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Research Article

Phytochemical Screening and Biological Activities of Some Species of *Alpinia* and *Convolvulus* Plants

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Abstract

Background and Objective: The safety, low toxicity and clinical effectiveness of naturally occurring compounds increased the attention of researchers to the biological activity of plants. Accordingly, the current study was carried out to determine the antimicrobial and anticancer activities of three different species of *Alpinia* and *Convolvulus* plants. **Materials and Methods:** Phytochemical contents and biological activity of *Alpinia calcarata* (*A. calcarata*), *Alpinia purpurata* (*A. purpurata*), *Alpinia zerumbet* (*A. zerumbet*), *Convolvulus arvensis* (*C. arvensis*), *Convolvulus austro-egyptiacus* (*C. austro-egyptiacus*) and *Convolvulus pilosellifolius* (*C. pilosellifolius*) extracts were determined. Antimicrobial, anticancer and toxic activities were assessed against clinically-isolated test organisms, different cell lines and laboratory animals, respectively. **Results:** The investigated plants contain carbohydrates and/or glycosides, flavonoids, sterols and/or triterpenes, protein and/or amino acids, tannins and alkaloids. Anthraquinones was only detected in *A. calcarata*, *A. purpurata*, *A. zerumbet*. All plant extracts exhibited very good antibacterial, antifungal and antitumor activities. However, the *C. austro-egyptiacus* exhibited remarkable antimicrobial activity. The *C. arvensis* and *C. pilosellifolius* demonstrated antitumor activity (6.1 ± 0.3) and (16.4 ± 0.3), respectively higher than the antitumor activity of vinblastine sulphate (30.3 ± 1.4) against CACO (colorectal carcinoma). Nevertheless, *A. purpurata* showed antitumor activity against HCT-116 (colon carcinoma), 4.3 ± 1.3 , similar to the vinblastine sulphate. **Conclusion:** Results of current study indicated that the alcoholic extracts of *Alpinia* sp. and *Convolvulus* sp. plants have antimicrobial and anticancer activities. Moreover, *C. arvensis* and *C. pilosellifolius* possess an excellent anticancer activity and should be used as therapeutic antitumor agents against colorectal carcinoma.

Key words: Antitumor, antimicrobial, sub-chronic toxicity, phytochemical contents, *Alpinia* sp., *Convolvulus* sp.

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Plant secondary metabolites have been used for centuries to cure various ailments. The uses of plant-derived compounds in traditional medicines have proved to be clinically effective and are much preferred due to their fewer side effects than drugs of synthetic origin^{1,2}. The family Zingiberaceae is a medicinally important family since it contains numerous plants with high potential biological activities. The genus *Alpinia*, the largest genus of family Zingiberaceae³, possess many bioactive compounds against many harmful microbes and different diseases like cancer, diabetes, ulcer and many neural disorders⁴.

The members of *Alpinia* have complex chemical profiles including flavonoids, alkaloids, steroids, tannins and other polyphenolics⁵. A great depth of antimicrobial activities has been reported from *Alpinia* species especially *A. galanga* which contain more bioactive compounds than the other species⁶. However, *Alpinia calcarata* has been used in traditional medicines as anti-inflammatory, analgesic and carminative agent⁷. Another species with very important traditional use is *Alpinia purpurata* which contains different phytochemical contents with various biological activities, flavonoids isolated from this plant may corroborate the potential medicinal value of this species⁸. *Alpinia zerumbet* also has medicinal use because it contains many active compounds especially flavonoids, essential oils, tannins, phenols and alkaloids which are responsible for some of its therapeutic effects⁹.

The Convolvulaceae is another important family which contains large number of medicinal plants used in treatment of many diseases in folklore medicine¹⁰. Many compounds have been isolated and identified from different members of the genus *Convolvulus*. *Convolvulus arvensis*, *Convolvulus austro-aegyptiacus* and *Convolvulus pilosellifolius* are of the most commonly used species of genus *Convolvulus* due to their use in folklore medicine especially in Asia and Africa^{11,12}. Keeping all the previous information in mind, the current study was carried out to screening the phytochemical constituents of *Alpinia* and *Convolvulus* species and to determine their antimicrobial and anticancer activities.

MATERIALS AND METHODS

Plant materials: The plant samples of *Alpinia calcarata* Roscoe, *Alpinia purpurata* K. Schum, *Alpinia zerumbet* Burt and Smith, *Convolvulus arvensis* L., *Convolvulus austro-aegyptiacus* L. and *Convolvulus pilosellifolius* Desr.

were collected from different localities of Saudi Arabia desert during April, 2016. The plants were identified by Dr. Jacob Thomas, Assistant Professor of Taxonomy, Botany and Microbiology Department, College of Science, King Saud University and compared with the published data¹³. Voucher specimen (KSU.NO.6012, 6016, 5019, 8122, 5026, 5022, 5024) was kept in the herbarium of Botany and Microbiology Department. The plant samples were air-dried in shade, reduced to fine powder, packed in tightly dark closed containers and stored for phytochemical and biological studies.

Phytochemical analysis

Qualitative phytochemical analysis: The air dried powders of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* were separately subjected to phytochemical screening according to the standard published methods¹⁴.

Quantitative phytochemical analysis: Three hundred gram powder of air-dried aerial parts of each plant was extracted by percolation in 1 L of ethanol (95%) (4 times × 4 days) till complete exhaustion¹⁰. The total ethanol extract was concentrated under reduced pressure at 35 °C.

The percentage yield of each plant extract was calculated according to the dry weight. The determination of moisture, total ash, acid insoluble ash and water soluble ash were carried out according to published method¹⁵. Quantitative analysis of percentage primary and secondary metabolites were carried out according published methods for carbohydrates¹⁶, proteins¹⁶, lipids¹⁷, phenols¹⁶, flavonoids¹⁸, alkaloids¹⁹ and tannins¹⁶⁻¹⁹.

Antimicrobial activity

Test organisms: Different clinically isolated microorganisms, *Escherichia coli* (RCMB 010056), *Klebsiella pneumonia* (RCMB 0010093), *Proteus vulgaris* (RCMB 010085), *Pseudomonas aeruginosa* (RCMB 0100243-5) and *Salmonella typhimurium* (RCMB 006 (1) ATCC 14028), *Bacillus subtilis* (RCMB 015 (1) NRRL B-543), *Staphylococcus aureus* (RCMB 010027), *Staphylococcus epidermidis* (RCMB 010024), *Stroptococcus pyogenes* (RCMB 010015), *Aspergillus fumigatus* (RCMB 02564), *Candida albicans* (RCMB 05035), *Candida tropicalis* (RCMB 05042), *Geotrichum candidum* (RCMB 05096), *Microsporium canis* (RCMB 0834) and *Trichophyton mentagrophytes* (RCMB 0925) were obtained from the Microbiology Laboratory, Regional Centre for Mycology and Biotechnology, Al-Azhar University, Cairo, Egypt and used as test organisms.

Antimicrobial assay: The antibacterial and antifungal activities of ethanolic extract of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* were determined using the well-diffusion method²⁰.

Determination of minimum inhibitory concentration

(MIC): The minimum inhibitory concentration (MIC) was determined by micro-dilution method using serially 2-fold diluted plant extracts²¹. The MIC of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* extracts were determined by dilution of concentrations from 0.0-10 mg mL⁻¹. Equal volume of each extract and nutrient broth were mixed in a test tube. The lowest concentration (highest dilution) of the plant extract that produced no visible microbial growth (no turbidity) when compared with the control tubes were regarded as MIC.

Antitumor activity: The antitumor activity of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* was determined against lung carcinoma (A-549), colorectal carcinoma (CACO), colon carcinoma (HCT-116), cervical carcinoma (Hela), larynx carcinoma (HEp-2), hepatocellular carcinoma (HepG-2) and breast carcinoma (MCF-7) cell lines. The tumor cell lines were suspended in medium at concentration 5×10^4 cell/well in Corning® 96-well tissue culture plates and then incubated for 24 h. The plant extracts were added into 96-well plates and vehicle controls with media and 0.5% DMSO were run for each 96 well plate as a control. After incubation for 24 h, the numbers of viable cells were determined by the MTT assay method²².

Plants toxicity

Animals: Swiss albino mice of both sex (25-32 g) and male Wistar rats (1700-220 g) were obtained from the animal house of King Saud University. Animals were kept in standard polypropylene cages and maintained under standard conditions (temperature $23 \pm 1.0^\circ\text{C}$, humidity $55 \pm 10\%$, 12 h light/12 h dark cycle). They were fed standard pellet diet with water *ad libitum* and allowed to adapt to the laboratory environment for 1 week before experimentation (procedures and protocols approved by the Research Ethics Committee in KSU No.241/2017).

Preparation of the extracts for biological studies: The *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* extracts were

freshly suspended in distilled water just before administration with the aid of few drops of Tween 80.

Acute toxicity (LD₅₀) test: Alcohol extracts of the plants were given orally to the animal for determined median lethal dose (LD₅₀) as described in literature²³.

Sub-chronic toxicity: For carrying the sub-chronic toxicity, rats were divided into 7 groups each of 6 rats. The 1st group was administrated with the vehicle orally and left as a control, while the groups (from 2-7) were separately administrated the total alcohol extracts in a dose of 200 and 400 mg kg⁻¹ for 15 days. After the examination period, the collected sera were used for determination of liver and kidney enzymes as published²³. Results obtained estimated as liver and kidney markers.

Statistical analysis: All values were expressed as Mean \pm SD. Statistical analysis was done by using SPSS 10 (IBM Corp. Released 2010. IBM SPSS Statistics for Windows, version 19.0. Armonk, NY: IBM Corp). The statistical significant differences between the two means were assessed by unpaired student's *t* test. Differences at $p < 0.05$, 0.01 and 0.001 were considered statistically significant²⁴.

RESULTS AND DISCUSSIONS

Phytochemical studies: The chemical constituents of the *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* were qualitatively and quantitatively analyzed using different analytical and spectroscopic methods, the results are recorded in Table 1-3.

Qualitative phytochemical analysis: The phytochemical screening of the plant extracts indicated the presence of carbohydrates and/or glycosides, flavonoids, sterols and/or triterpenes, protein and/or amino acids, tannins and alkaloids in *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* (Table 1). However, anthraquinones was detected in *A. calcarata*, *A. purpurata*, *A. zerumbet* only (Table 1). The variations of phytochemical constituents could be attributed to numerous environmental factors²⁵.

Quantitative phytochemical analysis: Quantitative analysis of the plant-alcoholic extracts showed variations in yield percentage (Table 2). The highest yield percentage, 17.29 ± 0.12 , 16.23 ± 1.29 and 15.9 ± 1.51 , was obtained by

Table 1: Qualitative phytochemical analysis of *Alpinia calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius*

Plant tests	<i>Alpinia calcarata</i>	<i>Alpinia purpurata</i>	<i>Alpinia zerumbet</i>	<i>Convolvulus arvensis</i>	<i>Convolvulus austro-aegyptiacus</i>	<i>Convolvulus pilosellifolius</i>
Sterols and/or triterpenes	+	+	+	+	+	+
Cardinolides	-	-	-	-	-	-
Carbohydrates and/or glycosides	+	+	+	+	+	+
Flavonoids	+	+	+	+	+	+
Tannins	+	+	+	+	+	+
Saponins	-	-	-	-	-	-
Antraquinones	+	+	+	-	-	-
Alkaloids and/or nitrogenous bases	+	+	+	+	+	+
Protein and/or amino acids	+	+	+	+	+	+

(+) present, (-) absence

Table 2: Percentage of yield, primary and secondary metabolites of the plants under investigations

Plants	Yield percentage (mg/dry plant weight)	Primary metabolites						Secondary metabolites			
		Carbohydrates	Lipids	Proteins	Alkaloids	Flavonoids	Phenolic compounds	Tannins			
<i>Alpinia calcarata</i>	15.22±0.21	5.27±1.9	9.25±1.9	6.29±1.7	0.96±1.02	6.15±1.3	10.12±1.7	7.20±1.1			
<i>Alpinia purpurata</i>	14.61±0.35	7.03±1.6	12.85±2.20	8.06±2.5	1.16±1.02	4.95±2.9	12.11±1.1	8.21±0.9			
<i>Alpinia zerumbet</i>	17.29±0.12	6.23±1.3	10.75±1.5	9.20±1.8	1.21±1.02	5.10±1.5	11.15±1.4	6.65±1.8			
<i>Convolvulus arvensis</i>	14.88±1.59	4.13±1.4	12.05±2.3	6.31±2.1	0.16±1.02	5.11±2.4	15.13±1.7	10.25±1.4			
<i>Convolvulus austro-aegyptiacus</i>	16.23±1.29	5.63±1.2	10.15±2.1	7.46±1.9	0.08±1.02	6.17±1.6	16.10±1.8	11.25±0.7			
<i>Convolvulus pilosellifolius</i>	15.90±1.51	7.01±1.8	11.05±2.9	8.56±1.3	0.06±1.02	7.11±2.3	15.19±1.5	6.21±1.15			

Values are the mean of triplicates ± standard deviation

Table 3: Quantitative phytochemical analysis of *Alpinia calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius*

Plants	Moisture (mg/dry weight)	Total ash	Acid insoluble ash	Water soluble ash
<i>Alpinia calcarata</i>	5.17±1.29	9.32±1.13	2.15±1.27	6.12±1.34
<i>Alpinia purpurata</i>	7.14±1.11	8.23±1.35	3.11±1.32	5.21±1.15
<i>Alpinia zerumbet</i>	6.87±1.99	9.65±1.61	3.98±1.71	6.29±1.78
<i>Convolvulus arvensis</i>	8.97±2.19	10.34±1.54	4.19±1.36	8.58±1.63
<i>Convolvulus austro-aegyptiacus</i>	9.77±2.43	8.35±1.22	3.16±1.23	5.22±1.54
<i>Convolvulus pilosellifolius</i>	10.15±2.01	9.17±1.37	5.33±1.46	5.28±1.42

Values are the mean of triplicates ± standard deviation

A. zerumbet, *C. austro-aegyptiacus* and *C. pilosellifolius*, respectively while the lowest yield, 14.61 ± 0.35 and 14.88 ± 1.59 was obtained by *A. purpurata* and *C. arvensis*, respectively (Table 2). The variations in the yields percentages are relevant to the presence and/or potentiality of active materials.

The results showed that the percentage of primary and secondary metabolites were varied and there was a significant difference according to the genus and species (Table 3). Moreover, the percentage of phenolic compounds was noticeably high in all the plants which indicate that the plant has potential biological activity and could be used as a source of useful drugs for treatment of infectious diseases²⁶.

The results of pharmacopoeia constants exhibited slight percentage variations among the plants (Table 3). The moisture contents ranged from 5.17 ± 1.29 (*A. calcarata*) to 10.15 ± 2.01 (*C. pilosellifolius*). The relatively high moisture content of the plants is attributed to their desert habitat. The highest total ash percentage (10.34 ± 1.54) was obtained in *C. arvensis*. The water-soluble ash percentage was also variable which indicate that the active principles in the plants are high. Determination of those constants is valuable to assisting the quality of plant material²⁷.

Antimicrobial activity: The antimicrobial results revealed that the alcoholic extract of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* have very good antimicrobial activity (Table 4, 5).

Among all the plant extracts, *C. austro-aegyptiacus* exhibited remarkable antibacterial and antifungal activities similar to the activity produced by standard antibiotic, gentamycin, ampicillin and amphotericin B against *Microsporum canis*, *Geotrichum candidum*, *Proteous vulgaris*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus fumigatus* (Table 4). The minimum inhibitory concentration results revealed that the MIC of the extract of *A. zerumbet* (0.002 mg mL^{-1}) against *Microsporum canis* was similar to the amphotericin B (Table 5). The potential activity of *Convolvulus austro-aegyptiacus* is attributed to its phytochemical constituents²⁶.

Anticancer activity: The results detected antitumor activity of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* against lung carcinoma (A-549), colorectal carcinoma (CACO), colon carcinoma (HCT-116), cervical carcinoma (Hela), larynx carcinoma (HEp-2), hepatocellular carcinoma (HepG-2) and breast carcinoma (MCF-7) cell lines (Table 6). With the

Table 4: Antimicrobial activity of *Alpinia calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* Diameter of the inhibition zone (mm)

Test organism	<i>Alpinia calcarata</i>	<i>Alpinia purpurata</i>	<i>Alpinia zerumbet</i>	<i>Convolvulus arvensis</i>	<i>Convolvulus austro-aegyptiacus</i>	<i>Convolvulus pilosellifolius</i>	Standard antibiotic
Gram negative bacteria							Gentamycin
<i>Escherichia coli</i>	18.9±0.58	19.4±0.39	22.3±0.39	23.6±0.42	25.4±0.29	21.2±0.58	31.3±0.18
<i>Klebsiella pneumoniae</i>	21.1±0.12	23.4±0.39	25.1±0.12	24.3±0.33	28.9±0.58	19.9±0.44	32.3±0.15
<i>Proteous vulgaris</i>	15.7±0.62	16.6±0.62	18.9±0.62	23.7±0.42	25.3±0.42	18.4±0.29	26.3±0.30
<i>Pseudomonas aeruginosa</i>	14.6±0.12	13.9±0.54	12.2±0.22	14.1±0.28	17.4±0.53	19.1±0.18	25.2±0.18
<i>Salmonella typhimurium</i>	15.8±0.34	15.1±0.32	13.0±0.18	15.9±0.14	18.1±0.21	16.3±0.11	27.6±0.15
Gram positive bacteria							Ampicillin
<i>Bacillus subtilis</i>	17.0±0.42	24.6±0.12	17.7±0.22	22±0.25	18.2±0.23	20.0±0.46	32.6±0.34
<i>Staphylococcus aureus</i>	23.8±0.33	24.3±0.21	26.4±0.33	25.7±0.39	29.4±0.28	22.8±0.28	30.9±0.14
<i>Staphylococcus epidermidis</i>	21.3±0.12	25.4±0.29	29.5±0.12	26.8±0.39	20.1±0.58	22.2±0.29	31.4±0.18
<i>Streptococcus pyogenes</i>	22.3±0.39	23.5±0.42	24.8±0.39	19.5±0.44	22.3±0.58	17.4±0.44	28.4±0.34
Fungi							Amphotericin B
<i>Aspergillus fumigatus</i>	21.3±0.28	24.9±0.34	26.9±0.28	26.4±0.24	27.4±0.24	24.3±0.58	29.7±0.10
<i>Candida albicans</i>	19.4±0.32	22.3±0.52	25.3±0.32	24.7±0.58	26.3±0.62	22.2±0.39	27.9±0.12
<i>Candida tropicalis</i>	20.0±0.21	24.2±0.58	24.9±0.21	23.2±0.42	25.8±0.42	21.9±0.58	31.4±0.16
<i>Geotrichum candidum</i>	23.1±0.25	27.9±0.31	30.6±0.25	28.1±0.28	31.6±0.16	25.3±0.39	32.4±0.20
<i>Microsporum canis</i>	21.8±0.58	25.8±0.33	26.8±0.28	26.4±0.58	28.4±0.24	22.8±0.44	28.2±0.34
<i>Trichophyton mentagrophytes</i>	20.2±0.28	23.4±0.32	23.9±0.32	22.3±0.62	26.1±0.62	18.4±0.29	30.1±0.18

Values are the mean of triplicates ± standard deviation

Table 5. Minimum inhibitory concentration (MIC) of *Alpinia calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* Minimum inhibitory concentration (mg mL⁻¹)

Test organism	<i>Alpinia calcarata</i>	<i>Alpinia purpurata</i>	<i>Alpinia zerumbet</i>	<i>Convolvulus arvensis</i>	<i>Convolvulus austro-aegyptiacus</i>	<i>Convolvulus pilosellifolius</i>	Standard antibiotic
Gram negative bacteria							Gentamycin
<i>Escherichia coli</i>	0.312	0.125	0.312	0.625	0.312	0.312	0.0004
<i>Klebsiella pneumoniae</i>	31.250	0.078	0.002	0.004	0.078	0.625	0.0001
<i>Proteus vulgaris</i>	1.000	1.000	1.000	0.004	0.002	0.250	0.0001
<i>Pseudomonas aeruginosa</i>	0.125	0.125	0.156	0.019	0.002	31.250	0.0001
<i>Salmonella typhimurium</i>	0.156	0.625	0.156	0.004	0.078	31.250	0.0002
Gram positive bacteria							Ampicillin
<i>Bacillus subtilis</i>	0.004	0.001	0.004	0.078	0.156	0.156	0.0003
<i>Staphylococcus aureus</i>	0.002	0.039	0.001	0.001	0.001	0.004	0.0003
<i>Staphylococcus epidermidis</i>	31.250	0.002	0.078	0.156	0.001	0.625	0.0003
<i>Streptococcus pyogenes</i>	0.156	0.078	0.002	0.625	0.156	1.000	0.0001
Fungi							Amphotericin B
<i>Aspergillus fumigatus</i>	31.250	0.002	0.002	0.001	0.002	0.004	0.0004
<i>Candida albicans</i>	0.625	0.156	0.002	0.001	0.001	0.156	0.0002
<i>Candida tropicalis</i>	0.625	0.004	0.002	0.078	0.002	0.156	0.0003
<i>Geotrichum candidum</i>	0.078	0.001	0.004	0.001	0.001	0.002	0.0001
<i>Microsporum canis</i>	0.078	0.001	0.002	0.001	0.001	0.004	0.0002
<i>Trichophyton mentagrophytes</i>	0.625	0.078	0.002	0.156	0.001	1.000	0.0004

exception of *C. arvensis* and *C. pilosellifolius*, all the extracts showed IC₅₀ values higher (lower antitumor activity) than the vinblastine sulphate (Table 6). Interestingly, the extract of *C. arvensis* and *C. pilosellifolius* possessed antitumor activity (IC₅₀ = 6.1±0.3) and (IC₅₀ = 16.4 ±0.3), respectively higher than the antitumor activity of vinblastine sulphate (IC₅₀ = 30.3±1.4) against CACO (colorectal carcinoma). Moreover, *A. purpurata* showed antitumor activity against HCT-116 (colon carcinoma), IC₅₀ = 4.3±1.3, similar to the vinblastine sulphate, IC₅₀ = 3.5±0.2 (Table 6). The obtained results strongly validate the using of medicinal plants in traditional medicine¹⁰.

Plants toxicity: The results of the alcoholic plant extracts LD₅₀ showed that the *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* are characterized by a low degree of toxicity. The obtained results indicated that different doses up to 5000 mg kg⁻¹ did not produce any symptoms of acute toxicity and none of the animal died during 24 h of observation. Accordingly, it was believed that the oral LD₅₀ of the tested extracts was higher than 5000 mg kg⁻¹ and the extract is considered safe²⁸.

The non-toxic nature of the alcohol extracts is well supported by the results of sub-chronic toxicity study. Oral dosing of the tested extracts, 400 mg kg⁻¹, for 14 days did not show any significant effect on the levels of ALT, AST, total bilirubin, total proteins, albumin, urea and creatinine in their sera as compared to control (Table 7). The serum transaminase level is most widely used as a measure of hepatic injury, due to its ease measurement and high degree of sensitivity. It is useful for the detection of early damage of hepatic tissue²⁹. Since the activity of ALT and AST are specific assayable liver enzymes, their normal levels in serum of rats treated for 14 days means that the investigated alcohol extracts are not hepatotoxic. Urea and creatinine are the most sensitive biochemical markers employed in the diagnosis of renal damage. In kidney damage, there will be retention of urea and creatinine in the blood, therefore, marked increase in serum urea and creatinine are indications of functional damage to the kidney³⁰. By these indicators, the investigated alcohol extracts are therefore, not nephrotoxic in rats.

From current findings, it was clear that this *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-aegyptiacus* and *C. pilosellifolius* can be used for the treatments of infectious diseases which caused by microorganisms in addition to possible use in treatment of some cancer cells with no limitation for their use because they are very safe for human use and have no toxicity on liver and kidney functions.

Table 6: IC₅₀ values of *Alpinia calcarata*, *A. purpurata*, *A. zerumbet*, *Convolvulus arvensis*, *C. austro-egyptiacus* and *C. pilosellifolius* on viability of different cell lines

Cell line	<i>Alpinia calcarata</i>	<i>Alpinia purpurata</i>	<i>Alpinia zerumbet</i>	<i>Convolvulus arvensis</i>	<i>Convolvulus austro-egyptiacus</i>	<i>Convolvulus pilosellifolius</i>	Vinblastine sulphate
A-549 (Lung carcinoma)	117±8.2	26.72±11.4	142.0±11.4	62.2±3.9	242±19.4	100±5.7	24.60±0.7
CACO (Colorectal carcinoma)	110±6.2	37.60±0.30	69.4±3.80	6.1±0.3	242±19.4	16.4±0.3	30.30±1.4
HCT-116 (Colon carcinoma)	122±9.1	4.30±1.30	30.1±1.60	30.1±1.6	242±19.4	55.9±6.9	3.50±0.2
Hela (Cervical carcinoma)	248±20.6	70.50±1.10	201.0±12.9	17.8±0.8	242±19.4	46.2±0.6	59.70±2.1
HepG-2 (Hepatocellular carcinoma)	102±9.4	44.39±0.40	87.0±6.30	6.1±0.5	242±19.4	11.4±0.4	2.93±0.3
MCF-7 (Breast carcinoma)	234±21.2	58.60±3.80	211.0±10.8	11.1±0.6	242±19.4	15.1±0.1	5.90±0.4
Pc3 (Prostate cancer)	164±9.2	57.00±247	462.0±32.7	53.3±4.2	242±19.4	109±8.9	21.20±0.9

Values are the mean of triplicates ± standard deviation

Table 7: Effect of the total alcohol extracts of plants under investigations on liver and kidney functions

Parameters	Control	<i>Alpinia calcarata</i>	<i>Alpinia purpurata</i>	<i>Alpinia zerumbet</i>	<i>Convolvulus arvensis</i>	<i>Convolvulus austro-egyptiacus</i>	<i>Convolvulus pilosellifolius</i>
ALT (U L ⁻¹)	63.33±2.4	64.30±2.50	65.10±2.01	63.30±2.5	64.30±2.3	64.10±2.6	64.30±2.5
AST (U L ⁻¹)	48.90±2.2	49.40±2.54	48.62±2.24	50.10±2.2	47.90±2.8	49.12±2.2	50.40±2.54
Tb (mg dL ⁻¹)	1.70±0.1	1.61±0.30	1.83±0.40	1.68±0.2	1.69±0.3	1.71±0.3	1.68±0.2
Tp (g dL ⁻¹)	8.20±0.3	8.90±0.40	8.65±0.20	8.87±0.3	8.32±0.4	8.63±0.5	8.54±0.3
Al (g dl ⁻¹)	3.70±0.1	3.90±0.20	3.50±0.50	3.80±0.7	3.90±0.2	3.60±0.5	3.80±0.8
Ur (mg dL ⁻¹)	35.06±2.3	36.13±2.09	34.23±2.19	34.85±2.5	36.23±2.1	35.98±2.2	36.65±2.3
Cr (mg dl ⁻¹)	0.43±0.4	0.61±0.60	0.52±0.40	0.52±0.7	0.61±0.4	0.56±0.4	0.51±0.6

All extracts (400 mg kg⁻¹) was administered to rats for 14 days, n = 10; sera were collected and different enzymes were measured. Tb: Total bilirubin, Tp: Total protein, Al: Albumin, Ur: Urea, Cr: Creatinine

CONCLUSION

The selected 6 plants under study proved to have very promising effects against microorganisms and some cancer cell lines (lung carcinoma, colorectal carcinoma, colon carcinoma, cervical carcinoma, larynx carcinoma, hepatocellular carcinoma and breast carcinoma cell lines).

Plants belonging to genus *Convolvulus* (*C. austro-egyptiacus*, *C. arvensis* and *C. pilosellifolius*) were better than those belonging to genus *Alpinia*. *C. austro-egyptiacus* is the best active plant with higher plant potential antimicrobial activity against Gram-negative, Gram-positive and fungi better than the activity produced by standard antibiotic while *C. arvensis* and *C. pilosellifolius* showed better antitumor activity much better than the vinblastine sulphate.

A. purpurata showed antitumor activity against HCT-116 (colon carcinoma similar to the vinblastine sulphate. All plants are safe for human use because they showed no toxicity on laboratory animals when they give orally for 15 days.

SIGNIFICANCE STATEMENT

This study discovers that the ethanol extracts of *A. calcarata*, *A. purpurata*, *A. zerumbet*, *C. arvensis*, *C. austro-egyptiacus* and *C. pilosellifolius* have a significant antimicrobial and anticancer activities. However, the anticancer activity of *Convolvulus arvensis* and *Convolvulus pilosellifolius* against colorectal carcinoma (CACO) cell lines was remarkably better than the vinblastine sulphate. These results strongly declare the significance of the medicinal plants and their valuable use for treatment of many diseases.

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