



Phytochemical analysis, antioxidant and anticancer activity of *Aerva javanica* growing in district Karak, Khyber Pakhtunkhwa Pakistan

SAJIDA AFZAL,¹ SIRAJ KHAN,² MAJID IQBAL,³ ANAM AKHTAR⁴

¹ Kohat University of Science and technology Kohat, Department of Zoology. ORCID: 0000-0002-6994-0039

² Department of Botany, Abdulwali khan university, Mardan, Pakistan.

³ Qarshi Herb Research Center (QHRC), Qarshi Industries (Pvt.) Limited. ORCID: 0000-0003-3657-5933

⁴ Institute of Geographic Science and Natural Resources Research UCAS China. ORCID: 0000-0003-0561-8494

⁴ Department of Plant Sciences, Quaid- i- Azam University Islamabad, 45320, Pakistan. ORCID: 0000-0002-3723-8907

Corresponding author email: sk3130249@gmail.com

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Abstract The purpose of this study was to evaluate the photochemical, antioxidants and anticancer activity of the medicinal plant *Aerva javanica*. This plant belongs to the Amaranthaceae family. Locally it is called “bui”. It is a shrub with a long tap root that grows all over India in the wild. The plant extracts were prepared using ethanol, methanol and distilled water as solvents. The antioxidant activity was determined using DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical scavenging activity and IC₅₀ was determined. The total flavonoids compounds found in *Aerva javanica* ethanolic extract were (0.90 ±0.16) while the total phenolic contents found in ethanolic extract were (0.78 ±0.16), followed by the methanolic and aqueous extract. The antioxidant results of methanolic extract of *Aerva javanica* showed 0.78 ±0.18 percent inhibition and SCV 49.10% at concentration of 1.5 mg/ml, ethanolic extract showed 0.54 ±0.12 percent inhibition with 64.28% SCV. Phytochemical analysis of best result oriented *Aerva javanica* extract was done with Gas Chromatography-Mass Spectrometry (GC-MS) technique. The results revealed the presence of different compounds predominantly Acetone (1.18%), Ethyl Acetate (38.95%), (20.77%), n-Propyl acetate (4.09%), Isobutyl acetate (2.71%), (3.84%), isoquinoline,1-[(3,4-diethoxyphenyl)methyl]-6,7-diethoxy- (3.36%), Cyclohexanone (1.43%), 1,1-Diisobutoxy-isobutane (2.02%), n-Hexadecanoic acid (5.61%), Phytol (3.57%), 9-Octadecenoic acid, 1,2,3-propanetriyl ester (10.72%), Octadecanoic acid (1.78%), Bis(2-ethylhexyl) phthalate (3.48%), Squalene (1.40%), 2,2-Dimethyl-3-(3,7,16,20-tetramethyl (1.12%) and 1,6,10,14,18,22-Tetracosahexaen-3-ol (1.195%). The study concludes that extensive research is required to detect more novel compounds in order to develop effective management approaches that significantly reduce the impact of the pathogens on human health as well as on environment.

Analiza fitochemiczna oraz antyoksydacyjna i przeciwzakrzepowa aktywność *Aerva javanica* z powiatu Karak, Khyber Pakhtunkhwa Pakistan

Słowa kluczowe fitochemia, potencjał antyoksydacyjny, potencjał przeciwnowotworowy, analiza GC-MS, *Aerva javanica*

Streszczenie Celem pracy była ocena fotochemicznej, antyoksydacyjnej i przeciwnowotworowej aktywności rośliny leczniczej *Aerva javanica*. Ta roślina należy do rodziny Amaranthaceae. Lokalnie nazywa się „bui”. Jest to krzew o długim korzeniu palowym, który dziko rośnie w całych Indiach. Ekstrakty roślinne przygotowano przy użyciu etanolu, metanolu i wody destylowanej jako rozpuszczalników. Aktywność przeciwutleniającą oznaczono za pomocą DPPH (2,2-difenyl-1-pikrylohydrazyl-hydrat) neutralizując wolne rodniki i wyznaczając IC50. Całkowita zawartość związków flawonoidowych znaleziona w ekstrakcie etanolowym *Aerva javanica* wynosiła (0,90 ± 0,16), podczas gdy całkowita zawartość fenoli znaleziona w ekstrakcie etanolowym wynosiła (0,78 ± 0,16), a następnie w ekstrakcie metanolowym i wodnym. Wyniki antyoksydacyjne ekstraktu metanolowego z *Aerva javanica* wykazały 0,78 ± 0,18 procent inhibicji, a SCV 49,10% przy stężeniu 1,5 mg/ml, ekstrakt etanolowy wykazał 0,54 ± 0,12 procent inhibicji z 64,28% SCV. Analizę fitochemiczną ekstraktu *Aerva javanica* przeprowadzono techniką chromatografii gazowej ze spektrometrią mas (GC-MS). Wyniki wykazały obecność różnych związków, głównie acetonu (1,18%), octanu etylu (38,95%), (20,77%), octanu n-propylu (4,09%), octanu izobutyli (2,71%), (3,84%), izochinoliny, 1-[(3,4-dietoksyfenilo) metylo]-6,7-dietoksy- (3,36%), cykloheksanon (1,43%), 1,1-diizobutoksy-izobutan (2,02%), kwas n-heksadekanowy (5,61%), fitol (3,57%), kwas 9-oktadecenowy, ester 1,2,3-propanotriylowy (10,72%), kwas oktadekanowy (1,78%), ftalan bis(2-etyloheksylu) (3,48%), skwalen (1,40%), 2,2-dimetylo-3-3,7,16,20-tetrametyl (1,12%) i 1,6,10,14,18,22-tetraoktadekano-3-ol (1,195%). Potrzebne są badania mające na celu wykrycie większej liczby nowych związków w celu opracowania skutecznych metod zarządzania, które znacznie zmniejszą wpływ patogenów na zdrowie człowieka, a także na środowisko.

Introduction

Plants are critical to human life on the planet earth. They can meet all the indispensable necessities of the individuals and also animals either as food stuff, energy, medication, ligneous, clothing, shelter, gums, and lubricants etc. (Karade et al., 2020). They are massive treasures not only from the global environmental perspective but also from the medicinal point of view. All plants ranging from a thorny bush to a tall evergreen tree are playing their role in serving the human. Among them medicinal and aromatic plants are of great importance which are serving as therapeutic agents since ancient times (Riaz et al., 2021). The history of plants as medicines is parallel to the development of life on earth. The use of plants for medicinal purposes is as old as human civilization. Since the advent of modern drugs, these plant resources have been used to treat various illnesses around the world. It's a long journey from a useless wild plant to a medicinal agent used to treat the disease (Dey, Nandy, Mukherjee, Modak, 2021).

Over the last few decades, there has been a great deal of awareness of traditional plants to their medical importance. The source of this interest lies in their economic feasibility, less toxicity and various pharmacological activities (Saha, Basak, 2020). The World Health Organization (WHO) estimates that approximately 80% of the population relies on local prescriptions to meet basic social security needs for minimal cost and availability. About 80% of Pakistani people are in urban households where herbal remedies are definitely available (Sani, Bello, Abdul-Kadir, 2014). Many plant extracts and plant components have been tested for their cytotoxicity and anti-cancer potential, and most of these plants tend to exhibit anti-cancer properties through

antioxidants (Russo et al., 2018). Some synthetic drugs are useful in treating cancer, but they are ineffective and dangerous. Currently, several known anti-cancer compounds such as taxol, podophyllotoxin, camptothecin, vinblastine, vincristine, and homoharringtonin have been isolated and purified from these herbal therapies (Kilcar et al., 2020). Cancer is the costliest and deadly infectious disease in the world. The most common cancers in Pakistan in 2016 were breast cancer (21.8%), leukemia (6.3%), Hodgkin lymphoma (4.9%), and non-Hodgkin lymphoma (4.7%) of the total number of reported cases. However, the actual prevalence of cancer can be higher than this because a proper registration system is not available in Pakistan. Currently, cancer management relies primarily on the availability of cancer treatments. In recent years, with the advent of new anti-cancer drugs, not only the repertoire, but also the average monthly cost of these treatments has expanded rapidly and significantly (Sarwar, Iftikhar, Saqib, 2018). The American Association for Cancer Research (ACS) estimates that 1,658,370 new cancer cases and 589,430 cancer deaths have been diagnosed in the United States in 2015. Its incidence will increase by 70% over the next 20 years. Currently, one in six deaths is due to cancer worldwide (Maqsood, Adiamo, Ahmad, Mudgil, 2020).

Biological activity from natural sources is due to a variety of chemicals that are widely distributed in nature. It has been shown that polyphenols can act as both antioxidants and oxidants, depending on their concentration and cellular environment. Medicinal plants have pharmacological activity and may be a source of new antitumor agents. Secondary plant metabolites and their derivatives have been used in the fight against cancer for the past half century (Janaki et al., 2018). Hence the present study is designed to analyze the biochemical compounds in the *Aerva javanica*, their antioxidant activity and anticancer property using MCF7 (Breast cancer), HepG2 (Liver cancer), HeLa (Cervical cancer) cell lines.

Material and methods

Sample collection

The experimental plant was collected from District Karak, Khyber Pakhtunkhwa (33°7'39.62"N; 71°5'50.33"E). The plant leaves were shade dried and grounded into fine powder using electrical grinder.

Extracts preparation

The sample was allowed to soak for 48 hours using methanol, ethanol and distilled water as the solvent, and Whitman filter paper No 1. Next, the filtrate was used under vacuum using a rotary evaporator until a sticky crude extract was obtained. The filtrate was then evaporated at a constant temperature of 50°C using vacuum rotary evaporator until a sticky mass of crude extract was obtained. The crude extract was dissolved in Dimethyl sulphoxide (DMSO) to make final working concentrations for biochemical analysis.

Phytochemical analysis

Quantification of total phenol (TPC) and total flavonoids (TFC) contents (quantitative test)

Determination of total flavonoids contents

Quercetin was taken as a standard for quantification of total flavonoids, plants extracts were taken in amount of 0.5 grams mixed with 4.3 milliliters of 80 percent methanol. Further 0.1 ml of 10% aluminum trichloride and potassium acetate was added to the mixture, volume reached up to 5 ml. The solution was placed at room temperature for 30 minutes of incubation; absorption was checked at 415 nm (Okafor, Ezejindu, 2014).

Determination of total phenols contents

Total Phenolic estimate was made by adding 0.5 gram of plants extracts to 80% of 1 ml ethanol. The resulting mixture obtained was centrifuged for 20 minutes at 10,000 rpm. Resulting supernatant was saved in test tube and the process was repeated 5 times. The supernatant was heated in water bath for dryness, than distilled water was added till its volume reached to 3 ml, 2 ml of 20% Na_2CO_3 was added to the solution and then 0.5ml of Folin ciocalteau reagent was done and after 5 minutes further 2 ml of 20% Na_2CO_3 was added to the solution, mixed thoroughly and test tube was placed in boiling water and absorbance was checked at 60 nm. Catechol has been used as a standard (Wang et al., 2018).

Antioxidant activity

The DPPH scavenging procedure was used to determine the antioxidant activity (Williams, Cuvelier, Berset, 1995). A DPPH solution was prepared by dissolving 24 mg of DPPH in 100 ml of methanol. A DPPH solution was prepared by dissolving 24 mg of DPPH in 100 ml of methanol. 1 mg/ml of plant samples was prepared to form stock solutions in methanol, and then diluted to 0.5, 1 and 1.5 mg/ml concentrations. Sample solutions and DPPH solution were combined in 1 : 1 ratio, incubated at 23°C to 30 minutes. Absorption was measured at 517 nm using spectrophotometer, and ascorbic acid was used as a standard (Wolfe, Wu, Liu, 2003). Percentage of scavenging action was determined using the equation:

$$\text{Scavenging effect \%} = \frac{\text{control absorbance} - \text{sample absorbance}}{\text{control absorbance}} \times 100$$

Anticancer activity

Cytotoxicity assay were performed to check the anticancer activity of ethanolic extracts of selected medicinal plants through MTT assay using MCF7 (Breast cancer), HepG2 (Liver cancer), and HeLa (Cervical cancer) cell lines as described earlier (Demirgan et al., 2016). The part of cytotoxicity assay were performed in School of Biological Sciences University of Punjab, Lahore. Cell lines were maintained in Dulbecco's modified Eagle's medium (DMEM), supplemented with 10% fetal bovine serum and 1% antibiotic. Cell lines were kept in incubator with moistened air with 5% CO_2 at 37°C. Cells were seeded in 96 wells plate and incubated for 48 hrs. After incubation MTT media were removed carefully and cells were treated with different concentrations of extract. All experiments were repeated 3 times with 3 replicates per experiment. Mean optical density value

of control was considered as 100% survival and treatment values were normalized accordingly by plotting barograph or table for cell survival.

GC-MS analysis of plant extracts

GC-MS analysis of ethanolic extracts of *Aerva javanica* showing most promising anticancer activity was carried out by using an Agilent 7890C gas chromatograph in tandem with a 5975C MSD and HP5MSI separation column whose length was 30 m, ID was 0.250 mm and its film thickness was 0.25 μm . Identification and quantification of compounds were conducted using AMDIS with a manually curated retention indexed GC-MS library while additional identification of compounds was performed using the NIST17 and Wiley 11 GC-MS spectral libraries (Ezhilan, Neelamegam, 2012).

Statistical analysis

One-way ANOVA was applied by using statistics 8.1 software programs followed by (Stefanelli, Barbini, Girolimetti, 2021). Mean and standard errors were calculated for three replicates. Means were compared by the LSD tests at significant level ($\leq 5\%$).

Results

Quantitative analysis of total phenolic and flavonoids contents

The Table 1 presented the TPC and TFC of methanolic, ethanolic and water extracts of the plant. The highest value of the flavonoids compounds was found in the ethanolic extract 0.90 ± 0.16 followed by the methanolic extract 0.63 ± 0.06 and distilled water 0.52 ± 0.08 . Using the Person's correlation analysis between standard Quercetin i.e. $\mu\text{g/g}$ and OD exhibited a strong positive relationship with adjusted $R^2 = 0.988$. The equation $y = 0.114x + 0.093$ is portrayed. The chemical Catechol was used as a phenol standard with a value of $y = 0.128x + 0.136$, $R^2 = 0.983$. The standard phenol curve in correlation analysis showed a significant linear positive correlation. The highest value of the phenolic contents was found in ethanolic extract 0.78 ± 0.16 followed by the methanolic extract 0.60 ± 0.13 and in aqueous extract was 0.64 ± 0.11 . Up to data no data is available about *Aerva javanica* phytochemical analysis both qualitatively and quantitatively (Table 1, Figure 2).

Table 1. Quantitative analysis of total Phenolic and Flavonoids contents of *Aerva javanica* in methanolic, ethanolic and aqueous extracts

Name of plant	Part of plant used	Extract of plant	Total Flavonoids mg/ml	Total phenols mg/ml
<i>Aerva javanica</i> Juss.	Whole plant	Ethanolic extract	0.90 ± 0.16	0.78 ± 0.16
		Methanolic	0.63 ± 0.06	0.60 ± 0.13
		Distilled water	0.52 ± 0.08	0.64 ± 0.11

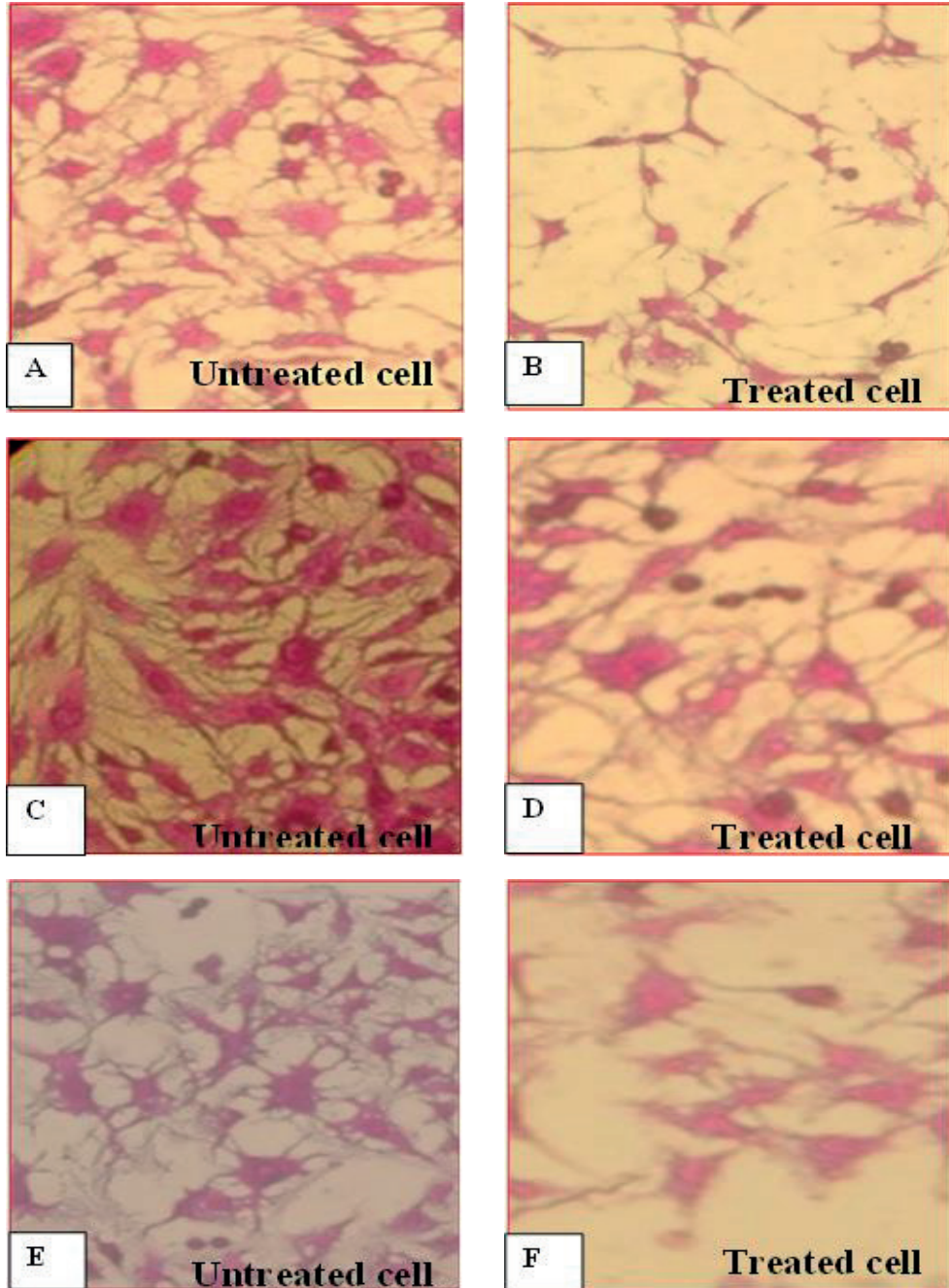


Figure 1. Showing different Cancer cell viability result before and after treatment of ethanolic extract of *Aerva javanica* i.e. MCF7 Brest cancer cell line (C) Untreated Cell, HeLa Cervical cancer cell line (E) Untreated Cell, (F) Treated Cell, HepG2 Liver cancer Cell line (A) Untreated Cell, (B) Treated Cell, (D) Treated Cell

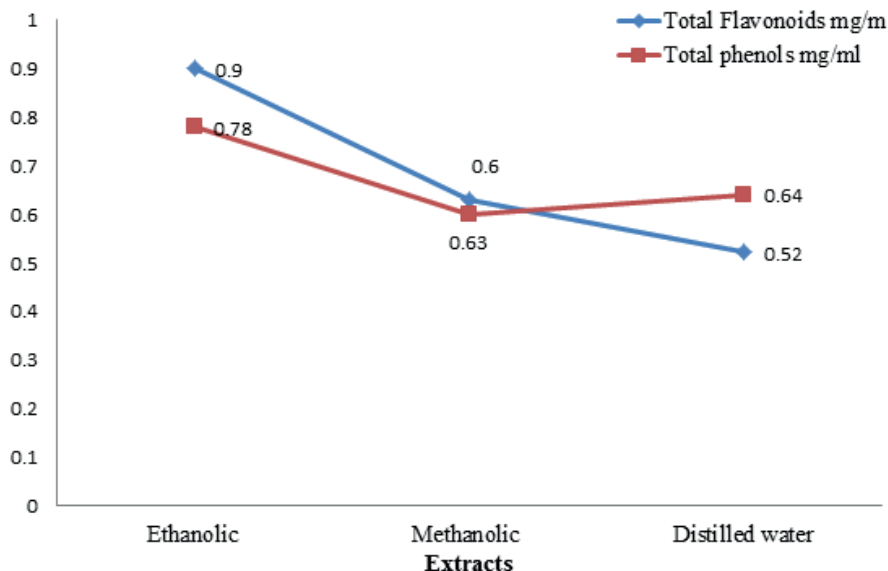


Figure 2. Total flavonoids and phenols in *Aerva javanica* different extracts

Antioxidant activity

The antioxidant activity (Table 2) showed that the DPPH free Radical scavenging assay of *Bassia indica* was carried out using different concentration of methanol, ethanol and distilled water. The ethanolic extract was the most potent extract with percent inhibition of 0.89 ± 0.02 at 0.5 mg/mL with IC_{50} of 47.94% followed by methanol and water respectively. The ethanol and water extract show least effective with IC_{50} 26.78% and 33.94% respectively. Overall the activity in the experiment was concentration dependent and ascorbic acid used as a standard (Figures 3–5).

Table 2. Antioxidant activity of *Aerva javanica*

The plant parts used	Extract	Concentrations (mg/ml)	Percent inhibition (means \pm SD)	Percent SCV	IC_{50} (%)
<i>Aerva javanica</i> Juss. whole plant	Methanolic	0.5	0.74 ± 0.35	36.60	33.94
		1.0	0.81 ± 0.10	37.5	
		1.5	0.78 ± 0.18	49.10	
	Distilled water	0.5	0.54 ± 0.12	64.28	26.78
		1.0	0.35 ± 0.14	57.14	
		1.5	0.30 ± 0.18	55.35	
	Ethanolic	0.5	0.89 ± 0.02	20.53	47.94
		1.0	0.77 ± 0.01	32.14	
		1.5	0.77 ± 0.26	33.93	

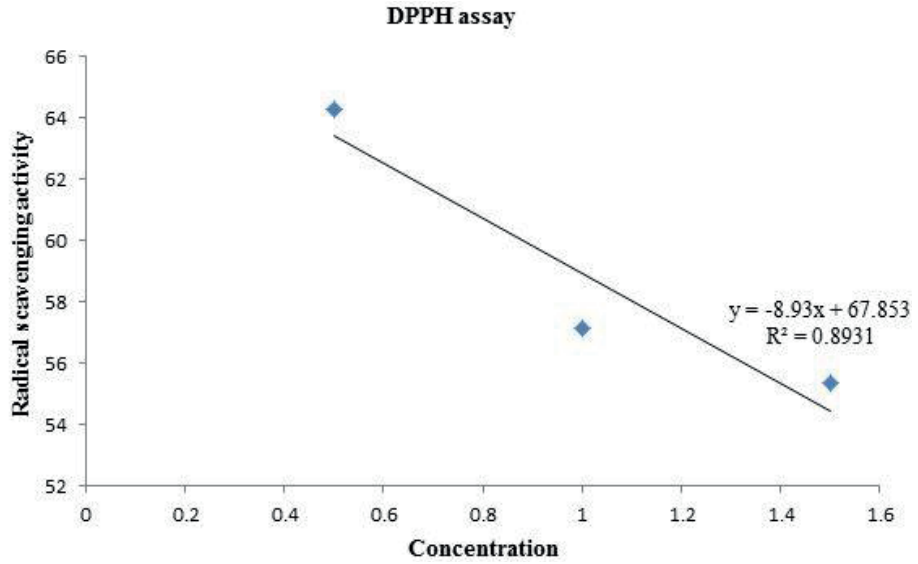


Figure 3. Standard curve of antioxidant activity of Distil water extract of *Aerva javanica*

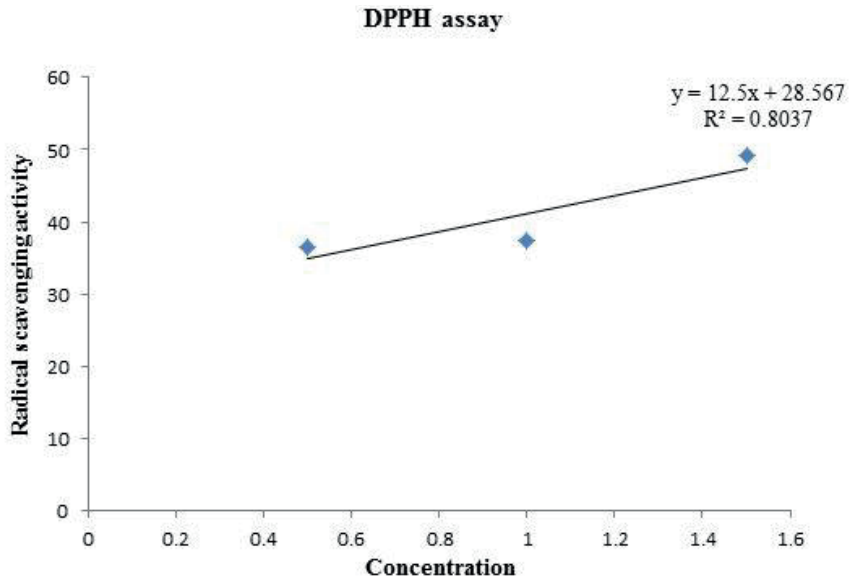


Figure 4. Standard curve of antioxidant activity of Methanolic extract of *Aerva javanica*

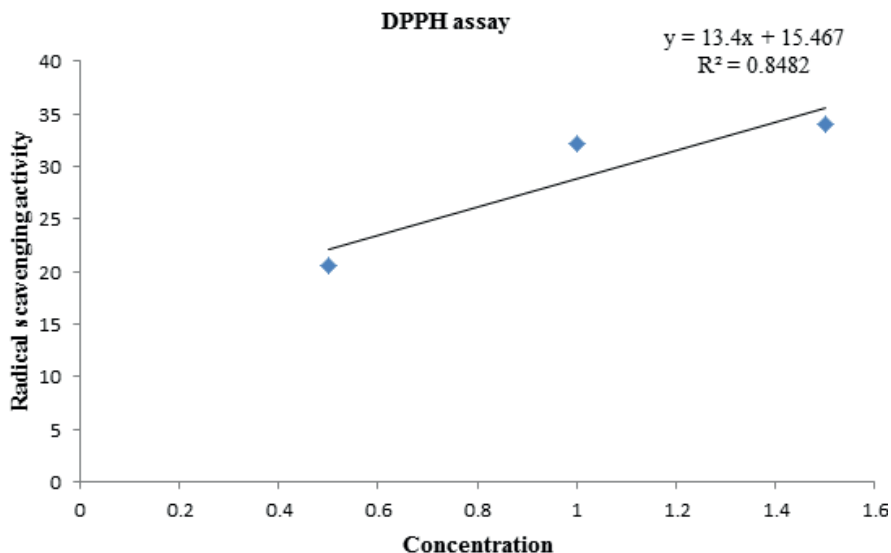


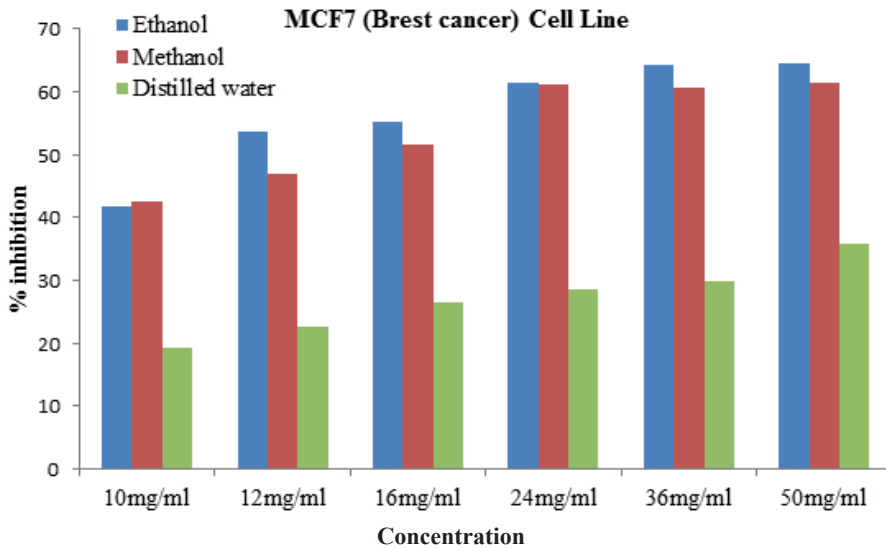
Figure 5. Standard curve of antioxidant activity of ethanolic extract of *Aerva javanica*

Anticancer activity

The anticancer activity (Table 3) of *Aerva javanica* against HePG2 (Liver cancer) cell lines indicated that the ethanol extract gave 64.6 ± 0.25 , 64.21 ± 0.77 , 61.40 ± 0.71 , 55.31 ± 1.67 , 53.60 ± 1.66 and $41.77 \pm 0.46\%$ inhibition with 50 mg, 36 mg, 24 mg, 16 mg, 12 mg and 10 mg concentrations, respectively. The results showed that the ethanol extract was the effective extract of *Aerva javanica*. The methanol and aqueous extract showed 42.47 ± 0.64 and 19.21 ± 0.44 inhibition at 10 mg concentration, 46.92 ± 0.15 and $22.56 \pm 0.28\%$ inhibition at 12 mg. While at 50 mg methanol showed 61.39 ± 0.76 and aqueous showed 35.86 ± 0.76 cell inhibition. The various concentrations of different extracts of *Aerva javanica* were also tested for anticancer activity against HeLa (cervical cancer) cell lines. The aqueous extract showed low inhibition at all concentrations 28.66 ± 0.41 (10 mg), 29.73 ± 0.45 (12 mg), 29.14 ± 0.07 (16 mg), 31.14 ± 0.22 (24 mg) and 35.78 ± 0.49 (100 mg). At 10 mg, methanol showed 58.19 ± 0.43 at 50 mg methanol showed 73.44 ± 0.39 . Ethanolic extract showed 52.64 ± 0.62 inhibition at 10 mg, and 70.22 ± 0.60 inhibition at 50 mg. The ethanol extract showed significant activity and was dose dependent activity against cancerous cells. Anticancer activity of *Aerva javanica* was carried out by using MCF7 (Breast cancer) cell lines. Different concentrations of different extracts were used. Showed that different concentrations of ethanol extracts have promising results against MCF7 (Breast cancer) cell lines, methanolic extract at 10 mg showed $44.11 \pm 0.73\%$ inhibition and 65.60 ± 0.19 at 50 mg. Ethanolic fraction showed 58.6 ± 0.36 at 10 mg, 72.69 ± 1.34 at 50 mg while aqueous extract showed 24.38 ± 0.16 , 66.72 ± 0.71 at 10 and 50 mg (Figure 1). All the extracts showed dose dependent activity (Figures 6–8).

Table 3. Anticancer activity of *Aerva javanica* through MTT assay using MCF7 (Brest cancer), HepG2 (Liver cancer), HeLa (Cervical cancer) cell lines

	Extract used	Percent inhibition (mean \pm SD) 10 mg/ml	Percent inhibition (mean \pm SD) 12 mg/ml	Percent inhibition (mean \pm SD) 16 mg/ml	Percent inhibition (mean \pm SD) 24 mg/ml	Percent inhibition (mean \pm SD) 36 mg/ml	Percent inhibition (mean \pm SD) 50 mg/ml	IC50
MCF7	Ethanol	41.77 \pm 0.46	53.60 \pm 1.66	55.31 \pm 1.67	61.40 \pm 0.71	64.21 \pm 0.77	64.6 \pm 0.25	27.18 \pm 0.30
	Methanol	42.47 \pm 0.64	46.92 \pm 0.15	51.66 \pm 0.47	61.11 \pm 0.35	60.63 \pm 0.36	61.39 \pm 0.76	28.55 \pm 0.23
	Distilled water	19.21 \pm 0.44	22.56 \pm 0.28	26.41 \pm 0.66	28.56 \pm 0.40	29.96 \pm 0.52	35.86 \pm 0.76	55.17 \pm 0.89
HeLa	Ethanol	52.64 \pm 0.62	55.42 \pm 0.28	56.80 \pm 0.16	70.96 \pm 0.29	70.96 \pm 0.29	70.22 \pm 0.60	25.24 \pm 0.78
	Methanol	58.19 \pm 0.43	58.67 \pm 0.38	59.63 \pm 0.37	61.16 \pm 0.86	71.77 \pm 0.53	73.44 \pm 0.39	24.44 \pm 0.03
	Distilled water	28.66 \pm 0.41	29.73 \pm 0.45	29.14 \pm 0.07	31.14 \pm 0.22	34.50 \pm 0.26	35.78 \pm 0.49	49.86 \pm 0.41
HeGP2	Ethanol	44.11 \pm 0.73	55.13 \pm 0.28	58.78 \pm 0.29	60.36 \pm 0.40	64.67 \pm 0.98	65.60 \pm 0.19	26.77 \pm 0.26
	Methanol	58.6 \pm 0.36	58.6 \pm 0.36	62.32 \pm 0.31	65.7 \pm 0.26	68.83 \pm 0.40	72.69 \pm 1.34	24.38 \pm 0.16
	Distilled water	24.38 \pm 0.16	59.19 \pm 0.17	60.68 \pm 0.37	63.57 \pm 0.35	65.63 \pm 0.23	66.72 \pm 0.71	25.74 \pm 0.10

Figure 6. Anticancer activity of *Aerva javanica* using MCF7 (liver cancer) cell lines

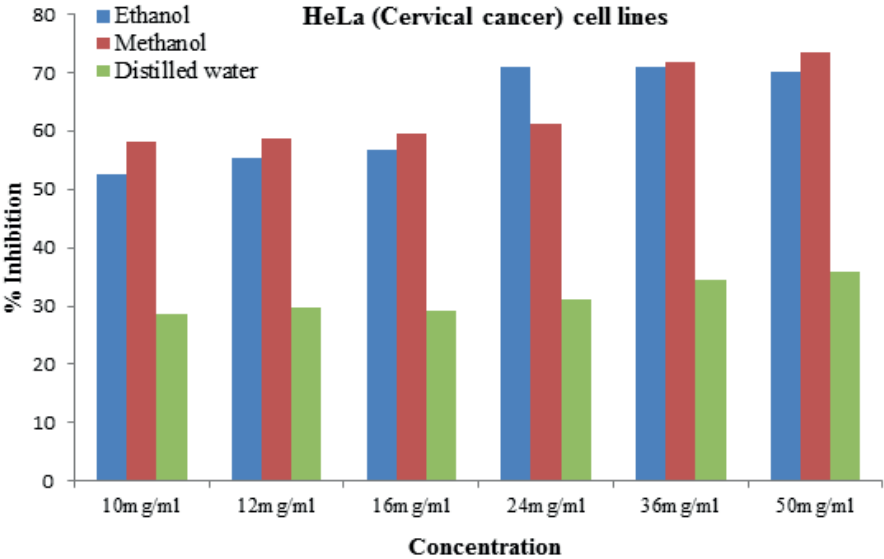


Figure 7. Anticancer activity of *Aerva javanica* using Hela (liver cancer) cell lines

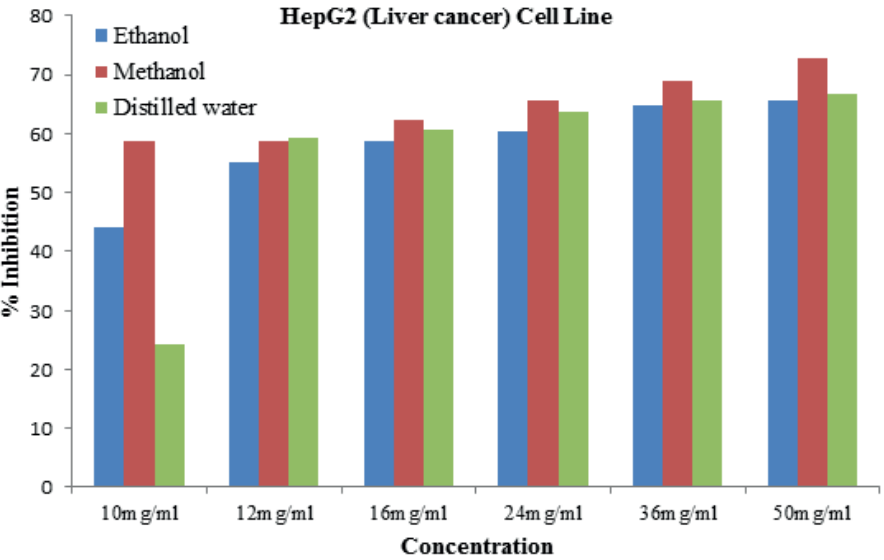


Figure 8. Anticancer activity of *Aerva javanica* using HepG2 (liver cancer) cell lines

GC-MS analysis

Superlative result-oriented extract of *Aerva javanica* was further subjected to GC-MS analysis in (Figure 9). The compounds identified from the extract along with their molecular weight, molecular formulae, retention time and peak areas (%) are presented in (Table 4). The dominant compounds present in ethanolic extracts in *Bassia indica* were Acetone (1.18%), Ethyl Acetate (38.95%), (20.77%), n-Propyl acetate (4.09%), Isobutyl acetate (2.71%), (3.84%), isoquinoline,1-[(3,4-diethoxyphenyl)methyl]-6,7-diethoxy- (3.36%), Cyclohexanone (1.43%), 1,1-Diisobutoxy-isobutane (2.02%), n-Hexadecanoic acid (5.61%), Phytol (3.57%), 9-Octadecenoic acid, 1,2,3-propanetriyl ester (10.72%), Octadecanoic acid (1.78%), Bis(2-ethylhexyl) phthalate (3.48%), Squalene (1.40%), 2,2-Dimethyl-3-(3,7,16,20-tetramethyl (1.12%) and 1,6,10,14,18,22-Tetracosahexaen-3-ol (1.195%). Different phenolic compounds were detected from *Aerva javanica* plant which proved their medicinal value.

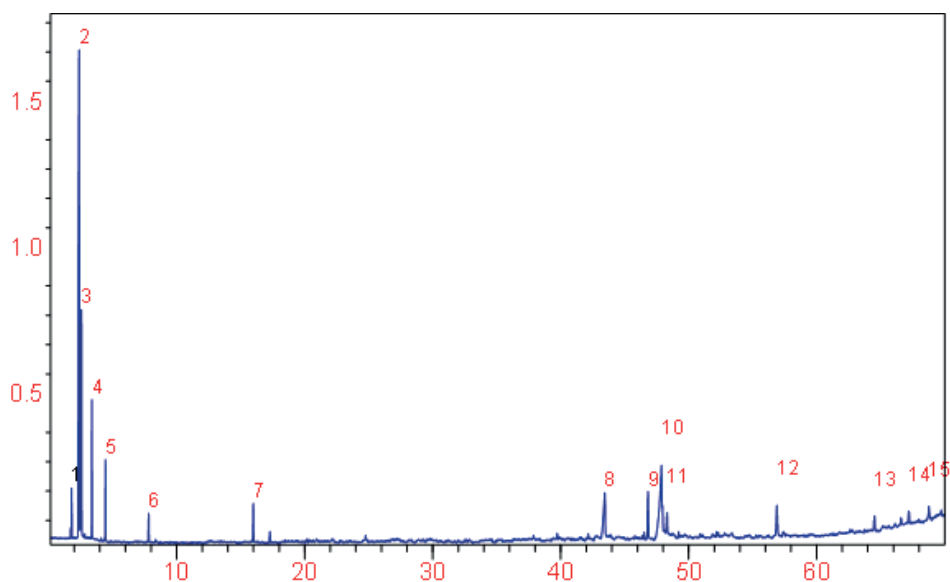


Figure 9. GCMS chromatograms of ethanolic extracts of *Aerva javanica*

Table 4. Compound identified from ethanolic inflorescence extract of *Aerva javanica*

S.NO	Names of Compounds	Formula	Mol. Wt.	Retention Time (Min)	Peak area (%)
1	2	3	4	5	6
1	Acetone	C ₃ H ₆ O	58	1.793	1.18
2	Ethyl Acetate	C ₄ H ₈ O ₂	88	2.379	38.95
3	1-Propanol, 2-methyl-	C ₄ H ₁₀ O	74	2.558	20.77
4	n-Propyl acetate	C ₅ H ₁₀ O ₂	102	3.378	4.09
5	Isobutyl acetate	C ₆ H ₁₂ O ₂	116	4.439	2.71

1	2	3	4	5	6
6	Cyclohexanone	C6H10O	98	7.815	1.43
7	1,1-Diisobutoxy-isobutane	C12H26O2	202	15.990	2.02
8	n-Hexadecanoic acid	C16H32O2	256	43.440	5.61
9	Phytol	C20H40O	296	46.821	3.57
10	9-Octadecenoic acid, 1,2,3-propanetriyl ester	C57H104O6	884	47.880	10.72
11	Octadecanoic acid	C18H34O2	282	48.308	1.78
12	Bis(2-ethylhexyl) phthalate	C24H38O4	390	56.896	3.48
13	Squalene	C30H50	410	64.520	1.40
14	2,2-Dimethyl-3-(3,7,16,20-tetramethyl	C29H48O	412	67.207	1.12
15	1,6,10,14,18,22-Tetracosahexaen-3-ol	C30H50O	426	68.770	1.19

Discussion

The medicinal plants are useful for healing as well as for curing of human diseases because of the presence of phytochemical constituents. Our results using three different extracts which show that the highest value of the flavonoids compounds was found in the ethanolic extract 0.90 ± 0.16 followed by the methanolic extract 0.63 ± 0.06 and distilled water extract 0.52 ± 0.08 . Using the Person's correlation analysis between standard Quercetin i.e. ug/g and OD exhibited a strong positive relationship with adjusted $R^2 = 0.988$. The equation $y = 0.114x + 0.093$ is portrayed. The chemical Catechol was used as a phenol standard with a value of $y = 0.128x + 0.136$, $R^2 = 0.983$. The present work is in accordance with the work of many previous researchers (Gopalakrishnan, Kalaiaarasi, 2014; Saidu, Oibiokpa, Olukotun, 2014; Mukherjee, Nema, Maity, Sarkar, 2013). The highest value of the phenolic contents was found in ethanolic extract 0.78 ± 0.16 followed by the methanolic extract 0.60 ± 0.13 and in aqueous extract was 0.64 ± 0.11 . Many reports support the results (El-Newary, 2016; Zhu, Nakagawa, Kishikawa, Ohnuki, Shimizu, 2015; Okafor, Ezejindu, 2014; Wang et al., 2018; Wolfe et al., 2003).

The human health can be enhanced by the use of vegetables and fruits rich in antioxidants which have the capability to neutralize the free radicals (Youdim, Shukitt-Hale, Joseph, 2004). In our result the *Bassia indica* were carried out using different concentration of methanol, ethanol and distilled water. The ethanolic extract was the most potent extract with percent inhibition of 0.89 ± 0.02 at 0.5 mg/mL with IC 50 of 47.94% followed by methanol and water respectively. The ethanol and water extract show least effective with IC50 26.78% and 33.94% respectively. In present study the results of DPPH free radical scavenging activity were expressed in percent inhibition with IC 50 values. Ascorbic acid was used as standard. Overall the activity was concentration dependent. Furthermore, our results are in accordance with results of (Rehman, Khan, Farid, Kamal, Aslam, 2013; Ismail et al., 2009; Veeru, Kishor, Meenakshi, 2009).

Plants provide opportunities of new medicines in the form of pure compounds or standardized extract. Biological activities gave clues about the plant or their extracts potency in the treatment of disease which led to the isolation of compounds for curing chronic and infectious diseases (Bajpai, Jadaun, Tiwari, 2016). GC-MS analysis is a potential means for identification of compounds present in the plant extracts. In this study, fifteen major compounds were identified from the ethanolic extract of *Aerva javanica*, while eleven other compounds shared minor peak area. In their studies Pandian and Noora (2019) observed sixteen photochemical present in the

methanolic leaf extracts of *Citrus medica* while (Hojjati, Barzegar, 2017) identified twenty-seven compounds including linalool, geraniol, α -terpineol and linalyl acetate.

In recent years there has been a steady revitalization of attention in the use of medicinal plants in the developing countries because herbal medicine has been reported safe and less or without any adverse effect especially when compared with synthetic drugs. Human beings have used the plants for medicinal purposes for centuries of the world including countries in the Indian sub-continent like India, Pakistan and Bangladesh (Shaikh, Shrivastava, Apte, Navale, 2016). About 70–95% people in rural areas of developing countries rely on local medicinal plants (Fridlender, Kapulnik, Koltai, 2015). In present study the anticancer activity of different extracts of *Aerva javanica* was carried out which indicate that the ethanolic extract showed 41.77 ± 0.46 , 53.60 ± 1.66 , 55.31 ± 1.67 , 61.40 ± 0.71 , 64.21 ± 0.77 and $64.6 \pm 0.25\%$ inhibition respectively at 10 mg, 12 mg, 16 mg, 24 mg, 36 mg and 50 mg concentrations against HepG2 (liver cancer) cell line. This showed that the ethanolic extract was the effective fraction of *Aerva javanica*. The methanol and aqueous extract showed 42.47 ± 0.64 and $19.21 \pm 0.44\%$ inhibition at 10 mg. While at 50 mg methanol showed 61.39 ± 0.76 and aqueous extract showed 35.86 ± 0.76 against HepG2 (liver cancer) cell line. All of the extract showed dose dependent activity, increase in concentration reduces cell viability. *Aerva javanica* was also tested for anticancer activity against HeLa (cervical cancer) cell lines. In HeLa (cervical cancer) cell lines the ethanolic extract of *Aerva javanica* show significant result at all concentrations. Ethanolic extract showed highest percentage of cell inhibition 70.22 ± 0.60 , methanolic extract 73.44 ± 0.39 and water extract 35.78 ± 0.49 at 50 mg concentrations. *Aerva javanica* against MCF7 lines also showed significance results, at 50 mg concentration water extract showed $66.72 \pm 0.71\%$ inhibition, ethanol showed 72.69 ± 1.34 , methanol $65.60 \pm 0.19\%$ inhibition at highest concentration 50 mg.

The result indicates that the different concentrations of ethanolic extract of *Aerva javanica* is more effective, significance and show more cell inhibition against HeLa (Cervical cancer) cell lines.

Conclusion

Current research shows that herbal remedies are a potential source of cure for human illness. (Firenzuoli, Gori 2007). This study fully supports the introduction of certain safe drugs to treat cancer and other infectious diseases. It has been concluded that *Aerva javanica* ethanol, methanol and aquatic extracts have shown potential botanical components, antioxidants and anti-cancer activity. Bassia extract was produced using gas (GC-MS) technology. The results revealed the presence of different compounds. Therefore, further research is needed to detect more compounds for the formation of stable products and to develop effective management approaches that significantly reduce the effects of pathogens on human health and the environment. Conclusion was also made based on the reliability of the present results that this species could be used in modern medicines that would be comparatively cheap and more effective on various infectious diseases with fewer side effects.

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