



Phytochemical analysis and antibacterial activity of methanolic and aqueous extracts of cumin (*Cuminum cyminum*) seeds

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Abstract

Cumin (*Cuminumcyminum*) is one of the most popular spices used in India. Cumin is a traditional and much used spice from Middle Ages. Various analyses of the cumin seeds reveals that it contains fixed oil, volatile oil, acids, essential oils, protein, pinene, cymene, terpinene, cuminaldehyde, oleoresin, thymol and other elements. The aim of the current study was to analyze the photochemical and antimicrobial activity of methanolic and aqueous extracts of cumin (*cuminumcyminum*) seeds. In this study various photochemical tests were analysed such as protein, carbohydrates, indole, phenol, alkaline, glycosides, steroids and saponins where the entire test showed positive except indole. Both methanolic and aqueous extracts showed antimicrobial activity against both gram positive and negative bacteria. However, the methanolic extract showed enhanced activity. From the study, it can be ascertained the use of cumin as an antimicrobial agent has a great potential.

Keywords: *Cuminum cyminum*, seeds, phytochemical analysis, antibacterial activity

Introduction

Medicinal plants have always been used in world-wise for treating various diseases from ancient time which is considered a healthy source of life for all people [1]. In the last few decades' antibiotic resistance is a very common problems in medical science which is a serious problems in treatment of various diseases. Inappropriate and overuses of antibiotics are the main causes of drugs resistance [5]. All that makes the use of medicinal plants is the best choice for the treatment of different diseases and infections [1]. One of these medicinal plants is Cumin (*Cuminumcyminum*) it's an herbaceous annual plant of the Apiaceae family and is used as a condiment and as an ingredient in many food industries [2]. It is believed that cumin is originated from Egypt and Syria but now India is major exporter of cumin [12]. From the most important medical uses of Cumin are to reduce inflammation increase, urination, prevent gas and suppress muscle spasms and it is also used as an aid for ingestion, jaundice, diarrhea and flatulence [1]. In India, cumin was also used for kidney and bladder stones, leprosy and eye disease [12]. Phytochemical analysis revealed that cumin seed contain various active phytochemicals, vegetable oils and essential oils which has tremendous antimicrobial properties against [2] gram positive and gram-negative bacteria [3]. Several studies have suggested the presence of cuminaldehyde which has antinociceptive, antineuropathic and anti-inflammatory effects [4]. The main aim of this study is to analysis of phytochemicals and antimicrobial activity of methanolic and aqueous extract against *Escherichia coli*, *Staphylococcus aureus* and *Klebsiellapneumoniae* which causes serious health issues in human.

Materials and methods

Materials

Collection of samples

Sample: Dried cumin seeds (*Cuminumcyminum*) were collected from local markets of Guwahati, Assam, India.

Test organism: *E. coli*, *Klebsiellapneumoniae* and *Staphylococcus aureus* were collected from down town hospital, Guwahati, Assam, India.

Glassware

Petri plates, Conical flask, Measuring cylinder, Test tubes, Beaker, Pipette, Reagent bottle, Parafilm, Inoculation loop, Spatula, Test tube-holder, Funnel, Filter paper.

Apparatus and Equipment

Laminar air flow, Incubator, Hot air oven, Hot plate, Colorimeter, Micropipette, weighing machine, Soxhlet apparatus

Reagents

Distilled water, Methanol, Fehlings reagent A and B, Nitric acid, Acetic acid, Molish reagent, Ferric chloride, Sodium hydroxide, Aluminium chloride, Sodium nitrate, Muller-hinton agar.

Methodology

1. Preparation of Cumin seed extract

a. Aqueous extract

5gm of crushed cumin seeds was mixed in 100ml of sterile distilled water. Then the mixture was boiled at 65 to 70°C for 40 minutes on water bath and then filtered by using Whatman filter paper. The filtrate was centrifuged at 5000 rpm for 10 minutes. The extract was collected and stored at -4°C for further use [10].

b. Methanol extract

10 gm of dried cumin seed powder was mixed with 100 ml of 80% methanol and kept in water bath for 1 hour 45 minutes at 65°C. The extract was centrifuge at 5000 rpm for 5 mins. And then supernatant was concentrated in vacuum evaporator and stored at -40°C. for further experimental use [11].

2. Qualitative analysis of Phytochemicals

By using standard methods various tests were carried out using both water extract and methanol extract of cumin seeds. The presence of phytochemicals was analysed by using chemical tests such as Millon's test for protein, Fehling's test for carbohydrate, Salkowski test for terpenoids, Alkaline reagent test for flavonoids, killer kiliani test for glycosides, Forthing test for Saponin, Liebermann-Burchard's test for steroids [13-20].

Antibacterial activity of the extract

Antibacterial activity of aqueous and methanolic extracts was determined by Agar Well Diffusion Assay. Using a sterile cork borer, nearly 8 mm diameter wells were bored in the seeded agar plates and a 100µl volume of cumin seed extract (equivalent to 33.33 mg dry seeds) diluted in 10% methanol was added into the wells. All the plates were incubated at 37°C for 24 hr. Antibacterial activity was determined by measuring the zone of growth inhibition around the well [3-7].

Result

Phytochemical analysis

The qualitative analysis of phytochemicals of the water and methanol extract of cumin seeds confirm the presence of protein, carbohydrate, flavonoid, saponin, glycoside and steroid. The phytochemical terpenoid is absent.

Table 1: The phytochemical analysis result of cumin seed extract

Phytochemicals	Water extract	Methanol extract
Protein	+ve	+ve
Carbohydrate	+ve	+ve
Terpenoid	-ve	-ve
Flavonoids	+ve	+ve
Glycosides	+ve	+ve
Saponin	+ve	+ve
Steroids	+ve	+ve

Determination of antimicrobial activity

To Estimate the antibacterial activity of the sample

After Incubating at 37°C for 24 hours, a clear zone of inhibition around the cumin seed extract was observed against different gram positive and gram-negative bacteria. Highest zone of inhibition was shown against *Staphylococcus aureus* as shown in the figure.

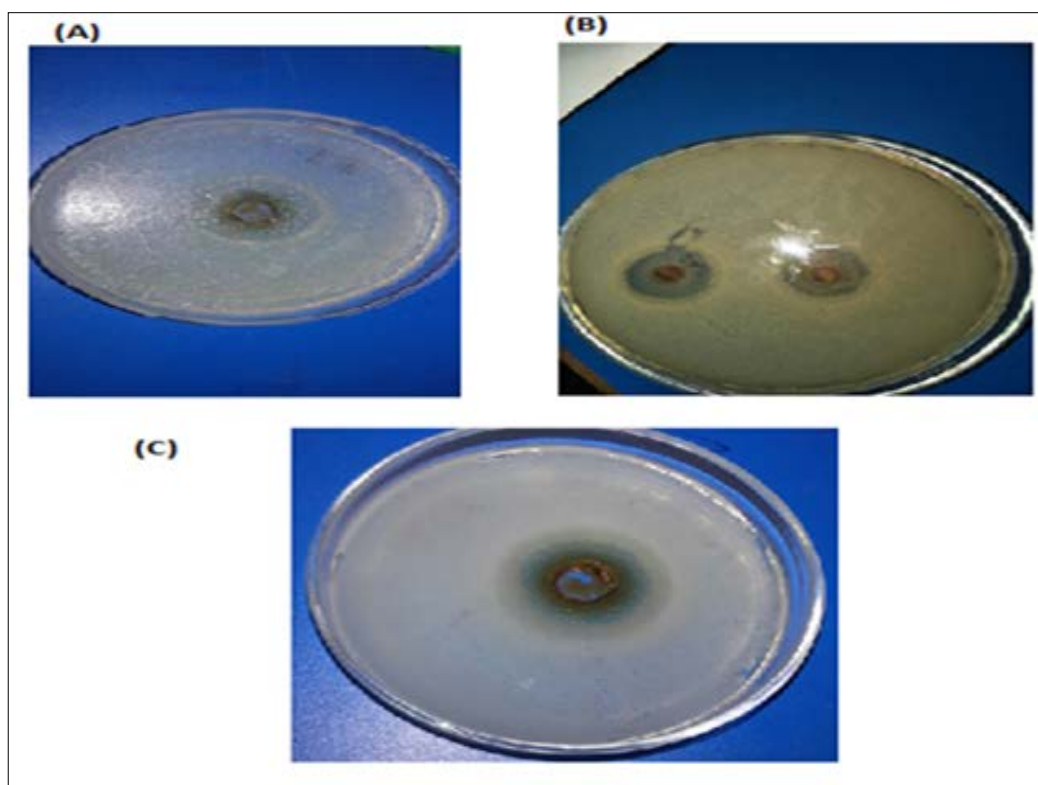


Fig 1: Antimicrobial activity of cumin extract. Zone of inhibition shown against (A) *E. coli*, (B) *Klebsiella pneumoniae*, (C) *Staphylococcus aureus*

Table 2: Antimicrobial activity results of *Cuminum cyminum*.

Bacterial Strain	Zone of Inhibition (Diameter) mm	
	Methanolic extract	Aqueous extract
<i>E.coli</i>	10.7	Nil
<i>Klebsiella pneumoniae</i>	13.1	10
<i>Staphylococcus aureus</i>	14	11

Discussion and Conclusion

Results of phytochemical analysis of *Cuminum cyminum* seed showed the presence of different phytochemicals present in methanolic and aqueous extract. Both the extract

showed positive results for steroid, Flavonoids, Sterols, Saponin which has antioxidant and antimicrobial properties. Most of the study showed the presence of vegetable oil and essential oil antineuropathic and anti-inflammatory effects [4]. In this study Methanolic extract showed antimicrobial activity against *Staphylococcus aureus* and *Klebsiella pneumoniae*. Against *E. Coli* methanolic extract show a moderate activity. Aqueous extract of *Cuminum cyminum* also show a good antibacterial activity against *Staphylococcus aureus*, moderate antibacterial activity against *Klebsiella pneumoniae*, and did not show

antibacterial activity against *E. coli*. The *Cuminum cyminum* seed extract also present antibacterial activity against *P.aeruginosa* and *B.pumilus*^[7]. Essential oil of *Cuminum cyminum* also has good antifungal activity against *Aspergillus flavus*, *Candida albicans* and *Cryptococcus sp*^[1]. The antimicrobial potential of the methanolic extract of cumin seeds against pathogenic bacteria *E.coli*, *Klebsiella pneumonia* causing gastroenteritis or urinary tract infections, stomach cramps, food poisoning and *S.aureus* causing pneumonia, food poisoning and toxic shock syndrome (TSS) were examined. Cumin extract has exhibited antimicrobial activity against all the three bacteria tested. Results in Figure show that all the bacterial strains are sensitive to the presence of cumin extract. The results showed that cumin extract is effectively inducing cell damage in both gram negative as well as gram positive bacteria. The used of plant and spice-based antimicrobials substances in food industry is common as natural agents for extending the shelf life of foods by reducing or eliminating pathogenic bacteria and increasing the overall quality of food products. So methanolic extract of *Cuminum cyminum* with good antibacterial activity against various pathogenic bacteria can be used as a spice-based antimicrobial substance in food industries.

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