IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 7, June 2026
Impact Factor: 8.2

Spiru Tea: Development and Nutritional Evaluation of Spirulina Infused with Gokarna Functional Herbal Tea

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Abstract: *Spirulina is a Blue Green alga that is widely used as super food. it is rich in high quantity of proteins, vitamins, amino acids, and antioxidants. Spirulina is known for its health benefits such as boosting immunity improving energy and helping in detoxification of body because of its high nutrient density it is commonly used in functional foods dietary supplements and health products*
Spirulina is blue green algae valued for its high nutritional content including proteins vitamins minerals and antioxidants and it is widely used as a dietary supplement to improve immunity energy levels and overall health.

Keywords: *Spirulina*

I. INTRODUCTION

Applications

Selection of plant:

1] Arthrospira Platensis (Spirulina)

1 Sample collection:

Location- sample were collected from a local area in Miraj

Collection method- Freshly harvested and dried algae was collected and it was collected during daytime to preserve its phytochemicals.

2 Preparation:

Cleaning - Fresh Algae was wash with clean water to remove impurities, after washing algae was dried in sunlight to prevent loss of proteins and other essential phytochemicals. it was dried for 2-3 days and then it was powdered for further use.

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Morphological characters of spirulina-

spirulina is a filamentous, photosynthetic cyanobacterium characterized by its spiral morphology. The organism consists of multicellular filaments known as trichomes which are composed of numerous of coiled cells arranged end to end in an unbranched chain these trichomes are typically coiled and the degree of coiling may vary depending on environmental conditions such as light, temperature, nutrient availability, pH. The cells consist of thin cell wall made primarily from peptidoglycan typically of prokaryotic organisms. As a cyanobacterium, spirulina lacks with true nucleus and membrane bounded organelles. The cytoplasm consists of photosynthetic pigments including chlorophyll-a, phytocyanin and carotenoids which gives the organism its blue green colour.

2] Clitoria ternatea (Gokarna)

1 Sample collection:

Location- sample were collected from a local area in Miraj

Collection method- Freshy collected flowers were harvested and dried using solar dryer and packed after drying to maintain its freshness and phytochemicals.

2 Preparation of sample:

Collection of sample freshly bloomed flowers were plucked by hand and dried under solar to maintain its phytochemicals and freshness.

Morphological Characters of Gokarna:

Objective

Clitoria ternatea commonly known as Gokarna in India it is a perennial herbaceous climber belonging to family Fabaceae. This plant typically shows leguminous morphological features with twinning stem that may grow up to 2-3 meters high.

Flowers are typically small, butterfly shape they are commonly deep blue, white and hybrid varieties also available mostly we choose deep blue colour flower because it is rich in phytochemicals such as anthocyanin. the plant has tap root system with serval lateral roots the seeds are in linear pod measuring about 5-7 cm in length which consist of serval black cooler seeds.

Preparation of Spirulina, Gokarna infused tea:-

Objective:

To prepare standardbred tea bags containing mixture of *Spirulina* and (Gokarna) flowers for extraction for phytochemical analysis.

Tea Bag Formulation:- Each tea bag was prepared with a total weight of 5 grams, composed of

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Procedure:

1. Preparation of Plant Materials: start procedure by weighing the dried powder individually using weighing balance for the spirulina and Gokarna flower. The large particles were broken down using a mortar and pestle allows ensure even distribution in the tea bags.

2. Blending: We combined 2.5 g of spirulina powder with 2.5 g of gokarna flower powder in a clean and dry container. The mixture was gently stirred using a spoon . This step ensured that both plant materials were evenly distributed throughout the bag.

3. Filling the Tea Bags:- Each tea bag was carefully filled with 5 g of the plant powder using a small funnel to ensure the material was distributed evenly, which could restrict water flow.

4. Sealing: Each tea bag was tied using a thread . Seal was checked to confirm it was secure and watertight. It prevent the plant material from escaping into the water during extraction.

5. Quality Control and Storage: Tea bags are stored in dry container away from sunlight and moisture to preserve phytochemicals.

Extraction process: For phytochemical analysis, two tea bags were used in aextraction. Each tea bag was dipped in 250 ml of hot water at 80-90C for 15 minutes. This allows to sufficient time for phytochemical compounds to dissolve into the ethanolic medium while avoiding denaturation of heat-sensitive phytochemicals. The tea bags were removed,the infusion was cooled to room temperature before further analysis.

Extraction by Soxhlet apparatus for the extraction of bioactive compounds from the dried powders of Spirulina platensis and Clitoria ternatea (Gokarna), the Soxhlet extraction method was used. This method ensures efficient extraction using ethanol as the solvent[9].

Materials and Solvents Used

Equipment

Principle of Soxhlet Apparatus

The Soxhlet extraction method uses the solvent reflux and siphon principle to continuously extract the solid material, which provides high efficiency of extraction. The powder is placed in a cone shape filter paper within the Soxhlet. The solvent is heat in the round bottom flask, and its vapor condenses and falls on the powder. As solvent mixes with phytochemicals, it reaches a maximum height in the siphon tube and automatically drains to the flask carrying the extract. This cycle continues for many time, ensuring complete extraction of phytochemicals.

Procedure:

1 Preparation of Plant Material:-

started by weighing both powder 10 g of dried plant powder—5 g each of *sample* using a balance powder was then carefully poured into a filter paper thimble cone to avoid any material from escaping during extraction.

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2 Assembly of Soxhlet Apparatus:-

Next, we assembled the Soxhlet apparatus by inserting the sealed thimble into the body. Then we connect the heating mantle to the round bottom flask, then we attach the condenser on top of the extractor and made sure both the water inlet and outlet connections were properly connected and to allow cooled water to pass through the condenser.

3 Addition of Solvent:-

We carefully poured 250 ml of ethanol into the Soxhlet extractor through the top opening, make sure to add it slowly to avoid any overflow. The solvent was ready to start the extraction cycle.

4 Extraction Process:-

Once everything was set up, we gently heated the system using the heating mantle. As the ethanol heated, it evaporated and traveled up to the condenser, where it cooled and condenses back into liquid, dripping down onto the plant material in the thimble. As the solvent accumulated and dissolved the bioactive compounds, it automatically siphoned back into the round bottom flask below. We allowed the apparatus to complete approximately 6 to 8 cycles, continuing until the ethanol in the thimble turned a golden or amber color, indicating that the extractable compounds had been effectively transferred to the solvent [9].

5 Collection of Extract:-

After the extraction cycles were complete, we carefully collected the ethanol extract from the round bottom flask and set it aside to cool to room temperature. Once cooled, we transferred the extract into a clean container for further analysis

Phytochemical Analysis of Spirulina platensis and Clitoria ternatea Extracts

When we dive into the world of phytochemical analysis, we're extracting our phytochemicals. Of *Spirulina platensis* and *Clitoria ternatea*, this means identification of various phytochemicals that give them their unique properties. For identification and detection that could lead to discovery of new medicines. This analysis is important as it helps us to understand the potential of plants for developing new treatments and therapies.

We typically approach this in two main ways:-

1 Qualitative Analysis: This is about finding out for the presence or absence of specific phytochemicals.

2 Quantitative Analysis: we measure how much of a specifically phytochemical is present in how much quantity.

Qualitative Analysis:-

Test for Glycosides

Glycosides are compounds that play many roles in plants, often linked to defense mechanisms.

Keller Killani Test: Imagine two layers of liquid in a test tube. When we added our test solution with a few drops of glacial acetic acid, ferric chloride solution, and concentrated sulphuric acid carefully to the side, we looked for a reddish-brown layer at the bottom and a bluish-green layer on top. This color separation hints at glycosides.

Raymond's Test: This test involves treating our solution with dinitrobenzene in a hot methanolic alkali. A violet color popping up would indicate the presence of glycosides.

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Legal's Test: Here, we mixed the test solution with pyridine and sodium nitroprusside. A color change to pink or red would suggest glycosides.

Test for Alkaloids

Alkaloids are often known for their potent pharmacological effects.

Mayer's Test: When we treated our test solution with Mayer's reagent (which is potassium mercuric iodide), a creamy precipitate forming would signal the presence of alkaloids.

Test for Flavonoids -

Ferric Chloride Test: A few drops of ferric chloride solution added to test sample forms green color to appear to flavonoids.

Shinoda Test: For this, we added magnesium ribbon and concentrated hydrochloric acid to our test solution shift to pink or magenta-red color is a sign of flavonoids.

Alkaline Reagent Test: The test solution is treated with sodium hydroxide solution makes a yellow color more intense. If adding a few drops of dilute acid then makes it colorless, flavonoids are present.

Test for Saponins

Foam Test: We mixed the saponins with water and gave it a good shake. If a stable foam (lasting at least 15 minutes) formed, saponins were there.

Raymond's Test: Just like with glycosides, a violet color with dinitrobenzene in methanolic alkali.

Test for Carbohydrates

Molisch's Test: we added a few drops of Molisch's reagent and concentrated sulphuric acid to our test solution, a purple ring appears where the two liquids meet would indicate carbohydrates.

Benedict's Test: Treating the test solution with Benedict's reagent and then boiling it in a water bath, resulted in a reddish-brown precipitate if carbohydrates were present.

Test for Proteins

Millon's Test: We treated the test solution with Millon's reagent and heated it in a water bath. If the protein stained yellow on warming, it was a positive result.

Xanthoproteic Test: Here, we added concentrated nitric acid to the test solution and boiled it. A yellow precipitate would form if proteins were present

Biuret Test: Mixing our test solutions with sodium hydroxide and copper sulfate solution, a blue color would develop if proteins were present.

Ninhydrin Test: If our test solution turned blue after being treated with ninhydrin reagent, it indicated the presence of proteins.

Test for Steroids

Chloroform Test: We dissolved the crude plant extract in chloroform and carefully added concentrated sulphuric acid to the side of the test tube. A red upper layer and a yellow-green fluorescent sulphuric acid layer pointed to steroids.

Salkowaski's Test: Adding a few drops of concentrated sulphuric acid to the test solution, shaking it, and letting it sit, would show a red lower layer if sterols (a type of steroid) were present.

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Test for Phenols

Ferric Chloride Test: A simple test! We took a small amount of ethanolic extract, mixed it with water, and added a couple of drops of Iron III chloride (FeCl3). A blue, green, red, or purple color would indicate phenols.

Test for Terpenoids

Salkowaski's Test: Similar to the steroid test, a red lower layer after adding concentrated sulphuric acid and shaking would indicate sterols, which are terpenoids.

Libermann-Burchard Test: After treating the test solution with acetic anhydride and mixing well, we added concentrated sulphuric acid drops to the side. A brown ring at the junction of the two layers and a green upper layer would suggest terpenoids.

Test for Starch

Starch is a common carbohydrate and energy storage for plants.

Starch Reagent Test: We added some of our extract to NaCl solution, heated it up, and then introduced the starch reagent. A blue-purplish color flashing up would confirm the presence of starch [9].

Test for Tannins

Gelatin Test: We dissolved the plant extract in distilled water, then added a 1% gelatin solution and 10% NaCl. A white precipitate forming would be a positive result for tannins.

NaOH Test: Adding 4 ml of 10% NaOH to a small amount of extract and shaking it to see emulsion formation would also suggest tannins.

Test for Flavanols

Ferric Chloride Test: Similar to the general flavonoid test, a few drops of ferric chloride solution indicating a strong color change would show flavanols [9].Shinoda Test: This test would yield a pink to magenta red color with magnesium ribbon and hydrochloric acid [9].Zinc-Hydrochloric Acid Reduction Test: This test would show a color change to red or pink in the presence of flavanols [9].Alkaline Reagent Test: An increase in yellow intensity followed by decolorization with dilute acid would point to flavanols [9].

Lead Acetate Solution Test: A precipitate formation would confirm flavanols [9].

Observations: What We Found

Here's a summary of what our tests revealed about the extracts:

Sr. no.	Chemical Constituents	Tests	Results
1	Glycosides	Keller Killani test	Negative
		Raymond's test	Negative
		Legal's test	Negative
2	Alkaloids	Mayer's test	Negative
3	Flavonoids	Ferric Chloride test	Positive
		Shinoda test	Negative

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Sr. no.	Chemical Constituents	Tests	Results
		Alkaline Reagent test	Positive
		Zinc- hydrochloric acid reduction test	Negative
		Lead acetate solution test	Positive
4	Saponins	Foam test	Positive
		Raymond's test	Negative
5	Carbohydrates	Molisch's test	Positive
		Benedict's test	Negative
6	Proteins	Millon's test	Positive
		Xanthoproteic test	Positive
		Biuret test	Positive
		Ninhydrin test	Negative
7	Steroids	Chloroform test	Positive
		Salkowaski's test	Negative
8	Phenols	Ferric Chloride test	Positive
9	Terpenoids	Salkowaski's test	Positive
		Libermann-Burchard test	Positive
10	Starch	Starch reagent test	Negative
11	Tannins	Gelatin test	Positive
		NaOH test	Positive
12	Flavanols	Ferric Chloride test	Positive

II. CONCLUSION

The phytochemical analysis of Spirulina platensis and Clitoria ternatea (*Gokarna*) extracts revealed a rich and diverse profile of phytochemicals. Our comprehensive testing identified the presence of several important phytochemicals in both plant extracts.

Nutritional Analysis:

Nutritional Analysis of Spirulina-Infused Tea Formulation: A Proximate and Bioactive Compound Evaluation

1. Introduction Spirulina (Arthrospira platensis) is a nutrient-dense cyanobacterium widely used in functional foods. It contains proteins, essential fatty acids, vitamins, minerals, and bioactive compounds such as phycocyanin and carotenoids. Paper-based tea bags are preferable to plastic alternatives, as plastic bags release approximately 11.6 billion microplastics during brewing. This analysis demonstrates how to conduct a proximate and phytochemical evaluation of a spirulina-infused tea product.

2. Materials and Methods formulation: 2.5 g spirulina + 2.5g Gokarna = 5 g total per tea bag (paper-based, plastic-free)

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Proximate Analysis: Use AOAC standard methods to measure protein, carbohydrates, lipids, ash, fiber, and moisture content.

Bioactive Compound Analysis:

2.1 Product Formulation the spirulina-infused tea formulation was prepared using the following specifications:

2.2 Proximate Analysis the nutritional composition of the formulated product was determined according to standard AOAC (Association of Official Analytical Chemists) methods.

Moisture Content: Samples heated at 105°C until constant weight; calculated as percentage loss

Ash Content: Samples incinerated at 550°C in a muffle furnace; residue weighed and expressed as percentage

Crude Protein: Determined using the Kjeldahl method (total nitrogen × 6.25 conversion factor)

Crude Fat (Lipids): Extracted using Soxhlet extraction with petroleum ether or hexane solvent

Crude Fiber: Digestion with dilute acid and alkali; calculated as organic matter loss after incineration

Carbohydrates: Calculated by difference: 100% - (protein + fat + ash + moisture + fiber)

Result

Name of Test	Result
Ash	0.154
Protein	1.428
Carbohydrates	0.820
Energy	8.995
Moisture	0.09
Mold And Fungus	Nill

3. Expected Nutritional Profile based on spirulina's documented composition [6], the 5-gram formulation should contain

4. Advantages of Paper-Based Packaging

5. Quality Control and Safety Considerations Test all raw materials for:

II. CONCLUSION

A spirulina-infused tea (2.5 g spirulina per 5 g formulation) in paper-based packaging offers a science-backed, microplastic-free functional beverage. The product combines spirulina's documented nutritional and bioactive benefits with consumer safety through plastic-free packaging, meeting market demand for clean label, health promoting foods.

DOI: 10.48175/IJARSCT-36809

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