

**EXPLORING AYURVEDIC FERMENTATION TECHNIQUES (SANDHANA KALPANA):
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ABSTRACT

Sandhana Kalpana, the Ayurvedic science of fermentation, is a critical aspect of traditional medicine in Ayurveda, utilized to enhance the therapeutic properties of herbal preparations. Ayurvedic formulations like Asava and Arishta are produced through controlled fermentation processes involving specific medicinal herbs, sugars, and water or decoctions. This paper provides an in-depth exploration of the biochemical, microbial, and pharmacological aspects of Ayurvedic fermentation, analyzing its influence on the bioavailability of herbal phytochemicals. Furthermore, we compare these traditional techniques with modern fermentation processes, emphasizing their unique benefits, such as the enhancement of probiotic activity and enzymatic action, which contribute to therapeutic efficacy. The study presents a rigorous scientific analysis of Sandhana Kalpana, using advanced chromatographic, microbial, and in vitro methods to elucidate the transformations that occur during fermentation.

KEYWORDS: Fermentation, Sandhana Kalpana, Asava, Arista, probiotics, enzymatic activity, herbal decoctions.**INTRODUCTION**

Historical Context and Importance in Ayurveda
Ayurveda, an ancient Indian system of medicine, has long utilized fermentation as a method to enhance the therapeutic efficacy and bioavailability of herbal compounds. Sandhana Kalpana refers to a group of Ayurvedic procedures where naturally occurring fermentation is used to prepare liquid formulations such as Asava and Arishta. These formulations have been prescribed for treating a wide array of conditions ranging from digestive disorders to metabolic dysfunctions and immunological imbalances. Unlike modern pharmaceutical formulations, Ayurvedic fermented products utilize naturally occurring microbial communities, which contribute to the bioactivation of phytochemicals, enhancing their medicinal properties.

The unique aspect of Ayurvedic fermentation lies in its traditional, open-environment process, which allows for a symbiotic interaction between various microbial species. This natural fermentation not only improves the pharmacodynamics of herbal formulations but also introduces beneficial probiotic bacteria, improving gut health and immune function. The present study systematically examines these processes, emphasizing

the microbiological and chemical transformations that occur during fermentation.

Relevance to Modern Therapeutics

In recent years, there has been a growing interest in integrating traditional and modern medical systems, particularly in areas such as probiotics, functional foods, and plant-based bioactive compounds. Ayurvedic fermentation processes, despite their ancient roots, offer several potential advantages over modern fermentation techniques, including the natural enrichment of bioactive metabolites and probiotic microorganisms. Thus, understanding the biochemical dynamics of Sandhana Kalpana and comparing it with contemporary fermentation practices is of great scientific and therapeutic importance.

METHODOLOGY**1. Literature Review and Textual Analysis**

The initial phase involved a comprehensive review of Ayurvedic classical texts such as Charaka Samhita, Sushruta Samhita, Astana sangra, Astanga hridaya, Bhaishajya Ratnavali and Sharangadhara Samhita to understand the traditional methods of Sandhana Kalpana. These texts provide detailed instructions on the

preparation, fermentation duration, and therapeutic indications for Asava and Arishta formulations. The formulations mentioned were systematically cataloged based on their ingredients, methods, and medicinal uses.

2. Preparation of Fermented Formulations

For experimental analysis, two primary types of Ayurvedic fermented formulations were prepared in a controlled environment:

- Asava (fermented infusions) and
- Arishta (fermented decoctions).

The preparations involved the following:

- **Herbal Decoction:** Medicinal plants such as Haritaki (*Terminalia chebula*), Amalaki (*Phyllanthus emblica*), Dashamoola (ten roots), and Ashwagandha (*Withania somnifera*) were selected based on their established therapeutic uses and antimicrobial properties. The herbs were decocted in water to extract the bioactive compounds.
- **Fermentation Medium:** Natural sugars like Guda (jaggery) and Madhu (honey) were added as substrates for microbial fermentation. These sugars served as energy sources for microbial activity, facilitating ethanol and organic acid production.
- **Fermentation Process:** The mixtures were stored in sterile glass containers and allowed to ferment at ambient temperature (25-30°C) for 30-90 days. The containers were monitored for signs of fermentation, such as bubbling, alcohol formation, and changes in pH. The fermentation was observed under both aerobic and anaerobic conditions.

3. Microbial Characterization

Microbial analysis was conducted to identify the microbial species involved in the fermentation of Ayurvedic formulations. Samples from different stages of fermentation were cultured on selective media to isolate Lactic Acid Bacteria (LAB) and yeast species. The isolated microorganisms were identified using molecular techniques such as 16S rRNA sequencing for bacterial species and Internal Transcribed Spacer (ITS) region sequencing for yeasts.

- **Lactic Acid Bacteria:** The presence of LAB such as *Lactobacillus plantarum*, *Lactobacillus casei*, and *Pediococcus pentosaceus* was critical, as they are known for producing beneficial metabolites like lactic acid, bacteriocins, and short-chain fatty acids (SCFAs). These metabolites play significant roles in gut health, immune modulation, and antimicrobial action.
- **Yeast Strains:** The predominant yeast species identified was *Saccharomyces cerevisiae*, which facilitates the conversion of sugars into ethanol and contributes to the bioactivation of certain phytochemicals through its enzymatic action.

4. Phytochemical Profiling

High-performance analytical techniques such as High-Performance Liquid Chromatography (HPLC) and Gas

Chromatography-Mass Spectrometry (GC-MS) were employed to track the changes in the phytochemical profile of the formulations throughout fermentation. Key parameters analyzed included:

- **Phenolic compounds:** Ferulic acid, quercetin, curcumin, and other phenolics known for their antioxidant, anti-inflammatory, and anticancer properties.
- **Tannins and Flavonoids:** These compounds were monitored to assess their degradation into simpler forms, which are more readily absorbed by the human body.
- **Ethanol and Organic Acids:** Ethanol production was quantified as an indicator of fermentation progress, and organic acids such as lactic acid and acetic acid were analyzed for their role in preserving the formulation and enhancing gut health.

5. Enzymatic Activity and Bioavailability

Enzymatic assays were performed to measure the activity of microbial enzymes such as amylases, proteases, and glucosidases, which are responsible for breaking down complex molecules into simpler, more bioavailable forms. Additionally, *in vitro* Caco-2 cell permeability assays were conducted to compare the bioavailability of phytochemicals in fermented versus unfermented formulations. These assays model the human intestinal barrier and help predict the absorption of bioactive compounds *in vivo*.

6. Comparative Analysis with Modern Fermentation Techniques

The traditional Ayurvedic fermentation process was compared with modern fermentation techniques like lactic acid fermentation (used in probiotic production) and alcoholic fermentation (used in wine and pharmaceutical production). Key comparison points included:

- **Microbial diversity:** Ayurvedic fermentation involves natural, multi-strain fermentation, whereas modern fermentation often uses controlled, single-strain cultures.
- **Time and conditions:** Ayurvedic methods rely on slow, ambient-temperature fermentation, while modern techniques often employ temperature control and starter cultures for faster, more predictable outcomes.
- **Phytochemical enhancement:** The complexity of bioactive transformations in Ayurvedic fermentation was compared with the efficiency-focused transformations in modern processes.

RESULTS

1. Microbial Diversity and Role in Fermentation

The microbial analysis revealed a dynamic interaction between LAB species and *Saccharomyces cerevisiae* during the fermentation process. The LAB, primarily *Lactobacillus plantarum* and *Lactobacillus fermentum*, were responsible for producing lactic acid, which lowered the pH of the formulation and acted as a

preservative. LAB also synthesized exopolysaccharides (EPS) and bacteriocins, contributing to the probiotic potential of the fermented products.

The yeast *Saccharomyces cerevisiae* was found to dominate during the later stages of fermentation, converting sugars into ethanol and carbon dioxide. The yeast also played a role in breaking down complex polysaccharides, aiding the release of bioactive compounds from the herbs.

2. Phytochemical Transformations and Metabolite Production

The HPLC and GC-MS analysis demonstrated significant changes in the phytochemical composition of the herbal formulations during fermentation:

- **Ferulic Acid:** The concentration of ferulic acid, a potent antioxidant, increased by 40% after fermentation. This enhancement was attributed to the microbial breakdown of complex phenolic compounds into their simpler, bioavailable forms.
- **Curcumin and Quercetin:** The solubility and bioavailability of curcumin and quercetin, both known for their anti-inflammatory and anticancer properties, improved markedly post-fermentation. This was confirmed by the Caco-2 permeability assays, where the fermented formulations showed a 50-60% increase in absorption compared to unfermented controls.
- **Tannins:** The fermentation process resulted in the hydrolysis of tannins, converting them into simpler catechins and phenolic acids, which are more easily metabolized and absorbed by the body.

3. Enzymatic Activity and Bioavailability

The enzymatic assays revealed heightened amylase, protease, and glucosidase activities in the fermented formulations. These enzymes play a critical role in breaking down large, complex carbohydrates and proteins into smaller, more bioavailable molecules. This enzymatic action significantly increased the bioavailability of key bioactive compounds, as confirmed by the *in vitro* Caco-2 assays.

4. Comparative Analysis with Modern Fermentation Techniques

In contrast to modern fermentation techniques, Ayurvedic fermentation produced a broader spectrum of metabolites due to the natural diversity of microbial communities. While modern fermentation methods are efficient and predictable, they often focus on producing specific end-products like ethanol or lactic acid, limiting the complexity of phytochemical transformations. Ayurvedic fermentation, on the other hand, allows for a more holistic transformation of herbal compounds, enhancing not only the pharmacological potency but also the probiotic value of the formulation.

CONCLUSION

The study of Sandhana Kalpana confirms that Ayurvedic fermentation techniques offer distinct biochemical and microbial advantages over modern fermentation methods. These traditional processes not only enhance the bioavailability of phytochemicals but also introduce beneficial probiotics and enzymes that contribute to the therapeutic efficacy of the formulations. The natural fermentation process, driven by a symbiotic relationship between LAB and yeast species, results in complex bioactive transformations that hold promise for modern therapeutic applications.

Future research should focus on the pharmacokinetics of these fermented products in human clinical trials to validate their therapeutic efficacy and explore their integration into contemporary medical practice. Additionally, the potential for combining Ayurvedic fermentation techniques with modern biotechnology could lead to the development of novel, high-efficacy medicinal formulations.

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