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Qualitative Phytochemical analysis & determination of antimicrobial activity of *Zingiber officinale*

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ABSTRACT

Numerous nations consume Zingiber officinale on a daily basis. Ginger (Zingiber officinale) is a popular tropical & subtropical shrub medicinal plant. The current study sought to assess the phytochemical composition and antimicrobial properties of Z.officinale extract. Qualitative evaluation of phytochemical tests revealed the existence of saponins, alkaloids, flavonoids, and steroids in ginger extracts in methanol and acetone, as well as the absence of carbohydrate, anthraquinones, and tannin in ginger extracts in methanol and acetone. The antimicrobial properties of Z.officinale extracts in solvents (aqueous, methanol) towards bacteria as well as fungus was investigated. When compared to other concentrations, the highest zone of inhibition was seen in 100µl concentrations of aqueous and methanolic extracts of Z.officinale towards Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Aspergillus flavus, A.niger, and Penicillium sp. This study discovered that the plant possesses excellent metabolites, proving that it has medicinal properties and may be used in daily life.

KEY-WORDS

Phytochemical, *Zingiber officinale*, Anti-microbial, Flavonoid, *Staphylococcus aureus*

INTRODUCTION

Therapeutic plants are plants that have active compounds that can be used to heal sickness or relieve suffering.[1] Conventional drugs and therapeutic plants are frequently used as therapeutic agents in most underdeveloped nations to maintain good health.[2] The modern pharmacopoeia still includes at least 25% of medications originated from plants, as well as numerous synthetic equivalents based on prototype chemicals extracted from plants. The growing expenses of prescription pharmaceuticals in the maintenance of human health and wellbeing, as well as the bioprospecting of novel plant-derived drugs, have fueled interest in medicinal plants as a re-emerging health assistance.[3] The increased

acknowledgment of therapeutic plants is attributable to a variety of factors, including growing confidence in herbal treatment.[4] In addition, a growing dependence on the usage of plants for medicinal purposes in industrialized countries may be traced back to the extraction and production of medications and chemotherapy drugs from these plants, as well as traditional herbal treatments.[5] Plants' therapeutic capabilities might be based on the antioxidant, antimicrobial, and antipyretic actions of their phytochemicals.[6,7] As stated by the World Health Organization, plants with medicinal properties are the best source of a wide range of medications. As a result, such plants should be studied in order to properly determine their qualities, safety, and efficacy.[8]

India is among of the nations that heavily relies on herbal medicine to suit its healthcare demands. Plants in the Zingiberaceae family are recognized for their preservation and therapeutic properties.[9] A variety of herbs from this family are utilized in traditional medicine.[10] Ginger is widely used as a natural remedy across India, and it originated in India and China, where it has been widely used in cooking for over 4000 years. Ginger is well-known because of its spicy and pungent flavor [11]. Zingiber is derived from the Greek zingiberis, which is obtained from the Sanskrit name of the spice, singabera; the Latin term, Zingiber, meaning fashioned like a horn and refers to the roots, which resemble deer antlers. The herb is referred to as Sringavera in Sanskrit [12]. Ginger is a type of rhizome from the Zingiberaceae family, which is a herbaceous perennial plant native to Southeast Asia. It consists of 47 genera & 1400 species [13]. The thick spiky rhizomes of this plant are used to make commercial ginger. Different divisions of the rhizome are known as "hands" because they branch with thick thumb-like protrusions. Rhizomes are 715cm long, 11.5cm wide, and compressed laterally. The branches grow obliquely from the rhizome and are around 13cm long, terminating in depress scars or underdeveloped buds. The exterior surface is buff in color and striated or fibrous lengthwise. The broken surface has a small cortex, a well-defined endodermis, and a broad stelle. [14]This complex combination of pharmacological compounds contains approximately a hundred recognized components, such as gingerols, betacarotene, capsaicin, caffeic acid, curcumin, and salicylate [15]. The strong flavor of ginger is due to non-volatile phenylpropanoid-derived compounds known as gingerols and shogaols. Gingerols are converted to shogaols when dried or heated [16]. The typical usage of fresh ginger root as a flavoring ingredient in India is estimated to be 8-10 grams daily. [17] Ginger may be included in a wide range of dishes. They may be boiled in boiling water to produce ginger tea, which is commonly sweetened with honey and can also include sliced orange or lemon fruit. Ginger root juice is quite potent and is usually utilized as a spice to flavor foods like shellfish, mutton, appetizers, or stew. Ginger powder (dry ginger roots

pulverized) is widely used to season ginger bread and other foods. Ginger flavoring is commonly used in biscuits, crackers, and cakes, as well as ginger ale, a sweet, fizzy, nonalcoholic drinks, gingerbread, ginger snaps, ginger cake, and ginger cookies [18]. A number of investigations have been conducted with the goal of discovering novel sources of bioactive chemicals with antioxidant and/or antibacterial characteristics from natural products and assessing their suitable utilization in meals such as ginger [19]. Ayurvedic practitioners employ ginger as a carminative, diuretic, antispasmodic, expectorant, periphery circulatory stimulant, astringent, hunger stimulant, antiinflammatory agent, diuretic medication, and gastrointestinal aid [20]. It's increasingly being regarded as a superior therapy for nausea and vomiting. It also has an effect on pregnant morning sickness and functions as an all-natural pain reliever and anti-inflammatory in the management of osteoarthritis & rheumatic gout. It also aids in the treatment of ulcers and the prevention of cardiac events such as strokes [21]. Saponins, tannins, flavonoids, alkaloids, anthraquinones, cardiac glycosides, and cyanogenic glycosides are the most regularly observed secondary metabolites of this plant. Thus, the goal of this work was to evaluate the phytochemical contents of Z. officinale extracts and assess their antibacterial efficacy against various clinical bacteria isolates.

MATERIALS AND METHODS

Sample Collection & extraction

In the month of August 2023, ginger was harvested from the local market of Jamuhar, Sasaram (Rohtas), Bihar, India. The fresh ginger was then wrapped in a plastic bag and taken to home, where it was stored at a comfortable temperature until processing. The obtained material was extracted in order to do qualitative analysis and research on the antimicrobial properties of ginger.

Micro-organisms Collection

Staph. Aureus, E. coli and Pseudomonas aeroginosa selectrd and collected as test bacteria.*Aspergillus flavus, A.niger, Penicillium* sp. selected and collected as test fungi and stored at favourable temperature.

Qualitative Analysis of Phytochemicals [22,23]

The plant extracts were screened for the presence of the phytochemicals like Alkaloids, Tannin, Flavonoids, Carbohydrates, Steroids, Saponin and protein etc.

1. **Test for Alkaloids**

Hager's Test: In 2 ml of 1% aqueous extract of ginger in acetone and methanol, add 2 ml of Hager's reagent. Yellow precipitates indicate the presence of alkaloids.

2. **Test for Flavonoids**

In 2 ml of 1% aqueous extract of ginger in acetone and methanol add 1 ml of 2N NaOH. Yellow color indicates the presence of flavonoid.

3. **Test for Tannin**

1 ml of 1% aqueous extract of ginger in acetone and methanol mixed with 2 ml of 5% FeCl3. Dark bluish/Greenish black color indicates the presence of the Tannin.

4. **Test for Carbohydrates**

2 ml of 1% aqueous extract of ginger in acetone and methanol was mixed with the 1ml of Molisch reagent. Few drops of concentrated sulfuric acid were slowly added from the side wall. Resulted solution was shaken carefully. Violet ring at the interface of the two liquid indicates presence of carbohydrates.

5. **Test for Steroid**

5 ml of chloroform added in the 1 ml of 1% aqueous extract of ginger in acetone and methanol. After that 5 ml of H2SO4 was added drop by drop in solution. Upper layer turns red and H2SO4 turns yellow-green.

6. **Test for Saponin**

2 ml of 1% aqueous extract of ginger in acetone and methanol was mixed with 2 ml of distilled water. The mixture was shaken for 15 min in a test tube. Formation of 1 cm foam layer indicates the presence of saponin.

7. **Test for Anthraquinones**

A Borntranger test , was performed whereby one millilitre of the plant filtrate was shaken with 10ml of benzene; the mixture was filtered and 5 ml of 10% (v/v) ammonia solutions was added shaken. A pinkish or violet solution indicated a positive test.

Screening for antimicrobial activity[24]

The antimicrobial action was evaluated employing 24 hour bacterial cultures (Escherichia coli, Pseudomonas aeruginosa, and Staphylococcus aureus) and 48 hour fungal cultures (Aspergillus flavus, A. niger, and Penicillium sp.) in different solvents of methanol and aqueous extracts of G. officinale. Individual bacterial and fungal strains were inoculated and maintained on nutrient agar and potato dextrose agar plates. A sterile cork borer was used to create a 6mm diameter borehole. In the well, different concentrations (50, 75, and 100µl) of various solvents were added separately with the extracts.

The plates were incubated at 37 2°C for 24 hours and antifungal assay plates were incubated at 28 2°C for 48 hours, with data recorded every 24 hours.

RESULT & DISCUSSION

This current phytochemical study revealed the existence of saponins, alkaloids, flavonoids, and steroids in ginger extracts in methanol and acetone, as well as the absence of carbohydrate, anthraquinones, and tannin in ginger extracts in methanol and acetone (**Table-1**).Alkaloids are well-known for their anesthetic, cardioprotective, and anti-inflammatory effects. Among the well-known alkaloids used in therapeutic settings include morphine, strychnine, quinine, ephedrine, and nicotine. Flavonoids have been shown to exhibit a variety of biological activities, including anti-microbial properties, cytotoxic, anti-inflammatory, and anticancer qualities; however, the ability to operate as strong antioxidants is the flavonoids' most famous attribute. Saponins have been proven to destroy protozoan organism as well as mollusks, impair protein digestion, and inhibit the stomach from accumulating vitamins and minerals as antioxidants, to produce hypoglycemia, and to have antifungal and antiviral effects. Steroids have analgesic qualities and stimulate the central nervous system.

The aqueous as well as methanolic extracts of Z. officinale were tested for antibacterial and antifungal activity against bacteria (Staphylococcus aureus, Escherichia coli, and Pseudomonas aeruginosa) and fungi (Aspergillus flavus, A. niger, and Penicillium sp). **Table 2** shows the antibacterial effect of Zingiber officinale, while **Table 3** shows the antifungal effect of Zingiber officinale. The current investigation found that Zingiber officinale had significant antibacterial properties against the examined microorganisms.

Table 1: Qualitative Phytochemical analysis of *Zingiber officinale* extract

S.N.	Name of compound	Ginger in acetone	Ginger in methanol
1.	Alkaloids	+	+
2.	Flavonoids	+	+
3.	Tannin	-	-
4.	Carbohydrates	-	-
5.	Steroid	+	+
6.	Saponin	+	+
7.	Anthraquinones	-	-

(-)means absent, (+) means present

Table 2: Effect of antibacterial activity of *Zingiber officinale* extract against bacteria

Bacteria	Zone of inhibition (mm)					
	Aqueous			Methanol		
	50µl	75µl	100µl	50µl	75µl	100µl
Staph. Aureus	14.3±0.21	15.0±0.40	16.4±0.83	15.7±0.35	16.3±0.21	13.3±0.54
E. coli	08.3±0.21	13.3±0.49	12.0±0.77	12.3±0.34	13.7±0.32	14.5±0.99
Pseudomonas aeroginosa	07.3±0.21	07.3±0.21	07.0±0.20	11.0±0.47	16.0±0.77	15.3±0.88

The values are expressed in terms of (Mean ± Standard deviation)

Table 3: Effect of anti-fungal activity of *Zingiber officinale* extract against fungi

Bacteria	Zone of inhibition (mm)					
	Aqueous			Methanol		
	50µl	75µl	100µl	50µl	75µl	100µl
Aspergillus flavus	08.6±0.65	08.3±0.54	13.0±0.20	09.0±0.77	07.7±0.29	14.5±0.20
A.niger	09.0±0.77	10.3±0.49	07.6±0.99	07.0±0.20	11.5±0.45	15.3±0.36
Penicillium sp.	12.0±0.77	13.3±0.21	14.7±0.65	06.6±0.36	12.3±0.34	13.2±0.32

The values are expressed in terms of (Mean ± Standard deviation)

DECLARATIONS

Conflict of Interest

There is no conflict of interest, The authors alone are responsible for the content and writing of the paper.

Ethical approval

Not applicable

Consent to participate

Not applicable

Availability of data and materials

Not applicable

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