Original article

# Antimicrobial Properties of Extracts from Cymbopogon Schoenanthus plant

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#### Abstract

In this study, Experiments were performed to explore the antibacterial properties of an extract derived from *Cympbogon Schonatus* with water. This study evaluated the antibacterial activity of alcoholic and aqueous extracts of *Cymbopogon Schoenanthus* against four distinct bacterial species using the well diffusion method: two Gram-negative (S. aur, B. sub) and two Gram-positive (E. coli, S. typhi). The primary biologically active substances found in *Cympbogon Schonatus* include tannin, anthraquinone, terpene, coumarin, alkaloid, and saponin. While some bacteria showed resistance to the extract, Staphylococcus aureus and Bacillus subtilis were inhibited by the alcoholic extract of Cymbopogon Schoenanthus. These results suggest that extract from Cymbopogon Schoenanthus might be a viable choice in the hunt for natural antimicrobials. This study offers scientific insight that aids in assessing antibacterial values and investigating other pharmacological characteristics. **Keywords**. Cymbopogon Schoenanthus, Natural plant, Extraction, Plant, Biological Activity.

#### Introduction

Medicinal plants have been utilized for thousands of years to treat illnesses and promote health, making them one of the oldest natural resources used in medicine. Numerous ancient cultures have documented these herbs, demonstrating a profound comprehension of their medicinal qualities [1]. Archaeological evidence, such as the Ebers Papyrus (circa 1550 BCE), shows that ancient Egyptians utilized a wide variety of plants for medicinal purposes. This papyrus contains hundreds of medical recipes based on herbs, indicating their profound knowledge of the effects of plants on the human body [2].

In Chinese civilization, physicians such as Emperor Shen Nong played a crucial role in documenting knowledge about medicinal plants. Texts like the classify plants according to their therapeutic properties, laying the foundation for traditional Chinese medicine [3]. In ancient India, Ayurvedic medicine developed, relying heavily on herbs and plants. Texts like the "Atharvaveda" and "Charaka Samhita" reflect this deep understanding of plant-based treatments, forming the basis for many traditional medical practices in India [4]. In Chinese civilization, physicians such as Emperor Shen Nong played a crucial role in documenting knowledge about medicinal plants. Texts like the classify plants according to their therapeutic properties, laying the foundation for traditional Chinese medicine [3]. In ancient India, Ayurvedic medicine developed, relying heavily on the use of herbs and plants. Texts like the "Atharvaveda" and "Charaka Samhita" reflect this deep understanding of plant-based treatments, forming the basis for many traditional medical practices in India [3]. These plants according to their therapeutic properties, in India [3]. These plants serve as characteristic flavor enhancers, craving stimulants, and additives, making them irreplaceable in culinary applications. The active components of these plants have medicinal properties that may result in specific bodily reactions.

This involves increasing disease resistance and improving both human and animal immune systems. Traditional medicine has successfully employed plant-based therapies to treat a range of illnesses by utilizing these benefits. The sustained usage of these natural therapies demonstrates their applicability and efficacy in fostering general health and well-being. The use of these crops in food production currently shows their many uses and shows how traditional methods are still essential to modern health plans. Through their biologically active components, they have a direct effect on health outcomes that extend beyond basic nutrition. These applications demonstrate the natural resources' lasting value in both historical and current circumstances [5]. Approximately 80% of people worldwide receive their primary medical treatment from traditional medicine, according to the World Health Organization. Particularly in light of the growing prevalence of antibiotic resistance, medicinal plants—known for their bioactive constituents—have become more and more important in the creation of novel medications [5].

Many bioactive substances found in medicinal plants, including phenolics, flavonoids, terpenoids, and alkaloids, are beneficial against microbial infections. These substances reduce the survival and reproduction of microbes by blocking vital activities through a variety of mechanisms. Nowadays, the development of novel medications that satisfy current medical standards depends on scientific analysis and biochemical research of these plants [6].

#### Alqalam Journal of Medical and Applied Sciences. 2025;8(1):401-407 https://doi.org/10.54361/ajmas.258158

It has been shown that active substances extracted from medicinal plants are more effective than those produced in a lab. Additionally, medicinal plants contain multiple active ingredients that operate in concert to treat the illness, something that pharmaceutical compounds made in a lab do not have. In addition to the negative impacts of numerous medicinal substances produced in labs. Chemical components found in many plants used in traditional medicine are byproducts of their metabolic processes and have significant therapeutic benefits. Primary and secondary metabolites [7], alkaloids, flavonoids, glycosides [8], phenolic acids [9], terpenes [10], saponins [11], and tannins [12]. Cymbopogon Schoenanthus, commonly known as lemon grass or camel grass, is known locally as Adhkhar in Libya and Halfaa Alber in Egypt. Both Alsakhbar in Saudi Arabia and Alsakhir in Morocco.



Figure 1. Cymbopogon schoenanthus (Fresh)



Figure 2. Cymbopogon Schoenanthus (Dried)

Cymbopogon Schoenanthus is an aromatic member of the Poaceae family that grows in rocky valleys and on mountain slopes throughout the Middle East, including the United Arab Emirates.

Its chemically aromatic leaves are used in food preparation, adding flavor to curries and custards, and lemongrass herbal tea is a popular substitute for traditional tea in North Africa, valued for its flavor and invigorating scent [13]. Cymbopogon Schoenanthus is a fragrant perennial herb that grows to a height of 30 to 100 cm and has several branches growing from the base. Rough, linear, and outwards curled, the leaves can grow up to 30 cm in length and 1-3 mm in width.[14]. Cymbopogon Schoenanthus has

The plant contains antirheumatic, antidiarrheal, emmenagogue, stomachic, diaphoretic, diuretic, carminative, and tonic properties [10]. The phytochemical constituents of Cymbopogon Schoenanthus, especially its essential oils rich in citral, limonene, and other terpenes, are widely known for their diverse biological activities. These activities include antibacterial and antifungal properties, antimicrobial activity [15], The plant exhibits powerful antioxidant effects, mostly due to the flavonoids and phenolic chemicals found in its extracts [16]. Through its ability to prevent the generation of pro-inflammatory mediators such as prostaglandins and cytokines, Cymbopogon schoenanthus essential oil decreases inflammation [17]. In addition to their analgesic and antispasmodic qualities, Cymbopogon schoenanthus aqueous and ethanolic extracts have anticancer effects [18].

The study aimed to evaluate the antibacterial qualities of many raw extracts made from C. Schoenanthus (Adkher). To do this, the extracts were tested against a variety of pathogenic bacteria to see how well they inhibited the development and survival of the bacteria.

# **METHODS**

#### Collection of plant material

The plant was collected in the spring of 2024 from the Bani Walid region of Libya. The Department of Botany at the Tripoli College of Medical Sciences in Tripoli, Libya, classified and verified it. It had been transported to the lab, where the entire plant was cleaned with purified water and allowed to dry for two weeks in the shade, away from moisture and sunshine.

#### Chemicals and their sources

High-grade analytical organic solvents, such as methanol and chloroform, are manufactured by Merck (Germany). Tetracycline and Vancomycin were among the antibiotic paper discs that Oxoid (England) produced.

#### Equipment:

Electric Grinder machine. Miniator Shaker (Fedmund Buhler KL2), Germany. Rotary evaporator (Heidolph300 LabroRota), Germany. Hot plate (0-450°C) with magnetic stirrer (0-1200 rpm/min) IKA labor technik, USA. Incubator with inner orbital shaking mechanism (15-50°C), a product of GFL, Germany. Spectrophotometer (Jenway UV-VIS 6305), Germany. UV light 366nm source CAMAG UV, Germany. Oven (50-500°C) Memmert D2800, were a product of Germany. Laminar flow Cabinet-Biosafe 5-130 EHRET, German made. Autoclave, Sterilization GMBH, KSG Germany. Ordinary Centrifuge (6000 revolution/min), Hettich EBA 20, Germany.

Table 1. Four types of bacteria were chosen to measure the biological Activity of plant

Gram (+ve)	Gram (-ve)
Bacillus subtilis (B.sub)	Salmonella typhimurium (Sal)
Staphylococcus aureus (S.aur)	Escherichia coli (E. coli)

# **Extraction Methods**

#### Crude plant extract preparation.

Using a clean electric blender, the dried material was ground into a powder and sieved through an acceptable mesh screen before being stored in an airtight, dark glass container (wrapped with aluminum foil sheets). The powdered material was subjected to the following wrapped in sheets of aluminum foil. This powdered substance was extracted using the following methods:

#### Aqueous extraction

**Water:** To make an aqueous infusion, 20 grams of ground plant material were immersed in 100 milliliters of distilled water for six hours at room temperature (25°C) in tightly sealed containers, with continuous shaking, Whatman no. 1 filter paper was used for filtering the crude extract, which was then stored at 4°C **Hot water**: To extract hot water, 20g of powdered plant material was boiled in distilled water for 10 to 15 minutes while being stirred. The extract was cooled, filtered through Whatman No. 1 filter paper, and stored in a sealed container at 4°C until it was needed.

#### **Organic Solvent Extraction**

To extract organic solvents, 20g of dry, milled plant material was extracted three times using 200 ml of the solvent (methanol) over 24 hours. The extract was filtered and dried using a rotary evaporator at 40°C and decreased pressure. Whatman filter paper was used to filter the crude extract, which was then stored at 4°C until it was needed.

# Phytochemical Screening

Phytochemical screening for major constituents was undertaken using standard qualitative methods alkaloids, anthraquinones, coumarins, flavonoids, saponins, tannins, and terpenes [19]. Table (2) shows the contents of the phytochemicals obtained from the extraction of Cymbopogon Schoenanthus rated from (+ ve) for faint to (+++ ve) for dense turbidity.

# Chemical testing (Color reactions)

Standard procedures were used to identify the contents by conducting chemical tests on the alcoholic and aqueous extracts of powdered specimens [20].

# Saponins Test

Saponins are naturally occurring amphiphilic glycosides composed of a hydrophobic aglycone (sapogenin) and a hydrophilic sugar moiety (glycone). A water bath was used to boil 2 g of the powdered sample in 20 ml of distilled water and filter. A consistent, long-lasting froth was achieved by rapidly shaking 2.5 ml of distilled water with 5 ml of the filtrate. After adding three drops of olive oil to the foam and giving it another good shake, the emulsion was observed to form. Emulsion formation indicates the presence of Saponins [21].

# Anthraquinone Test

After 0.5g of extract and 10 ml of benzene were shaken and filtered, a 10% ammonia solution was added to the filtrate, and the mixture was shaken once more. Anthraquinones are indicated by the production of a pink, crimson, or violet color in the ammoniacal phase [22].

# Alkaloids Test

Alkaloids are nitrogen-containing substances that occur naturally and are typically obtained from plants. They are extensively researched for their pharmacological qualities and display a variety of biological actions. By precipitating alkaloids using acidic reagents, qualitative tests for alkaloids identify the presence of their basic nitrogenous structure [22].

# Tannins Test

Many plants contain polyphenolic chemicals called tannins, which are distinguished by their capacity to precipitate and bind proteins and alkaloids. They are divided into two primary categories: condensed tannins (flavonoid polymers) and hydrolyzable tannins (derivatives of gallic or ellagic acid). Usually, qualitative analysis based on the tannins' reactivity with particular reagents is used to test for tannins. A test tube containing roughly 0.5g of each of the dried powdered samples was filled with 10ml of water, boiled, and then filtered. The filtrate was treated with a few drops of 0.1% ferric chloride, and its coloration was checked for brownish green or blue-black hues. The intensity of color indicates the tannins' richness [22].

# Flavonoids Test

One class of polyphenolic chemicals that is commonly present in plants is flavonoids. They have important functions as plant pigments, antimicrobials, and antioxidants. Flavonoids are categorized into multiple subclasses, such as flavones, flavonols, flavanones, isoflavones, and anthocyanidins, and are distinguished by their C6-C3-C6 backbone structure. Based on their unique chemical characteristics, flavonoids are tested using both qualitative and quantitative techniques. After adding 5 milliliters of diluted ammonia solution to a fraction of each plant extract's aqueous filtrate, concentrated H2SO4 was added. Each extract contains flavonoids, which are indicated by a yellow tint that disappears while the extract is standing [21].

# Terpenoids Test (Salkowski test)

Terpenoids, commonly referred to as isoprenoids, are a broad and varied class of organic compounds that occur naturally and synthesized from isoprene units. Based on the quantity of isoprene units, they are categorized into various classes (monoterpenoids, sesquiterpenoids, diterpenoids, etc.). The qualitative and quantitative identification of terpenoids in biological or chemical samples can be accomplished using a variety of techniques, which are explained in detail below.

Five milliliters of each plant's aqueous extract were combined with two milliliters of chloroform, and three milliliters of concentrated H2SO4 were carefully added to create two layers. When there is a reddish-brown tint at the interface, it means that terpenoids are present [23].

# Assay for Antimicrobial Properties of Extracts Hole-plate diffusion methods:

Using "hole-plate diffusion methods," the antibacterial activity of the crude extracts was evaluated. For testing, each test organism was grown in nutrient broth (No. 2, Biolab, Difco) for 24 hours at 37°C after being maintained on nutrient agar slant [24].

# Result and dissection Phytochemical screening

The active phytochemical components of *Cymbopogo Schoenanthus* (Table 2) have been identified through screening. Tannins, alkaloids, terpene, coumarin, and anthraquinone are either abundant or moderately present in the plant, according to the results. Other phytochemical components, such as flavonoids, were found to be present in varying levels in the sample, with few exceptions [24].

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Table 2. Cymbopogon	Schoenanthus's o	qualitative	screenina	findinas	for common	hutochemicals.
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Phytochemicals	Water extract	Methanol extract
Alkaloid	+ve	++ve
Flavonoid	-ve	-ve
Tannin	+ve	+++ve
Terpene	+ve	++ve
Coumarin	-ve	++ve
Saponin	+ve	+ve
Anthraquinone	-ve	++ve

+++ve, High; ++ve, Medium, +ve, low, -ve, None

#### Plant productivity percentage

According to the crude extract percentage table listed below, the saving facility has a 35.4% productivity rate.

Table 3. Results of percentages of crud extracts					
Plant	Plant productivity percentage				
Cymbopogo Schoenanthus of methanol extraxts	%35.4				

# Antimicrobial activity

# Aqueous extract

*Cymbopogon Schoenanthus* aqueous extract's antibacterial characteristics (Table 2) are investigated for their active phytochemical constituents. According to the results, the plants have a rich or moderate number of tannins, alkaloids, terpene, coumarin, and anthraquinone. Other phytochemical components, such as flavonoids, were found to be present in various quantities in the sample, with few exceptions.

From Table 4, it can be concluded that the water extracts demonstrated antibacterial activity was weak to extremely weak against both G +ve and G -ve bacteria.

# Table 4. The cold aqueous plant extract's in vitro antibacterial effectiveness against sensitive,narrow-spectrum bacteria.

		Inhibition Zone (mm) *			
Aqueous extract	Local name	Gram positive		Gram negative	
		S. aur	B. sub	E. coli	S. typhi
Cymbopogo Schoenanthus	Aladkhar	10	10	10	8

\*8mm means no observed inhibition, # MP extracts are of 200mg/ml concentration. S. aur: Staphylococcus aureus, B. sub: Bacillus subtilis, E. coli: Escherichia coli, S. typhi: Salmonella typhi

According to the results in Table (5), the plant's hot water extract exhibits almost the same level of activity against every bacterial strain that was tested as its cold-water extract.

# Table 5. The hot aqueous plant extract's in vitro antibacterial effectiveness against sensitive,narrow-spectrum bacteria

		]	Inhibition	n Zone (mm)*	
Hot aqueous extract	Local name	Gram positive		Gram negative	
		S. aur	B.sub	E. coli	S. typhi
Cymbopogon Schoenanthus	Aladkhar	8	8	10	8

\*8mm means no observed inhibition, # MP extract are of 200mg/ml concentration, B. sub: Bacillus subtilis coli: Escherichia coli,S. typhi: Salmonella typhi, S. aur: Staphylococcus aureus

# Methanolic extract

Based on the results obtained from Table 7, it is evident that: a) Cymbopogon Schoenanthus methanolic extract has modest antibacterial activity (DIZ= 14-18 mm on G +ve / 10-12 mm on G-ve e.g., b) The remaining tested methanolic extract showed remarkable antibacterial activity against Gram-positive bacteria. Since *Cymbopogon Schoenanthus* extract offers a DIZ range of 14–18 cm on G+ve bacteria, it

appears to have relatively superior action when compared to the reference antibiotic Vancomycin (at 30  $\mu$ g/disc) (Table 6).

 

 Table 6. The methanolic extract's in vitro antibacterial effectiveness against sensitive, narrowspectrum bacteria

		Diame	ter of Inhib	oition Zone (mm) *		
Methanolic extract of #	Local name	Gram p	Gram positive		Gram negative	
		S. aur	B. sub	E. coil	S. typhi	
Cymbopogon Schoenanthus	Aladkhar	18	14	12	10	
Vancomycin (30µg)		10	15	24	21	
Tetracycline (30µg)		26	24	27	25	

\*8mm means no observed inhibition, # MP extracts are of 200mg/ml concentration while diluted (as negative control) gave negative results on all test bacteria, S. aur: Staphylococcus aureus B. sub: Bacillus E. coli: Escherichia coli S. typhi: Salmonella typhi

The antibacterial activity of these plants' aqueous extracts varied from mild to considerable. This activity is probably caused by the alkaloids it contains. Because of this, alkaloids' ability to deactivate microbial adhesions, enzymes, and cell membrane transport proteins may be linked to their effectiveness against different microbial species [25]. In comparison to other solvents, including water, ethanol, and hexane, methanol was found to be the most effective solvent for the reliable extraction of antimicrobial compounds from medicinal plants in several studies [26]. Additionally, Stanojevic et al. (2009) demonstrated that alcoholic extracts of Hieracium pilosella had a higher level of phenolic content than water extracts [27]. Methanol was found to be the most effective extraction solvent in this investigation, and the presence or lack of inhibition zones served as a qualitative indicator of antimicrobial activity [Table 7]. The use of these dyes will greatly contribute to achieving a safe, eco-friendly, and green environment.

# Conclusion

It was found from this study that the traditional Libyan plants under investigation, such as Cymbopogon Schoenanthus, have demonstrated a dual function and are being regarded as candidates for additional indepth research on the toxicity and molecular makeup of their active compounds. It can be successfully used as an Antibacterial activity. These results give us different data to evaluate antibacterial effectiveness against sensitive, narrow-spectrum bacteria for other plants

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المستخلص

في هذه الدراسة، أُجريت تجارب لاستكشاف الخصائص المضادة للبكتيريا لمستخلص مائي مُشتق من نبات الإذخر (Cymbopogon Schoenanthus). قيّمت هذه الدراسة النشاط المضاد للبكتيريا للمستخلصات الكحولية والمائية من نبات الإذخر ضد أربعة أنواع مختلفة من البكتيريا باستخدام طريقة الانتشار في الآبار: اثنتان سالبة الغرام ( S. aur, B. (sub) واثنتان موجبة الغرام (E. coli, S. typhi). تشـمل المواد الفعالة بيولوجيًا الرئيسية الموجودة في نبات الإذخر التانين، والأنثراكينون، والتربين، والكومارين، والقلويدات، والصابونين. بينما أظهرت بعض البكتيريا مقاومة للمستخلص، تم تثبيط المكورات العنقودية الذهبية (Staphylococcus aureus) والعصوية الرقيقة (Bacillus subtilis) بواسطة المستخلص الكحولي من نبات الإذخر. تشير هذه النتائج إلى أن المستخلص من نبات الإذخر قد يكون خيارًا قابلاً للتطبيق في البحث عن مضادات الميكروبات الطبيعية. تقدم هذه الدراسة رؤى علمية تساعد في تقييم القيم المضادة للبكتيريا والتحقيق في الخصائص الدوائية الأخرى.