

Sustainable Grain Protection: Combatting Rice Weevils (*Sitophilus oryzae*) with Natural Plant Powders

Abstract

Grain storage plays a pivotal role in ensuring food security and sustaining agriculture for future generations. However, a wide array of insect pests, including the rice weevil (*Sitophilus oryzae*), pose a significant threat to stored grains. This study was conducted at the Laboratory of the Department of Seed Science and Technology, Bangladesh Agricultural University, Mymensingh, from October 2019 to March 2020, to evaluate the efficacy of plant-based powders for managing rice weevils. The experiment was designed using a Completely Randomized Design (CRD) with three replications and seven treatments. These treatments included five botanical powders—*Ocimum tenuiflorum* (Tulsi), *Azadirachta indica* (Neem), *Polygonum hydropiper* (Biskatali), *Nicotiana tabacum* (Tobacco), and *Lantana camara* (Lantana)—applied at three dosage levels: 1 g, 3 g, and 5 g per 100 g of grain. Additionally, a chemical insecticide (Sevin 85 SP @ 0.25%) and an untreated control were included for comparison. Results revealed that neem leaf powder was the most effective, achieving the highest adult insect mortality and the lowest adult emergence. In contrast, Lantana powder exhibited the least efficacy, allowing 94.38% more adult insects to emerge compared to neem powder. Untreated grains experienced a 20% weight loss due to rice weevil infestation after one month of storage, whereas treatment with 5% neem leaf powder reduced weight loss to a mere 0.14%. This reduction was statistically significant compared to all

other treatments. These findings suggest that neem leaf powder at 5% is a highly effective, eco-friendly, and safe alternative to chemical insecticides for controlling rice weevils in stored grains.

Keywords: Rice weevil, *Ocimumtenuiflorum* (Tulsi), *Azadirachta indica* (Neem), *Polygonum hydropiper* (Biskatali), *Nicotiana tabacum* (Tobacco), and *Lantana camara* (Lantana), Sevin 85 SP

1. Introduction

Grain storage is crucial for ensuring food security, as it safeguards the essential resources required for human survival. Proper preservation techniques are indispensable to meet future food demands, particularly during disasters or famines. These techniques also support sustainable development and humanity's future [1]. Consequently, the preservation of stored grains is of paramount importance, not only for ensuring food security but also for mitigating economic losses caused by insect pest infestations.

Among stored grain pests, the rice weevil (*S. oryzae*) is a significant challenge to the post-harvest management of grains worldwide [2]. For instance, a study conducted in Benin revealed that weight loss after six months of storage ranged from 5.47% in the south to 4.07% in the center, and 1.64% in the north [3]. Similarly, other studies reported an average grain weight loss of 13.63% and physical grain damage of 7.22% [4]. Infested rice grains not only suffer quantitative losses but also experience a significant reduction in carbohydrate content (from 68.15% to 58.43%) and alterations in mineral content, thereby diminishing their nutritional value [5].

To address these challenges, synthetic chemical pesticides, such as organophosphates and carbamates, have been widely used for many years to control stored grain pests. However, their extensive use has resulted in several problems, including the development of pest resistance, environmental degradation, and health risks to humans [6]. For example, phosphine and chlorpyrifos-methyl, commonly used for stored grain pest management, can adversely affect human health and contribute to environmental pollution [7]. Moreover, over-reliance on synthetic insecticides has exacerbated environmental damage and resistance among pest populations, further complicating pest management strategies [8]. These issues are particularly

severe in developing countries, where stored grain pests contribute significantly to economic losses[9].

Given these challenges, there is an urgent need for alternative pest management strategies that are readily available, cost-effective, less toxic, and environmentally friendly [10]. Botanical insecticides offer a promising solution, as they are biodegradable, non-residual, equally effective, and widely available. Furthermore, they have the potential to control storage pests without compromising grain or seed quality or harming the environment [11]. For example, neem leaf powder demonstrated a mortality rate of up to 90% in rice weevils, significantly higher than other treatments [12]. Among neem's bioactive compounds, azadirachtin stands out for its multifaceted insect control mechanisms. For instance, it deters feeding by insects, disrupts their growth and molting processes, causing developmental delays and premature death, and interferes with their hormonal systems, hindering growth and reproduction [13,14,15]. Additionally, azadirachtin functions as a natural insect repellent, further reducing the likelihood of infestations [16]. Despite these advantages, the development of botanical pesticides has been historically overlooked, with research and development focusing predominantly on synthetic pesticides. Additionally, previous studies have largely tested botanical insecticides at single concentrations, limiting the understanding of their dose-dependent efficacy. The lack of research on varying doses creates a critical gap in determining optimal application levels for maximum effectiveness. Exploring this gap is essential to refine botanical pest management strategies and promote their practical adoption in diverse storage conditions.

Therefore, this study was undertaken to assess the efficacy of selected medicinal plant powders as protectants of rice grains against infestation by *S. oryzae*. By exploring multiple doses of botanical powders, this research aims to contribute to the development of safer and more sustainable pest management practices.

2. Materials and methods

2.1 Experimental Site

The study was conducted in the Laboratory of the Department of Seed Science and Technology at Bangladesh Agricultural University, Mymensingh, from October 2019 to March 2020. The experimental site is geographically located at a latitude of 24°43'26" N and a longitude of 90°25'48" E.

2.2 Experimental Design and Treatments

The experiment was conducted using a Completely Randomized Design (CRD) with three replications. A total of five plant-based repellent powders at 3 different doses and one chemical insecticide were used as treatments, as detailed in Table 1.

The leaves of the plant materials were collected from the botanical garden of Bangladesh Agricultural University, Mymensingh. The collected samples were thoroughly washed and subsequently air-dried using an oven. Once dried, the samples were ground into fine powder using an electric grinder to prepare the treatments.

Table 1. Experimental plant powders and application doses

Powders used	Family name of the plant	Plant parts used	Doses (g/100 g grain)
<i>Ocimum tenuiflorum</i> (Tulasi)	Lamiaceae	Leaf	1,3,5
<i>Azadirachta indica</i> (Neem)	Meliaceae	Leaf	1,3,5
<i>Polygonum hydropiper</i> (Biskatali)	Polygonaceae	Leaf	1,3,5
<i>Nicotiana tabacum</i> (Tobacco)	Solanaceae	Leaf	1,3,5
<i>Lantana camara</i> (Lantana)	Verbenaceae	Leaf	1,3,5
Sevin 85 SP			0.25

2.3 Collection, Culturing of *S. oryzae*, and Experimental Procedure

Infested rice grains containing *S. oryzae* were collected from the Agricultural Farm of Bangladesh Agricultural University, Mymensingh. Male and female insects were separated using a magnifying glass and a simple microscope. The insects were then reared in the laboratory of the Department of Seed Science and Technology under controlled conditions of 27–30°C temperature and 70–75% relative humidity. To establish the culture, 10 pairs of adult rice weevils were introduced into sterilized jars containing rice grains pre-treated at 60°C for 30 minutes to ensure sterility. The jars were covered with cheesecloth secured by rubber bands to prevent contamination and insect escape. The weevils were allowed to oviposit freely for seven days, after which the adults were removed. The jars were then placed in a growth chamber for 30 days to allow eggs to develop into adults. Emerging adults were collected by sieving and sorted as one-day-old adults. These were regularly transferred to separate jars with fresh rice grains and maintained under the same environmental conditions. For the experiments, three to seven-day-old adult insects were selected.

2.4 Data Collection and Calculation

Data were recorded on the following parameters: percent mortality, adult emergence, and seed weight loss.

2.5.1 Percent mortality

The number of dead and alive insects was monitored regularly, and mortality was recorded at 7, 14, 21, and 28 days after treatment (DAT). The percentage of weevil mortality was calculated using the following formula, as described by [17]:

$$\% \text{ Weevil mortality} = \frac{\text{Number of dead insects}}{\text{Total Number of insects released}} \times 100$$

2.5.2 Percent of Seed Weight Loss

The final weight of the seeds was measured to determine the weight loss. Clean grains, excluding those with visible holes, were weighed separately for each Petri dish. The percentage of weight loss was calculated using the formula provided by [18]:

$$\text{Seed weight loss (\%)} = \frac{\text{Initial weight of seeds} - \text{Final weight of seeds}}{\text{Initial weight of seeds}} \times 100$$

2.5.3 Adult Emergence

The counting of emerged adults began 7 days after the release of insects and continued daily from the first day of emergence until the final day. After each count, the adults were removed from the plastic jars to prevent further egg-laying.

2.6 Statistical analysis

The data obtained from the experiments were statistically analyzed using a one-factor Completely Randomized Design (CRD) with the help of RStudio software (version: 2024.04.0+735). Mean separation was performed using Duncan's Multiple Range Test (DMRT) and Least Significant Difference (LSD) tests, where applicable, at a 5% level of probability.

3. Results and Discussion

3.1 Efficacy of Different Plant Powders on Mortality of Rice Weevil Under Laboratory Condition

Significant variations were observed in the mortality of *S. oryzae* due to the application of botanicals and one synthetic insecticide (Table 2). These findings align with the observations of [19]. Among the botanical powders, neem powder at 5% concentration induced the highest mortality, a result consistent with [20], who reported that neem-based pesticides exhibited superior performance, achieving a mortality rate of 66.67% compared to the untreated control. [21] also reported that among the treatments, neem oil achieved the highest mortality rate, with 83.33%. This clearly highlights the potential of neem as an effective grain protectant, as also demonstrated by [22].

Furthermore, [23] observed that neem powder led to a mortality rate of 45% in certain cases while significantly mitigating rice damage. Complementing this, [22] suggested that neem powder could achieve up to 40% mortality in rice weevils within 48 hours when applied at specific concentrations (6g and 12g per 20g of rice). This efficacy can be attributed to the bioactive compounds in neem (*Azadirachta indica*), particularly azadirachtin. According to [24], azadirachtin disrupts the pest's physiological and reproductive processes, including protein synthesis and hormonal regulation, thereby impairing reproduction and growth. [25] also emphasized that azadirachtin interferes with ecdysone, a hormone critical for insect development, thus disrupting their lifecycle.

In addition, azadirachtin, abundantly present in neem kernels, acts as a natural antifeedant, sterilant, and insect growth regulator. As noted by [26], this compound reduces insect feeding, mating, molting, and fecundity, further enhancing its effectiveness as an eco-friendly insecticidal agent. Furthermore, azadirachtin disrupts insect development by inhibiting prothoracicotropic and allatotrophic hormones, impairing ecdysone and juvenile hormone synthesis needed for molting [27]. In *Spodoptera frugiperda*, it reduces HR3 expression, blocking ecdysis, and suppresses apoptosis-related genes, causing tissue damage and halting growth [28,29]. Taken together, these findings underscore the role of neem powder as a sustainable solution for managing *S. oryzae* infestations in stored grains.

Table 2. Efficacy of Different Plant Materials on Mortality (%) of *S. oryzae* under laboratory condition

Treatments	Doses (g/100g)	Mortality (%)			
		7DAT	14DAT	21DAT	28DAT
Biskatali	1	16.34e	23.64f	46.34e	56.34g
	3	20d	40c	55.44d	64.34e
	5	33.64a	50b	70b	94.64b
Lantana	1	0i	13.64h	26.34j	33.64m
	3	13.64f	23.64f	40g	40k
	5	20d	36.34d	56.34d	61.64f
Neem	1	20d	26.34e	46e	56.34g
	3	23.64c	40c	61.34c	78c
	5	30b	53.64a	81.34a	90a
Tobacco	1	6.34g	16.34g	30i	36.34l
	3	20d	23.64f	43.64f	51.34i
	5	30b	40c	56.34d	74d
Tulsi	1	3.66h	13.66h	36.34h	43.67j
	3	16.35e	26.34e	46.35e	53.64h
	5	20d	36.34d	60c	63.34e
Sevin 85 SP	0.25	40a	60.33a	86.55a	93.45a
Control	-	1.66h	1.66i	3.34k	4.66n
SEm(±)	-	0.59	2.96	0.66	0.43
CV (%)	-	5.67	0.53	2.26	1.28
Level of significance	-	***	***	***	***

Means with the same letters or without letters within the same column do not differ significantly.

*** = Significant at 0.1 % level of probability($P < 0.001$); SEM= Standard Error of Mean, CV= Coefficient of Variance; DAT= Days After Treatment

3.2 Evaluating the Combined Effect of Chemical and Botanical Treatments on Seed Weight Loss Caused by Infestation

The results demonstrated a significant reduction ($P < 0.001$) in the percentage of weight loss of grains infested by *S. oryzae* when treated with various plant powders and chemical insecticides, compared to the control, across different powder concentrations (Figure 1; Appendix 1). This finding was consistent with the observations of [30]. The minimum weight loss occurred when the insects were exposed to neem powder, which reduced grain loss by 91.89% compared to the control. This reduction was significantly lower among all botanical treatments and statistically comparable to Sevin 85 SP.

[23] similarly observed that neem powder substantially reduced rice damage, with weight loss as low as 3.14% in treated samples compared to untreated grains, which experienced over 21% weight loss. Additionally, [31] reported that neem-treated unhusked rice grains exhibited weight losses as low as 16.67%, compared to losses exceeding 70% in untreated samples over a three-month storage period. This effectiveness was attributed to the bioactive compounds in neem, such as azadirachtin, salannin, and nimbin, which interfered with the weevil's ability to feed and reproduce, disrupted protein synthesis, altered biological fitness, impaired sexual communication, and inhibited chitin synthesis, as noted by [25].

Furthermore, [32] reported that neem compounds disrupted the pest's hormonal and digestive systems, leading to reduced grain consumption and delayed population growth. These findings underscored the efficacy of neem as a sustainable and potent grain protectant against *S. oryzae*.

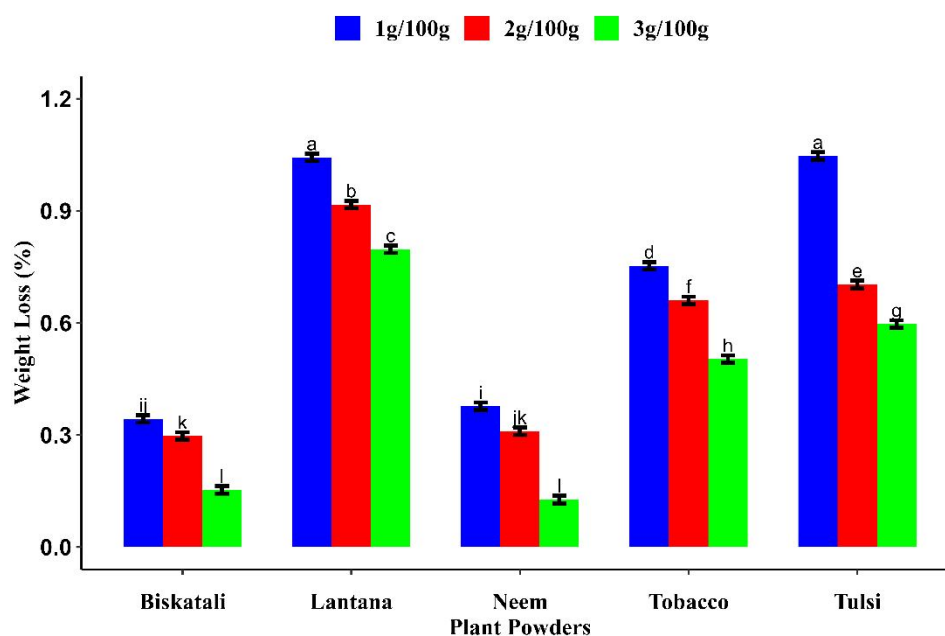


Figure 1. Efficacy of three different application rates of botanical powders in reducing seed weight loss caused by *S. oryzae* infestation (Data are presented as mean \pm SEM; $n = 3$; $P < 0.001$).

3.3 Effect of Different Treatments on Adult Emergence

Figure 2 and Appendix 1 depict the number of adults that emerged 28 days after treatment with various botanicals and one chemical insecticide. The results exhibited notable variations among the different doses of powders ($P < 0.001$). The untreated control treatment recorded the highest number of adult emergences (56.57), which was significantly greater than all other treatments. Following this, Lantana powder applied at 1% exhibited the second-highest value, with 41.33 adults emerging.

In contrast, the lowest number of adults was observed in the neem powder treatment at 5%, which reduced adult emergence by approximately 95.88% compared to the control. This outcome was statistically comparable to the Sevin 85 SP treatment. According to [23], neem leaf powder significantly suppressed adult rice weevil (*S. oryzae*) emergence, yielding the lowest survival rates of 78.75% in local rice and 55% in Thailand rice. Similarly, as noted by [33], neem

leaf powder at concentrations of 1%, 2%, and 3% killed 61.13%, 68.76%, and 77.75% of adult weevils, respectively.

This reduction in adult insect emergence can be attributed to the active compound azadirachtin present in neem powder, which, as explained by [34], disrupts insect growth and development by affecting larval stages. It inhibits successful maturation into adults by interfering with protein synthesis and chitin formation, both of which are essential for insect growth and development. Furthermore, as supported by [25], azadirachtin plays a crucial role in impeding the life cycle of weevils, thereby reducing their survival rate.

Additionally, [35] suggested that neem-based products effectively suppressed adult insect emergence due to their toxic effects. These toxic effects not only caused mortality but also led to deformities in emerging adults, which disrupted their survival and reproductive abilities. Consequently, this significantly decreased progeny production and infestation levels in treated grains.

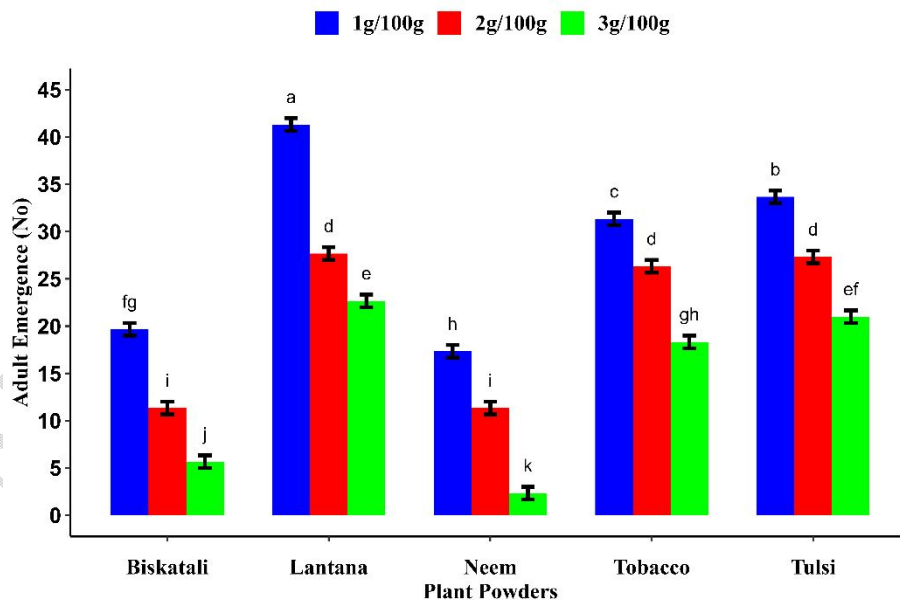


Figure 2. Influence of various plant powders at different application rates on the emergence of *S. oryzae* adults (Data are presented as mean \pm SEM; n = 3; $P < 0.001$).

5. Conclusion

The findings of this study highlight the effectiveness of botanical insecticides in enhancing adult mortality, suppressing adult emergence, and minimizing percentage weight loss in stored grains. However, among the tested botanicals, neem leaf powder (*Azadirachta indica*) demonstrated superior performance, making it a promising candidate for sustainable post-harvest grain protection. The insecticidal properties and the widespread availability of these botanicals present an opportunity to improve traditional storage practices, especially in resource-limited settings. Future research should focus on optimizing application methods and determining appropriate concentrations of *A. indica* extracts for various storage materials and environmental conditions. Additionally, investigating the residual effects and long-term storage stability of these treatments would provide critical insights into their feasibility and scalability for broader adoption in grain storage systems.

Disclaimer (Artificial intelligence)

Option 1:

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

6. References

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Appendix 1

Treatments	Doses (g/100g)	Weight Loss (%)	No. of adult emergence
Biskatali	1	0.343ij	19.67fg
	3	0.297k	11.33i
	5	0.153l	5.67j
Lantana	1	1.043a	41.33ab
	3	0.917b	27.67d
	5	0.797c	22.67e
Neem	1	0.377i	17.33h
	3	0.31jk	11.33i
	5	0.127l	2.33k
Tobacco	1	0.753d	31.33c
	3	0.66f	26.33d
	5	0.503h	18.33gh
Tulsi	1	1.047a	33.67b
	3	0.703e	27.33d
	5	0.597g	21ef
Sevin 85 SP	-	0.102l	2.02k
Control	-	1.567a	56.57a
SEm (±)		0.01	0.67
CV (%)		4.11	5.49

Means with the same letters or without letters within the same column do not differ significantly.
*** = Significant at 0.1 % level of probability ($P < 0.001$); SEM= Standard Error of Mean, CV= Coefficient of Variance; DAT= Days After Treatment

UNDER PEER REVIEW