

Evaluation of antifungal and antimicrobial activities of *Caralluma adscendens* var. *Fimbriata* RoxB as potential medicinal herbs

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Abstract

Caralluma adscendens var. *fimbriata* is a species used as traditional medicines in many parts of India as an anti-diabetic, anticancer, anti-oxidant, anti-inflammatory and etc. The aim of this study was to evaluate the anti-microbial and antifungal properties of the aqueous extract of the plant. The study showed antibacterial activity against Gram positive and Gram negative bacteria making it a better chemotherapeutic agent. The extracts and its compounds may be used as a safe nutraceutical for the treatment of diseases. In several regions of India, *Caralluma adscendens* var. *fimbriata* is a species that is used in traditional medicine for its anti-inflammatory, anti-cancer, anti-diabetic, and antioxidant properties. The investigation demonstrated antifungal activity against *Aspergillus niger* [ATCC 16404] and *Candida albicans* [NCIM 3102]. The extracts and their constituents could be employed as a secure nutraceutical to address medical conditions. The need for novel approaches is justified by the rise in fungal resistance to conventional medications, the high expense of treatment, and the fact that the majority of antifungal medications on the market only have fungistatic effects. Since its discovery, the identification of medicinal substances found in plants has been the focus of attention.

Keywords: Anti-microbial; Anti-fungal; Fungistatic effects; *Caralluma adscendens*.

1. Introduction

Plants have the healing properties since time immemorial. Around the globe the therapeutic usage of plant originated products is used from generation to generation and various medicines have been developed. The development of micro-organisms resistance against various drugs/antibiotics has resulted in major cause of infectious disease and clinical therapeutic problems¹. Hence, recent trend, requires understanding and awareness on traditional medicine and the presence of natural bioactive molecules have been drawing much attention in order to develop alternate antibiotics from various plants. Synthetic antimicrobial drugs are costly, insufficient for the treatment and furthermore produce side impacts². This present circumstance gave the need to the pursuit for new antimicrobial drugs from medicinal plants. The therapeutic properties of plants are due to the presence of phyto-chemicals and bioactive compounds like phenol, flavanoids, alkaloid and tannin that makes the plants acceptable for curing the infectious disease. *Caralluma adscendens* var. *fimbriata* known as "Yugmaphallottatna" in Sanskrit is an edible thick succulent cactus and is perennial herb growing abundantly in some villages of India and in dry hill regions of Satara districts of Maharashtra. It belongs to Asclepiadoideae family and its local name in Maharashtra is "Makadshenguli/ Shengulmakad. *Caralluma* species uses are becoming popular in various tribes of India as the species is used as an appetite suppressant and it has an ability to control weight and lowers blood sugar level. It has been found that the plant has some medicinal properties against cancer rheumatism, gastric ulcer and skin infections. The *Caralluma* species found in India are the part of the medicine system of the country. The species are rich in triterpenes, alkaloids, Glycosides, Saponins and Tannins. The crucial phytochemical constituents of the species make them distinctive and diversified are pregenane glycoside, Flavone glycoside, Saponin glycoside, Megastamine glycoside, Sitosterol and Tomentoganin. (Fig. 1). Hence, in the present study,

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an effort is made to identify the antibacterial and antifungal activities of ethanolic and methanolic extracts of *Caralluma adscendens* var. *fimbriata* against *Escherichia coli*, *Pseudomonas*, *Coliform*, *Staphylococcus aureus*, *Aspergillus niger* and *Candida albicans*^{3,4}.

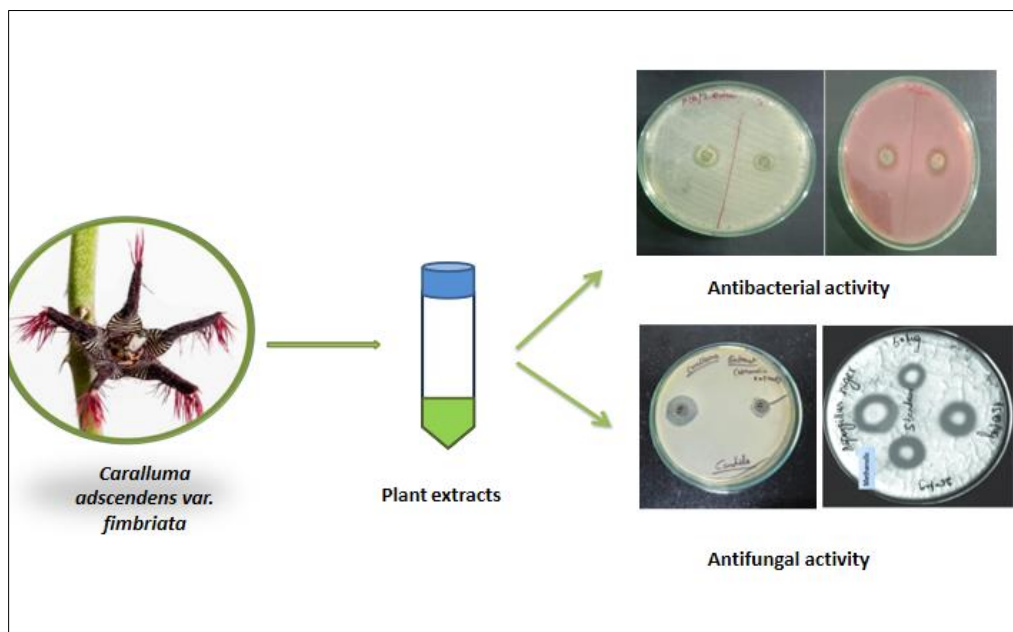


Figure 1 Pictorial representation of experimental work

It is referred to locally in Maharashtra as "Makadshenguli/Shengulmakad" and is a member of the Asclepiadoideae family. Indian tribes are starting to utilise the caralluma species more frequently because it suppresses hunger, helps people maintain their weight, and reduces blood sugar. The herb has been discovered to possess certain therapeutic qualities against skin diseases, stomach ulcers, rheumatism, cancer, and rheumatism. Medicinal herbs are a rich source of therapeutic applications and are essential for preserving good health and treating a wide range of illnesses in people and animals worldwide. India is one of the world's leading hubs for plant-based pharmaceuticals. Folk medicine is thought to employ over 25,000 plant-based medicinal compositions that are effective. They are also reportedly quite well-known in India's rural communities. India is a vast repository of medicinal plants that have gained popularity in conventional medicine. Many medical systems, including Ayurveda, Siddha, and Unani, use a variety of plant-based remedies to treat a wide spectrum of illnesses^{5,6,7}.

The current article discusses the pharmacological properties of the several species of the *Caralluma* genus. Anti-infection agents are antibacterial specialists that are obtained from common sources. The experts in antibacterial treatment may be either biocidal, eliminating microscopic organisms, or biotic, merely blocking the growth of germs. The microbes and growths deal with a vast and diverse array of living organisms, covering a major portion of the living world. The world's biotic sector includes a vast variety of living organisms. The biotic component of the biological system, which includes all living things with five domains, and the abiotic component, which is affected by climate, are interconnected and interact with one another. Even though the growths were originally given the term "plants," they have now been segregated since they are heterotrophs. This suggests that they use carbon fixed by another organic entity for digesting instead of fixing their own carbon through photosynthesis. Growth is classified alongside creatures in the monophyletic group of opisthotonos and are currently believed to be more strongly identified with animals than with plants^{8,9}.

2. Material and Methods

2.1. Preparation of Methanolic and Ethanolic extract

The powdered plant material extracts were prepared using solvent of high polarity. A portion of 25g of powdered material was extracted using 250ml of Methanol and Ethanol in a Soxhlet apparatus for 24 hours. Consequently, the solvent extract was evaporated to dryness at 44°C in an incubator. The yield of each extract constituents was calculated and stored at 4°C for prior use. The extract was used for assessment of antimicrobial activity.

The standard concentration of ethanolic/methanolic extract was obtained as 100 mg/ml of stock. Sterilisation of the extract was achieved by keeping it in an incubator overnight at about 37°C. *Caralluma fimbriata* ethanolic and methanolic extracts were tested for antibacterial activity using the agar well diffusion method. Using a sterile swab, the inoculum of every bacterial culture that was going to be tested was spread out on agar plates^{10,11}.



Figure 2 Ethanolic and Methanolic extraction process using Soxhlet of *Caralluma adscendens* var. *Fimbriata* Plant

2.2. Preparation of Plant extracts standard concentration

100mg/ml of stock was obtained as a standard concentration of ethanolic/methanolic extract (Fig. 2). The extract was sterilized by overnight incubation in an incubator at temperature ~37°C¹². *Well diffusion assay of plant extract:* Agar well diffusion method was used for screening the antibacterial activity of ethanolic extract of *Caralluma fimbriata*. Inoculums of each bacterial culture to be tested were spread on agar plates with a sterile swab. Subsequently, wells were punched into the agar medium and filled with 150µl (100mg/ml) of plant extract. The plates were then incubated in the upright position at 37° for 24 h. Two replicates were carried out for the extract against each of the test organism. After the incubation, the wells were observed and the zone of inhibition was noted with the help of measuring scale¹³.

2.3. Disc diffusion assay of standard antibiotics disc

Standard antibiotic discs of Penicillin, Vancomycin and Ampicillin were used as a control. The antibiotic discs were placed on the plates that were swabbed by particular bacterial culture on agar medium. After incubation for 24hrs, the diameters of the growth inhibition zones were observed and measured with the help of measuring scale in mm. The ethanolic extract of *Caralluma fimbriata* was tested for antibacterial activity using the Agar well diffusion method. Each bacterial culture that was going to be examined had its inoculums spread out on agar plates using a sterile swab¹⁴.

2.4. Microorganisms and Fungus

Hence, in the present study, an effort is made to identify the antibacterial activities of ethanolic extracts of *Caralluma adscendens* var. *fimbriata* against *E. coli*, *Pseudomonas*, *Coliform* and *Staphylococcus aureus*. Then for antifungal analysis *Aspergillus niger* [ATCC 16404] and *Candida albicans* [NCIM 3102] was used. In Chandigarh, India, the Microbial Sort Culture Assortment and Quality Bank (MTCC) collected the *Aspergillus niger* and *Candida albicans* societies. The collected boundaries were identified using standard microbiological techniques¹⁵.

2.5. Assay of Antifungal Activity

An antifungal activity test was conducted subsequent to the modification of the initial approach as presented by references. Once prepared, Potato Dextrose Agar (PDA) was autoclaved for 20 minutes at 15 pounds of pressure and cooled to 45 degrees Celsius. In order to act as antibacterial experts, 10 ml/L of tartaric corrosive (10%) was added to the cooled solution before being put onto sterile Petri plates and given time to harden. Using a sterile swab, the individual microbial suspension was cultured on the media-containing plates. In addition to placing control and standard (Amphotericin B (100 Units/Circles)) plates, the various solvents extracted and arranged circles separately

on each Petri plate. The plates were incubated for 72 hours at 28°C. Following the period of brooding, the distances across the zone that matched the paper circle were estimated and communicated in mm^{16,17}.

3. Results and Discussions

3.1. Extract yield

We used ethanol solvent for extraction of compounds from *Caralluma adscendens* var. *fimbriata* to see the antibacterial activity against various strain of bacteria. The yield percentage of the ethanolic extracts was 32.3%.

Table 1 Results for Zone of Inhibition of market available antibiotic disc

S. No.	Bacterial Strains	Zone of inhibition in diameter(mm) of Control		
		Penicillin (10mg)	Ampicillin (10mg)	Vancomycin (30mg)
1	<i>Staphylococcus aureus</i>	6mm	20mm	5mm
2	<i>Pseudomonas aeruginosa</i>	6mm	16mm	15mm
3	<i>Escherichia coli</i>	8mm	13mm	18mm
4	<i>Coliform</i>	1mm	10mm	4mm

Table 2 Results for zone of inhibition of ethanolic plant extract.

S. No.	Sample	Bacterial strains	Zone of inhibition in diameter(mm)
			Ethanolic extract
1	<i>Carallumafimbriata</i>	<i>Staphylococcus aureus</i>	13.5mm
2		<i>Pseudomonas aeruginosa</i>	13.5mm
3		<i>Escherichia coli</i>	18mm
4		<i>Coliform</i>	16mm

Table 3 Antifungal activities of *Candida albicans* and *Aspergillus niger* in ethanolic and methanolic extract at different concentration

S. No.	Sample name	Agar well 1 (Extract)	Agar well 2 (Control)
1.	<i>Caralluma fimbriata</i> Ethanolic extract (<i>Candida albicans</i>)	18 mm	No Zone
2.	<i>Caralluma fimbriata</i> Methanolic extract (<i>Candida albicans</i>)	20 mm	10 mm
3.	<i>Caralluma fimbriata</i> Ethanolic extract (<i>Aspergillus niger</i>)	14 mm	12 mm
4.	<i>Caralluma fimbriata</i> Methanolic extract (<i>Aspergillus niger</i>)	15 mm	13 mm

3.2. Evaluation of antibacterial activity

By disc method the effectiveness of three antibiotics was determined against the bacterial strains (Fig. 3). Ampicillin have shown highest zone of inhibition (20mm) against *Staphylococcus aureus* and Penicillin showed minimum zone of inhibition against *Coliform*.

3.3. Evaluation of plant extracts bioactivity

In present study the antibacterial activity of the ethanolic extract of *Caralluma fimbriata* was assayed *in-vitro* by agar well diffusion method against 4 bacterial strains (Fig. 4). Among the four bacterial strains *Escherichia coli* have shown highest zone of inhibition in ethanolic extract while *Staphylococcus aureus* and *Pseudomonas aeruginosa* have shown lower zone

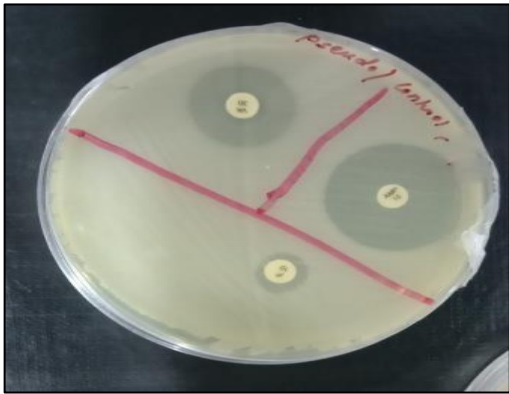
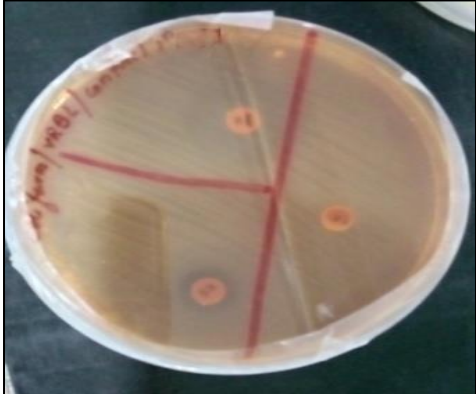
of inhibition in ethanolic extract compared to *E. coli*. The *coliform* bacterial have shown intermediate result between the above bacterial strains.

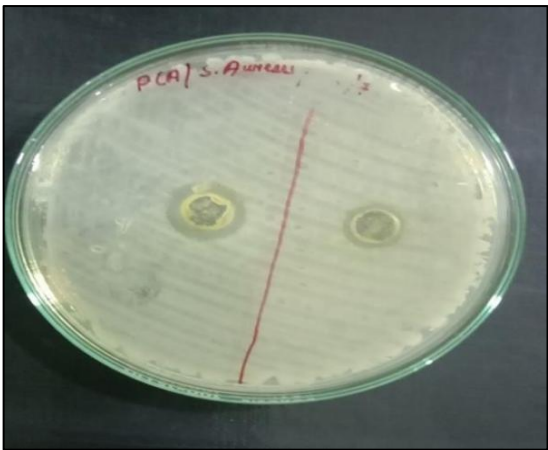
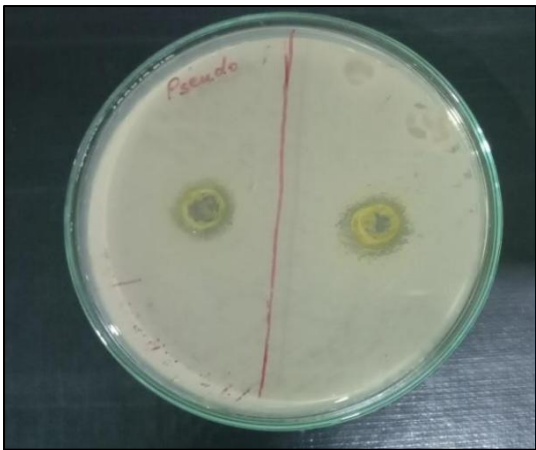
3.4. Antifungal activity

In the current investigation, the antifungal activity of *Caralluma fimbriata*'s ethanolic and methanolic extract was evaluated in vitro against a single fungus strain using the agar well diffusion method.

3.5. Against *Candida albicans* and *Aspergillus niger*

The table number 3 displays the outcomes of the ethanolic plant extract test against *Candida albicans* using 100 μ l of the extract. It is demonstrated that the 18 mm zone of inhibition exists in the ethanolic extract. When 100 μ l of the methanolic plant extract was tested against *Candida albicans*, the results indicated that the extract had a 20 mm zone of inhibition shows in figure 5.

	
(a) Inhibition zone of antibiotics against <i>Escherichia coli</i> .	(b) Inhibition zone of antibiotics against <i>Staphylococcus aureus</i> .
	
(c) Inhibition zone of antibiotics against <i>Pseudomonas aeruginosa</i> .	(d) Inhibition zone of antibiotics against <i>Coliform</i> .
Figure 3 The result of Inhibition zone of marketed antibiotic disk	

	
(a) Zone of inhibition shown by Ethanolic plant extract <i>Staphylococcus aureus</i>.	(b) Zone of inhibition shown by Ethanolic plant extract against <i>Pseudomonas aeruginosa</i>
	
(c) Zone of inhibition shown by Ethanolic plant extract against <i>Escherichia coli</i>.	(d) Zone of inhibition shown by Ethanolic plant extract against <i>Coliform</i>
Figure 4 The inhibition zone of Plant extract: a), b), c), d).	

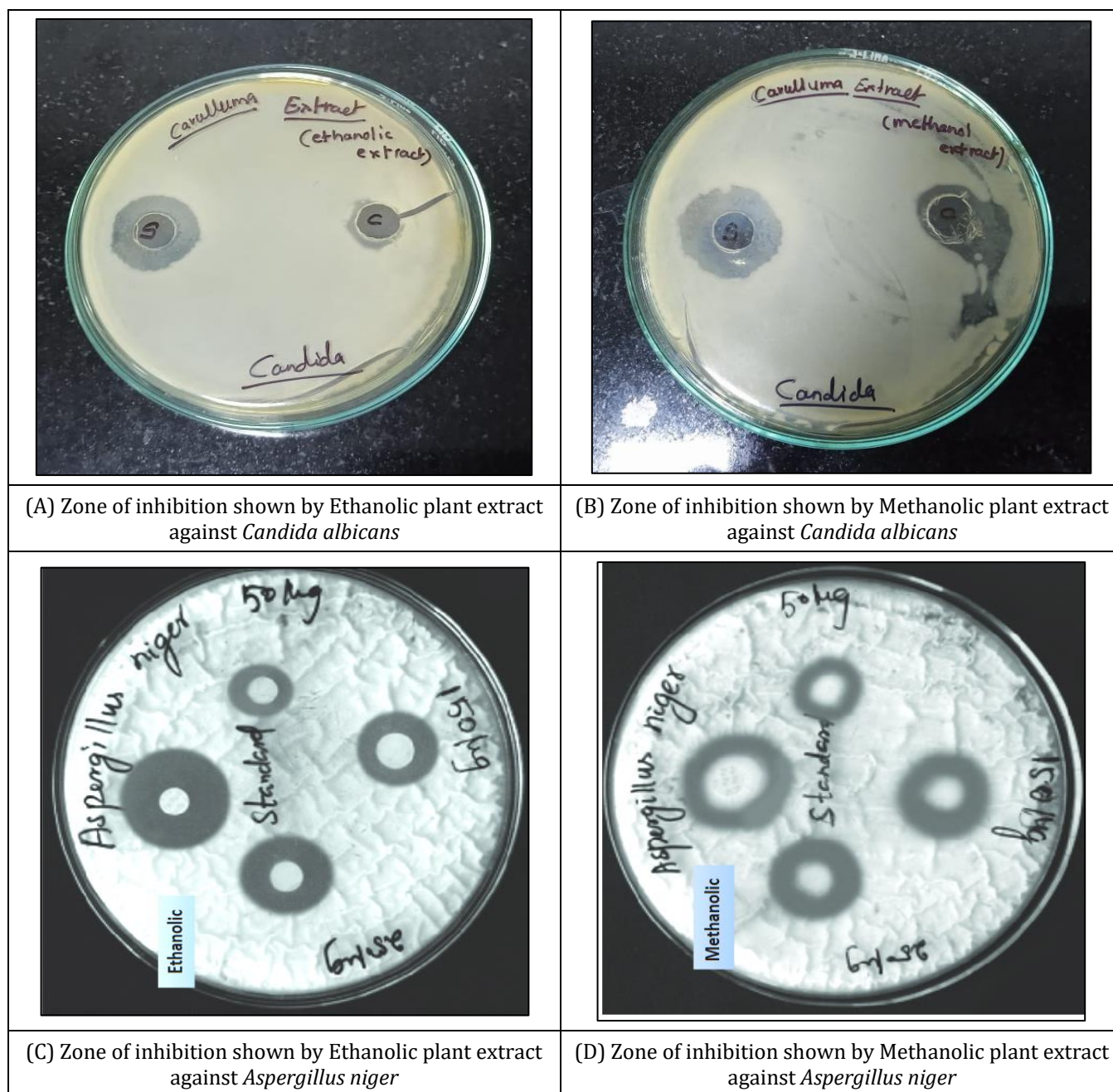


Figure 5 Zone of inhibition shown by Ethanolic and Methanolic plant extract against *Candida albicans* and *Aspergillus niger*.

4. Conclusion

This study presents an overview of the *Caralluma adscendens* var. *fimbriata* that can be used to treat against obesity, diabetes, hypertension, ulcers, and cancer. The *Caralluma adscendens* var. *Fimbriata* genus is a promising source for phyto-chemicals with therapeutic usage and suggests possible leads for researchers to discover herbal effects but most of the evidence remains suggestive but not conclusive so further phytochemical and pharmacological research is needed. The demonstration of antibacterial activity of *Caralluma adscendens* var. *fimbriata* against Gram positive (*Staphylococcus aureus*) and Gram negative bacteria (*Coliform*, *Pseudomonas*, *E. coli*) provides the scientific basis for its use in the traditional treatment. Further studies are required to establish the exact mechanism of antibacterial activity of phytochemicals extracted from so *Caralluma adscendens* var. *fimbriata* that better and safer chemotherapeutic agents can be developed from this plant. Further phytochemical and pharmacological research with more work should be done to address critical health concerns prevailing in developed as well as developing countries for designing a safer nutraceuticals approach for these diseases. An overview of *Caralluma adscendens* var. *fimbriata*, a plant that can be used

to treat cancer, diabetes, obesity, ulcers, and high blood pressure, is given in this paper. Further phytochemical and pharmacological research is required because *Caralluma adscendens* var. *Fimbriata* genus is a promising source of phyto-chemicals with therapeutic applications and offers potential leads for researchers to discover herbal effects. However, the majority of the evidence is suggestive rather than conclusive, so more research is needed. *Caralluma adscendens* var. *fimbriata*'s antibacterial activity against *Aspergillus niger* and *Candida albicans* has been demonstrated, which offers scientific support for the plant's usage in traditional medicine. To create safer and more effective chemotherapeutic medicines, more research is needed to determine the precise mechanism of action of the phytochemicals isolated from *Caralluma adscendens* var. *fimbriata*.plant. Numerous issues in practically all bio study domains can be resolved with the use of bioinformatics investigations on proteins, nucleic acids, and other biomolecules. In order to address serious health issues that are present in both developed and developing nations, more phytochemical and pharmacological research should be conducted in order to provide a safer nutraceuticals method for treating these disorders.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declare that they have no conflicts of interest.

Statement of ethical approval

The present research work does not contain any studies performed on animals/humans subjects by any of the authors.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

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