



Behavioral Responses of *Rhyzopertha dominica* to Different Food Bait Attractants in Paddy Storage Godown

M. Sathiyaseelan and K. Balaji

Department of Agricultural Engineering, Sethu Institute of Technology, Virudhunagar- 626 115, India
E-mail: sathiyaseelanm1998@gmail.com

Abstract: This study aimed to evaluate behavioural response of *Rhyzopertha dominica* to crushed groundnut, wheat flour, cracked corn, sorghum flour, rice flour, pearl millet flour, rice bran, rice bran + rice flour, cracked sorghum and control were tested. It showed that wheat flour, cracked sorghum and pearl millet flour were found to be the more attractive. Cracked sorghum exhibited the highest attraction levels, with 26.11% at 5 days after placement (DAP). Wheat flour showed a relatively higher attraction, with 12.79% at 10 DAP. The attraction index is 11.25% at 15 DAP, over the observation period in pearl millet flour. Overall, the results indicate that cracked sorghum (85.07%), wheat flour (57.41%) and pearl millet flour (48.04%) are particularly attractive to *R. dominica*, while other food sources exhibit varying levels of attractiveness. The bait effectiveness was confirmed through testing, revealing varying ratios. Notably, the chamber retained the highest number of *R. dominica* which contained 2:1:1 ratio of wheat flour: sorghum flour: pearl millet flour. The effective baits were also test verified through four-arm olfactometer and found the highest orientation in the arm containing wheat flour and sorghum flour.

Keywords: *Rhyzopertha dominica*, Wheat flour, Sorghum flour, Pearl millet flour, Paddy storage godown

Rice is one of the crucial food crops for over half of the global population. The impact of insect-related losses in storage significantly affects food availability. Most commonly the insects, mites, birds, rodents, fungi and moisture are the major problems in storage godowns. Adult beetles, especially stored product insects, exhibit a propensity for seeking shelter in the cracks and crevices of warehouses and storage godowns due to their harborage seeking behavior. Worldwide, chemical control stands out as the most commonly utilized method for managing pests of stored products (Fields and White 2002; Nayak et al., 2020). However, resistance among several species of stored product insects to conventional pesticides has increased over the last few decades (Hagstrum and Phillips 2017, Nayak et al., 2020). Certain populations of the lesser grain borer, *Rhyzopertha dominica* (Coleoptera: Bostrychidae), have been discovered to exhibit over a 1500-fold increase in resistance to the fumigant phosphine compared to susceptible strains (Opit et al., 2012). *R. dominica* has been documented as a significant pest of stored foods on a global scale, capable of infesting fresh and processed food resources derived from 53 plant species, which can be categorized into 31 families (Edde 2012, Buonocore et al., 2017, Dissanayaka et al., 2020). *R. dominica* feeds on whole kernels of stored cereals, in addition to other commodities such as legumes and tubers. The extent of feeding and damage inflicted by *R. dominica* varies across different types of stored products (Cinco-Moroyoqui et al., 2006, Naseri and Majd-Marani 2022). In principle, this species is regarded as a

particularly significant pest of stored cereals, such as wheat, barley, rice and to a lesser extent in maize (Edde 2012). The process of host selection in insects typically involves chemical stimuli, such as host plant volatiles. Olfactory cues play crucial roles in food-searching by stored product insects, offering vital information to discriminate among different substrates (Hagstrum and Phillips 2017). In this study, the behavioral responses of *R. dominica* to volatiles from different grain commodities, i.e. crushed groundnut, wheat flour, cracked corn, sorghum flour, rice flour, pearl millet flour, rice bran, rice bran + rice flour, cracked sorghum and control (without bait) were assessed in storage godown and four arm olfactometer bioassays. Hence, this study will prove valuable for the advancement of new attractants by enhancing the efficacy of pheromone lures with volatile semiochemicals.

MATERIAL AND METHODS

Bait traps, filled with attractants, are strategically positioned between the stacked bags in the godown. Designed with a 4mm entrance, they guide insects into the lower polyethylene receptacle. The trap catches were documented at intervals of 5, 10, 15, 20 and 25 days after the placement (DAP) of baits. The quantity of insects captured in the control group, lacking bait material, was juxtaposed with the count of insects captured in the other treatment groups. The trap catches were recorded on 5, 10, 15, 20, 25 days after placement of baits. The number of insects caught in control (without bait material) was compared with the number of insects caught in other treatments (Sathiyaseelan et al.,

2022). The attraction index was calculated by using the formula (Smith et al., 1993).

$$\text{Attraction Index} = \frac{T-C}{N} \times 100$$

T- Number of insects drawn to the treatment.

C- Number of insects drawn to the control

N - Total number of individuals

Rearing of test insects: Adults of the lesser grain borer, *R. dominica*, were reared in plastic jars using wheat flour and grains as their diet. Each jar contained 20 to 30 pairs of insects and 250 g of grains, with a piece of kada cloth secured on top with rubber bands. The rearing environment maintained a controlled setting with a 12:12 hour light: dark photoperiod, temperatures ranging between 26 and 28°C, and relative humidity levels maintained at 60 to 65 percent. All experiments were conducted under uniform conditions to ensure consistency in culture maintenance.

Trapping efficiency of different food bait mixture: Depending on bait attractiveness, three promising bait sources were selected for further studies. An experimental chamber was arranged with four numbers of container 150 g capacity attached to two litre containers at the centre through PVC pipes with an angle of 45°. The selected food bait materials viz. wheat flour, sorghum flour and pearl millet flour were mixed at the ratios of 1:1:1, 2:1:1, 1:2:1, 1:1:2 (20g each) and filled in the chambers. The test insect *R. dominica* (700) were released in the central arena and the movement of insects towards the food bait mixtures was observed 24 hours after release (HAR).

Behavioural / Orientation studies: The test insects, including the lesser grain borer, *R. dominica*, underwent a 24h period of starvation in Petri plates prior to initiating the olfactory bioassay. Fifty adult insects, their sexes undisclosed, were introduced into the central chamber of the olfactometer, where a 7mm aperture was present. The setup was covered with cloth to reduce the insect's attraction to light. At 5, 10, 15, 20, 25 Minutes After Release (MAR), the location of the *R. dominica* was observed (Vijay et al., 2020). Each treatment was replicated 5 times. The reaction of *R. dominica* was evaluated using wheat flour, sorghum flour and pearl millet flour.

Statistical analysis: The data on attraction index and behavioural response/orientation of the *R. dominica* beetle were statistically analysed using completely randomized design by one-way ANOVA subjecting the data to arcsine transformation and were separated by using Duncan's multiple range test (DMRT) with IBM SPSS statistics 22.0 software and differences were regarded as significant at $p < 0.05$.

RESULTS AND DISCUSSION

Different food baits used to study the attraction index for

trapping *R. dominica* revealed that cracked sorghum was the most effective, achieving an attraction index of 26.11% at 5 DAP. Bait traps filled with wheat flour and a mixture of rice bran and rice flour collected 12.79% and 10.94% respectively. The catches for cracked corn and pearl millet flour were 8.86% and 7.73%, while rice bran was the least effective, attracting only 3.89%. By 10 DAP, the attraction index for *R. dominica* was in descending order, with cracked sorghum at 19.41%, followed by wheat flour at 12.79%, and cracked corn at 10.46%, which was comparable to rice bran + rice flour at 10.37%. Crushed groundnut (7.9%), pearl millet flour (7.53%) and sorghum flour (7.17%) also attracted the pests, while rice bran remained the least effective at 3.97%. At 15 DAP, the highest trap catches were recorded for cracked sorghum at 14.66%, followed by wheat flour at 12.76% and pearl millet flour at 11.25%. The lowest catches continued to be observed in rice bran and rice flour. At 20 DAP, cracked sorghum again showed the greatest effectiveness with 12.92%, followed by pearl millet flour (11.15%) and wheat flour (10.82%). Rice bran continued to yield the lowest catches. Finally, at 25 DAP, cracked sorghum recorded the highest trap catches at 11.97%, closely followed by pearl millet flour (10.38%), crushed groundnut (10.35%) and wheat flour (9.52%). Once again, rice bran and rice flour showed the least effectiveness. In total, cracked sorghum achieved an impressive attraction index of 85.07%, followed by wheat flour (57.41%), pearl millet flour (48.04%) and rice bran + rice flour (47.22%), with rice flour having the least attraction at 26.48% (Table 1).

Wheat flour are more attractive than other flours, while millet flour or cracked millet volatiles are more attractive than whole millet kernels Sathiyaseelan et al. (2023). The orientation behavior of the lesser grain borer, *Rhyzopertha dominica*, larger grain borer, *Prostephanus truncatus* and maize weevil, *Sitophilus zeamais* was studied in response to crushed white maize (food odor) combined with color cues (white, yellow, blue, green). While *S. zeamais* responded to both color and odor stimuli, *R. dominica* and *P. truncatus* showed no response (Arnold et al., 2015). The behavioral responses of the lesser grain borer, *Rhyzopertha dominica* and the red flour beetle, *Tribolium castaneum* to sorghum, wheat, and cotton seeds were observed in the field, both near and away from the storage godown. It was found that *T. castaneum* was more attracted to cottonseed than to sorghum or wheat, while *R. dominica* showed a preference for wheat and did not respond to cottonseed (Ahmad et al., 2013).

The *R. dominica* (180 Nos.) 2:1:1 ratio was found to be highly attractive followed by 1:1:1 (165 Nos.) and 1:1:2 (162 Nos.) and both the bait ratio was on par with each other. The

less numbers of *R. dominica* (158 Nos.) retained in 1:2:1 ratio (Table 2). *Rhyzopertha dominica* spent more time inside and entered more areas containing both winter wheat and maize, spending more time in and making more entries into the zones containing these seeds (Ukeh and Umoetok 2007).

Orientation of stored product insect in four-way olfactometer: Olfactometer bioassay revealed the significant variations on orientation/behavioural response of *R. dominica* towards wheat flour, sorghum flour, pearl millet flour and control (without food bait) in a four-arm olfactometer. The olfactometer bioassay demonstrated significant variations in the orientation and behavioral responses of *Rhyzopertha dominica* towards wheat flour, sorghum flour, pearl millet flour and a control (without food bait) in a four-arm olfactometer. At 5 minutes after release (MAR), the highest orientation was recorded towards wheat flour at 24%, which was significantly greater than the other flours. This was followed by sorghum flour at 17.3% and pearl millet flour at 16.6%. At 10 MAR, wheat flour continued to attract the most *R. dominica*, with an orientation of 32.1%, followed by sorghum flour at 28.1%. By 15 MAR, the maximum orientation shifted slightly, with wheat flour reaching 36.2%, followed by pearl millet flour at 28.2% and sorghum flour at 22.2%. At 20 MAR, *R. dominica* showed a strong preference for wheat flour at 30.1%, with sorghum

flour following at 26.1%. Finally, at 25 MAR, 38.6% of *R. dominica* settled in wheat flour, succeeded by sorghum flour at 26.6% and pearl millet flour at 18.6% (Table 3).

The four-way olfactometer is utilized to investigate the behavior of *Rhyzopertha dominica* (Bashir et al., 2003). Vijay et al. (2020) reported that the highest orientation of *S. oryzae* was recorded towards sorghum (53.33% and 48.67%) in 20 MAR. According to Trematerra et al. (2000), *O. surinamensis*, *T. castaneum*, and *T. confusum* use grain volatile odours to determine whether stored wheat grain kernels have been damaged mechanically or by insects and these studies are

Table 2. Effect of bait source ratio on the arrest of insect movement (wheat flour: sorghum flour: pearl millet flour)

Bait ratio	Number of percent recaptured*
1:1:1	165±3.9863 ^b
2:1:1	180±5.5113 ^a
1:2:1	158±3.2251 ^c
1:1:2	162±1.7636 ^b
Total	665
Not responded	35

* Based on 700 insects
Means followed by the same letter (s) in a column are not significantly different by DMRT (P=0.05)

Table 1. Comparative response of *Rhyzopertha dominica* to various food attractants

Attractants	Relative attraction index (%)					Total attraction (%)
	5 DAP *	10 DAP	15 DAP	20 DAP	25 DAP	
Crushed groundnut	5.34±0.053 ^g	7.9±0.116 ^d	9.29±0.016 ^e	9.66±0.108 ^d	10.35±0.256 ^b	42.54
Wheat flour	11.57±0.079 ^b	12.79±0.305 ^b	12.76±0.122 ^b	10.82±0.317 ^c	9.52±0.291 ^c	57.41
Cracked corn	8.86±0.006 ^d	10.46±0.142 ^e	8.84±0.180 ^f	8.73±0.042 ^e	8.46±0.248 ^e	45.35
Sorghum flour	6.29±0.120 ^f	7.17±0.117 ^f	9.41±0.275 ^e	8.77±0.149 ^e	8.79±0.084 ^d	40.43
Rice flour	4.54±0.148 ^b	6.16±0.130 ^g	5.27±0.104 ^g	5.43±0.129 ^b	5.08±0.076 ^b	26.48
Pearl millet flour	7.73±0.110 ^e	7.53±0.097 ^e	11.25±0.299 ^c	11.15±0.243 ^b	10.38±0.099 ^b	48.04
Rice bran	3.89±0.119 ^g	3.97±0.005 ^h	5.38±0.143 ^g	6.72±0.146 ^g	7.96±0.087 ^f	27.92
Rice bran + Rice flour	10.94±0.089 ^c	10.37±0.000 ^c	9.8±0.267 ^d	8.41±0.195 ^f	7.7±0.042 ^g	47.22
Cracked sorghum	26.11±0.764 ^a	19.41±0.647 ^a	14.66±0.020 ^a	12.92±0.273 ^a	11.97±0.277 ^a	85.07
Control	0.00	0.00	0.00	0.00	0.00	0.00

*DAP – Days after placement; Means followed by the same letter (s) in a column are not significantly different by DMRT (P=0.05)

Table 3. Behavioural/orientation response of *Rhyzopertha dominica* to various food attractants

Food attractants	<i>Rhyzopertha dominica</i> settled (%)				
	5 MAR*	10 MAR	15 MAR	20 MAR	25 MAR
Wheat flour	24.6 ±0.347 ^a	32.1±0.226 ^a	36.2±1.002 ^a	30.1±0.656 ^a	38.6±0.318 ^a
Sorghum flour	17.3 ±0.565 ^b	28.1±0.267 ^b	22.2±0.513 ^c	26.1±0.603 ^b	26.6±0.325 ^b
Pearl millet flour	16.6±0.169 ^c	14.1±0.182 ^c	28.2±0.287 ^b	20.1±0.136 ^c	18.6±0.101 ^c
Control (without food)	6.6±0.004 ^d	9.5±0.181 ^d	4.9±0.106 ^d	7.4±0.211 ^d	8.0±0.152 ^d
Unsettled	48.0	26.8	19.5	26.8	21.3

*MAR- Minutes after release

corroborative to our findings. Sathiyaseelan et al. (2024), who demonstrated odour-based host searching behaviour of *R. dominica*, *T. castaneum* where all of them were attracted by the trap odour emanated from wheat flour. The highest orientation recorded was 28.5% toward wheat flour, which was significantly greater than that of other flours. This was followed by sorghum flour at 21.16% and pearl millet flour at 17.5% (Sathiyaseelan et al. 2024).

CONCLUSIONS

This study highlights the significant behavioral responses of the lesser grain borer, *Rhyzopertha dominica*, to various food bait attractants in storage environments. Cracked sorghum and wheat flour emerged as the most attractive substrates, demonstrating the importance of specific grain volatiles in guiding the orientation of *R. dominica*. The findings indicate that the attraction indices varied over time, with distinct preferences observed for different bait materials. This knowledge can be instrumental in developing effective monitoring and control strategies for managing *R. dominica* populations in storage facilities. By incorporating attractive volatile semiochemicals into pheromone lures, it is possible to enhance the efficacy of pest management practices, ultimately contributing to improved food security and reduced losses in stored products. The insights gained from this research could inform future studies on sustainable pest management approaches and promote the development of more effective and environment friendly attractants for stored product insects.

AUTHOR CONTRIBUTION

The study was conceptualised, designed and carried out the experiments by M. Sathiyaseelan. K. Balaji assisted with the data collection and data analysis. The article was read and approved by all the authors.

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