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EXPLORING THE PHYTOCHEMICAL AND ANTIOXIDANT POTENTIAL OF *ALLIUM SATIVUM* L. BULBS FOR MEDICINAL APPLICATIONS

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Abstract

This study examines the phytochemical and antioxidant properties of Allium sativum (garlic) bulbs, highlighting their potential for plant-based medicine. Qualitative analysis of ethanolic extracts identified key bioactive compounds, including alkaloids, flavonoids, coumarins, terpenoids, phenolics, and proteins, while saponins and glycosides were absent. The DPPH radical scavenging assay revealed significant antioxidant activity, with the ethanolic extract showing 93.21% scavenging, surpassing ascorbic acid. These findings underscore garlic's therapeutic potential and the importance of medicinal plants in developing new therapeutic agents.

Key words: Antioxidant properties, Bioactive, Metabolized, Coumarins

Introduction

The Human beings have always depended on nature to provide for their basic necessities, including food, clothes, shelter, transportation, fertilizer, flavors, scents, and—most importantly—medicines. Plants, in particular, have played a pivotal role in providing life-saving medications, forming the foundation of traditional medical systems that have persisted for millennia and continue to offer innovative treatments today (Sharma and Dubey, 2011). Many modern drugs have been derived from natural sources, often inspired by the traditional uses of these plants in healing practices (Farombi, 2003). With approximately 80% of the global population still relying on plant-based medicine for primary healthcare (Owolabi *et al.*, 2007), herbal remedies remain an essential component of global health systems. The therapeutic properties of plants were likely discovered through careful observation and trial-and-error, leading to the development of well-defined herbal pharmacopoeias that have become the cornerstone of traditional medicine. Given their broad biological benefits, high safety margins, and relatively low costs, herbal medicines continue to be in high demand and are widely used in both developed and developing countries (Kamboj, 2000). Investigating the phytochemical constituents in plants or plant products is important for research across various fields. Numerous studies have been conducted and published on this topic in the past (Mikail, 2010, Senthamil *et al.,* 2016).

This study focuses on *Allium sativum* L, commonly known as garlic, due to its widespread cultivation, nutritional benefits, and significant role in both culinary and medicinal applications, making it an ideal subject for further investigation. Garlic, *Allium sativum* L. is a member of the Alliaceae family, has been widely recognized as a valuable spice and a popular remedy for various ailments and physiological disorders. The name garlic may have originated from the Celtic word 'all' meaning pungent. Cultivated practically throughout the world, garlic appears to have originated in central Asia and then spread to China, the Near East, and the Mediterranean region before moving west to Central and Southern Europe, Northern Africa (Egypt) and Mexico. The active constituents are several

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complex sulfur-containing compounds that are rapidly absorbed, transformed and metabolized. Pooled data from numerous randomized trials suggest that garlic lowers total cholesterol concentrations by approximately 10% and favorably alters HDL/LDL ratios. Randomized trials also support garlic's effectiveness as a mild antihypertensive which lowers blood pressure by 5-7%. Garlic also inhibits platelet aggregation and enhances fibrinolytic activity, reducing clots on damaged endothelium. In vitro data suggest antibacterial effects, but these have not been evaluated in controlled trials in humans.

Materials and Methods

Sample collection

For the present study, bulbs of *Allium sativum* were collected from a local market near Sree Narayana College, Chempazhanthy, Thiruvananthapuram, Kerala, India. The bulbs were carefully washed under running water to remove any impurities, and the outer layers of garlic were peeled off before being washed again.

Preparation of plant Extract

i)Aqueous extract

The 100 mg bulbs were cut the small pieces and was made into a crude paste. The paste was soaked in 100 ml of sterile distilled water for 72 hours at 4oC and it was then filtered thrice using a sterile muslin cloth. The filtrate was poured into a beaker and concentrated on a water bath at 100°c to obtain semi-solid residues and they aqueous extract was weighted and this was immediately subjected to preliminary phyto-chemical and antioxidant analysis using standard method.

ii) Ethanolic extract

After cleaning 100 mg of garlic as described earlier they were made into a paste was soaked in 100ml of ethanol for 72 hours at room temperature than it were filtered using sterile muslin cloth and the filtrate was poured into a beaker and concentrated on a water bath at 70-80oC to obtain semi-solid residue. The weight of the yield was noted and this was subjected in preliminary phyto-chemical and antioxidant analysis using standard method.

iii) Chloroform extract

After making a paste of 100 mg garlic of different varieties as described earlier, they were separately soaked in 100 ml of chloroform for 72 hours at room temperature the filtered using sterile muslin cloth and filtrate was concentrated in a beaker at 60-62°C to obtain semi-solid residue. This have weighted and subjected to preliminary phyto-chemical and antioxidant analysis.(Prakash. *et al.,* 2016)

iv) Petroleum ether extract

Following the earlier procedure, garlic bulb of different varieties was prepared and soaked in 100ml of petroleum ether for 72 hours at room temperature. It was filtered and the filtrate we concentrated at 40-60oc. The extract was weighted and subjected to preliminary phyto-chemical and antioxidant analysis standard method.

Phytochemical Investigations

Preliminary phytochemical analysis of crude extract of *H.hispidimus* Griff., were carried out according to the standard procedures. In accordance with the established standard procedures, a series of phytochemical tests were conducted to detect the presence of various compounds, including alkaloids, flavonoids, carbohydrates, coumarins, saponins, glycosides, proteins, phenols, and tannins, to evaluate the diverse chemical constituents of the plant materials used in the study.

Antioxidant assay of experimental plant

DPPH free radical Scavenging Assay

Free radical scavenging potential of ethanolic extract of experimental plant sample was determined by 2, 2-diphenyl-1-picrylhydrazyl (DPPH) assay. DPPH is a stable free radical. On accepting hydrogen from a corresponding donor, its solutions lose the characteristic deep purple (λmax515-517 nm) color. For getting different concentrations, the sample was diluted with 0.2 ml of extract solution in ethanol (95%) at

different concentration (20,40,80,160) was added to 8ml of 0.004% (w/v) stok solutions of DPPH in ethanol (95%0. The scaveging activity on the DPPH radical was determined by measuring the absorbance at 517 nm until the reaction reached the steady state, using a spectrophotometer (Mini Spec Sl-207, Elico Limted India). As a positive control, ascorbic acid (150 µg/ml) was used. All determinations were performed in triplicate.

The percentage of DPPH radical scavenging activity (S %) was calculated by using the following equation.

S % = (A control – A sample)/ A control) x 100
A control is the absorbance of the blank control (containing all reagents except the extract solution). A sample is the absorbance of the test sample.

Results

Phytochemical analysis refers to the study of plant chemicals, and plant-based medicine offers great potential as an accessible source for treating various diseases. In this study, the phytochemical properties of *Allium sativum* (garlic) bulbs were analyzed. The qualitative phytochemical analysis revealed the presence of a variety of bioactive compounds in the plants. The preliminary screening of the ethanolic extracts of *Allium sativum* demonstrated significant results. Out of the eleven tests conducted, the presence of seven key phytochemical compounds: alkaloids, flavonoids, carbohydrates, coumarins, terpenoids, phenolic compounds, and proteins. Table 1 shows that phytochemical characters of *Allium sativum* from that Coumarins, alkaloids and Flavonoids shows positive results followed by saponin and glycosides are showed negative result.

In vitro Antioxidant studies

DPPH radical scavenging activity

Antioxidants are substances that prevent oxidation and have the ability to neutralize potentially harmful oxidizing agents in living organisms. Many phytochemicals found in plants help reduce or prevent oxidative damage to human cells, which can even lead to cancer. Understanding the antioxidant properties of plants and the specific phytochemicals responsible is crucial. In our study, the antioxidant activities of *Allium sativum* (garlic) were assessed using the DPPH radical scavenging method, which produced significant results.

DPPH (1,1-diphenyl-2-picrylhydrazyl) is commonly used to evaluate free radical scavenging due to its simplicity and reliability. In this study, ethanolic extracts of *Allium sativum* demonstrated effective scavenging activity against DPPH radicals. Among the different extracts tested, ethanolic extracts showed the highest scavenging activity. The antioxidant potential of these extracts was compared to that of ascorbic acid, the standard reference. While ascorbic acid exhibited strong scavenging activity, the ethanolic extracts of *Allium sativum* also showed notable effects. Notably, *A. sativum* exhibited the highest antioxidant activity, with an ethanolic extract scavenging 93.21% of the radicals, (Table 2). This confirms that the extracts have significant antioxidant potential, with *A. sativum* showing the most potent activity.

Table 1. Qualitative phytochemical analysis of *Allium sativum L.*

SL No.	Phytochemicals	PE	CHL	ETH	DW
1	Alkaloids	+	+	+	+
2	Flavonoids	+	+	+	+
3	Carbohydrates	-	+	+	+
4	Coumarins	+	+	+	+
5	Glycosides	-	-	-	-
6	Saponins	-	-	-	-
7	Proteins	-	-	+	-
8	Phenols	+	+	+	-
9	Terpenoids	+	-	+	-
"+"present;"-" Absent; PE- petroleum ether; CHL- Chloroform; ETH- Ethanol; DW- Distilled water					

Discussion

Phytochemicals are naturally occurring compounds found in plants. Medicinal plants are particularly rich in bioactive phytochemicals

Table 2. DPPH free radical scavenging activity of different exracts of *Allium sativum* L.

Sl. No.	Concentration(µg/ml)	97.08±0.18			
1	Control Ascorbicacid(100)				
		PE	CHL	ETH	DW
2	20	1.04±0.02	9.08±0.02	19.11±0.02	12.46±0.02
3	40	19.23±0.004	25.19±0.004	48.03±0.004	28.83±0.004
4	80	38.74±0.01	45.43±0.01	79.54±0.01	55.3±0.01
5	100	55.66±0.01	60.32±0.01	93.21±0.01	79.12±0.01

and other bio-nutrients, which have been shown to play a crucial role in preventing various diseases. These compounds include a wide range of secondary metabolites found in plants, each with distinct structures and functions. Traditional herbal medicines have garnered significant interest as a source of new therapeutic drugs. These plants have been carefully selected and used by humans over generations, not only for their healing properties but also as a valuable source of chemical constituents that could potentially be developed into drugs with targeted therapeutic effects.

The qualitative phytochemical analysis plays a major role in identifying the therapeutically active compounds present in medicinal plants. Understanding these chemical constituents is essential for evaluating their pharmacological properties. The therapeutic effects of traditionally used medicinal plants are largely attributed to the phytochemicals they contain, which are primarily secondary metabolites such as alkaloids, tannins, and Phytochemical screening is crucial for discovering and developing novel therapeutic agents with enhanced efficacy. Among the various phyto constituents, phenolic compounds are one of the most abundant and widespread groups of plant metabolites (Singh *et al.*, 2007). A comparative preliminary phytochemical analysis of different fractions and crude extracts of *A. sativum* revealed a rich presence of beneficial bioactive constituents.

The phytochemical screening in the present study, has revealed the presence of terpenoids, steroids, glycosides, flavonoids, tannins, carbohydrates and coumarins in the bulbs extract. Further the presence of different phytoconstituents in the four different extracts may be responsible for the therapeutic properties of garlic. Comparatively ethanolic extracts of *A. sativum* contain more phyto constituents. Similarly reported that garlic contain bioactive compounds such as organosulfur compounds, saponins, phenolic acids, flavonoids and polysaccharides (Szychowski *et al*.,2018; Bradley *et al*., 2016)

Ascorbic acid, the standard anti-oxidant used in the present study, act as a chain breaking scavenging agent that impairs the formation of free radicals in the process of intracellular substances formation throughout the body, including collagen bone matrix and tooth Several methods have been developed to estimate the antioxidant capacity of different plant materials. Hence in the present study, different extract of *A. sativum* were investigated for their antioxidant activity using DPPH radical scavenging assay.

The DPPH radical scavenging assay is commonly used to assess the antioxidant potential of natural products due to its stability in the radical form, along with the accuracy and simplicity of the method. In its radical form, DPPH decolorizes when reduced by an antioxidant compound or radical species, transforming into a stable diamagnetic

molecule. This results in a color change from purple to yellow, which indicates the hydrogen-donating ability of the tested samples (Oktay *et al.*, 2003; Lee *et al.*, 2012).

In the present study, the ethanolic extracts of *A. sativum* (garlic) exhibited strong DPPH radical scavenging activities, with *A. sativum* showing a highest antioxidant activity. These findings suggest that the plant extracts, through their proton-donating ability, could act as effective free radical inhibitors or scavengers, potentially serving as primary antioxidants. Several studies have been conducted on Allium species in relation to their medicinal properties (Prakash *e t al.*, 2007, , Bagheri *et al*, 2011).

Conclusion

This study underscores the notable phytochemical and antioxidant properties of *Allium sativum* (garlic) bulbs, highlighting their potential as a valuable source of bioactive compounds for medicinal applications. The qualitative phytochemical analysis identified several key compounds, including alkaloids, flavonoids, coumarins, and phenolic compounds, which are likely responsible for garlic's therapeutic effects. The antioxidant activity, assessed using the DPPH radical scavenging assay, revealed that ethanolic garlic extracts exhibit strong free radical scavenging activity, achieving an impressive 93.21% scavenging rate, surpassing that of ascorbic acid. These results emphasize garlic's potential as a potent natural antioxidant and therapeutic agent, supporting its use in the development of plant-based medicines. With its rich phytochemical profile, garlic offers promising opportunities for further research, particularly in addressing oxidative stress-related health conditions.

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