The Screening on Phytochemical Analysis and Anti Oxidant Activity Of *Syzygiumaromaticum* .

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Abstract

India has a rich culture of medicinal herbs and spices which includes about more than 2000 species and has a vast geographical area with high potential abilities fof Ayurvedic, Unani and Siddha traditional medicines. But only few have been studied chemically and for, pharmacologically for their potential medicinal values. Plants are used medicinally in different countries and are a source of many potent and powerful drugs. The study investigates the Screening on Phytochemical Analysis and Anti Oxidant Activity Of *Syzygiumaromaticum.*

Key Words: Medicinal plant, *Syzygiumaromaticum,* Anti Oxidant Activity.

I.Introduction

Medicinal plants have been identified and used throughout the human history of plant makes many chemical compound for biological functions and including defence against insects, fungi and herbivorous mammals. Herbal spices have beneficial pharmacology, it also gives them the potential as conventional pharmaceutical drugs to cause harmful side effects. The herbs are used widespread to treat disease in non-industrialized societies [1].

Spices have been used as medicines throughout history. Indeed, studies of wild animals show that they also instinctively eat certain plants to treat themselves for certain illnesses. In Asia, the practice of herbal medicine is extremely well established and documented; as a result, most of the medicinal plants that have international recognition come from China and India Many people now take medicinal spices products on a daily basis, to maintain good health as much as to treat illness.[2]

II Materials And Method

2.1 Sample collection

Three different types of spices were collected from various places in and around Tiruppur District, Tamilnadu.

2.2 Sample extraction

The fresh dry samples were collected, dried, powdered and stored for further analysis. 16g of each sample was taken and mixed with 80ml of different solvents (Water, Acetone, Ethanol) the contents were mixed well and kept in a shaker for 24 hours. Then the samples were filtered using 25µm pore sized whatman No.1 filter paper and the clear solution was stored in a brown bottle for future use.

Figure of *Syzygiumaromaticum*powder

2.3 Phytochemical Analysis

Tests for carbohydrates

To this extract is dissolved in 5.0ml of water and filtered. The filtered was subjected to the following tests.

Molisch's test

To 2.0ml of filtered, 2 drop of alcoholic solution of a-naphthol were added. The mixer was shaken well.1.0ml of concentrated sulphuric acid is added slowly along the sides of the test tubes and allowed to stand. A violet colour ring is indicates the presence of carbohydrate [3].

Test for Reducing Agent

Fehling's Test

To 1.0ml of filtrate was boiled on water bath with 1.0ml of each Fehling's solution A & B red Precipitate indicates the presence of sugars**.**

Tests for alkaloids

Mayer's Test

Take 5 ml of extract, few drops of Mayer's reagent is added by the side of the test tube. A green coloured precipitate indicates the test as positive.

Wagner's Test

Take 5 ml of extract, few drops of Wagner's reagent along with 1.5% HCL is added by the side of the test tube. A reddish brown precipitate confirms the test as positive.

Hager's Test

Take 5 ml of extract, 1 ml of Hagers reagent was added. A prominent yellow/orange precipitate indicates the test as positive.

Test for Saponins

Foam Test

5ml of the extract is vigorously shaken with 8 ml of distilled water in a test tube for 30 sec and was left undisturbed for 20 min. Formation of foam layer indicating the presence of saponins.

Test for Tannins

Lead Acetate Test

To the 2ml of extract, few drops of lead acetate solution is added. A Yellow precipitate indicates the presence of phenolic compounds.

Test for Flavanoids

Add few drops of Dilute H_2SO_4 to a small quantity of extract.

Test for Terpenoids

To 2ml of Extract, add 2ml of acetic acid along with few drops of H_2SO_4. Blue ring formation indicates the presence of Terpenoids.

Test for Phlobotannins

The extract is boiled and add 1% HCL. The deposition of Red precipitate indicates the presence of phlobotannins.

Test of coumonin

To 2ml of extract, add 10% of 3ml of NaOH. The yellow colour formation indicates the presence of coumonin

Test for cycloglycosides

Take 5ml of extract and add 2ml of Acetic acid, a drop of $FeCl_2$, 1ml of H_2SO_4 are added in test tube. Which forms brown violet and greenish ring it indicates presence of cycloglycosides.

Test for Total Phenols

Take a few ml of extract and 3% ferric chloride are added in the test tube. Formation of blue colour indicates presence of phenols.

Test for Quinone

Take a few ml of extract and 5ml of HCL are added in the test tube. The formation of yellow precipitate indicates the presence of Quinone.

Test for arthoquinone

Take a 2ml of extract added with 2ml of 10% ammonium hydroxide in the test tube. The bright pink colour formation indicates presence of arthoquinone.

Test for steroids

To a 2ml of extract,2ml of chloroform,acetic acid are added in the test tube.The formation of reddish brown indicates the presence of steroids.

Test for carotenoids

To a10ml off extract was evaporation to dryness and added with 2 to 3 drops of concentrated sulphuric acid and chloroform. Which produce blue colour indicates presence of carotenoids.

Test for fatty acids

To a few ml of extract added with aqueous NaOH solution and concentrated sulphuric acid in the test tube. The test tube was shaken well. Small portions of solvent and extract the Fatty Acid is evaporated it residue only. It indicates the presence of fatty acid.[4]

Test for Amino acids

The extract (100 mg) is dissolved in 10 ml of distilled water and filtered through Whatmann No. 1 filter paper and the filtrate is subjected to test for Amino acids.[5]

Ninhydrin test

Two drops of ninhydrin solution (10 mg of ninhydrin in 200 ml of acetone) are added to 2 ml of aqueous filtrate. Appearance of purple colour indicates the presence of amino acids.

Test for Proteins

The extract (100 mg) is dissolved in 10 ml of distilled water and filtered through Whatmann No. 1 filter paper and the filtrate is subjected to test for proteins.

Millon's test

To 2 ml of filtrate few drops of Millon's reagent are added. A white precipitate indicates the presence of proteins.[6]

Biuret test

2 ml of filtrate is treated with 1 drop of 2% copper sulphate solution. To this 1 ml of ethanol (95%) is added, followed by excess of potassium hydroxide pellets. Pink colourethanolic layer indicates the presence of protein.

Test for Cholesterol

To a 2 ml of extract and 2 ml of chloroform are added in test tube. After adding reagent to dry the test tube, then added 10 drops of acetic anhydride and two to three drops of concentrated H2SO4 in the test tube. The red rose color then appear the bluish green color, it indicates the presence of Cholesterol.

Test for cardiocglycosides

To a 5ml of extract, 2ml of glacial acetic acid, 1 drop of ferric chloride and concentrated sulphuric acid are added with sides of test tube. To formation of violet ring it indicates presence of cardiac glycosides.

Detection of Phlobotannis

To a few ml of extract boiled with 1% HCL gives deposition of red precipitate. It indicates presence of phlobotannis.

Test for Arrthoganins

To a few ml of extract, 2ml of 2N ammonium chloride and ammonia are added in test tube. To appearance of pink-red to blue-violet colour it indicates presence of arrthoganins.

Test for Leucoarthocyanin

To a 5ml of extract and 5ml of isoamyl alcohol are added in the test tube. Formation of red colour appear the upper layer it indicates presence of leucoarthocyanin.

Test for Phenols

Ferric Chloride test

The extract (50 mg) is dissolved in 5 ml of distilled water. To this few drops of neutral 5% ferric chloride solution are added. A dark green colour indicates the presence of phenolic compound

Gelatin test

The extract (50 mg) is dissolved in 5 ml of distilled water and 2 ml of 1% solution of Gelatin containing 10% NaCl is added to it. White precipitate indicates the presence of phenolic compounds.

Lead acetate test

The extract (50 mg) is dissolved in of distilled water and to this 3 ml of 10% lead acetate

Test for Emodins

To a 2ml of ammonium hydroxide, 3ml of benzene and few ml of extract. It appearance a red colour to presence of emodines.

Test for Glycosides

For 50 mg of extract is hydrolysed with concentrated hydrochloric acid for 2 hours on a water bath, filtered and the hydrolysate is subjected to the following tests.

Borntrager's test

To 2 ml of filtered hydrolysate, 3 ml of choloroform is added and shaken, choloroform layer is separated and 10% ammonia solution is added to it. Pink colour indicates presence of glycosides.

Legal's test

50 mg of extract is dissolved in pyridine, sodium nitro prusside solution is added and made alkaline using 10% NaOH. Presence of glycoside is indicated by pink colour solution is added. A bulky white precipitate indicates the presence of phenolic compound

2.4 Antioxidant Activity

The detection of anti-radical substances of the extracts was performed by the method of DPPH

DPPH free radical scavenging activity

The diluted working solutions of the test extracts were prepared in ethanol. Ascorbic acid was used as standard in 530µg/ml solution. 0.002% of DPPH (Diphenyl-2-picrylhydrazyl) was prepared in ethanol and 1 ml of this solution was mixed with 1 ml of sample solution and standard solution separately. These solution mixtures were kept in dark for 30 min and optical density was measured at 517 nm using UV spectrophotometer. Ethanol (1 ml) with DPPH solution (0.002%, 1 ml) was used as blank. [7]

Calculation

The optical density was recorded and % inhibition was calculated using the formula,

Percent (%) inhibition of DPPH activity = 100- (A-B/A) x100

Where,

A = optical density of the blank and

B = optical density of the sample.

III. Results And Discussion

3.1 Phytochemical Analysis

Phytochemicals are found in spices and are nutrient in nature. They have certain disease preventing properties. It offers protection against pathogens [8]. Previous studies reported that the *Syzygiumaromaticum* carbohydrate, Alkaloids, saponins, Flavonoids, Glycosides in the extraction of *Syzygiumaromaticum.* [9].These can also act as anti-bacterial and hormonal stimulation.

The phytochemical screening of various extracts shows that the presence of certain important components such as carbohydrate, Alkaloids, saponins, Flavonoids, Glycosides, cycloglycosides and total phenols. The above phytochemical constituents were highly present in the acetone and chloroform extraction. The major types of phenolic compounds found were phenolic acids (gallic acid), flavonol,glucosides, phenolic volatile oils (eugenol, acetyl eugenol) and tannins. It was highlighted the huge potential of clove as radical scavenger and as a commercial source of polyphenols.

S.no	Constitution	Clove (*Syzygiumaromatigum*)		
		Aqueous	Acetone	Ethanol
1	Carbohydrates	+	-	-
2	Reducing sugar		-	-
3	Alkaloids	+	+	+
	(a)Hager test	-	-	-
	(b)Mayer test	-	-	-
	(c)Wagner test	+	+	+
4	Saponins	+	-	+
5	Tannins	+	-	+
6	Flavonoids	+	+	-
7	Terpeniod	+	-	+

The present study, the extraction of *Syzygiumaromatigum* showed the alkaloids, which is used allophatic system.

8	Phlobotannin	-	-	-
9	Coumorin	+	-	-
10	Cycloglycosides	+	-	+
11	Phenol	+	+	+
12	Quinone	+	-	-
13	Anthroquinones	-	-	-
14	Steroids	+	-	+
15	Carotenoids	-	-	-
16	Fatty acids	-	-	-
17	Amino acids	+	+	+
18	Proteins	+	-	+
19	Cholesterol	-	+	-
20	Cardiac glycosides	-	-	-
21	Arrthocyanins	-	-	-
22	Leucoarthocyanin	-	-	-
23	Phenols	+	+	+
24	Emodins	-	-	-
25	Glycosides	+	+	+

Results for Phytochemical Screening of Clove (*Syzygiumaromaticum*)

3.2 Antioxidant Activity

phenol and tannins acts as antioxidants. It also has biological property like anti carcinogen, anti-inflammation, cardiovascular protection and cell proliferation activities reported that glycosides are useful in lowering blood pressure. In the present study, the extracts of spices showed the presence of alkaloids, which is used in allophatic systems. The present studies of spices is proved the antioxidant properties have a possible application in medicinal purpose. The current investigation deals with the spices such as, Clove *(Syzygiumaromaticum*), extraction has the high amount of antioxidant. The presence of antioxidant including secondary metabolites to studies of spices biochemical activity.

Conclusion

Phytochemicals are found in spices and are nutrient in nature. They have certain disease preventing properties. It offers protection against pathogens These can also act as anti-bacterial and hormonal stimulation.

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