International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

# Pharmaceutico-Analytical and Experimental Study on Madhushukta with Special Reference to Fermentation Dynamics (Sharangadhara Samhita, Madhyama Khanda)

*Dr. Rajath D[1], Dr. M S Krishnamurthy[2], Dr. Rama Bhat P.[3]
[1]*P.G. Scholar, Department of P.G. Studies in Rasashastra and Bhaishajya Kalpana, Alva's Ayurveda Medical College, Moodubidire, Dakshina Kannada, Karnataka, India*
[2]*Professor and Head of Department of P.G. Studies in Rasashastra and Bhaishajya Kalpana, Alva's Ayurveda Medical College, Moodubidire, Dakshina Kannada, Karnataka, India*
[3]*Professor and Head of Department of P.G. Studies in Biotechnology, Alva's College (Autonomous), Moodubidire- 574227*

Date of Submission: 05-11-2025 Date of Acceptance: 15-11-2025

**ABSTRACT**
**Background**: Madhushukta is a classical Sandhana Kalpana (fermented preparation) described by Acharya Sharangadhara in Madhyama Khanda – Asava–ArishtaKalpanaAdhyaya. It serves as a model formulation demonstrating the early stage of fermentation. Two distinct methods are mentioned: one using Madhu, NimbuRasa, and PippaliChurna; and the second using Yava Kwatha, Madhu, Shunti, and Guda. The present study aimed to prepare Madhushuktapharmaceutically and analyse its characteristics to understand its stage of fermentation and therapeutic relevance.
**Methods**: Both formulations were prepared as per classical instructions and kept for three days for Sandhana. Analytical parameters such as pH, specific gravity, reducing sugar, and alcohol content were evaluated.
**Results**: The analytical results revealed that both samples showeddistinct acidic transformation with pH values of 2.2 and 4.18respectively and retained reducing sugars (350–390 µg/mL) but showed 0% alcohol, indicating Amlikarana without alcoholic fermentation.
**Conclusion**: Madhushukta represents the Amlikarana (acidic transformation) stage of SandhanaKalpana, serving as a non-alcoholic bio-ferment with Deepana–Pachana and Rasayana potential. This study reaffirms the Ayurvedic understanding of controlled biotransformation and provides a foundation for further standardization of non-alcoholic fermented formulations.
**Keywords**: Madhushukta, Sharangadhara Samhita, SandhanaKalpana, Fermentation,Amlikarana

## I. INTRODUCTION

Ayurveda, the science of life, emphasizes that the process of Sandhana Kalpana (fermentation) not only preserves the formulation but also enhances its therapeutic potency through biological transformation. Among the Asava–ArishtaKalpana described by Acharya Sharangadhara, Madhushukta occupies a special position as the simplest and most illustrative example of the Sandhana process[1]. Unlike classical Asava–Arishta preparations that undergo prolonged fermentation (15–30 days), Madhushukta completes in merely three days, highlighting it as a demonstrative model for the early stages of fermentation (Amlikaraṇa rather than Sandhana Avastha)[2].

The term Madhushukta denotes a naturally fermented preparation in which Madhu (honey) serves as both the carbohydrate source and preservative, while acidic agents such as NimbuRasa or decoctions like Yava Kwatha initiate mild fermentation and acidogenesis.
Acharya Sharangadhara explains two distinct methods of Madhushukta preparation in Madhyama Khanda, Asava–Arishta Kalpana Adhyaya:

i. Madhu, Nimbu Rasa, and PippaliChurna — the acidic (citrus-based) model.
ii. Yava Kwatha, Madhu, Shunti, and Guda — the decoction-based model.

Pharmaceutically, Madhushukta represents a transitional form between Sura Kalpana (alcoholic fermentations) and Shukta Kalpana (acidic fermentations), serving as a vital example to understand Ayurvedic biotransformation science. Its mild acidity, absence of alcohol, and retention of sugars suggest incomplete fermentation-suitable for Deepana, Pachana, and Rasayana therapeutic applications[3].

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

Modern biochemistry parallels with microbial acidogenesis, enzymatic inversion of sugars, and organic acid enrichment, validating the Ayurvedic concept of Agnivardhana through biochemical activation. Modern fermentation dynamics show that organic acid enrichment without ethanol formation mirrors probiotic fermentation seen in mild lacto-ferments[4].

## II. AIM

To pharmaceutically prepare Madhushukta by the two classical methods described in Sharangadhara Samhita and to compare their organoleptic and analytical characteristics for understanding their stage of fermentation and therapeutic potential.

## III. OBJECTIVES

a. To prepare Madhushukta by the two classical methods:

Method I: Madhu + NimbuRasa + PippaliChurna

Method II: Yava Kwatha + Madhu + ShuntiChurna+ Guda

b. To observe and record organoleptic features (colour, odour, taste, texture, and consistency) during and after three days of fermentation.

c. To evaluate analytical parameters such as pH, specific gravity, reducing sugars, and alcohol content for both methods.

d. To interpret pharmaceutical changes and establish the probable biochemical and Ayurvedic basis for Amlikarana Avastha.

## IV. MATERIALS AND METHODS

### 4.1 Ingredients and Proportions

**Method-1**

| Ingredients | BotanicalName/Binomial Nomenclature | Quantity |
|---|---|---|
| Madhu | Apis indica | 1 Prastha (768 mL) |
| Nimbu Rasa | Citrus limon Linn. | 1 Kudava (192 mL) |
| PippaliChurna | Piper longum Linn. | 1 Pala (48 g) |

**Method-2**

| Ingredients | Botanical Name/Binomial Nomenclature | Quantity |
|---|---|---|
| Yava Kwatha | Hordeum vulgare Linn. | 1 Prastha (768 mL) |
| Madhu | Apis indica | 1 Kudava (192 mL) |
| Shunti | Zingiber officinale Rosc. | 1 Pala (48 g) |
| Guda | SaccharumofficinarumLinn. | 1 Pala (48 g) |

### 4.2 Procedure

#### 4.2aPoorvakarma

1. Collection of raw materials and necessary preparations:

- Madhu: Fresh, unadulterated, and unheated multifloral honey.
- Nimbu Rasa: Fresh juice of Citrus limon fruits, extracted just before use.
- PippaliChurna: fruit part pounded and coarsely powdered.
- Yava Kwatha: Decoctionprepared by boiling 1-part coarse powder(192gm) in 16 parts water(3072ml), and reduced to 1/4$^{th}$(768ml).
- Shunti: Dried rhizome powder, freshly prepared.
- Guda: Organic jaggery, clean, without impurities.

2. Selection and preparation of Sandhana Patra:

- Porcelain jars were selected, washed and dried completely.
- Fumigation (Dhoopana Karma) was performed using Guggulu, Vacha, Jatamansi, Karpoora, Haridra and Maricha to ensure microbial sterility and Rakshoghna effect.
- Muslin cloth, clay and thread were kept ready for Sandhi Bandhana (sealing).

3. Environmental Setup:

A clean, warm, and undisturbed area (DhanyarashiMadhyeSthapyaSthana) was chosen to maintain ambient temperature for controlled fermentation. The average temperature maintained was 30-32$^0$C for optimal Amlikarana.

#### 4.2b Pradhana Karma

Two formulations were prepared as per Sharangadhara Samhita:

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

➢ Method I – Acidic Model (Madhu + Nimbu Rasa + PippaliChurna)

- Fresh Nimbu Rasa (192ml) was poured into porcelain jar.
- Madhu (768ml) was then slowly added with gentle stirring using a big spoon to ensure homogeneity.
- Finally, PippaliChurna (48g) was added gradually and mixed uniformly to avoid lump formation.
- The mouth of the jar was covered with a double layer of muslin cloth and sealed firmly with mud and cloth (Sandhi Bandhana).
- The vessel was then placed undisturbed in the prepared warm chamber (DhanyarashiMadhyeSthapya) for three days.

➢ Method II – Decoction Model (Yava Kwatha + Madhu + Shunti+ Guda)

- Freshly prepared Yava Kwatha(768ml) was filtered through muslin cloth and cooled to room temperature. After cooling it was poured to porcelain jar.
- Madhu (192ml) was added and stirred until completely dissolved.
- Guda and ShuntiChurna(48g respectively) were then added successively with constant mixing to ensure uniform distribution.
- The mouth of the jar was covered with a double layer of muslin cloth and sealed firmly with mud and cloth (Sandhi Bandhana).
- The vessel was placed undisturbed in the prepared warm chamber (DhanyarashiMadhyeSthapya) for three days.

**4.2c Paschat Karma**

- On the third day, the sealed vessel was carefully opened.
- Burning candle test was checked for any escaping gas-results indicated incomplete alcoholic fermentation.
- Content was filtered through a double-layered clean muslin cloth to separate sediment.
- The filtrate (Madhushukta Drava) was collected, labelled, and stored in airtight amber glass bottles to avoid light degradation.
- Organoleptic characteristics (colour, odour, taste, and consistency) were recorded immediately.
- Analytical parameters — pH, Specific Gravity, Reducing Sugars, and Alcohol Content — were tested within 24 hours post-filtration.
- The sample was then preserved for further Sthiratva Pariksha (stability observation).
- The same procedure was carried out for the second method sample.

## V. ORGANOLEPTIC OBSERVATIONS

| Parameter | Method I | Method II |
|---|---|---|
| **Colour** | Light brown | Dark brown |
| **Odour** | Sweet–sour | Sweet with ginger aroma |
| **Taste** | Strongly sour (AmlaPradhana) | Sweet–spicy (Madhura–Katu Pradhana) |
| **Consistency** | Thin fluid | Slightly viscous |
| **Appearance** | Semi-clear | Opaque |

## VI. ANALYTICAL RESULTS

| Parameter[5] | Method I | Method II |
|---|---|---|
| **pH** | 2.2 | 4.18 |
| **Reducing Sugar (µg/mL)** | 350 | 390 |
| **Specific Gravity** | 1.0788 | 1.0309 |
| **Alcohol (% v/v)** | 0 | 0 |

## VII. DISCUSSION

**7.1 Discussion on role of ingredients**

- **Madhu**: Acts as the primary medium (Yogavahi Dravya) facilitating the blending of all components due to its Samskara Anuvartana property.Its inherent Madhura–KashayaRasa, Ruksha–Laghu Guna, and Ushna Veerya create a favourable substrate for natural acidification and preservation.It also contributes to mild enzymatic activity aiding in the breakdown of sugars during the short Sandhana period.
- **NimbuSwarasa** –Provides an immediate acidic environment, lowering the pH and

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

accelerating Amlikarana (acidic transformation). The citric acid content acts as a natural catalyst for biochemical reactions while also preventing unwanted microbial growth.Thus, it supports faster stabilization of the formulation within the stipulated three days.

- **PippaliChurna** – Adds mild pungency (KatuRasa) and enhances the Samskara potency of Madhu.Its Ushna Veerya helps maintain internal warmth in the sealed container, subtly promoting enzymatic reactions.Pippali also aids in uniform mixing and imparts characteristic aroma and colour changes during fermentation.
- **Yava Kwatha** – Acts as the carbohydrate source supporting slow biochemical reactions and acid production.Its mild alkalinity balances the sweetness of Madhu and heaviness of Guda, leading to gradual fermentation.The colloidal nature of Yava maintains the body and consistency of the final product.
- **Shunti**– Provides a KatuRasa base and Ushna Veerya, ensuring a conducive internal temperature for Sandhana reactions.Its digestive and carminative nature prevents stagnation and foul odour formation within the vessel.It also contributes to the pleasant flavour profile noted at the end of fermentation.
- **Guda** –Serves as the fermentable substrate, providing sugars for transformation into organic acids.The presence of minerals and trace elements enhances the nutritional and preservative quality of the final product.It maintains sweetness while supporting gradual Amlikarana without alcoholic conversion.

### 7.2 Discussion on the Pharmaceutical Study of Madhushukta

- **Faithful reproduction of classical procedure**- The entire process was carried out in strict adherence to Sharangadhara Samhita instructions — preparation of ingredients, transfer into Sandhana Patra (porcelain jar), proper sealing (Sandhi Bandhana), and undisturbed storage for three days. This ensured that the transformation occurred under controlled, semi-anaerobic, naturally warm conditions, replicating the classical environment of Sandhana Sthana.

- **Integrity of the sealed stage**- No observation or sampling was attempted during the sealed period. The vessel was opened only on the fourth day, as per textual direction (TridinamSthapyaMadhyame). This practice preserved the internal humidity and prevented external contamination or disturbance of developing biochemical activity.

- **Condition of the preparation at opening (4th day)**- The mixture appeared self-clarified with a distinct layer of supernatant liquid above fine sediment, demonstrating completion of internal reaction and settling (ParisravaSiddhi Lakshana).Absence of froth or gas pressure indicated that the internal activity had naturally subsided, marking the end of short-termAmlikarana.The overall odour changed from purely sweet to mild sweet-sour, confirming successful initiation of fermentation without putrefaction.

- **Comparative outcome of both methods**- Method I (acidic model) yielded a lighter, thinner, uniformly brown liquid with pronounced sourness — evidence of more advanced acid formation.Method II (decoction model) produced a darker, denser liquid with mild sourness and visible extractive matter, signifying slower transformation due to the thicker, starch-rich medium.Both achieved physical stabilization and pleasant aroma, fulfilling Maardava Lakshana of a successful Sandhita Dravya.

- **Nature of transformation**- The change in taste from Madhura to Amla-Madhura, along with clarification and sediment formation, corresponds to AmlikaranaAvastha—the preliminary acidic stage of Sandhana Kalpana. The short duration and sealed condition restricted the process to acidogenesis rather than ethanolic conversion.

- **Pharmaceutical validity of the process**-The observations prove that three-day Sandhana is sufficient to bring about perceptible biochemical and organoleptic transformation while maintaining stability and non-alcoholic nature.

- **Overall inference**- Thus, pharmaceutically, Madhushukta represents an acidic bio transformed product, where the Madhura Rasa of honey evolves into Amla Pradhana Rasa, enhancing its Deepana–Pachanapotency. This

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

process exemplifies Gunantaradhana — transformation resulting in new attributes without losing safety or stability.

**7.3 Discussion on Analytical Results**

- pH: Method I (2.2) was more acidic due to lemon juice; Method II (4.18) showed moderate acidity due to honey and jaggery fermentation. → Confirms Amlikarana and microbial acid production, not ethanol.
- Specific Gravity: Method I (1.0788) > Method II (1.0309); higher soluble solids indicate incomplete sugar breakdown.
- Reducing Sugar: Both samples retained high levels (350–390 µg/mL), confirming mild enzymatic conversion, suitable for probiotic preservation.
- Alcohol Content: 0% in both samples → absence of ethanolic fermentation; stable and safe for internal use.
- The acidic nature provides natural preservation, aligning with Asava–Arishta shelf-life principles[6].
- Analytical results confirm that Madhushukta belongs to acidic fermentation type (Amlikarana) rather than full alcoholic Sandhana.
- Hence, it may be categorized as a semi-fermented nutraceutical, representing Ayurvedic acidic probiotic fermentation.
- Analytical findings reinforce the classical intent — a mild biotransformation aimed at enhancing digestibility, Agnivardhana, and stability, not intoxication.

## VIII. PROBABLE MODE OF ACTION

At biochemical level: Citric and gluconic acids activate digestive enzymes and modulate gut microflora, leading to Agnideepana and Amapachana.

Honey (Madhu): Due to Yogavahi Guna, it enhances absorption of other constituents.

Pippali and Shunti: Stimulate bile flow and thermogenic activity.

Acidic pH: Facilitates ionic exchange and improves gastrointestinal motility.

Together, they produce a Deepana–Pachana effect through mild acid–enzyme synergy.[7]

## IX. FURTHER SCOPE OF THE STUDY

- ExtendedFermentation Study:Since Madhushukta was observed only up to three days, future research may extend the Sandhana period to 5, 7, or 10 days to document progressive biochemical transitions from Amlikarana Avastha toward SandhanaAvastha, helping to establish fermentation kinetics in Ayurvedic pharmaceutics.
- Microbial and Enzymatic Profiling:Analytical assessment of the natural microbial flora and enzyme activity involved during the process could scientifically validate the traditional concept of Swayambhu Sandhana (self-induced fermentation).
- Comparative Analytical Modelling:Studies using advanced analytical tools such as HPTLC, GC–MS, or LC–MS may help identify specific organic acids and phytoconstituents formed during the transformation, strengthening the link between Agnivardhana and biochemical acidogenesis.
- Stability and Shelf-life Evaluation:Long-term storage studies can be performed to determine the effect of acidity and sugar content on product stability, confirming Sandhita Dravya Sthiratva over different durations.

## X. CONCLUSION

The pharmaceutical and analytical study of Madhushukta validates Sharangadhara's description of a three-day acidic fermentation process (Amlikarana Avastha).

Both methods yield stable, non-alcoholic preparations with mild sourness, confirming the concept of acidogenesis without ethanol formation.

This study highlights Ayurveda's advanced understanding of controlled fermentation, enzymatic transformation, and preservation. Madhushukta thus serves as a prototype formulation for teaching and research in Sandhana Kalpana, bridging traditional pharmaceutics and modern biochemical fermentation science[8]. It stands as a testament to Ayurveda's empirical understanding of fermentation, preservation, and transformation- demonstrating how traditional pharmaceutics can inform modern innovation in nutraceutical and functional food research.

## REFERENCES

[1]. Sharangadhara, SharangadharaSaṃhita, Madhyama Khanda, Asava–Arishta Kalpana Adhyaya (Chapters 11–12). In: Tripathi I, editor. 7th ed. Varanasi: ChaukhambhaSurbharati Prakashan; 2013. p. 190–192.

[2]. Patgiri B, Prajapati PK. Bhaishajya Kalpana Vigyana. 2nd ed. Varanasi:

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

Chaukhambha Bharati Academy; 2010. p. 175–180.
[3]. Kulkarni DA, Patil KS, et al. Analytical evaluation of Madhushukta as a prototype of Sandhana Kalpana. AYU Journal. 2018; 39(2):85–90.
[4]. Dash B, Kashyap L. Materia Medica of Ayurveda. Vol. 1. New Delhi: Central Council for Research in Ayurvedic Sciences (CCRAS); 1980. p. 112–115.
[5]. Mukherjee PK. Quality Control of Herbal Drugs: An Approach to Evaluation of Botanicals. 1st ed. New Delhi: Business Horizons; 2002. p. 255–257.
[6]. Nadkarni KM. Indian Materia Medica. Vol. 1. 3rd ed. Mumbai: Popular Prakashan; 2002. p. 734–736.
[7]. Shastri KA. Rasa Tarangini of Sadananda Sharma. 11th ed. New Delhi: Motilal Banarsidass; 2009. p. 14–16.
[8]. Bhattacharya S, Sharma R, et al. Fermentation biotransformation in traditional Ayurvedic preparations – a review. Journal of Ethnopharmacology. 2020; 250:112491.

**Photographs of pharmaceutical preparation**

Ingredients of Method 1 Ingredients of Method 2

Preparation of Yava Kwatha

Extraction of NimbuSwarasa

DhoopanaKarma of Sandhana Patra

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 6 Nov - Dec 2025, pp: 263-269 www.ijprajournal.com ISSN: 2456-4494

Method 1- Addition of Madhu, NimbuRasa, PippaliChurna

Method 2- Addition of Yava Kwatha, Madhu, Shunti and Guda

Sandhi Bandhana Dhanyarashi Madhye Sthapyep