

Phytochemical Analysis and Biological Activity of *Celosia Argentea* from Iraq

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Abstract

Scientists from all over the world are studying plants that can be used to make medicines. People have been using medicinal plants for a long time, so they are an important and inexpensive source of medicines. The top part of the *C. argentea* herb was taken off using hexane, methanol, and ethanol, in that order. Gas chromatography-mass spectrometry (GC/MS) was used to look at the chemicals. Important phytochemicals were found in *C. argentea*. These included triterpenoids, flavonoids, alkaloids, saponins, steroids, and tannins. Researchers need a full report on how *C. argentea* is used economically, medically, ethnobotanically, pharmacologically, phytochemically, and in zoology. People who are interested in *C. argentea*'s phytochemical analysis, antibacterial activity, and antifungal activity will find this review helpful.

Keywords: *C. argentea*, Methanolic extract, Ethanolic extract.

1. Introduction

Herbal medicines are used as the primary form of healthcare by almost 80% of the world's population, primarily in developing countries. The shortcomings of the healthcare system in addressing the problem of synthetic drugs will become more glaring in the coming years. Ayurveda, an antiquated Indian medical system, has been practiced for ages. Numerous studies have been conducted on the pharmacology, chemistry, pharmacology, and clinical therapeutics of ayurvedic medicinal plants. Natural cures from plants, animals, and minerals have been used to treat human diseases. Allopathy, or modern medicine as it is now known, was gradually developed over time through the scientific and observational work of scientists. Traditional medical practices, however, still form the basis for its advancement. A methodical, scientific approach for biologically assessing plant products based on their use in conventional medical systems is ideal for developing new plant-based medications (1).

The Amaranthaceae family includes the species *Celosia*. The generic name alludes to the flame-like flower heads and is derived from the Greek word *kelos*, which means "burned." More than 70 different species have been identified, and *C. argentea* is one of many that is frequently used as a leafy vegetable (2). As a result, *Celosia argentea* has been chosen as our sole medicinal plant. Information on phytochemical and antimicrobial activities will be provided in this study.

A quantitative short-day plant, *Celosia* species alternate entire or infrequently lobed leaves. *C. argentea* is a typically 0.5 to 1.5 m tall, but can occasionally grow much taller, erect, coarse, simple, branched, smooth annual herb. It doesn't have many

branches, at least not until the time of flowering. The light green, alternately entire or infrequently lobed leaves. Although those on flowering shoots are a little bit longer, they are usually 2 X 6 cm. There may be significant amounts of betalain pigments even in the green foliage.

Plant produces small, frequently pinkish or white flowers that are dense, erect spikes that are 8 to 12 millimeters in length. These flowers are borne in solitary, erect, stout, dense, white, purple, or pink, glistening spikes. They lack petals and range in size from 3 to 30 centimeters in length and 1.5 to 2 centimeters in thickness. Because sepals are 6 millimeters long, they are longer than bracts. It has fruits with membranes. Large quantities of seeds, which are typically black and have a diameter of 1 mm, are produced by *C. argentea* flowers. Late summer to late fall are prime blooming times for the cockscomb flower. An annual dicotyledon plant called *C. argentea* (3).

The alcohol extract of *C. argentea* showed sensitivity in the order *Shigella* sp., *Pseudomonas* sp., *Staphylococcus* sp., *Streptococcus* sp., *Vibrio* sp., *Klebsiella* sp., *E. coli*, and *Salmonella* sp. when investigated the antibacterial activity of *C. argentea*. Unfortunately, the active antibacterial compounds of the promising antibacterial compounds are unclear, so this study will include a portion of that goal (4).

The n-hexane extract of *C. argentea* plant seed oil was used to study fungi. *Aspergillus fumigatus*, *Candida tropicalis*, and *Trichophyton mentagrophytes* could not grow in the presence of *C. argentea* seed oil at a concentration of at least 50%. This means that the food supplement, drug, and cosmetics industries should use these qualities (5).

Taxonomy

Division : Magnoliophyta

Kingdom : Plantae
 Clade : Angiosperms
 Order : Caryophyllales
 Family : Amaranthaceae
 Genus : Celosia
 Species : Argentea

Morphology

Flower : In spikes, dense, cylindric, pink turning white
 Fruit : A Capsule, globose .seeds, reticulate
 Leaf Apices : Acute
 Leaf Arrangement : Alternate spiral
 Leaf Bases : Cuneate
 Leaf Margins : Entire
 Leaf Shapes : Elliptic
 Leaf Types : Simple
 Habit : An erect, glabrous profusely branched annual herb

2. Material and Method

2.1. Collection of plant

Fresh aerial portions of *Celosia argentea* were collected from a Baghdad garden from August 8 to December 15, 2021. After washing the aerial parts with tap water and de-ionized water, they were allowed to air-dry at room temperature. The dried plant was then chopped into small pieces and stored in a dry, dark location until it was time to create an extract.

2.2. Chemical Solvent

One of the simplest extraction methods is maceration, in which coarse and powdered plant material is soaked in solvents like methanol (99.8%), ethanol (99.99%), and n-Hexane (99%). It's one of the most typical and low-cost strategies for extracting bioactive compounds from plants. Methanol and ethanol are used for extracting polar compounds, while hexane and other nonpolar solvents are used for extracting nonpolar compounds (6).

2.3. Preparation of Extracts by Maceration Method

Maceration was used to extract the essential oils from the *celosia argenta* plant's aerial parts, and a series of solvents of varying polarities were tried. To sum up, we subjected 100 grams of powder to 500 ml of n-hexane, 96 grams to 500 ml of ethanol, and 91 grams to 500 ml of methanol. The supernatant was filtered out after 48 hours at 25 °C in the dark. To further purify the extracts and isolate the bioactive compounds, the process was repeated, and the solvent in the supernatant was evaporated in a vacuum rotary evaporator (7).

2.4. GC-MS specification

1- Injectors: 150 °C injector temperature and in the 5975-SMB we used a Restek deactivated sky liner that did not have glass wool. The standard GC-MS injector temperature was also 150°C.
 2- Columns: In Cold EI it was 8 m 0.25 mm ID, 0.25μ

film of DB-5MS UI. In standard EI it was 30 m 0.25 mm ID, 0.25μ film of DB-5MS.

3- Column flow rate: In Cold EI it was 2 ml/min for 1 minute followed by flow programming to 12 ml/min at 4 ml/min. This flow program enabled the elution of volatile compounds shortly after the solvent elution yet lowered the elution temperature of labile sample compounds with the high 12 ml/min column flow rate. Standard EI flow rate was 1 ml/min.

4- Injection volumes: 1 uL with split 20 in the 5975-SMB GC-MS with Cold EI.

5- Oven: In the 5975-SMB GC-MS with Cold EI it was 80 °C with wait 0.5 min followed by 20 °C/min to 300 °C and wait 2 min for total of 13.5 min.

6- Electron Energy: 70 eV for both systems.

7- 5975-SMB Mass spectral range: 50-500 amu with 3.2 Hz scan rate

2.5. Bioassay Techniques

The antibacterial and antifungal activities of *Celosia argentea* essential extracts are evaluated as shown below:

2.5.1. Preparation of Microorganism

The pathogenic bacteria and pathogenic fungi were both obtained from the BPC laboratory analysis center.

1. Gram positive bacteria
2. *Bacillus subtilis*
3. *Staphylococcus aureus*
4. 2- Gram negative bacteria
5. *Klebsiella pneumonia*
6. *proteus vulgaris*
7. 3- Fungi
8. *candida albicans*

2.5.2. Preparation of Potato Dextrose Agar (PDA)

Dissolve 39 g in 1 liter of ultra-purified or distilled water. The medium must be brought to a boil in order for the substance to dissolve. To sterilize for 15 minutes, autoclave at 121 degrees Fahrenheit (15 psi). Keep in mind that vials need to cool to 45-50 degrees Celsius before being dispensed or used. When working at a pH of 3.5 is essential, sterile 10% tartaric acid is added to the medium. Sterile and refrigerated media each call for about 1 mL of acid. When acid is added to the medium, it does not heat up (8).

2.5.3. Preparation of Petri –plates

After being brought to a boil, the solid medium was chilled to 45 °C and placed in Petri dishes (8 cm in diameter) for use in subsequent tests. Strict biosafety measures were taken before inoculating the sterile, stabilized, refrigerated medium in Petri dishes with the experimental fungi. For three days, Petri dishes were kept at 31 degrees Celsius.

2.5.4. Preparation of Mueller Agar Media

38.5 grams of media and one liter of distilled water were used to create Mueller agar media. The mixture was boiled in a water bath until the agar melted, the pH was adjusted to 7.1–7.5, and it was then poured into conical flasks with cotton plugs and aluminum foil. The samples

were then adjusted in an autoclave at 121 °C and 15 psi of pressure for 15 minutes.

2.5.5. Preparation of Petri –plates

Mueller agar media was boiled, cooled at 45 °C, and Pour into Petri dishes (8 cm in diameter) for use in sub-cultivation and for further testing. The test organism (bacteria) was incubated at room temperature (37 °C).

2.5.6. Agar well diffusion method

Put MHA or PDA in a petri dish and wait for it to set. Include 0.1 mL of either a 24 or 72-hour-old bacterial and fungal culture. Employ a sterile spreader to apply the fungal inoculum to the agar surface. Use a cork borer to drill four holes into an agar surface. Put 25, 50, 75, and 100 [microgram/ml] of each extract into each of the 4 holes. Incubation at 37 °C for 24 to 72 hours Different-sized inhibition zones around each extract were observed. Place the metric ruler across the zone of inhibition at its widest diameter, hold the plate upright, and measure the distance between its edges. The size of the inhibition zones surrounding each extract varies. If no zone has been reported, enter a negative. Milliliters are used to measure zone diameters (8).

3. 3. Results

3.1. Maceration Method

The air-dried whole plants (weighing 100 g) were extracted with n-Hexane, ethanol, and methanol. Evaporation of the organic solvent under reduced pressure at 30–40 °C yielded a crude extract. The yield was 1 gram of light green hexane extract, 2.44 grams of green ethanol extract, and 3 grams of green methanol extract.

3.2. GC-MS of hexane extracts

The chromatograms of the plant's upper portion reveal the presence of compounds with distinct retention times. According to Figure (1), the GC-MS chromatogram of the hexane extract contains (21) distinct compounds. The bioactive compound found in the aerial portion of *Celosia argentea* can contribute to the plant's medicinal properties.

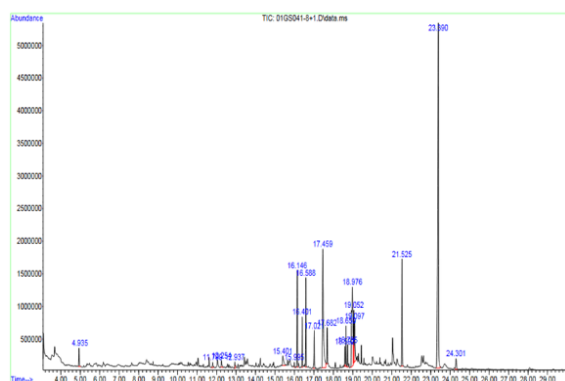


Figure (1): GC-MS of n-Hexane extract.

According to the curve's peak area percentage in Figure (1), the most important phytochemicals in *C. argentea* n-Hexane extract are demonstrate as table (1) below :

Biological activity	Corr. % max.	RT.T.min	Compound
antinociceptive and antioxidant activities (10) as well as anti-inflammatory and ant allergic effects (11)	12.01	16.588	Phytol
Antimicrobial (12)	2.8	23.301	Squalene
Anti-oxidant (13)	6.2	17.021	Hexadecanoic acid

3.3. GC-MS of ethanolic extracts

The chromatograms of the plant's upper portion reveal the presence of compounds with distinct retention times. According to Figure (2), the GC-MS chromatogram of the hexane extract contains (23) distinct compounds. The bioactive compound found in the aerial portion of *Celosia argentea* can contribute to the plant's medicinal properties.

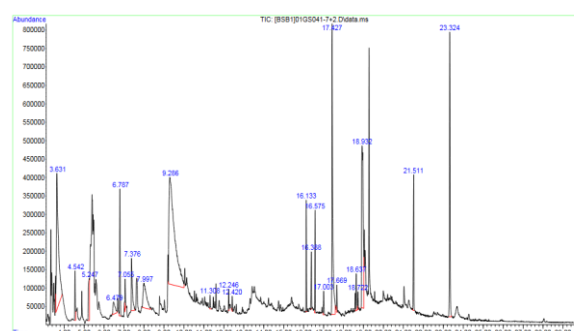


Figure (2): GC-MS of ethanolic extract.

According to the curve's peak area percentage in Figure (1), the most important phytochemicals in *C. argentea* ethanolic extract are demonstrate as table (2) below :

Biological activity	Corr. % max.	RT.T.min	Compound
antinociceptive and antioxidant activities (10) as well as anti-inflammatory and ant allergic effects (11)	7.67	16.575	Phytol
Anti-oxidant (12)	43.4	17.003	Hexadecanoic acid
Antibacterial antifungal antioxidant decrease blood cholesterol (14)	43.8	18.932	Octadecenoic acid

3.4. GC-MS of methanolic extract

The chromatograms of the plant's upper portion reveal the presence of compounds with distinct retention times. According to Figure (3), the GC-MS chromatogram of the hexane extract contains (20) distinct compounds. The bioactive compound found in the aerial portion of *Celosia argentea* can contribute to the plant's medicinal properties.

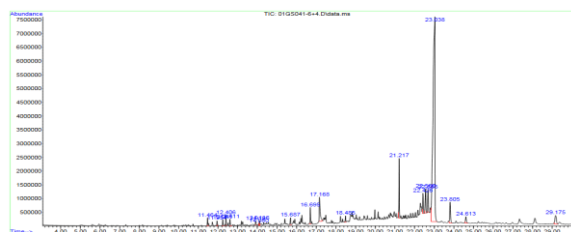


Figure (3): GC-MS of methanolic extract

According to the curve's peak area percentage in Figure (1), the most important phytochemicals in *C. argentea* ethanolic extract are demonstrate as table (3) below :

Biological activity	Corr. % max.	RT.T.min	Compound
Antimicrobial (12)	4.1	23.805	Squalene
Anti-oxidant (13)	3	29.175	Vitamin E
Anti-oxidant (12)	5.1	17.168	Hexadecanoic acid
antinociceptive and antioxidant activities (10) as well as anti-inflammatory and ant allergic effects (11)	7	21.217	phytol

3.5. Agar Well diffusion method⁴

The agar-well diffusion method was used to study the biological activity. The ethanolic extract of *Celosia argentea* was more effective against four different types of bacteria (*Klebsiella*, *Staphylococcus aureus*, *Bacillus subtilis*, and *Proteus vulgaris*) and fungi (*Candida albicans*).

The methanolic extract was more effective against seven different types of bacteria (*Bacillus subtilis*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Proteus vulgaris*), and fungi (*Candida albicans*).

With increasing concentration, each extract's biological activity against bacteria and fungi became more potent a concentration of 100 $\mu\text{g/ml}$ was more potent than a concentration of less than 75 $\mu\text{g/ml}$ and the latter had a greater potency than 50 $\mu\text{g/ml}$ and 25 $\mu\text{g/ml}$. The ethanol extract displayed more activity in *Bacillus*. The ethanol extract was more active in *Bacillus*. All the extracts have an effect on the *Candida*, but the ethanolic extract was more effective than the methanolic extract. Figures 4, 5 demonstrate this.

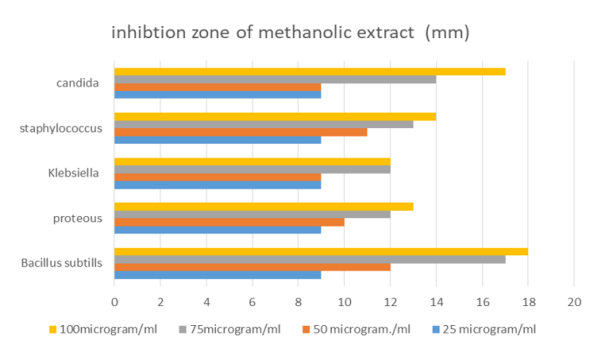


Figure (4): inhibition zone of methanolic extract.

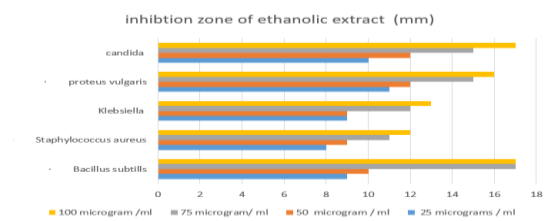


Figure (5): inhibition zone of ethanolic extract.

4. Discussion

The GC-MS analysis of the *C. argentea* extract revealed that different compounds (Phytol, squalene, Hexadecanoic acid, Octadecenoic acid and vitamin E) have distinct biologic properties. To investigate biological activity, the agar-well diffusion method was used. The activity was variable and varied according to the concentrations. *C. argentea* ethanolic extract was effective against four types of bacteria (*Klebsiella*, *Staphylococcus aureus*, *Bacillus subtilis*, and *Proteus vulgaris*) and fungi (*Candida albicans*).

Four bacteria (*Klebsiella*, *Staphylococcus aureus*, *Bacillus subtilis*, and *Proteus vulgaris*) and one fungus (*Candida albicans*) are more likely to be killed by methanolic extracts.

The biological activity of each extract against bacteria and fungi increased with increasing concentration (a concentration of 100 $\mu\text{g/ml}$ was more effective than a concentration lower than 75 $\mu\text{g/ml}$), and the latter had a higher effectiveness than 50 and 25 $\mu\text{g/ml}$). Both extracts were highly effective against *Bacillus*. The ethanolic extract showed higher activity against *Candida albicans* than the methanolic extract.

5. Conclusion

The aerial part of *C. argentea* was used in the current study for a variety of phytochemical analyses and biological activity purposes. n-Hexane, ethanol, and methanol were used as solvents to extract the material from the dried and powdered aerial part. The extract's phytoconstituents, antibacterial, and antifungal properties were examined. The presence of phytoconstituents such as (Phytol, squalene, Hexadecanoic acid, Octadecenoic acid and vitamin E) in the extract was determined using GC-MS analysis. Antibacterial activity and antifungal activity of the extract were determined by using the agar well diffusion method against four pathogenic bacteria and one fungus. Ethanolic and methanolic extracts showed higher activity in *Bacillus*. All the extracts showed efficacy against the fungi, but the ethanolic extract was more effective than the methanolic extract.

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