid, butyl 2-methylpropyl ester	278.3435	$C_{16}H_{22}O_4$	22.91	3
14.	Nonadecane, 2,3-dimethy	296.5741	$C_{21}H_{44}$	24.53	0.53
15.	Phytol	296.531	$C_{20}H_{40}O$	24.82	1.67
16.	Heptadecylheptafluorobutyrate	452.4902	$C_{21}H_{35}F_7O_2$	25.76	1.19
17.	Bis(2-ethylhexyl) phthalate	390.5561	$C_{24}H_{38}O_4$	29.93	3.36
18.	Nonahexacontanoic acid	999.836	$C_{69}H_{138}O_2$	30.33	1.31
20.	Indolizine, 2-(4-methylphenyl)-	207.27	$C_{15}H_{13}N$	34.03	0.52
21.	Fumaric acid, 2-decyl tridecyl ester	477.849	$C_{23}H_{31}Cl_3O_4$	34.28	0.51
22.	13-Methyl-Z-14-nonacosene	420.8	$C_{30}H_{60}$	35.3	0.69
23.	4,6-Cycloheptatrien-1-one, 3,5-bis-trimethylsilyl-	250.48	$C_{13}H_{22}OSi_2$	35.73	0.49
24.	1,4-Bis(trimethylsilyl)benzene	222.4741	$C_{12}H_{22}Si_2$	35.92	0.31
25.	beta-Sitosterol	414.7	$C_{29}H_{50}O$	37.69	2.29
26.	beta-Amyrin	426.7	$C_{30}H_{50}O$	38.16	2.54
27.	Hydrocinnamic acid, benzyldimethylsilyl ester	298.5	$C_{18}H_{22}O_2Si$	39.95	1.67
28.	Taraxasterol	426.7	$C_{30}H_{50}O$	40.11	4.1

Table 3: List of compounds obtained from GCMS from methanol extract *in vitro* cytotoxic activity.

Sr. No.	Identified Compound Names	Molecular Weight	Molecular Formula	RT	Area%
1.	Tridecanoic acid	214.1932	$C_{13}H_{26}O_2$	12.54	4.36
2.	Undecanoic acid	186.16198	$C_{11}H_{22}O_2$	14.74	6.74
3.	Pentadecanoic acid, 14-methyl-, methyl ester	270.2558	$C_{17}H_{34}O_2$	15.84	12.3
4.	n-Hexadecanoic acid	256.2402	$C_{16}H_{32}O_2$	16.08	72.4
5.	9-Octadecenoic acid (Z)-, methyl ester	296.2715	$C_{19}H_{36}O_2$	16.76	10.7
6.	cis-Vaccenic acid	282.2558	$C_{18}H_{34}O_2$	16.95	17.1
7.	Glycidylpalmitate	312.2664	$C_{19}H_{36}O_3$	17.66	74.8
8.	Z-(13,14-Epoxy) tetradec-11-en-1-ol acetate	268.2038	$C_{16}H_{28}O_3$	18.68	1.1
9.	2-Undecanol palmitate	410.4123	$C_{27}H_{54}O_2$	18.94	0.28
10.	Eicosanoic acid, 2,3-bis(acetyloxy)propyl ester	470.3607	$C_{27}H_{50}O_6$	20.02	4.34
11.	1,1,1-Trifluoroheptadecen-2-one	308.2327	$C_{17}H_{31}F_3O$	21.19	2.36
12.	Di-n-decylsulfone	346.2905	$C_{20}H_{42}O_2S$	23.72	4.22

Table 4: The effect of three various solvent extracts from the plant extracts of *procumbens* on four different cancer cell lines.

Plant name	Extract	HepG2	NCIH-460	MDAMB-231	K562
		IC_{50}(µg/ml)			
Launaea procumbens	LPA	190.53	165.82	235.86	118.98
	LPC	193.45	35.33	55.08	213.65
	LPM	166.72	30.79	42.44	190.54

To understand the anticancer activity of *Launaea procumbens* plant extracts, cytotoxicity assays were conducted on the aerial part. The extracts tested included *L. procumbens* methanol (LPM), *L. procumbens* chloroform (LPC), and *L. procumbens* aqueous (LPA). The assays were performed on four different cancer cell lines: MDAMB-231 (Human breast cancer cell), HePG2 (Human liver cancer cell), K562 (myelogenous leukemia cancer cell), and NCIH-460 (Lung cancer cell line). Various concentrations were used, positive control received Camptothecin (at C9911 sigma) for MDAMB-231, HepG2 cell lines and Doxorubin (at D515 sigma) for NCIH-460, K562 cell lines. The cytotoxicity of four cancer cell lines was assessed through the MTT (3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyltetrazolium bromide) assay with concentrations of plant extracts (12.5 µg/ml, 25 µg/ml, 50 µg/ml, 100 µg/ml, and 200 µg/ml) were applied to the cells, and the incubation period was 48 HR. The concentration-dependent inhibition of cell proliferation was observed over the designated time duration.

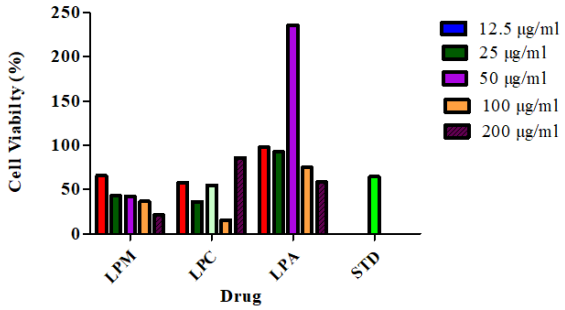


Fig. 2. Anti proliferative effects of *Launaea procumbens* Methanol (LPM), *Launaea procumbens* Chloroform (LPC), *Launaea procumbens* Aqueous (LPA) on MDAMB-231 cell line on 48h treatment, Camptothecin (STD; 8µg/ml) used as a positive control.

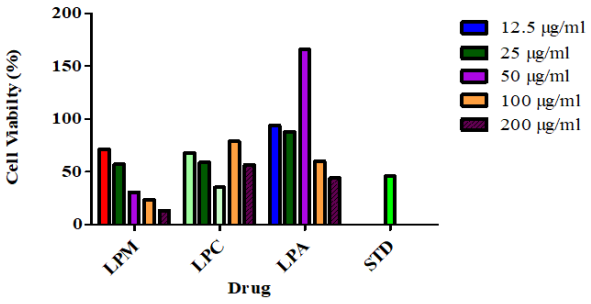


Fig. 3. Anti proliferative effects of *Launaea procumbens* Methanol (LAM) *Launaea procumbens* Chloroform (LAC) *Launaea procumbens* Aqueous (LAA) on NCIH-460 cell line on 48h treatment, Doxorubin (STD; 5ug/ml) Employed as a positive control.

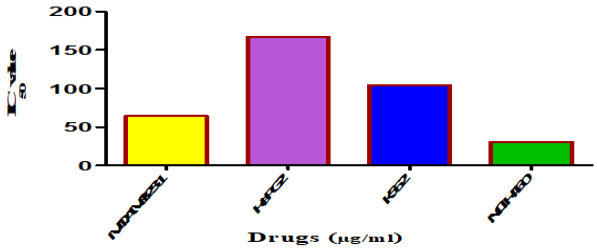
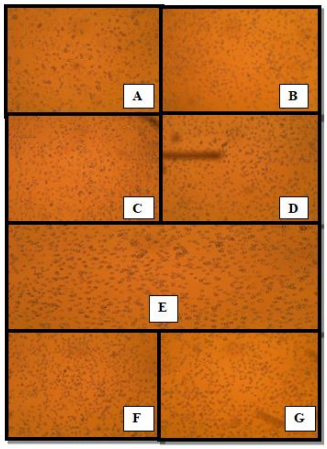
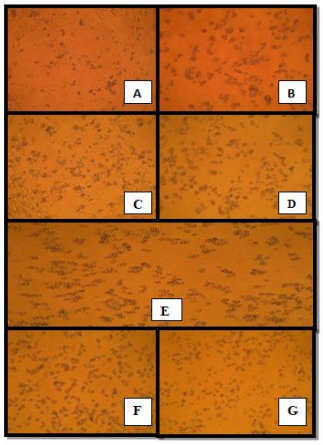


Fig. 4. Cytotoxicity activity of *Launaea procumbens* Methanol (LPM), *Launaea procumbens* Chloroform (LPC) *Launaea procumbens* Aqueous (LPA) on four different MDAMB-231, HePG2, K562 and NCIH-460 cell lines on 48h treatment.



A-control, B-Untreated, C-12.5mg/ml, D-25mg/ml, E-50mg/ml, F-100mg/ml and G-200mg/ml

Fig. 5. Cytotoxic activity MDAMB -231 cell line show different concentration of *Launaea procumbens* in Methanol extract observed under contrast microscope.



A-control, B-Untreated, C-12.5mg/ml, D-25mg/ml, E-50mg/ml, F-100mg/ml and G-200mg/ml

Fig. 6. Cytotoxic activity on NCIH-460 cell line shows different concentration of *Launaea procumbens* in Methanol extract observed under contrast microscope.

DISCUSSION

The present study focuses on aerial part of *Launaea procumbens*, a wild plant from the Asteraceae family, was used to carry out phytochemical analysis revel the presence of phytoconstituents alkaloids, flavonoids, aponins, glycosides, terpenoids, tannins, proteins and carbohydrates are shown the Table 1. Similarly, according to some literature survey phytochemical *Launaea procumbens* for various fractions prepared from methanolic extract showed major bioactive molecules responsible for anticancer activity (Rawat *et al.*, 2016).

That GCMS analysis leaves part of methanolic extract possessing phthalate, fatty acids and triterpenoids (Rawat *et al.*, 2021), present our work. In GCMS of crude aerial methanolic and chloroform extracts of *Launaea procumbens* to determine its volatile compounds. According to our knowledge concerned this are first time noted compounds in aerial part of *Launaea procumbens* chloroform and methanol extract. The literature on heptadecane indicates its presence in *Spirulina platensis*, a blue-green alga, and highlights its anti-inflammatory properties (Kim *et al.*, 2013). Recently, hentriacontane and octadecane, 3-ethyl-5-(2-ethylbutyl)-, were discovered in *Polygonatum verticillatum*, showing promising anti-inflammatory and anticancer effect (Singh & Patra 2018). Indolizine, 2-(4-methylphenyl)-, found in this

source, exhibits various bioactivities, including cytotoxicity (Sharma & Kumar 2014) Fumaric acid derivative, specifically Fumaric acid, 2-decyl tridecyl ester, known for its anti-inflammatory and analgesic properties, was identified in extracts from *Fumaria indica* (Shakya *et al.,* 2014). Additionally, recent findings highlighted the neuroprotective and anti-inflammatory properties of fumaric acid derivatives (Cateni *et al.,* 2021) beta-Sitosterol, structurally similar to cholesterol and with multiple bioactivities, including cytotoxicity, was identified in this study. Other research demonstrated its efficacy against HeLa, MCF-7, and NIH/3T3 cancer cell lines (Ayaz *et al.,* 2019). Some other research work showed its hepatocellular anti-cancer activity from the plant *Indigofera zollingeriana* (Vo *et al.,* 2020). Taraxasterol, identified in this study, exhibits various bioactivities, including potential anticancer properties, as recently reviewed by Jiao *et al.* (2022)

In the present section, the compounds isolated from the methanol extract of *Launaea procumbens* are discussed with a focus on their potential anticancer properties. Among these compounds, hexadecanoic acid, also known as methyl palmitate, was identified. This compound has been previously reported in studies involving *Hibiscus sabdariffa* (Ajoku *et al.,* 2015) and *Juglans regia* (Wang *et al.,* 2009). Research has indicated that methyl palmitate exhibits anti-phagocytic activity, modulates immune responses, prevents Kupffer cell activation in liver transplanted rats, and possesses anti-inflammatory and antifibrotic properties (El-Demerdash, 2011; Mantawy *et al*., 2012). On literature survey it was noted that only few work are undertaken for cytotoxic activity on *Launaea procumbens* with reference to this Recent work on leaves part on various cell lines Its IC_{50} values were 42 µg/ml for HeLa, 56.70 µg/ml for K562, and, 62 µg/ml for MCF-7 (Rawat *et al.,* 2016). Others studies on Launaea species like *Launaea capitata* on human cancer cell line (A549). The findings from the MTT cytotoxicity assay of n-butanol indicated IC_{50} 69.9 mg/ml and ethyl acetate fraction showed IC_{50} value 35.15mg/ml respectively (Khalil *et al.,* 2020). The anti-proliferative activities of ethanol extracts from *Launaea fragilis* and *Launaea nudicaulis* were investigated against various cancer cell lines. IC_{50} value was found to be the greatest level of inhibition, reaching 81.7 µg/ml, was observed against Hep G2 with *L. fragilis*, whereas the least inhibition, at 60.4 µg/ml, was noted for PC3. *Launaea nudicaulis* demonstrated the highest inhibition percentage 77.2 µg/ml against MCF-7, whereas the lowest percentage 70.8 µg/ml was observed for PC3 (El-Darier *et al.,* 2021). *Launaea mucronata's* species cytotoxic studies on 24 hr of incubation period reveal the IC_{50} value found to be for stem part extract 76.70 µg/ml for HePG2, 53.2 for A549 µg/ml, for HCTII6 97.20µg/ml for MCF-7 109.40 µg/ml, 204.83 for MRC-5 respectively. Similarly leaves part extract of *L. mucronata's* the IC_{50} value found to be 48.10 µg/ml for HePG2, 123.2 µg/ml for A549, 112 µg/ml for HCT116, 180.80 µg/ml for MCF-7, 412 µg/ml for MRC-5 respectively (Abouzied *et al.,* 2021).

In our research the aerial part extracts showed cytotoxic effects after 48 hr of incubation time the most potent and had the strongest effect of the four possible plant extracts and these cell line was first time reported as per literature. Its IC_{50} values were 30.79 µg/ml of (LPM) and 35.33µg/ml of LPC for NCIH-460. In MDAMB-231 the IC_{50} value is found to be 55.08µg/ml of LPC and 42.44 µg/ml of LPM. In K562 and HePG2 cancer cell lines also shows anticancer activity with highest IC_{50} value. In our research, we extended this investigation by selecting and examining cell lines that have not been previously reported.

In our present work the most potent activity against all tested cancer cell lines is exhibited by aerial part extract show the good activity on MDAMB-231 and NCIH-460 cell lines, respectively (Fig. 2 & 3). Therefore, it is concluded that *Launaea procumbens* of aerial part exhibits significant anticancer activity as if know it was first report in aerial part work undertaken into cytotoxic work. This study contributes novel insights into the anticancer properties of different extracts on these cell lines, presenting a comprehensive exploration of their cytotoxic effects and Subsequent research should focus on isolating the specific compound responsible for this anticancer activity, which could facilitate more effective treatments in herbal medicine.

CONCLUSIONS

The current study regarding to cytotoxic and anticancer activity of aerial part of *Launaea procumbens* methonlic extract showed strong anticancer activity. Among four cancer cell lines, two cell lines such as MDAMB-231 and NCIH-460 showed good cytotoxic activity. The GCMS analysis provides evidence that presence of some bioactive compounds in methonlic and chloroform extract.

FUTURE SCOPE

In future research in the isolation and characterization of specific compounds and their mode of action on cancer cell lines. Furthermore *In vitro* and pathway studies may leads to development good herbal medicine in treatment of cancer.

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