

Comparative Evaluation of *Rasa* (Taste), Phytochemical Characterisation, HPTLC Fingerprinting and Antioxidant Activity of *Taruni peya* (Modified Rose Tea) Brewed using Five Methods: A Research Protocol

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ABSTRACT

Introduction: A “rose” is usually a flowering shrub of the genus *Rosa* that has thorny stems and fragrant blooms. Humans have been using roses and their preparations since ages for health benefits. Rose tea is an infusion prepared from rose buds and petals that is popular for its mild flavour and possible health benefits, including its antioxidant content. It can be drunk as a hot or cold infusion.

Need of the study: No research has been done to date on the effects of brewing procedures on *Rasa*, phytochemical characterisation, High-performance Thin-layer Chromatography (HPTLC) fingerprinting, and the antioxidant properties of modified rose tea. A new field for standardising the brewing process for beverages like modified rose tea will be made possible by the study of these factors.

Aim: To evaluate of *Rasa* (taste), phytochemical characterisation, HPTLC fingerprinting, and antioxidant activity of *Taruni peya* {Modified Rose (*Rosa centifolia* Linn.) tea} prepared by five different brewing methods.

Materials and Methods: An experimental study will be conducted in the Department of Dravyaguna Vigyan, Mahatma Gandhi Ayurved

College and Hospital and Research Centre (MGACH and RC), Salod, Wardha, Maharashtra, India, from April 2024 to June 2025. Roses (*Rosa centifolia* Linn.) will be collected from the natural habitat of the plant. The plant material will be authenticated and identified from Foundation for Revitalisation of Local Health Traditions (FRLHT), Bengaluru, Botanical Survey of India (BSI), or the Botany Department or by an authorised person of the *Dravyaguna* (Pharmacology) Department. *Rasa Nirdharana* (Taste) of the samples will be analysed using parameters mentioned in *Samhitas*. *Charaka*, *Sushruta*, and other *acharyas*, have described the method of the perception and determination of taste by putting a substance on the tongue in which the *Rasa* perceived soon after the substance comes in contact with the tongue is called *Rasa*. Standard Pharmacognostical character and phytochemical characterisation will be performed on all samples using standard methods like macroscopy-microscopy and HPTLC fingerprinting will be studied as given in Ayurvedic Pharmacopoeia of India. Antioxidant properties will be studied by using 2, 2-diphenyl-1-picrylhydrazyl (DPPH), Ferric ion Reducing Antioxidant Potential (FRAP), and phosphomolybdenum assay. Analysis of Variance (ANOVA) test will be used for statistical analysis with a level of significance at p-value <0.05.

Keywords: Cardamom, High-performance thin-layer chromatography, Pharmacognostic, Phosphomolybdenum

INTRODUCTION

Herbal teas are widely utilised as antioxidants all over the world [1]. There is no standard method of brewing. All across the world, various brewing techniques are employed to prepare these herbal beverages [2,3]. Various beverages in the market are made from herbal sources that have already been quoted in Ayurvedic literature for their possible therapeutic advantages. *Aparajita* (butterfly pea), *Taruni* (rose) and *Shyamaparni* (tea) are a few of these plants that have therapeutic potential. The best antioxidant beverages, according to market claims are herbal teas. Rose tea is regarded as the finest antioxidant, at par with or even superior to green tea [4], which is advertised as the best antioxidant globally. But in the current situation, there are no standard brewing techniques, because these techniques vary according to the geographical locations, cultures and nutritional preferences of the communities/tribes in question [3]. There may be a change in the antioxidant property of the tea if various brewing techniques are employed to prepare it. Various rose tea recipes use various components as an addition for flavour, taste, etc., such as cardamom, *daalchini*, clove, etc., [5].

Rose tea is naturally devoid of caffeine, making it a fantastic alternative to some of the more popular caffeinated drinks. Every medicine in

Ayurvedic pharmacology functions by the *Rasapanchaka*, or *Rasa*, *Guna*, *Vipaka*, *Veerya*, etc.,

The antioxidant content and general properties of modified rose tea may be affected by five different brewing techniques, including soft infusion, hard infusion, ambient method, cold infusion and decoction [3]. The selection of five different brewing methods is predominantly used in Asian cultures as in India, Pakistan, China and Bangladesh. An explanation may be sought for the variations in antioxidant capabilities of these five tea brewing techniques employed in various Asian nations [3]. The effects of varying brewing procedures on the antioxidative properties of modified rose tea have not been thoroughly investigated. The present research was aimed to contribute to a better understanding of how different brewing techniques impact the antioxidant content, phytochemical makeup, HPTLC fingerprinting, and overall properties of the tea, consequently improving our knowledge about how these factors affect human health.

The aim of the present study was the comparative evaluation of the effect of five different brewing methods on *Rasa*, phytochemical characterisation, HPTLC fingerprinting and antioxidant activity of modified rose tea.

Primary objectives:

- To evaluate and compare the *Rasa* of modified rose tea prepared by five different brewing methods.
- To analyse the phytochemical characterisation of modified rose tea prepared by five different brewing methods.
- To prepare HPTLC fingerprint and evaluate the effect of modified rose tea prepared by five different brewing methods.
- To evaluate the effect of brewing methods on antioxidant properties.

Secondary objectives:

- To evaluate the pharmacognostical characters (Macroscopic, Microscopic, and powder microscopy) of rose flowers as per API [6].
- To evaluate parameters of identity, purity, and strength as per API [6].

Research hypothesis:

Null hypothesis: (H_0): There will be no significant difference in *Rasa*, phytochemical characterisation, HPTLC fingerprinting and antioxidant properties of *Taruni peya* (modified rose tea) prepared by five different brewing methods.

Alternate hypothesis: Brewing methods will affect the *Rasa*, phytochemical characterisation, HPTLC fingerprinting and antioxidant properties of modified rose tea.

REVIEW OF LITERATURE

Rose flowers belong to the kingdom Plantae, division tracheophytes, class angiosperms, family Rosaceae, genus *Rosa* and species *Rosa centifolia*.

The Indian Ayurvedic pharmacopoeia also contains a monograph on the *Shatapatrika* [6]. Since ancient times, roses, also referred to as edible flowers, have been utilised as culinary ingredients, either in their fresh form or in processed goods like beverages and confections [4]. The creation of these drinks, which are frequently sold under the name of tea, involves a variety of brewing techniques including boiling, chilling, fermenting, etc., [3]. According to reports, the tea made from rose blossoms has the most antioxidant power [5]. The term "*Shatapatri*" or "*Saptaparni*," which refers to a plant with 100 petals, is used in Ayurveda to refer to rose plants. The cooling qualities of the rose plant according to Ayurveda, help to lessen redness, irritation and inflammation of the skin. In Ayurvedic skincare products, rose petals are frequently used to hydrate the skin, minimise acne, and enhance skin texture. Polyphenols, flavonoids and other antioxidants found in abundance in rose plants can shield the body from oxidative stress [7]. Rose petals can aid in preventing oxidative damage because of their strong antioxidant activity [8].

MATERIALS AND METHODS

An experimental study will be conducted in MGACH & RC, Salod, Wardha, Maharashtra, India, from April 2024 to June 2025. The present study has been approved by Institutional Ethical Committee (IEC) (wide reference number: MGACHRC/IEC/Sep-2023/730, Dated 18/09/2023). Roses (*Rosa centifolia* Linn.) will be collected from the *Prakriti* herbal garden of the study Institute. The plant material will be authenticated and identified by FRLHT Bengaluru or BSI or Botany Department or by an authorised person of the *Dravyaguna* Department.

Sample size: 15

Rasa of the samples will be analysed using previously reported validated structured questionnaires based on parameters mentioned in *Samhitas* (GAU, Jamnagar) [9,10] preferably enrolling Ayurveda scholars as volunteers in the study.

Inclusion criteria:

- Volunteers with written informed consent;
- Healthy Ayurveda scholars of either sex and aged from 18-45 years.

Exclusion criteria:

- Volunteers with any habit of tobacco/gum/gutka etc., chewing.
- Volunteers with any diseases affecting the tongue, mouth or taste perception.

Study Procedure

Mature rose petals will be cleaned and washed under tap water, then dried in shade and coarsely powdered.

↓

Dried cardamom pods will be powdered

↓

Both drugs will be separately powdered in a mixer and sieved

↓

Both will be stored in air-tight containers separately.

↓

Take 5 gm dried rose petal powder + 1 gm cardamom pods powder

↓

Take 100 mL water. Boil it and turn off the flame

↓

Add 6 gm powdered (will contain a mixture of 5 gm dried rose petals and 1 gm cardamom) in boiled water and let it steep

↓

Strain out the powdered drug using fine mesh strainer

↓

Add 2.5 gm *Sita* (candy sugar) for taste

Determination of *Rasa*: Taste with tongue रसो निपाते द्रव्याणाम्.... (Charakasamhita, Sutrasthana 26/66) [11] criterion for determining the *Rasa* of the drugs.

Brewing methods:

Rationality of selecting below five methods- These five methods are predominantly used in Asian cultures as in Pakistan, China, India and Bangladesh [3].

1. **Soft infusion [2,3]:** In this preparation method, a tea bag containing five grams of dried rose flower powder will be soaked for 3-5 minutes in warm water that is between 75°C and 85°C. Chinese people prepare tea using this technique.
2. **Hard infusion [2,3]:** This brewing method involves infusing a tea bag for 25-30 minutes in warm, distilled water that ranges in temperature from 75-85°C.
3. **Ambient infusion [2,3]:** This method involves dipping of tea bag in distilled water at room temperature 25±2°C for 30-40 minutes.
4. **Cold infusion [2,3]:** In this method, the tea bag will be infused with distilled water and maintained at room temperature for 15 minutes. After that, the prepared infusion will be refrigerated for an hour.
5. **Decoction method [2,3]:** In this recipe, the tea bag will be placed in distilled water and boiled for 3-5 minutes.

Outcomes:**i) Drug analysis: Organoleptic characters:**

- a) *Shabda* (Sound)
- b) *Sparsha* (Touch)
- c) *Rupa* (Vision)
- d) *Rasa* (Taste)
- e) *Gandha* (Smell)

ii) Pharmacognostical study

Rose flower will be studied under the following heads [6]

- A macroscopic study will be done by observing the characteristic features of the rose flower, as mentioned in the Ayurvedic pharmacopoeia of India [6].
- Microscopic study will be done by taking a transverse section of a rose flower and microscopic features will be observed under the digital microscope as per mentioned in API [6].
- Powder microscopy will be done by taking dried powder of the whole flower and structures will be observed under the digital microscope as per mentioned in API [6].

iii) **Study of parameters of identity, purity and strength:** Rose flower will be studied for:

- Foreign matter: Any additional material discovered in drugs, whether it be biological, chemical, or physical;
- Total ash is what remains after a sample is completely burned;
- Acid-insoluble ash is the portion of ash that is insoluble in acid, or more precisely, insoluble in a diluted HCl solution;
- Alcohol-soluble extractive is usually given as a percentage of the entire amount of the material being removed;
- Water-soluble extractives how much of the drug's content is soluble in water;
- HPTLC fingerprinting.

iv) **Rasa Nirdharana:** *Rasa* of the drug will be analysed by previously reported method. (*Charaka*, *Sushruta* and other *acharyas* have described the method of the perception and determination of taste by putting a substance on the tongue in which the *Rasa* perceived soon after the substance comes in contact with the tongue is called as *Rasa*) [9,10].

v) **Phytochemical characterisation [12,13]:** Qualitative tests will be performed on all samples using standard methods. Phytochemical analyses, including the *Salkowski* (for steroid), *Molisch* (for carbohydrate), *Keller-Killiani* (for glycoside), *Shinoda* test (for flavonoid), *Dragendorff* (for alkaloids) and *Lead acetate* solution tests (for tannin), will be carried out following *Harbone's* standard protocol [14].

vi) **HPTLC fingerprinting:** HPTLC fingerprinting will be obtained on a single plate and a comparative chromatogram will be analysed using suitable techniques/methods [15].

vii) **Antioxidant property evaluation:** Antioxidant properties will be studied by using DPPH, FRAP and phosphomolybdenum assay [16,17].

STATISTICAL ANALYSIS

GraphPad InStat for Windows version 3.06, La Jolla, CA, USA will be used for statistical evaluation. ANOVA test will be applied for comparison.

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