International Journal of Recent Innovations in Academic Research
E-ISSN: 2635-3040; P-ISSN: 2659-1561
Homepage: https://www.ijriar.com/
Volume-9, Issue-3, July-September-2025: 167-173

Research Article

An Eco-Friendly Approach to Produce Dhoop Sticks by Utilizing Silkworms Litter and Assessment of Their Antimicrobial Activity

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Received: July 19, 2025 **Accepted:** August 09, 2025 **Published:** August 15, 2025

Abstract
Appropriate utilization of by-products is a multifarious approach in resource management that offers many advantages in any industry, including economic, environmental, and social advantages. Systematic utilization of waste generated in any activity reduces waste, creates new avenues for production of value-added products for boosting economic productivity, and reduces the environmental impact that contributes to the sustainable development of any business enterprise. The by-products of sericulture have several uses and generate additional income for the sericulture farmers. Litter of silkworm *Bombyx mori* is a valuable by-product that can be utilized as a nutritional supplement and in traditional medicine and also for the production of several value-added products. In view of the burning properties and sticky nature of the litter, it is proposed to develop dhoop sticks by adding the herbal ingredients along with cow urine to provide a refreshing environment. In view of the significance of by-product utilization, the researchers planned to convert silkworm litter, the waste generated during silkworm rearing, into a natural value-added product, i.e., dhoop sticks for the sustainable development of the sericulture industry, and to study the antimicrobial activity of the dhoop sticks for the green environment. Transfiguring litter of silkworm *Bombyx mori* to dhoop sticks signifies an optimistic innovation addressing the critical need for the effective utilization of by-products of the industry for sustainable sericulture development.
Keywords: Antimicrobial Activity, By-Product, Sustainable Development, Innovation.

1. Introduction

The silkworm *Bombyx mori* L. is a monophagous insect, which feeds on the leaf of the *Morus* species, commonly known as mulberry. Silk cocoons from this species are the source of commercial silk. Silkworm is one of the most economically important domesticated insects, which produces luxuriant silk thread in the form of cocoon by consuming mulberry leaves during larval period. In Indian context sericulture has many advantages like high employment potential, provides vibrancy to village economics, low gestation, high returns, women friendly occupation, ideal program for weaker section of the society and eco-friendly activity. By-products generated out of all the major sericultural activities play a significant role in generating additional income. Optimum utilization of sericultural wastes will generate handsome income for the sericulturists. This will help the sericulture industry stand on an economically sound footing. Many by-products presently discarded as waste can be put to better use for financial gains and generation of value-based products, thereby driving the industry to more profitable and economically viable position. The major by-products generated during silkworm rearing include excess of the harvested mulberry leaves accounting for 10-20%, unfed leaves comes around 20-30 percent, and silkworm litter and moulting skin.

Silkworm litter, the waste that is generated during the culturing of silkworms, has various usages, such as compost making, biogas production, as a substance for mushroom cultivation, etc. The litter (feces) of silkworms, which contains organic matter, can be used as a component in dhoop sticks. Incense sticks (dhoop sticks) can be produced by utilising stickiness and burning properties of silkworm excreta. Appropriate utilization of the litter can provide additional income to the sericulture practitioners during their leisure time. Twenty to twenty-five percent of the non-pathogenic microorganisms are omnipresent. The pathogenic microbes, additionally found in our surroundings, have the potential to cause diseases [1]. To reduce the pathogenic microbes in the environment, a number of conventional methods, including

dhoopana, have been employed in the fumigation process. The literature also explains that fumigation should be used for wound care and hospital operating room sterilization [2].

In the modern era, environmental issues have become a major concern. Human survival depends on a clean environment, which includes clean air, water, land, and energy. Airborne infections are caused by the presence of microbes in the air. Pathogens that cause diseases are transferred from one individual to another via coughing, talking, sneezing, breathing, and even laughing [3]. Although dhoopana is reported to be quite efficient in both preventive and curative aspects, there is still a need for improvement in this area because its practice is rarely employed. The current study aims to produce dhoop sticks made up of silkworm litter and herbs and evaluate the effectiveness of the dhoop sticks made against microorganisms. Ayurveda traditionally placed a strong emphasis on promoting and preserving people's health. Ayurveda functions by keeping a person's body, mind, soul, and spirit in harmony. According to Ayurveda, the fumigation technique dhoopana is a way to administer medication by inhalation. The dhoops developed smells good, natural and inexpensive. Therefore, the dhoop sticks can be used as an alternative to chemical-based dhoop sticks and synthetic air fresheners, in a variety of settings, including homes, offices, hospitals, etc.

Litter of silkworm *Bombyx mori,* is a valuable by-product that can be utilized for the production of value-added products. In view of the burning properties and sticky nature of the litter, it is proposed to develop dhoop sticks by adding the herbal ingredients along with cow urine to provide refreshing environment (Figure 1).

2. Materials and Methods

For the preparation of dhoop sticks, silkworm litter has been collected from the silkworm rearing laboratory of the Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam, Tirupati, Andhra Pradesh. Marigold flowers were collected from the herbal garden of the Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam, Tirupati. Desi cow urine was collected from TTD Goshala, Tirupati. Lemongrass oil, dasangam powder, camphor, cow ghee, and guar gum powder were purchased from the market. Silkworm litter-based dhoop sticks have been prepared based on sensory analysis. Based on the sensory scores, the ingredients and their proportions used for the preparation of the dhoop sticks are presented in the table below.

2.1. Preparation of Dhoop Sticks from Silkworm Litter

For the preparation of dhoop sticks, dried mulberry leaves and exuvia were separated from the silkworm litter collected and shade-dried. The shade-dried silkworm litter has been made into powder by using a mixer grinder. Collected marigold flowers were also shade-dried and then made into powder by using a mixer. Then all the ingredients were added in the proportions as presented in Table 1 in a systematic way. Then the mixture was loaded into dhoop stick moulds and air-dried at room temperature for 4 days. These dhoop sticks were stored in airtight containers for further use.

2.2. Evaluation of Dhoop Sticks for Antimicrobial Activity

The dhoop sticks' antimicrobial efficacy was assessed by using the approach suggested by Lad and Palekar [4] with few adjustments. For the study, 3 sets of petri plates were taken, and in each petri plate 20 ml of nutrient agar media was poured. After solidification, the petri plates with media were exposed to three different environmental conditions. The first set (2) of petri plates were kept open outside the laboratory, the second set was exposed to laboratory conditions, and the third one was kept open in the laminar flow for 10 minutes. After 10 minutes of exposure in three different environments, all the petri plates were kept in the sterilized laminar flow, and these petri plates were exposed to the fumes of the dhoop sticks prepared with silkworm litter for half an hour. After exposure to fumes of the dhoop sticks, they were incubated for 24 to 48 hours; each plate was incubated at 37 degrees Celsius. Then the microbial colonies were enumerated, and the results were reported as a percentage decrease in the microbial burden [4].

2.3. Sensory Evaluation of Dhoop Sticks

Sensory evaluation studies were carried out among 50 volunteers who have used the dhoop sticks and observed the smell, irritability, ease of using at home, ease of handling, and whether the product is eco-friendly and suitable for recommending.

2.4. Mosquito Repellent Activity

The spatial repellency method was followed to evaluate the dhoop sticks made up of silkworm litter, cow urine, and other herbs against mosquitoes. An insect cage was taken that contained 50 mosquitoes, then a

dhoop stick was burnt for 30 minutes, and the number of mosquitoes incapacitated was counted. The experiment was repeated thrice, the values were recorded, and the average of the paralyzed mosquitoes was enumerated.

3. Results

It is clear from the results (Table 2) of the current investigation that the dhoop sticks made from silkworm litter exhibit the antibacterial property. Plates exposed to the fumes of dhoop sticks displayed a reduced number of colonies in petri plates subjected to various environmental conditions, including outside the laboratory, in the main lab, and within a laminar air flow system, compared to the colonies that were not exposed to the fumes of the dhoop sticks (Figure 2: 2.1 to 2.4).

% Reduction of microbial load = Number of microbial colonies after exposure to dhoop/Number of colonies before exposure to dhoop x 100

3.1. Sensory Evaluation of Dhoop Sticks

The majority of the volunteers did not experience eye irritation during the study, and the majority said the dhoop fragrance was noticeable. Since the dhoop had demonstrated antibacterial activity, they all agreed that it looked appropriate and that they would like to use and suggest it (Table 3).

3.2. Mosquito Repellent Activity

The results (Table 4) indicate that 86 percent of mosquitoes exposed to vapors of the dhoop stick developed from silkworm litter for 30 minutes became immobilized and mutilated.

3.3. Mosquitoes Repellant Activity

The repellency activity by the dhoop sticks to the mosquitoes shown successful results when burning in a corner of home having mosquitoes which shows that natural insecticidal preparation are always effective than synthetic repellant. During the burning of dhoop it was shown that up to 85% of mosquitoes number was reduced. Lemon grass oil and camphor shows the maximum mosquito repellant activity.

4. Discussion

Only 20 to 25% of the microorganisms that are prevalent all around us are nonpathogenic. Our surroundings are also home to pathogenic bacteria that can lead to diseases. The fumigation process uses a number of traditional methods, including Dhoopana and Havana, to reduce the microbial burden in the environment to nonpathogenic levels. The literature also explains that fumigation can be used for wound care and hospital operating room sterilization [2]. One of the items that is frequently utilized in religious ceremonies in both rural and urban environs of India is dhoop. One of the key methods for reducing the microbial burden in particular locations that is outlined in the Indian ayurveda system is dhoopana. Our surroundings contain a variety of germs that can lead to a number of health issues. Dhoopana can help patients with swine flu, bronchial spasms, bronchitis, asthma, dyspnea, rhinitis, and foul mouth and nose odor.

Ayurveda claims that according to Sushrutha Samhita and Charka Samhita dhoopana, also known as fumigation, is used to treat a variety of illnesses and to disinfect abiotic surfaces and objects as well as the environment [5]. The results of the present investigation showed that exposure to dhoop in a variety of environments significantly reduced the microbial burden. A reduction (66.6%) of microbial colonies was recorded in the nutrient agar plates that were kept outside the laboratory and that were exposed to fumes of dhoop sticks developed in the study. A drop of fifty percent of microbial load was noticed in the microbial colonies on nutrient agar plates after exposure to smoke of the dhoop sticks in both the environments, i.e., in the laboratory and laminar flow. Similar observations were made by Prabhu et al. [6].

Sumitha and Prasad assessed their formulation DHUP's antibacterial efficacy, and the results showed that dhoopana had a strong antibacterial impact against bacteria and fungi during the investigation and up to six days of follow-up when compared to the control [2]. Godeshwar *et al.*, observed a consistent drop in microbial colonies in hospital kitchen samples on agar plates over the course of three days that were exposed to formaldehyde and dhoopsticks [7]. In the study, the dhoop sticks were made using ingredients such as silkworm litter, dasangam powder, turmeric, marigold petals, lemongrass oil, and cow products such as ghee and cow urine. All the components selected have health-promoting and therapeutic properties.

Silkworm excreta is comprised of chlorophyll, flavonoids, and organic compounds which have antioxidant and curative properties. Dasangam powder has a pleasant scent that helps improve sleep quality, reduce

stress and anxiety, and create a pleasant atmosphere. The active ingredient in turmeric, curcumin, has demonstrated antimicrobial properties [8]. Additionally, marigold oil has a strong, albeit dose-related, antifungal and insecticidal effect on *Odontotermes obesus* Rhamb. (Isoptera: Termitidae), the white termite [9]. Lemon grass oil (*Cymbopogon citratus*), has a reputation for being an efficient insect repellent. Studies have shown that it also works well to keep mosquitoes away [10, 11]. Gomutra, or cow urine, is a traditional ingredient in the production of dhoop (incense), especially in India, where it is thought to have aromatic and cleansing qualities.

Herbal dhoop was made for the current study using silkworm litter, dasangam powder, turmeric, marigold petals, lemon grass oil and cow products such as ghee, and cow urine. The antibacterial activity of the dhoop against the airborne microorganisms was further assessed. Good antibacterial action against airborne microorganisms has been discovered in our study. After exposing the plates to dhoop in laminar airflow, we have noticed a considerable decrease in the microbial load of 66.12% in the environment, 55.38% in the main lab, and 60% in the LAF chamber. During mosquito repellent activity it was observed that up to 80 % of mosquitoes number was reduced. Earlier studies shows that lemon grass oil [12-14] and camphor shows the maximum mosquito repellant activity [15].

5. Conclusion

After the air was fumigated by burning organic dhoop sticks, the formulated dhoop sticks demonstrated a significant decrease in the microbial count of the air. The findings indicate that the microbial burden in the investigated environment may be decreased by the dhoop sticks potential antibacterial action. Human accessibility was also established by sensory examination. This herbal dhoop has a pleasant scent and is made from inexpensive, readily accessible raw materials like silkworm litter. In addition, it is inexpensive, non-toxic, and simple to make, making the herbal dhoop a viable alternative to the riskier fumigation techniques.

Declarations

Acknowledgments: The authors gratefully acknowledge that the funding for this publication was provided by the Pradhan Mantri Uchchatar Shiksha Abhiyan (PM-USHA), under the Multi-disciplinary Education and Research Universities (MERU) Grant Sectioned to Sri Padmavati Mahila Visvavidyalayam, Tirupati.

Author Contribution: All authors equally contributed in designing the experiment, collection of data, data analysis, overall research work and preparation of manuscript. All authors reviewed and approved the final manuscript.

Conflict of Interest: The authors report no financial or any other conflicts of interest in this work.

Consent to Publish: The authors agree to publish the paper in International Journal of Recent Innovations in Academic Research.

Data Availability Statement: All the data is available with the authors and shall be provided upon request.

Funding: The funding for this publication was provided by the Pradhan Mantri Uchchatar Shiksha Abhiyan (PM-USHA), under the Multi-disciplinary Education and Research Universities (MERU) Grant Sectioned.

Institutional Review Board Statement: This study does not involve experiments on animals or human subjects.

Informed Consent Statement: Not applicable.

Research Content: The research content of manuscript is original and has not been published elsewhere.

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Citation: Shanthi Sree, K.S., Shobha Rani, A., Sujathamma, P. and Savithri, G. 2025. An Eco-Friendly Approach to Produce Dhoop Sticks by Utilizing Silkworms Litter and Assessment of Their Antimicrobial Activity. International Journal of Recent Innovations in Academic Research, 9(3): 167-173.

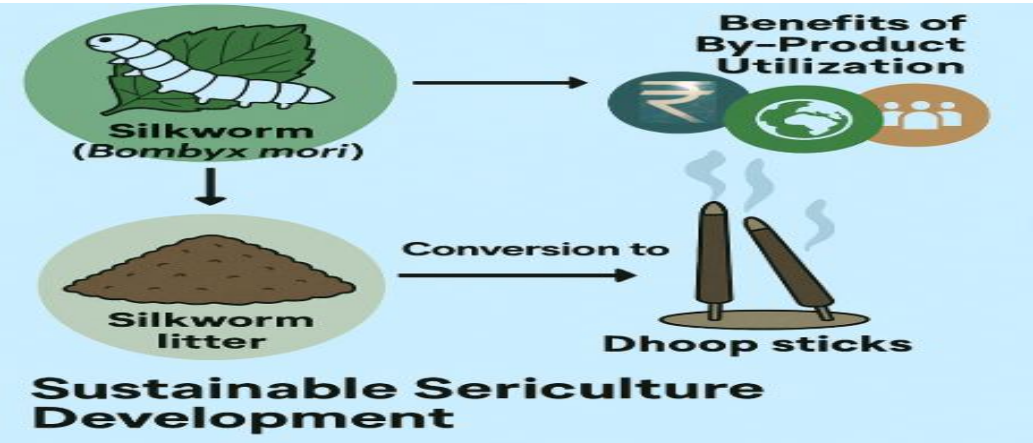

Figure 1. Sustainable sericulture development (Author view).

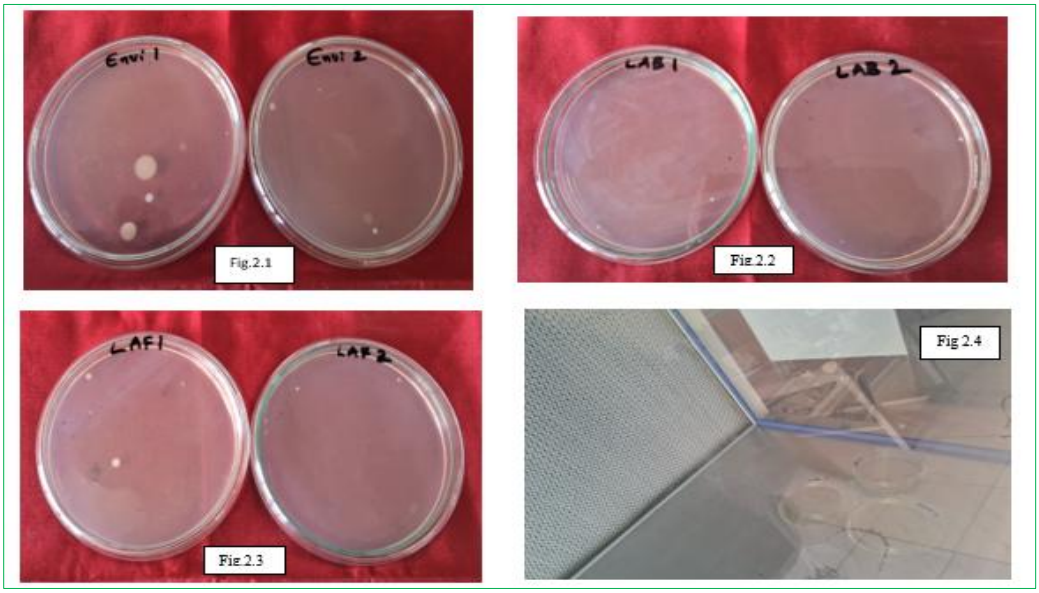

Figure 2. 2.1: Colonies on agar plates (environment) before exposure of plates to dhoop and after exposure of plates to dhoop; **2.2:** Colonies on agar plates (main lab) before exposure of plates to dhoop and after exposure of plates to dhoop; **2.3:** Colonies on agar plates (LAF room) before exposure of plates to dhoop and after exposure of plates to dhoop; **2.4:** Exposure of agar plates to dhoop in laminar air flow.

Table 1. Composition of dhoop sticks.

S/N	Ingredients	Percentage (%)
1	Silkworm litter	26
2	Marigold flowers (petals)	6
3	Lemon grass oil	20
4	Dasangam powder	18
5	Desi cow urine	8
6	Camphor	6
7	Ghee	13
8	Guar gum powder	4

Table 2. Antimicrobial effect of prepared dhoop.

S/N	Area	Microbial colonies on nutrient agar plates without exposure to dhoop	Microbial colonies on nutrient agar plates after exposure to dhoop	Reduction of microbial load (%)
1	Environment	06	04	66.66
2	Main Lab	02	01	50.00
3	Laminar air flow	04	02	50.00

Table 3. Sensory evaluation of dhoop sticks.

S/N	Details	Yes	No	Percentage (%)
1	Is the smell appreciable	50	00	100
2	Smoke is irritating	49	01	98
3	Use the product at home	49	01	98
4	Will you recommend the product	50	00	100
5	Ease of handling	50	00	100
6	Ease product ecofriendly	50	00	100

Table 4. Mosquito repellent activity.

S/N	Number of mosquitoes	Number of mosquitoes incapacitated	Percentage of mosquitoes incapacitated
1	50	43	86