Pharmacognostical Standardization of *Zingiber officinale* Roscoe plant family *Zingiberaceae*

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Abstract
The primary standardization studies are the basic and gives evidence of the raw drugs. It includes crude drug macroscopic, microscopic as well as identity of herbal drugs. Quality controls of herbal drugs remain most imperative and effective. In this current study we studied standardization parameter such as morphological, microscopically, physicochemical, and phytochemical evaluation of *zingiber officinale roscoe* family *zingiberaceae*. The study revealed that rhizomes were found light brown to reddish brown with characteristic aroma, astringent taste, hard texture and splintery fracture. T.S. of the rhizome displayed an Orangish/reddish-brown colour throughout. Rhizome powder was observed under microscope, it shows various elements such as fibers, oil globules, starch grains, and calcium oxalate crystals and yellow coloured lignified fibres. The heavy metal analysis was determined the contents of Pb-0.584, As-0.021, Cu-0.375, Cd-0.020, Hg-0.247, Mg-0.421, Zn-0.033 and Fe-1.296 in the powdered sample. The results of the study showed that the ethanol extract from the specified plants included alkaloids, saponins, amino acids, tannin, flavonoids, carbohydrates, cardiac glycosides, phenol, protein, sterol, and tri-terpenoid. **Keywords:**
Zingiber officnale, macrosocpic, microscopic, phytochemical, physiological,

Introduction

Since ancient times medicinal and aromatic plants have known for their nutritional, ethanomedicinal and therapeutic value. In last few years the main focus of scientific researcher's was the use of plant derived molecules. Plant contains a wide range of bioactive molecules that show multiple effects (1). Ginger is a culinary plant having biological name *Zingiber offcinale* belonging to family *zingiberaceae*. It is a monocotyledonous plant and their rhizomes also known as root contain secondary metabolites. It is a well-known spices in world used in foods and cultivated in a large number of countries due to its medicinal properties. Ginger is found in the tropical and subtropical countries. It is now cultivated with a purpose as a commercial crop in America, Africa, Australia, China, India, as well as in South East Asia. Ginger is cultivated during April-May month and collected when the leaves

appear yellow colour after planting 7-8 months in December-January. Cultivation is done by directly digging of rhizomes. After collection ginger is properly washed and then dries. It is used in the form of fresh paste as spices in foods, dried form of powder, and candy used for flavouring agent in tea, and also used as herbal remedies (2-5). Traditionally ginger is used for the cure of asthma, appetite stimulant, constipation, cold and flu, digestion, diarrhea, dyspepsia, inflammation, nausea, pain, and vomiting (6-7).It is a characterstics odour and flavour because it contains shagols, zingerone and gingerols (5). Ginger contains 1 to 2% essential oils. Essenstial oil of ginger comprises a mixture of chemical constituents such as sesquiterpenes and monoterpenes (ar-curcumene, α-zingiberene, β-bisabolene, β-sesquiphellandrene, borneol, camphene, citral, cineole, citronellol, limonene, linalool, and phellandrene, zingiberol and zingiberenol also contains some aliphatic aldehydes and alcohols (5). Volatile oil composition varies to various factors such geographical region, temperature, harvesting time, post harvesting treatment, dry condition (8-9). Previous study on *zingiber officinale* indicates that ginger have antioxidant, anti-inflammatory, anti-apoptotic, antiemetic, anti-hyperglycemic, anti-hyperlipidemic etc effects (10, 25)

The primary standardization studies are the basic and gives evidence of the raw drugs. It includes crude drug macroscopic, microscopic as well as identity of herbal drugs. Quality controls of herbal drugs remain most imperative and effective. In this current study we studied standardization parameter such as morphological, microscopical, physicochemical, and phytochemical evaluation (11).

MATERIAL AND METHODS

Plant collection and Authentication

The plant was collected from local market Rohtak, Haryana and a voucher specimen was authenticated by Dr. Kailash C. Bhatt, Principal Scientist ICAR- National Bureau of Plant Genetic Resources, Division of Plant Exploration and Germplasm Collection, National Herbarium of Cultivated Plants (NHCP), Pusa Campus, New Delhi 110012, vide reference no, AC-261/2024 dated 19.06.2024. The plant rhizome extract and oil were selected for the proposed work. The plant was identified as *Zingiber officinale* Roscoe Family Zingiberaceae.

Pharmacogonostical studies

The *Zingiber officinale* Roscoe r was analyzed for pharmacognostical studies like morphology, microscopic evaluation, total ash, water-soluble ash value, acid-insoluble value,

foreign organic matter, and moisture content and heavy metal analysis using standard protocols. (12-13).

Morphological Studies

It involves evaluation of plant materials using the sensory organs. It is an efficient method to determine the identification and purity of a raw material, therefore ensuring its quality. The morphology includes the shape, size, color, aroma, taste or flavour, and fracture of the plant material (11,14).

Microscopical Study

Fresh sample was taken and observed the features of their internal surface. Additionally, desiccated specimens from the herbarium were utilized for comparative analyses. The specimens for microscopy examinations were mounted on different staining agent. In Powder Microscopy fine dried powder was taken instead of fresh plant part (14-16).

Other physiochemical studies

i) Determination of Ash Contents

The Total Ash value is incinerated carbon free ash of plant material. Physiological ash is direct plant parts ash and non-physiological ash is from extraneous matter like sand or soil. About 2g powdered plant material was placed in a silica crucible and then temperature was slowly increased to 500-600°C until the material turned white and free of carbon. The crucible was then allowed to cool down, and the weight of the material was recorded. Total ash content was calculated by formula:

$$\% \text{ Total Ash} = \left(\frac{\text{Weight of } Ash}{\text{Amount of sample taken}}\right) \times 100$$

Acid insoluble ash, residual matter was obtained by getting boiling with dilute hydrochloric acid and igniting the insoluble substance. Crucible containing total ash treated with 25 ml of HCl and covers it. Boil it for about 5 min. dried the sample on hotplate by ignition to get constant weight. Acid insoluble ash was calculated by formula:

$$\% \text{ Acid insoluble ash} = \left(\frac{\text{Residue weight}}{Total\ Ash}\right) \times 100$$

Water-soluble ash was assessed by calculating the difference between the total ash content and the residue generated after treating it with water, including sand and siliceous ground matter. Add water to the crucible with total ash and boiled. The insoluble matter was collected and washed with hot water. The water-soluble ash content was calculated, keeping in mind that ignition temperature should not exceed 450°C (17).

ii) Determination of Foreign organic matter (FOM)

In this study carefully inspected the presence of foreign matter and also observed discoloration that could damage the plant components. The plant sample about 2g was weight and analyzed for foreign organic matter. The FOM was calculated by using formula:

$$\% \text{ FOM } = \left(\frac{\text{Organic matter weight}}{\text{Total weight}}\right) \times 100$$

iii) Determination of Moisture content

In this study plant sample was carefully dried to determine their moisture content. About 2gm powdered plant sample was weighed and dried to a constant condition. The sample cooled with hot air oven at low temperature. The dry weight of the plant sample was recorded again after cooling. Finally, the moisture content was calculated using formula:

$$\% \text{ Moisture content} = \left(\frac{A - B}{B}\right) \times 100$$

Where, W – weight of the moisture in the given sample in %

A – Wet sample weight in grams; B – Dry sample weight in grams

iv) Detection of the presence of microbial pathogens

Tests have been developed to ascertain the quantity of viable aerobic bacteria and to identify the presence of specific microbial species in medicinal compounds. The term 'growth' is employed to denote the existence and presumed multiplication of live microorganisms.

a) *Escherichia coli*

Escherichia coli is the prevailing coliform, characterized by its gram-negative rod shape and facultative anaerobic nature. The culture media employed in this study consisted of Mac Conkey agar medium, Mac Conkey broth medium, and Agar peptone broth.

i) Primary test

Approximately 10 grams of the sample were aseptically transferred into a medium consisting of 100 ml of lactose broth/soybean casein digest broth. The mixture was then incubated at a temperature of 37°C for a duration of 24 hours. Following the incubation period, the flask was assessed for the presence of growth and gently agitated through mild shaking, but only if growth was detected. The 1 ml of the enrichment culture was transferred by pipette into tubes containing 10 ml of Mac Conkey's broth, each equipped with a Durham's tube. The tubes were then incubated at a temperature of 35°C for a duration of 24 hours. The secondary test was conducted just in cases when the contents exhibited acidity or gas. Conversely, a loop

containing an enrichment culture was streaked separately onto the surface of Mac Conkey's agar medium. The plates were subjected to incubation at a temperature of 37°C for duration of 24 h. The presence of *E. coli* was deemed to be present alone if a pinkish red hue appeared.

ii) Secondary test

The suspicious colonies were transferred to the surface of Levine eosin-methylene blue agar medium using an inoculation loop on an individual basis. The plates were incubated in an inverted posture at a temperature of 37°C for duration of 24-48 h.

iii) Confirmatory test

Aseptically, a loop was used to transfer a full culture obtained from Levine eosin-methylene blue agar to 5 ml of Tryptophan broth. The broth tubes were then incubated in a water bath at a temperature of 43.5°C for a duration of 24 hours. Following the incubation period, the broth tubes were analyzed to determine the presence of indole.

Indole test

Approximately 0.5 ml of Kovac's reagent were introduced into peptone broth and vigorously agitated. The mixture was afterwards left undisturbed for duration of one minute. The observation of a red colour ring forming within the reagent layer can be interpreted as evidence for the presence of indole, which is associated with the presence of *Escherichia coli*.

b) *Salmonella typhi*

The culture media employed in this study included Selenite F broth, brilliant green agar medium (BGA), bismuth sulphite agar medium (BSA), xylose-lysine-desoxycholate agar medium (XLD), deoxycholate citrate (DCC) agar, and urea broth.

i) Primary test

A volume of 1.0 ml of the enrichment culture was introduced into tubes containing 10 ml of Selenite F broth. The tubes were then incubated at a temperature of 35°C for duration of 48 h, during which they were monitored for the occurrence of turbidity. The secondary test was conducted alone in the presence of turbidity.

ii) Secondary test

The content of the selenite broth medium was extracted and afterwards streaked across the surfaces of BGA, BSA, XLD, and DCC agar plates. The samples were subjected to incubated at 35°C for duration of 24 h. Subsequently, the resulting colonies were carefully examined and compared

iii) Confirmatory test

A loopful of culture was obtained from the plates displaying positive colony features. This culture was then streaked over the surface of a medium known as triple sugar-iron agar. Additionally, the same culture was injected into a tube containing urea broth. The tubes were subjected to incubation within a temperature range of 36-38°C for duration of 18 to 24 h. The presence of *Salmonella typhi* can be inferred from the occurrence of gas production, with or without concurrent blackening, in the stab culture. Additionally, the absence of acidity in the surface growth of the triple sugar-iron agar and the lack of red color in the urea broth.

c) *Pseudomonas aeruginosa*

The culture mediums employed in this study consisted of cetrimide agar medium, *pseudomonas* agar specifically designed for the detection of pyocyanin, and pseudomonas agar optimized for the identification of fluorescein.

i) Primary test

Approximately 10 grams of the specimen were aseptically put into a 100 ml solution of buffer NaCl-peptone. The mixture was then incubated at a temperature of 37ºC for duration of 24 h. A volume of 1 ml of the enrichment culture was introduced into a volume of 100 ml of fluid soybean-casein digest medium. The mixture was thoroughly blended and thereafter subjected to incubation at a temperature range of 35-37ºC for duration of 24 to 48 h. The medium was assessed for growth, and if any growth was observed, a section of the medium was streaked over the surface of cetrimide agar media, which was then plated in a petri dish. Subsequently, the oxidase and pigment tests were performed.

ii) Secondary test

The procedure involved streaking colonies, which were representative suspects, from the agar surface of cetrimide agar onto the surface of pseudomonas agar medium. This was done and detected the presence of fluorescein. Additionally, pseudomonas agar medium was used and detected the presence of pyocyanin. The media that had been injected was covered and inverted, and afterwards incubated within a temperature range of 33°C to 37°C for a minimum duration of three days. The plates were then observed for the presence of colony features.

iii) Oxidase test

Upon the detection of suspicious colonies, a small amount of a recently made 1% w/v solution of N,N,N′,N′-tetramethyl-4-pheneylenediamine dihydrochloride was applied to filter paper. This solution was then spread onto the colony for further analysis. The colour

transition from pink to purple occurring within a time frame of 5 to 10 seconds serves as an indicator for the presence of *P. aeruginosa*.

d) *Staphylococcus aureus*

The culture media employed in this study consisted of Mannitol salt agar and Baird-Parker agar medium (BPA).

i) Primary test

Upon the observation of growth in the enrichment culture, a part of the medium was afterwards streaked onto the surface of BPA. The specimen was subjected to incubation at a temperature of 35°C for a duration of 24 hours. Upon the occurrence of growth, the secondary test was conducted. The observation of black colonies consisting of Gram-positive *cocci*, which are frequently encircled by a clear zone, may potentially suggest the existence of *Staphylococcus aureus*.

ii) Secondary test

The colonies present on the agar surface were subsequently transferred to a tube containing 0.5 ml of either rabbit or horse plasma, with or without other substances. The specimen was subjected to incubation at a temperature of 37°C for duration of 3 hours. The presence of *Staphylococcus aureus* was established through the utilization of the coagulase test, which involved the observation of coagulation to a certain extent.

iii) Coagulase test

The representative suspicious colonies were transferred from the agar surface of the Baird-Parker agar or mannitol salt agar to individual tubes. Each tube contained 0.5 ml of mammalian plasma, preferably rabbit or horse plasma, with or without appropriate additions. This transfer was performed using an inoculating loop. The tubes were subjected to incubation within a water bath maintained at a temperature of 37°C. The tubes were subjected to examination after a duration of 3 hours, followed by additional evaluations at suitable intervals extending up to a period of 24 hours. The presence of *Staphylococcus aureus* in a given sample was indicated by a positive test, specifically the formation of a clot in the test tube within the designated time frame.

v) Heavy metal analysis

Heavy metal of the powdered plant material was done using nitric-perchloric acid digestion method using the procedure recommended by the AOAC (1990). For this analysis, one gram of powdered plant material was boiled gently with 10 ml of concentrated nitric acid for 30-45

min. This mixture was cooled down and 5 ml of 70% perchloric acid was added to it. Further, the mixture was boiled gently until the appearance of dense white fumes. This solution was cooled down and 20 ml of distilled water was added to it and boiled to release the white fumes. After cooling, the solution was filtered through whatman No. 42 filter paper (18). The samples obtained after filtration were analyzed in Atomic Absorption Spectroscopy (AAS) (GBC 932 plus). An atomic absorption spectrophotometer with hollow cathode lamp for lead (Pb), arsenic (As), copper (Cu) cadmium (Cd), mercury (Hg), magnesium (Mg), zinc (Zn) and iron (Fe) was used. The instrument was calibrated by using the standard solutions of Pb, As, Cu, Hg, Cd, Zn, Fe and Mg at various wavelengths 283.5, 228.8, 193.7, 253.7, 283.5, 324.8, 213.9, 248.3, 242.1 nm respectively. Then, the standard calibration curves of these elements were prepared. The instrument was optimized as per requirement and results were obtained in ppm levels (24).

Extraction of plant material

The *Z. officinale* Roscoe rhizomes were cleaned and then dried in shade and cut into small pieces. The dried rhizomes were powdered with a suitable electric blender and stored in an air tight container at cool and dry place. The powder *Z. officinale* Roscoe rhizome was extracted using a method called successive soxhletion. This process involved using various solvents such as petroleum ether, ethyl acetate, chloroform and ethanol at a temperature of 60°C in a soxhlet apparatus. The *Z. officinale* Roscoe rhizomes extracts obtained from this process were then filtered and concentrated using a rotary vacuum evaporator. The yield of the dried leaves extracts was calculated and stored in dessicator (19-21).

Extraction of oil

The *Zingiber officinale* Roscoe rhizome was shade dried, chopped and crushed by mechanical grinder and placed in air tight container. The oil was extracted by Clevenger apparatus and percentage yield of extracted oil was calculated. Oil was further used for phytochemical and physicochemical characterization.

Phytochemical screening (17, 22-23)

1. Test for carbohydrates

Dissolve small portion of extracts and added 4 ml of distilled water (DW) and filtered it. The filtrate was observed for the following tests:

i) Molish's test: few drops of α-napthol (20% w/v) in ethyl alcohol were added in 2 ml of filtrate. Added 1 ml of concentrated sulfuric acid and a ring of roseate violet color between the two layers confirmed the presence of carbohydrates.

ii) Benedict's test: The filtrate was mixed with 1 ml of Benedict's solution and boiled. Brick red precipitate appeared confirmed the presence of reducing sugar.

iii) Fehling's test: Extracts filtrate was boiled on a water bath and added 1 ml of Fehling's reagent (copper sulphate under alkaline conditions) which showed red brick precipitates.

2. Test for alkaloids

Taken the prepared extracts and added 3 ml of 1.5% v/v hydrochloric acid, and the filtrate was examined using subsequent reagent:

i) Dragendroff's test: The prepared extracts were added potassium bismuth iodide (Dragendorff's reagent). An orange red precipitate formed which indicates the presence of alkaloids.

ii) Mayer's test: A little amount of potassium mercuric iodide solution (Mayer's reagent) was added to extract, and the development of a pale creamy-colored precipitate was observed.

iii) Hager's reagent: After adding two to three drops of the picric acid-saturated solution (Hager's reagent) to the prepared extracts, yellow precipitate forms were observed.

iv) Wagner's reagent: The prepared extracts were mixed with a few drops of Wagner's reagent, and a reddish-brown precipitate was observed.

3. Glycosides

i) Keller Killiani test: The prepared extracts were treated with glacial acetic acid, ferric chloride, and concentrated sulfuric acid. If produced characteristic color change detects the presence of cardiac glycosides.

ii) Legal test: To the prepared extracts add equal volume of water and 0.5 ml of strong lead acetate solution. Shaked and filtered the mixture. Filtrate was extracted with equal volume of chloroform and dissolved in 2 ml of pyridine. The sodium nitropruside 2 ml was added followed by addition of NaOH solution. The formation of pink colour indicated presence of glycosides.

iii) Fluorescence test: Taken prepared extracts and mixed with 1N NaOH solution (one ml each). If development of blue-green fluorescence, which indicates presence of glycosides.

4) Test for saponins

i) Frothing test: Each extract solution about 1 ml, and shaken for 15 minutes in a cylinder. Saponins were confirmed by layer of foam that is one centimeter thick.

ii) RBC haemolysis test: After centrifuging 5 ml of blood, RBCs suspended in 10 ml of regular saline. Each prepared extracts were added to the solution (1 ml) and mixed. Saponins are present when froth that resembles honeycomb forms.

5) Test for flavonoids

i) Ammonia test: The prepared extracts solution were used to dip filter paper strips, and ammoniated. When filter paper turned into orange instead of white, confirmed the presence of flavonoids.

ii) Shinoda test: Two drops of concentrated hydrochloric acid were added to one ml of prepared extracts and then added a piece of metallic magnesium or zinc. The incidence of flavonoids is specified by the appearance of red color.

6. Test for phenolic compound and tannins

i) Ferric chloride test: After heating the prepared extracts, added 2 ml (5%) ferric chloride solution, then appearance of green and blue color confirmed presence.

ii) Silver nitrate test: Test tubes were heated on water bath for 25 to 30 minutes and then added one drop of ammonium hydroxide and 10% silver nitrate solution. The deposition of silver mirror is a positive sign.

7. Test for sterols

The prepared extracts refluxed with alcoholic potassium hydroxide solution. Diethyl ether used to dilute saponification mixture. After the ether extract evaporated, other tests were carried out using the residue.

i) Salkowski reaction: A few milligrams of each prepared extract's residue were combined with 2 ml of concentrated sulfuric acid, and the mixture was then observed for the development of a yellow ring between two layers that becomes red after one minute.

ii) Liebermann-Burchard’s test: Add prepared residue in chloroform and then added a little amount of acetic anhydride. The concentrated sulfuric acid was introduced from the tube's side. Sterol and triterpenes are indicated by the development of blue to brick red hue.

8. Fixed oils and fats

i) Solubility Test: Prepared extracts were dissolved in organic solvents like chloroform, alcohol as well as in water. When observed the samples, it formed an oily layer above the

surface of the water then fat is present. If partially soluble in alcohol and fully soluble in chloroform than the presence of fat is confirmed.

ii) Translucent Spot Test: In this, test the prepared extracts tested by rubbing between the folds of filter paper. The appearance of translucent spot confirms the presence of fats in the tested extracts.

Results and Discussion

Morphological Studies

The morphology of *Zingiber officinale* Roscoe rhizome including the shape, size, colour, aroma, taste, and fracture of the rhizomes were assessed. The study revealed that rhizomes were found light brown to reddish brown with characteristic aroma, astringent taste, hard texture and splintery fracture. The young to mature stem size varies and shown in Plate 1.

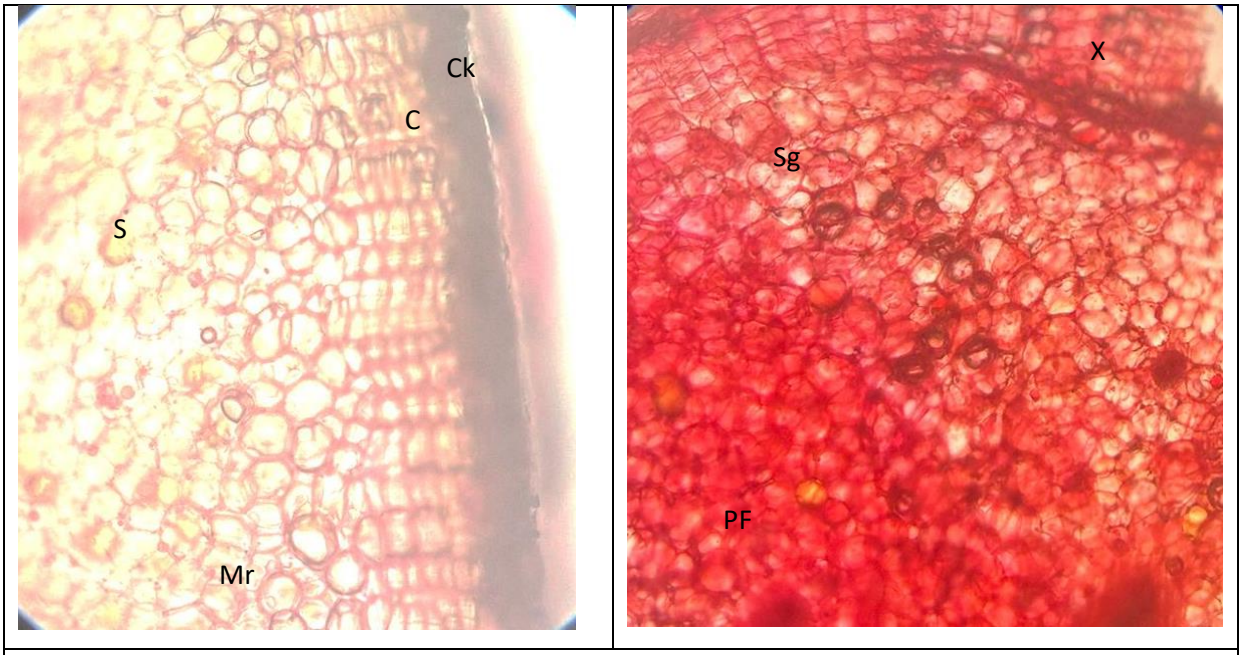
Plate 1: Morphology of *Zingiber officinale* Roscoe rhizome

Microscopical studies

The microscopical study of *Zingiber officinale* Roscoe rhizome were performed using transverse sections and powder study.

i) Transverse section

T.S. of the rhizome displayed an Orangish/reddish-brown colour throughout. The presence of outer layer as cork and after that cortex presents. The vessels were arranged in a diffuse pattern, with a few groups arranged in radial rows of 2-3 vessels. The medullary rays were uni-seriate and run almost straight and parallel. The medullary rays contain starch grains and prismatic crystals of calcium oxalate (Fig 2).



Ck- Cork, C- Cortex, S- Starch, PF- Pericyclic fiber, X-Xylem, Mr- Medullary ray, Sg- Starch grains

Plate 2: Transverse Section of *Z. officinale* Roscoe rhizome

ii) Powder Microscopy

When *Z. officinale* Roscoe rhizome powder was observed under microscope, it shows various elements such as fibers, oil globules, starch grains, and calcium oxalate crystals and yellow coloured lignified fibres which are shown in Plate 3.

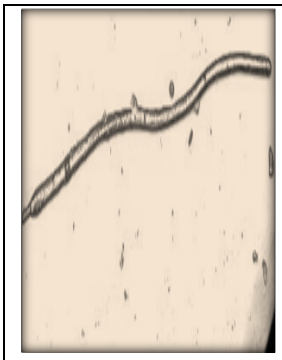
			
Fiber	Oil Globules	Starch grain	Calcium oxalate crystal

Plate 3: Powder Microscopy of *Z. officinale* Roscoe rhizome

Physicochemical standardization

Various physicochemical parameters like Total Ash value, foreign organic matter, moisture content, were studied and results are shown in Table 1.

Determination of Ash Contents

In the current study the ash contents were evaluated for total ash content, acid-insoluble ash and water-soluble ash. Various ash values of *Z. officinale* Roscoe rhizome samples are given in Table 1.

Table 1: Ash Value of *Zingiber officinale* Roscoe rhizome

S. No.	**Type of Ash**	**Ash (% w/w)**
		Rhizome
1.	Total ash	5.23 ± 0.16
2.	Acid insoluble ash	4.09 ± 0.02
3.	Water soluble ash	1.55± 0.02

Table 2: Physicochemical characters of *Z. officinale* Roscoe rhizome

S. No.	**Parameter**	**Rhizome**
1.	**Foreign organic matter**	3.21 ± 1.03 %w/w
2	**Moisture content**	3.5 ± 0.2% w/w
3	**Loss on drying**	37 ± 1.8 %w/w

Values: % w/w mean ± SEM; n=3

Physicochemical characters of *Z. officinale* Roscoe rhizome oil

Z. officinale Roscoe rhizome oil physicochemical characteristics were observed including color, odour, taste, refractive index, relative density, boiling point, acid value, saponification value. The results are shown in table 3.

Table 3: Physicochemical characters of *Z. officinale* Roscoe rhizome oil

S.No.	Oil Parameter	Value
1.	**Color**	Pale yellow to brownish
2.	**Odour**	Characteristic
3.	**Appearance**	Oily consistency
4.	**Refractive index**	1.462-1.494
5.	**Relative density**	0.892 ± 0.08 % wt./ml
6.	**Boiling point**	172 ±1.4°C
7.	**Optical Rotation**	-28º to -45º
8.	**Specific Gravity**	0.881-0.912± 0.14

Values: % w/w mean ± SEM; n=3

Microbiological studies

The microbiological studies in *Z. officinale* Roscoe rhizome were determined. The extracts of the plant were free from bacterial and fungal growth.

Heavy metal analysis

The heavy metal analysis in *Z. officinale* Roscoe rhizome sample was determined the contents of Pb, As, Cu, Cd, Hg, Mg, Zn and Fe in the powdered sample and outcomes are shown in the Table 4.

Table 4: Elemental analysis of *Z. officinale* Roscoe rhizome

Metal	Concentration (ppm)
	***Z. officinale* Roscoe rhizome**
Lead	0.584
Arsenic	0.021
Copper	0.375
Cadmium	0.020
Mercury	0.247
Magnesium	0.421
Zinc	0.033

Iron	1.296

Preparation of extracts

The yield of *Z. officinale* Roscoe rhizome extracts were found in various solvents such as petroleum ether (10.2%), ethyl acetate (9.2%), chloroform (4.2%), ethanol (15.4%) and cold extraction in aqueous (20.4%).

Extraction of oil

The *Z. officinale* Roscoe rhizome oil was extracted by Clevenger apparatus and percentage yield of extracted oil was 5.7%. Oil was further used for physicochemical characterization.

Preliminary phytochemical screening

Qualitative analysis

Preliminary phytochemical analysis of *Z. officinale* Roscoe rhizome indicated the presence of glycosides, carbohydrates, saponins, phenols, tannins, flavonoids, steroids and terpenoids in different extracts. The results of preliminary phytochemical screening of *Zingiber officinale* Roscoe rhizome are shown in Table 5.

Table 5: Preliminary phytochemical screening of *Z. officinale* Roscoe rhizome extracts

Test	**Petroleum ether extract**	**Ethyl acetate Extract**	**Chloroform Extract**	**Ethanol Extract**	**Aqueous Extract**
Carbohydrates					
Molish test	+	+	-	+	+
Benedict's test	-	-	-	+	+
Fehling's test	-	-	-	+	+
Barfoed test	-	-	-	+	+
Alkaloids					
Dragendroff's test	-	+	+	+	+
Mayer's test	-	-	+	+	+
Hager's test	-	+	+	+	+
Wagner's test	-	+	+	+	+
Glycosides					

Keller Killiani test	-	-	+	+	+
Legal test	-	-	+	+	+
Fluorescence test	-	-	+	+	+
Saponins					
Frothing test	-	+	-	+	+
RBC haemolysis test	-	+	-	+	+
Flavonoids					
Ammonia test	-	-	-	+	+
Shinoda test	-	-	+	+	+
Phenolic compound and tannins					
Ferric chloride test	-	-	+	+	+
Silver nitrate test	-	-	+	+	+
Steroids and triterpenoids					
Salkovaski Test	+	+	+	+	-
Libermann Burchard's test	+	+	+	+	-
Fixed oils and fats					
Spot Test	+	+	-	+	-
Translucent Spot Test	+	+	-	+	-

+= present, -= absent

The results of the study showed that the ethanol extract from the specified plants included alkaloids, saponins, amino acids, tannin, flavonoids, carbohydrates, cardiac glycosides, phenol, protein, sterol, and tri-terpenoid. Numerous pharmacological actions were attributed to these secondary metabolites. Plant extracts are screened for phytochemicals shown the presence of alkaloids, saponins, amino acids, flavonoids, protein and sterol. These active constituents put an accountable step over the course of therapy for innumerable types of abnormalities. These standardization procedures provide valuable information about both the plants. The information assembled from the existing research could be valuable in endorsement, characterization, standardization and evaluation of herbal drugs.

Conclusion

The Researcher's from last some years are deeply going to study many ethanomedicinally used plants/herbs, because of easily available, cost effective, high potential effect to treat disease, significant action, and less adverse and toxicity level. The rhizhome of zingiber officinale used in many disorder. In current study it was indicating that given results can be employed as suitable quality control measures to ensure the quality, safety, and efficacy of this herbal drug. The pharmacolognostical standardization of this plant gives an idea about identification, standardization and the monograph of plant. It is also valuable in long term study of plant to evaluate their therapeutic and medicinal effect of this plant.

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Declaration of interest

The author declares no conflict of interest.

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