Think India Journal
ISSN: 0971-1260 Vol-22, Special Issue-31
National Conference ETDAB-2019
Held on 23th & 24th December 2019
Organized by: Deptt. of Botany, Deogiri College, Aurangabad, M.S

Study of invitro Antioxidant, Antibacterial Activity and Phytochemical Analysis of *Coriander Sativum*

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ABSTRACT

Antioxidant and Antibacterial activities of coriander leaf extract were studied with their phytochemical analysis. Antioxidant activity was performed by DPPH (1,1 diphenyl-2-picryl hydroxyl) radical scavenging method for different extracts. They exhibited strong antioxidant DPPH radical scavenging activity with absorbance 0.0880, 0.0410 for ethanolic leaf extract and ascorbic acid respectively. To investigate the antibacterial activity, leaf extract was tested against *E.coli, Salmonella typhi, Staphylococcus aureus*, *Bacillus subtilis*. The Ethanolic and Aqueous extract inhabited the growth of selected bacteria. A phytochemical analysis of Coriander leaf extract shows the presence of flavonoids, alkaloids, phenolic compounds, tannin, saponin and terpenoids. This study suggest that Coriander consist of important phytochemical constituents which leads to its good potential of antioxidant and antibacterial properties.

Key Words :*Coriander Sativum,* Antioxidant, Antibactrial, DPPH, Flavonoids, phenolic compounds

INTRODUCTION

Coriander sativum is an annual herb of the family Apiaceae. All parts of coriander plant are edible. Their leaves are used as spice in food. The leaves are variable in shape, lobed at the base of plant. Fresh juice of coriander is extremely advantageous in curing many deficiencies related to vitamin and iron. Its juice is beneficial in curing many diseases. Fresh leaves can also be eaten as such because of various health benefits [1]. In Indian traditional medicine, C. sativum is used in treating the disorders of digestive, respiratory and urinary systems, as it has diaphoretic, diuretic, carminative and stimulant activity [2]. Many of the medicinal plants display antioxidant and antimicrobial properties which can protect the human body against both cellular oxidation reactions and pathogens [3]. Medicinal plants are known to produce certain bioactive molecules which are responsible for their antimicrobial properties [4]. The present study reporting the antioxidant and antibacterial activity of *Coriander sativum*with its phytochemical analysis.

MATERIALS AND METHODS

Collection of Plant :

Fresh samples of *Coriander sativum* were purchased from local market of Osmanabad. The plants were collected in plastic bag which was washed two to three times with tap water and then rinsed with distilled water.

Prepareation of Plant Extract :

Aqueous Extract : 5 gm of dry powder was taken in conical flask having 100 ml of deionized water flask was heated at 90^0c in water bath for 1 hour. After 1 hour flask was taken out from the water bath and kept at room temperature for cooling purpose. Then the extract was filtered with the help of Filter paper and stored at 4^0c[5].

Ethanolic Extract : 5 gm of powdered plant sample was soaked with 50 ml of ethanol. Themixture was incubated at 4 ^{0}c for 48 hrs. After incubation period was over, the mixture wasfiltered and centrifuged at 10000 rpm at 40 ^{0}c. The extracts were concentrated to dryness inrotary evaporator and stored at 40 ^{0}c [6].

Antioxidant DPPH Assay :

The radical scavenging activities of different extracts were determined by 1,1-diphenyl-2-picryl hydroxyl (DPPH), the method described by Tailor Shekhar and Goyal Anju. [7]. 1ml of 0.1mM DPPH solution was added to 3ml of different extracts at concentrations 5,10,15,20,25,30 $\mu g/ml$. The mixture was shaken vigorously and allowed to stand at room temperature for 30 min. then absorbance was measured at 517 nm. Reference standard compound being used was Ascorbic acid and experiment was done in triplicate [8]. Lower absorbance of the reaction mixture indicated higher free radical activity [9]. The percent DPPH scavenging effect was calculated by following equation:

$$Percent\ Inhibition = \frac{A0 - A1}{A0} \times 100$$

A_0= Absorbance of control reaction

A_1= Absorbance of test

Preparation of Bacteria Inoculums:

A 24 hrs old culture of bacterial isolate was emulsified in sterile normal saline and adjusted to 0.5 Mcfarland standard (by compared the desired inoculums with 0.5 Mcfarland standard) [6].

Antimicrobial Activity:

Disc Diffusion Method- For each ethanolic extract and aqueous extract a solidified Muller Hinton agar plate was assigned to it and aseptically each plate was seeded by flooding with test

organisms (*Escherichia coli, Salmonella typhi, Staphylococcus aureus* and *Bacillus subtilis*)and then allowed to settle for 10 min. Then sterile disc containing 20 μleach extract were applied on bacterial lawn. Incubated at 37^0c for 24 hrs.

Phytochemical Analysis:

Phytochemical analysis of the test sample was carried out according to standard methods [10].

Test for Carbohydrates

Molisch Test- 1ml of aqueous extract was treated with 2drops of alcoholic alpha-naphthol solution in a test tube and then 500 ul of concentrated sulphuric acid was added carefully along sides of the test tube. Formation of the violet ring at the junction indicates the presence of carbohydrates.

Test for Proteins and Amino Acids

Ninhydrin Test- One ml of the test solution was heated with 1 drop of 5% Ninhydrin solution ona water bath for 10 minutes. Formation of the blue color indicates the presence of amino acids.

Tests For Alkaloids : To the extract, dilute hydrochloric acid was added, shaken well andfiltered.

Mayer's Reagent Test - To 1 ml of filtrate, few drops of Mayer's reagent was added along sidesof the tube. Formation of creamy precipitate indicates the presence of alkaloids.

Test for Flavonoids

Alkaline reagent test - One ml of the extract was treated with few drop of sodium hydroxidesolution separately in a test tube. Formation of intense yellow color, which becomes colorless onthe addition of drops of dilute acid, indicates the presence of flavonoids.

Tests for Glycosides

Borntrager's Test - To 1 ml of test solution, dilute sulphuric acid was added, boiled for 5minutes and filtered. To the cold filtrate, 1 ml of benzene or chloroform was added and it wasshaken well. The organic solvent layer was separated and ammonia was added to it. Formationof pink to redcolor in ammonical layer indicates the presence of anthraquinones glycosides.

Tests for Tannin and Phenolic Compounds

Ferric chloride test 5% - A small amount of extract was dissolved in distilled water. To thissolution 500µl of 5% ferric chloride solution was added. Formation of blue, green or violet colorindicates the presence of phenolic compounds.

Test for Saponins

Foam test- One ml of the extract was diluted with 2ml of distilled water and shaken in agraduated cylinder for 15 minutes. The formation of layer of foam indicates the presence ofsaponins.

Tests for Terpenoids

Salkowski's Test- One ml of the extract was treated with 1ml of chloroform and filtered. Thefiltrate was added with few drops of concentrated sulphuric acid, shaken and allowed to stand. Ifthe lower layer turns red, a steroid is present. Presence of golden yellow layer at the bottomindicates the presence of triterpenoids.

RESULTS

The ethanolic extract of coriander leaves showed higher antioxidant activity compared with standard Ascorbic acid by DPPH scavenging method. The absorbance at 517 nm were 0.880 and 0.0410 for ethanolic extract and ascorbic acid respectively (Table-1, Graph-1). The percent inhibition was 65.21 and 83.79 for ethanolic extract and ascorbic acid (Table-2, Graph-2). It shows that at higher concentration ethanolic extract captured more free radicals formed by DPPH which leads to decrease in absorbance.

Table-1 : Absorbance of Ethanolic and Aqueous extracts of *C.sativum*with standard ascorbic acid at 517 nm

Sr. No.	Concentration ($\mu g/ml$)	Ascorbic acid (Absorbance)	Ethanolic extract (Absorbance)	Aqueous extract (Absorbance)
1.	05	0.2470	0.2331	0.2315
2.	10	0.1829	0.2311	0.2322
3.	15	0.0478	0.2071	0.2350
4.	20	0.0424	0.1510	0.2217
5.	25	0.0421	0.1311	0.2110

6.	30	0.0410	0.0880	0.2290

Table-2: Percent Inhibition Ethanolic and Aqueous extracts of *C.sativum*with ascorbic acid

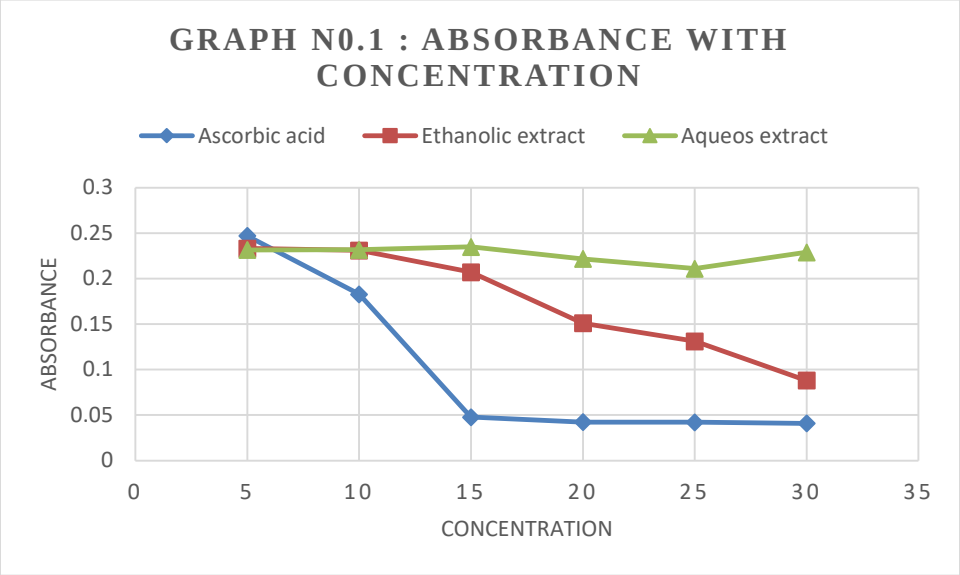
Sr. No.	Concentration ($\mu g/ml$)	Ascorbic acid (% inhibition)	Ethanolic extract (% inhibition)	Aqueous extract (% inhibition)
1.	05	2.37	7.86	8.49
2.	10	27.70	8.65	8.22
3.	15	81.10	18.14	7.11
4.	20	83.24	40.31	12.37
5.	25	83.35	48.18	16.60
6.	30	83.79	65.21	9.48

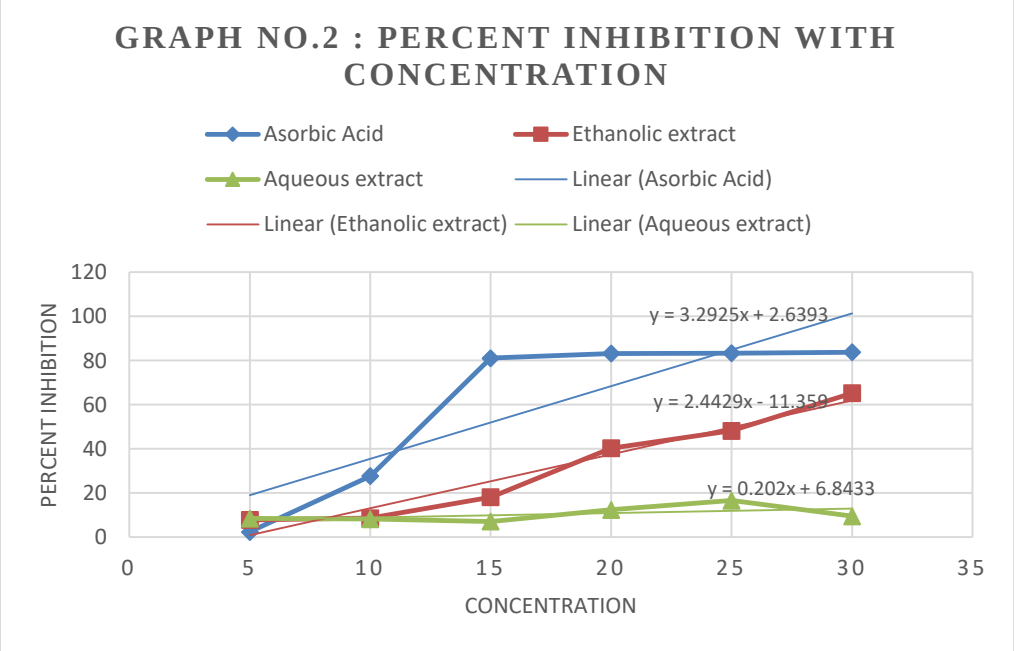
Table-3: Antibacterial activities of *Coriander sativum* leaf

Test Organisms	Inhibition zone diameter (mm)	
	Ethanolic extract	Aqueous extract
Escherichia coli	11	16
Salmonella typhi	09	10
Staphylococcus aureus	18	12
Bacillus subtilis	10	14

The antibacterial activity of ethanolic extract and aqueous extract of C.sativum is shown in Table-3. The ethanolic extract was more effective against Staphylococcus aureus while aqueous extract was more effective against E.coli. The antibacterial activity of ethanolic and aqueous extract was least against *Salmonella typhi.*

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The result of the phytochemical test carried out on the ethanolic extracts and aqueous extract was recorded as shown in Table-4. Ethanolic extract of coriander leaves showed high concentration of alkaloids, flavonoids, Tannin, phenolic compounds and terpenoids while in aqueous extract alkaloids, flavonoids, Tannin, phenolic compounds and terpenoids was absent. Ethanolic and

aqueous extract was containing low concentration of carbohydrates, protein, glycosides and saponin.

Table-4 : Phytochemical analysis of *Coriander sativum* leaves

Sr.No.	Photochemical Test	Ethanolic extract	Aqueous extract
1	Test for Carbohydrates A)Molisch Test	+	+
2	Tests for Protein &Amino acids A) Ninhydrin Test	+	+
3	Test for Alkaloids A) Mayer's Reagent Test	++	-
4	Test for Flavonoids A) Alkaline Regent Test	++	-
5	Test for Glycosides A) Borntrager Test	+	+
6	Test for Tannin and Phenolic compounds A) Ferric chloride Test	++	-
7	Test for Saponin A) Foam test	+	+
8	Tests for Terpenoids A) Salkowski's test	++	-

++ = High concentration, + = Low concentration, - = Absent

DISCUSSION

The present study shows that ethanolic leaf extract of *C.sativum*has better antioxidant potential by DPPH scavenging method. Numerous in vitro models exist for the assay of the antioxidant capacity of plant extracts. DPPH assay remains the most acceptable model owing to its simplicity and reproducibility. This model involves the reduction of DPPH radical by the

antioxidant compounds which are evident from the colour change from purple to yellow [11]. The ethanolic and aqueous extract of coriander leaf shows antimicrobial activity against both Gram positive as well as Gram negative bacteria. Medicinal plants are known to produce certain bioactive molecules which are responsible for their antimicrobial properties [12]. The ethanolic extract shows high concentration of alkaloids, flavonoids, Tannin, phenolic compounds and terpenoids. These secondary metabolites are absent in aqueous extract. The phenolic and flavonoid content may contribute directly to the antioxidant activity [13].

CONCLUSION

The ethanolic extract of coriander in Osmanabad region selected for present study contains strong antioxidant potential. The antibacterial activity of different extract of *C.sativum* is observed significant. The phytochemical analysis shows the presence of active constituents in ethanolic extract. These active constituents may increases the antioxidant potential of ethanolic extract of *Coriander sativum.*

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