



TOTAL PHENOLIC CONTENT AND PHYTOCHEMICAL ANALYSIS OF CUMIN AND CLOVE

Deepali J. Gore, Z. H. Khan*, S. M. Mular and Nazia D. Khan

Department of Biochemistry, Shri Shivaji College of Arts, Commerce and Science, Akola (M.S.), India 444003.

***Author for Correspondence: Ziaulhasan Khan**

Department of Biochemistry, Shri Shivaji College of Arts, Commerce and Science, Akola (M.S.), India 444003.

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ABSTRACT

Spices are building block of flavor in food. Their primary functions are to provide aroma, texture and color to food. In addition they also act as preservative and provide nutritional, and health benefits. Cumin (*Cuminum cyminum*) locally known as 'Zeera' is a flowering plant in a family Apiaceae. Clove (*Dendrophthoe Pentandre* (L). Is one of semi-parasitic plants belonging to the Loranthaceae family. A large no of spices are claimed to possess the antioxidant and antibiotic properties in the traditional system and are also use extensively by the tribal people world wide. The purpose of the study was to determine the content of secondary metabolites in cumin and clove extract through phytochemical screening and total phenolics compounds for this methnolic extract of cumin and clove were prepared by soxhlet extraction method. After extraction, the extraxt was taken in beaker and kept on hot plate and heated at 30-40 °c till all the solvent got evaporated. Dried extract was used for the further study. The highest total phenolics content was found in cumin i.e 78.37mg GAE/mg as compare to clove that is 38.97 mg GAE/mg .Where as flavonoid was present in cumin and absent in clove.

KEYWORDS: Cumin, clove, phytochemical screening, total phenolic content.

INTRODUCTION

The seeds and plants are a treasure house of potential drugs and in the recent years there has been increasing awareness about the importance of medicinal plants and their seeds. Plant-derived substances have recently become of great interest owing to their versatile applications.^[1] Spices are natural compounds derived from many parts of plants: seed, flowers, fruits, leaves, bark, roots and rhizomes etc. Spices not only used for dietary purposes like aroma, color, taste, flavor and preservations of foods but also used as a medicine in traditional system of medicine. Spices have their own unique aroma and flavor which derived from phytochemical compounds.^[2]

Dried clove and cumin is rich in polyphenolic compound, a large class of plant based compound through to impart antioxidant properties. In 2010 scientists at Miguel Henandez University in Spain reported that clove rank highest as a natural antioxidant due to it's phenol content and demonstrated ability to inhibit several damaging oxidative processes. Cumin is good source of iron and keep immune system healthy. Water boiled with cumin seed is good for coping with dysentery. One study found that cumin was protective against memory lost and the damaging effect of stress on the body.^[3] Another study evaluated the antioxidant

content of cumin and found it more effective than other antioxidant content that it might have a role in fighting cancer. Clove are used as carminative, to increase hydrochloric acid in stomach and to improve peristalsis cloves are said to be nature anthelmintic, balch, Phyllis and balch, James prescription for nutritional healing.^[4] Clove oil is beneficial for coping with tooth ache and sore gum. It is also beneficial remedy for chest pain, fever, digestive problem, cough and cold. In addition, clove oil is used in preparation of some tooth paste and clove vaccine solution, which is local anaesthetic use in oral ulceration and inflammation^[5] Present study is undertaken to find out phenolic content and phytochemical study of cumin and clove.

MATERIAL AND METHODS

The cumin and clove were collected from the Akola market.

Preparation of seeds extract

The seeds of cumin and clove were washed under running tap water. It was then dried under shade and ground into coarse powder in the electronic grinder.

Solvent extraction

Crude cumin and clove seeds extract was prepared by Soxhlet extraction method. About 20gm of powdered

cumin and clove seed material was separately uniformly packed into a thimble and extracted with 250 ml of methanol. The process of extraction was continued till the solvent in siphon tube of an extractor became colorless. After that the extract was taken in a beaker and kept on hot plate and heated at 30-40°C till all the solvent got evaporated. Dried extract was kept in freezer at 4°C until further use.

Total phenolic compounds

The total phenolic compounds of the methanolic extracts were determined by using Folin-Ciocalteu reagent), according to the procedure described by Dewanto et al. (2002). Briefly, 200 µL of properly diluted sample extract or standard solution were dissolved in 400 µL of Folin-Ciocalteu reagent. The total volume of the solution was made 4.6 mL with the addition of deionized water. The mixture was shaken, kept for 10 min at room temperature. Afterwards 1 mL of 10% Na₂CO₃ was added and the mixed thoroughly. After incubation in the dark for 90 min, the absorbance was measured at 760 nm using a spectrophotometer. Total phenolic amounts were expressed as mg of gallic acid equivalents per gram of dry weight (mg GAE/g DW), using a calibration curve with gallic acid.

Test for alkaloids

Crude extract was mixed with 2ml of 1% HCl and heated gently. Mayer's And Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids.^[2]

Test for flavonoids

Crude extract was mixed with 2ml of 2% solution of NaOH. An intense yellow colour was formed which turned colourless on addition of few drops of diluted acid which indicated the presence of flavonoids. Another test was performed by mixing crude extract with few fragments of magnesium ribbon. Then concentrated HCl was added drop wise. The development of pink scarlet coloration after few minutes indicated the presence of flavonoids.(2)

Test for saponins

Crude extract was mixed with 5ml of distilled water in a test tube and shaken vigorously then was left to stand for 10 minutes and the result was noted. No thick persistent froth was observed. This indicated the absence of saponins.(2)

Test for glycosides

For detection of glycosides, crude extract was hydrolysed with concentrated HCl for 2 hrs on water bath, filtered and the hydrolysate was subjected to the following test. Borntrager's test: To 2 ml of filtered hydrolysate, 3 ml of chloroform was added and shaken, chloroform layer was separated and 10% ammonia solution was added to it. The development of pink coloration indicated the presence of glycosides.(2)

Test for proteins

Crude extract when mixed with 2 ml of Millon's reagent, white precipitate appeared which turned red upon gentle heating that confirmed the presence of proteins.(2)

Test for steroids

Crude extract was mixed with 2ml of chloroform and concentrated H₂SO₄ was added sidewise. A red color produced in the lower chloroform layer indicated the presence of steroids.(2)

Test for phenolic compounds and tannins

Crude extract was mixed with 2ml of 2% solution of FeCl₃. A blue-green coloration indicated the presence of phenolic compounds and tannins.(2)

Test for fixed oils and fats

A few drops of 0.5 N alcoholic KOH were added to a small quantity of crude extract along with a drop of phenolphthalein. The mixture was heated on water bath for 2hrs. Partial neutralization of alkali indicated the presence of fixed oils and fats.(2)

Test for carbohydrates

Molisch's test: Crude extract was mixed with 2ml of Molisch's reagent and the mixture was shaken properly. After that, 2ml of concentrated H₂SO₄ was poured carefully along the side of the test tube. Appearance of a violet ring at the interphase indicated the presence of carbohydrates.

Fehling's test: Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and gently boiled. A brick red precipitate appeared at the bottom of the test tube indicated the presence of reducing sugars.

RESULTS AND DISCUSSION

Total phenolic content in Clove and Cumin

Total phenol content (TPC) was determined in comparison with standard gallic acid and the results are expressed in terms of mg GAE/g dry sample. Total phenolic content (TPC) of clove and cumin was 78.37, 38.97 mg (GAE)/mg DW respectively. TPC in clove i.e. 560 mg GAE/100 ml.

Further efforts were made to analyse the waste portion of clove and cumin i.e. clove and cumin extract. Highest total phenolic of 74.28 mg Gallic acid equivalent (GAE/mg). Clove and cumin show a TPC of 30 mg gallic acid equivalent (GAE/mg) DW. Phenolic content in clove observed was 371.56 mg of Gallic acid equivalent (GAE/mg) such as organic acid which interferes the determination of total phenolic contents by the Folin Ciocalteu method which leads to overvaluation of the phenolic content. Furthermore different phenolic might present different response with the Folin.

Phytochemical Analysis of Cumin and Clove

The preliminary screening carried out on methanolic extract of cumin and clove revealed the presence of alkaloid, tannin, protein, and amino acid. Flavonoid are

absent in the clove and present in cumin. Methanolic extract of cumin and clove have shown a positive result. In positive result for alkaloid, protein and tannins.

Table 1 : Phytochemical analysis of clove and cumin

Sr. No.	Microchemical Test	Coloration	Cumin	Clove
1	Alkaloid by -			
a.	Mayer's reagent	Dark brown	++	++
b	Wagner's reagent	Dark brown	++	++
2	Flavonoid	Yellow	++	-
3	Glycosides by -			
a.	Modified Borntragers test	Rose Pink colour	++	++
	Legal's test	Pink to Blood Red	++	++
4	Saponin	No formation of foam	-	-
5	Tannins	Bluish green	+++	+++
6	Protein by-			
a)	Xanthoproteic test	Yellow colour	+++	+++
b)	Ninhydrin Test	Blue colour	+++	+++

REFERENCE

1. Prasad MP *et al.*,. Phytochemical analysis and antioxidant potential of Piper species and its Molecular Characterization by RAPD Markers. *International Journal of Fundamental & Applied Sciences*, 2012; 1(4): 71-73.
2. Prashant Tiwari, Bimlesh Kumar, Mandeep KAUR, Gurupreet Kaur, Harleen Kaur. Phytochemical screening and Extraction: *International Pharmaceutica Scientia*, 2011; 1(1): 98 – 99.
3. Ramaya BS, Ganesh P.. Phytochemical analysis and comparative effect of *Cinnamomum zeylanicum*, *Piper nigrum* and *Pimpinella anisum* with selected antibiotics and its antibacterial activity against enterobacteriaceae family. *International Journal of Pharmaceutical & Biological Archives*, 2012; 3(4): 914-917.
4. Remington G, P.. Remington: The science and practice of pharmacy, 21st edition, Lippincott William and Wilkins, 2005; 773-774.
5. Youngken, H.W: Text book of pharmacognosy 6th ed., 1950; 86-90.