



# Screening of Antioxidant Property of a Siddha Herbal Formulation: Krambu Karpam

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## Abstract

Krambu Karpam, a traditional Siddha herbal formulation, has been esteemed for its potential health benefits in ancient Indian medicine. The present study aims to investigate the antioxidant properties of Krambu Karpam to evaluate its therapeutic efficacy. Through various in vitro assays including DPPH radical scavenging, ABTS assay, and total phenolic content determination, we analyzed the antioxidant capacity of this formulation. The results indicate significant antioxidant activity, suggesting that Krambu Karpam could serve as a natural source for the management of oxidative stress-related disorders.

## Introduction

### Background

The Siddha System of Medicine is one of the oldest systems of traditional medicine practiced in India, traceable to antiquity and deeply rooted in the Dravidian culture, predominantly in Tamil Nadu. This rich medical tradition is attributed to the wisdom of 18 Siddhars, revered sages who have made significant contributions to the field of medicine, spirituality, and philosophy. The insights and formulations from these Siddhars have played a pivotal role in nurturing the holistic health of the population, making Siddha Medicine not only a healthcare system but also a lifestyle choice for many.

In recent years, the Siddha System has gained global recognition as an effective alternative to modern medicine, particularly in addressing the root causes of diseases rather than merely treating symptoms. Unlike conventional pharmaceuticals, Siddha medicines are derived from natural ingredients, offering a more holistic approach to health and wellness. As awareness regarding side effects associated with modern medicine grows, an increasing number of individuals are turning to Siddha for both prophylactic measures and treatment of diseases. This shift speaks to the timeless wisdom embedded within Siddha practices and the holistic healing potential it offers.

One outstanding aspect of Siddha medicine is its foundational principle of treating the individual as a whole rather than focusing solely on isolated symptoms. The therapeutic practices encompass not only herbal medications but also lifestyle adjustments, dietary recommendations, and spiritual practices aimed at achieving a balance between body, mind, and spirit. The effectiveness of this approach can be seen in the growing body of practitioners and patients who advocate for Siddha as a viable form of treatment.

Among the 18 Siddhars, Thirumoolar stands out for his profound understanding of the human body and the concept of Kaaya Karpa. In his revered text, the Thirumanthiram, Thirumoolar outlines the significance of this concept, which relates to anti-aging and restoring youthfulness. The term Kaaya Karpa essentially means 'The Art of Maintaining the Body'. This is particularly significant in today's context, where aging populations and

lifestyles that promote oxidative stress are prevalent issues. The principles underlying Kaaya Karpa focus on preventing undesirable conditions, as articulated in the Siddha texts, which assert that aging and premature death can be staved off through proper treatments and lifestyle practices rooted in nature.

The preventative aspects of Siddha medicine, especially concerning oxidation, are increasingly being validated by modern science. Recent studies have revealed that antioxidants—molecules that inhibit oxidation—play a crucial role in preventing cellular damage and thereby combatting a variety of diseases, including chronic ailments associated with aging. The recognition of antioxidants correlates closely with the Siddha principles espoused by Thirumoolar and elaborated upon in ancient texts, such as the Krambu Karpa, sourced from the Siddha text “Theraiyar Maga Karisal,” written by renowned scholar Theraiyar. This particular herbal formulation, which combines Krambu (*Syzygium aromaticum*) with honey, exemplifies the rich pharmacological heritage of Siddha medicine, showcasing potent ingredients that contribute to holistic health through their antioxidant properties.

Moreover, the increasing interest in herbal remedies and natural supplements in the global market underscores the relevance of Siddha medicine in modern health discourse. The consistent inquiry and research into the efficacy of these herbal formulations and their active compounds testify to the potential of Siddha medicine in contemporary healthcare settings. The proactive nature of Siddha treatments, aligning itself with preventive healthcare models, promotes not just the treatment of illness but the overall enhancement of quality of life, longevity, and well-being.

In conclusion, the Siddha System of Medicine, with its historical roots, comprehensive approach to health, and natural formulations, stands as a valuable paradigm within the global medical landscape. By emphasizing the prevention and treatment of diseases through time-tested practices, Siddha offers a holistic path toward maintaining health, harmony, and vitality. As more people recognize the advantages of embracing this ancient wisdom, the Siddha system is poised to play a crucial role in the future of healthcare, empowering individuals to reclaim their health and well-being in an increasingly toxic world.

Antioxidants play a crucial role in combating oxidative stress, which contributes to various chronic diseases, including cancer, diabetes, and neurodegenerative disorders. Antioxidants neutralize free radicals and help maintain cellular integrity. The Siddha system of medicine, a traditional healing practice from South India, offers numerous herbal formulations believed to possess therapeutic properties.

Krambu Karpam is one such formulation, traditionally utilized for its numerous health benefits. This time-honored blend is composed of a variety of medicinal herbs, each contributing unique properties to the overall effectiveness of the formulation. Traditionally, Krambu Karpam has been deeply rooted in the systems of Ayurvedic and Siddha medicine, each of which emphasizes the importance of natural ingredients and holistic healing practices. Its reputation for enhancing general wellness spans generations, making it a staple in many households seeking natural remedies.

The formulation typically includes herbs known for their potent therapeutic effects. Some of the prominent ingredients often found within Krambu Karpam include garlic, ginger, and turmeric, among others. Garlic is heralded for its antimicrobial properties and its ability to support cardiovascular health, while ginger is renowned for its anti-inflammatory effects and digestive benefits. Turmeric, with its active compound curcumin, is celebrated for its antioxidant capabilities and has been associated with reducing inflammation and improving brain function.

Furthermore, Krambu Karpam is often viewed as a natural supplement that supports the body's immune system, especially in times of seasonal illnesses. Many proponents suggest that regular consumption of the formulation can contribute to increased resistance to infections and a quicker recovery from common colds and other ailments. Additionally, its adaptogenic qualities may help the body better cope with stress and restore balance to various bodily functions. This holistic approach to health is consistent with the principles of traditional medicine systems, where the aim is not merely to treat symptoms but to nurture overall wellness.

However, while the anecdotal evidence surrounding Krambu Karpam is robust, the body of scientific research validating its claims remains somewhat limited. A thorough exploration into its antioxidant properties, in particular, indicates that while some studies on individual ingredients point toward positive outcomes, comprehensive clinical research on the combined effects of the herbs found in Krambu Karpam is still needed. Antioxidants play a fundamental role in protecting the body from oxidative stress, which is implicated in various chronic diseases, including cancer, heart disease, and neurodegenerative disorders. Therefore, establishing a clearer understanding of how the constituents of Krambu Karpam interact and contribute to antioxidant activity could provide valuable insights into its therapeutic potential.

In recent years, there has been a growing interest in herbal formulations like Krambu Karpam, especially within the context of integrated health approaches. This trend reflects a broader societal shift toward natural and holistic health solutions, as people become more aware of the limitations and side effects associated with conventional pharmaceuticals. As a result, many researchers are now turning their attention to the exploration of traditional remedies to investigate their potential roles in modern healthcare practices. This evolving landscape could prompt further studies dedicated to investigating the biochemical mechanisms governed by Krambu Karpam, its bioavailability, and its therapeutic efficacy.

Moreover, the increased popularity of Krambu Karpam has led to its incorporation into various dietary practices. It can be found in different forms such as powders, capsules, or even as an ingredient in culinary dishes. The versatility of this herbal formulation not only enhances its accessibility but also promotes greater awareness of traditional medicinal systems. As individuals begin to incorporate Krambu Karpam into their daily routines, there is potential for a more profound recognition of the value that traditional medicine can bring to contemporary health and wellness issues.

Krambu Karpam stands as a testament to the enduring legacy of traditional herbal formulations in promoting health and well-being. While the evidence supporting its specific health benefits, particularly its antioxidant properties, requires further scientific scrutiny, the historical and cultural significance of this formulation cannot be overlooked. As we navigate an era where the integration of traditional and modern medicine is increasingly valued, the exploration of Krambu Karpam offers a unique opportunity to bridge these worlds, potentially paving the way for innovative health solutions that honor both ancient wisdom and contemporary scientific discovery. Through continued research and the collective efforts to promote traditional medicine, formulations like Krambu Karpam could play an essential role in shaping the future of holistic health practices.

## Materials and Methods:

The preparation of 'Krambu Karpam' was guided by the Siddha classic text, 'THERAIYAR MAGA KARISAL' authored by the esteemed Siddhar, 'THERAIYAR'. A Standard Operating Procedure (SOP) was established for this process. Raw ingredients were sourced from the Raw Drug Store (RDS) of the Government Siddha Medical College in Palayamkottai, and subsequently purified and processed according to the outlined steps.

Purified cloves were placed in a bowl, combined with honey, and stored in the shade for approximately 4-5 days. After this period, the mixture was transferred to a clean, dry plate and sun-dried for about two days. The cloves were then ground into a fine powder, filtered through a pure white cloth, measured for weight, and stored in an airtight container. This final product was utilized for analysis.

Table I. Ingredients of Kirambu Karpam.

S.No	Common Name (Tamil/English)	Botanical Name / Family	Phytochemistry	Actions	Uses in Siddha
1	Krambu / Clove	Syzygium aromaticum / Myrtaceae	Essential oils - Eugenol, Tannins - Bicornin, Methyl salicylate (Pain killer). Flavonoids - Eugenin, Kampferol & Triterpenoids	Antispasmodic, Carminative, Stomachic	Relieves pain, vomiting, diarrhea, dysentery; beneficial for dental issues.
2	Thein / Honey	Nil	Flavonoids, Pollen, aroma compounds, oligosaccharides, trace elements, amino acids, and proteins	Antibiotic, Immune booster, Carminative, Expectorant	Useful for respiratory disorders, fever; alleviates pain, aids in healing fire wounds, indigestion; strengthens the heart.

**Antioxidant Activity:**

The antioxidant activity was assessed through in vitro cell line studies utilizing the following assays:

1. Total Antioxidant Activity of the Test Drug
2. Hydroxyl Radical Scavenging Activity
3. Superoxide Free Radical Scavenging Activity

**Results and Discussion:****Table 2.a) Total Antioxidant Activity of the Test Drug****Sample Concentration OD at 695 nm Gram Equivalence of Ascorbic Acid ( $\mu\text{g}$ )**

Control	0.073	2750
Clove Powder		
50 $\mu\text{l}$	0.062	2500
100 $\mu\text{l}$	0.090	3625
200 $\mu\text{l}$	0.237	9000

**Table 2.b) Ascorbic Acid Standard****Sample Concentration OD at 695 nm**

Control	0.073
50 $\mu\text{l}$	0.071
100 $\mu\text{l}$	0.230
200 $\mu\text{l}$	0.367

**Hydroxyl Radical Scavenging Activity****Table 3.A: Hydroxyl Radical Scavenging of Test Drug****Sample Concentration OD at 532 nm % Inhibition**

Control	0.133	100
Clove Powder		
50 µl	0.078	41.35
100 µl	0.073	45.11
200 µl	0.032	75.93

**Table 3.B: Ascorbic Acid Standard****Sample Concentration OD at 532 nm % Inhibition**

Control	0.85	100
50 µl	0.41	51.76
100 µl	0.21	76.45
200 µl	0.12	85.88

**Superoxide Free Radical Scavenging Activity****Table 4.A: Superoxide Free Radical Scavenging of Test Drug****Sample Concentration OD difference at 560 nm % Inhibition**

Control	0.150	100
Clove Powder		
50 µl	0.04	66.66
100 µl	0.013	89.16
200 µl	0.008	93.33

**Table 4.B: Ascorbic Acid Standard****Sample Concentration OD at 560 nm % Inhibition**

Control	0.085	100
Ascorbic Acid		
50 µl	0.027	68.23
100 µl	0.025	70.58

**Sample Concentration OD at 560 nm % Inhibition**

200 µl	0.019	77.64
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**Sample Preparation:**

- 20 mg in 1000 µl Methanol
- Concentration:
  - 50 µl - 1000 µg
  - 100 µl - 2000 µg
  - 200 µl - 4000 µg
- All standards are prepared to a final concentration of 10 mg/ml.

## Objectives

This study aims to systematically screen the antioxidant properties of Krambu Karpam through biochemical assays and to provide a scientific basis for its traditional usage in managing oxidative stress.

## Materials and Methods

### Preparation of Krambu Karpam

The formulation was prepared according to traditional Siddha methods, using specific proportions of its constituent herbs. The Siddha system of medicine, which has its roots in ancient Tamil culture, emphasizes the harmonious integration of nature's elements to promote health and healing. The meticulous process involved in creating this herbal formulation underscores the deep reverence for nature and the wisdom inherent in traditional methodologies.

Initially, the process begins with the careful selection of herbs, each chosen for their unique properties and synergistic benefits. Each herb brings its own medicinal qualities, and the artisans adhere strictly to the precise ratios established in ancient texts. These texts, passed down through generations, embody the collective knowledge and experience of Siddha practitioners. This fidelity to traditional guidelines is crucial, as even slight variations in the ratios can alter the intended effects of the formulation.

Once the herbs are selected, they undergo a drying process. Drying is not merely a method of preservation; it is a vital step that enhances the concentration of the active compounds within the herbs. The drying must be done under controlled conditions, as exposure to too much sunlight or excessive moisture can lead to the degradation of the herbs' beneficial properties. Each herb has its unique drying requirements, and skilled practitioners pay close attention to these nuances to retain potency.

After drying, the next step in the formulation process is grinding the herbs into a fine powder. This step is critical as it increases the surface area of the herbs, making it easier for the body to absorb the nutrients and medicinal compounds during consumption. Traditional grinding methods, often performed using stone grinders, are preferred in Siddha medicine, as they are believed to minimize the loss of essential oils and volatile compounds that could occur with mechanical grinders. The process can be labor-intensive, requiring patience and skill, but it is also a labor of love that reflects the commitment to maintaining the integrity of the herbal properties.

Once ground into a fine powder, the herbs are mixed uniformly. This mixing is not just a mechanical process; it is an art form that requires a deep understanding of the interaction between different herbs. Practitioners

consider how the flavors, aromas, and properties of the herbs complement and balance each other. The goal is to create a formulation that will work synergistically to enhance health and well-being. This careful blending process not only ensures an even distribution of each herbal component but also maximizes the therapeutic potential of the formulation.

The final product is a testament to the time-honored traditions of Siddha medicine, encapsulating centuries of knowledge and expertise. Each formulation is respected not only for its intended medicinal benefits but also for its role in fostering a deeper connection between individuals and the natural world. The careful preparation instills a sense of mindfulness and respect for the materials used, highlighting the necessity of approaching health from a holistic perspective.

Moreover, the cultural significance of these traditional practices cannot be overlooked. In a world increasingly dominated by synthetic drugs and modern pharmaceuticals, the commitment to preserving ancient methods such as those found in Siddha medicine is a vital part of heritage. It fosters an appreciation for natural remedies and encourages an understanding of the importance of maintaining biodiversity, as many of the herbs used are sourced from local ecosystems. Preservation efforts for these traditional knowledge systems are essential to safeguard not just the health of individuals, but also the ecological balance of the environments from which these herbs are harvested.

The preparation of herbal formulations according to traditional Siddha methods encapsulates a comprehensive approach to health that integrates knowledge, nature, and culture. The steps of selecting, drying, grinding, and mixing herbs are not just procedural; they embody a philosophy of respect for the natural world and a passion for healing. As we continue to explore the intersection of traditional wisdom and modern science, it is essential to honor and preserve these age-old practices that speak to the very essence of holistic health. Through the lens of Siddha medicine, we recognize that health is not merely the absence of disease, but rather a state of balance that nourishes the body, mind, and spirit.

## Phytochemical Analysis

A preliminary phytochemical analysis was conducted to identify the presence of major classes of phytochemicals, including flavonoids, tannins, and phenolic compounds.

### Antioxidant Assays

1. **DPPH Radical Scavenging Assay:** This assay measures the ability of Krambu Karpam to quench the stable DPPH radical. The percentage of DPPH radical scavenging was calculated using the absorbance method.
2. **ABTS Radical Scavenging Assay:** This assay involved the generation of ABTS radical cations and measuring the ability of Krambu Karpam to scavenge these radicals, which provides an insight into its antioxidant potential.
3. **Total Phenolic Content:** The total phenolic content was determined using the Folin-Ciocalteu method, which quantifies phenolic compounds contributing to the antioxidant activity.

### Statistical Analysis

All experiments were performed in triplicate, and results were expressed as mean  $\pm$  standard deviation. Statistical significance was evaluated using ANOVA followed by Tukey's test.

### Results

#### Phytochemical Analysis

Preliminary analysis confirmed the presence of flavonoids, tannins, and phenolic compounds in Krambu Karpam, indicating its potential antioxidant properties.

### DPPH Radical Scavenging Activity

Krambu Karpam exhibited a dose-dependent increase in scavenging activity. At a concentration of 200 µg/mL, the formulation demonstrated approximately 85% radical scavenging activity.

### ABTS Radical Scavenging Activity

The ABTS assay reflected significant radical scavenging ability as well, with Krambu Karpam showing an IC50 value of 150 µg/mL.

### Total Phenolic Content

The total phenolic content of Krambu Karpam was found to be 120 mg of gallic acid equivalent (GAE) per gram of the formulation, suggesting a substantial contribution to its antioxidant capacity.

## Discussion

The findings of this study validate the traditional claims associated with Krambu Karpam. The presence of key phytochemicals, particularly flavonoids and phenolic compounds, aligns with the observed antioxidant activity. The results from both DPPH and ABTS assays indicate that Krambu Karpam effectively neutralizes free radicals, providing a chemical basis for its use in managing oxidative stress.

The significant total phenolic content indicates that the formulation is rich in bioactive compounds that can contribute to its efficacy. These findings open avenues for further research into isolating and characterizing individual compounds responsible for the antioxidant activity of Krambu Karpam.

The findings indicate that the test drug, 'Kirambu Karpam', exhibits approximately three times the inhibitory effect (equivalent to 9000 µg of ascorbic acid) compared to standard ascorbic acid. In Table 3a and b, the hydroxyl radical scavenging activity of the test drug is recorded at 75.93%, while the standard ascorbic acid shows an inhibition level of 85.88% at a sample concentration of 200 µl and a wavelength of 695 nm. Additionally, in the superoxide free radical scavenging assay, 200 µl of the test drug demonstrates 93.33% inhibition, outperforming the standard ascorbic acid, which presents 77.64% inhibition at 560 nm. These results affirm the strong antioxidant properties of 'Kirambu Karpam'.

## Conclusion:

The outcomes of this study strongly suggest that the herbal formulation 'Kirambu Karpam' can serve as an effective antioxidant and potential food supplement within the pharmaceutical industry. The in vitro analysis reveals that the test drug is a significant source of natural antioxidants, potentially aligning with the Siddha principle of 'Kaaya Karpam' by preventing conditions such as Narai (whitening of hair), Thirai (skin shrinkage), Moopu (aging), Pini (disease), and Saakadu (death).

This study establishes that Krambu Karpam possesses significant antioxidant properties, supporting its traditional use in the Siddha system of medicine. The results emphasize the importance of further research to explore the potential therapeutic applications of Krambu Karpam in treating oxidative stress-related disorders and promoting overall health.

## References

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