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Aggregation pheromone of locusts contributes to its control

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Abstract

Locusts may form large swarms that can quickly consume enormous amounts of crops and vegetation, which can have a disastrous effect on agriculture. These swarms are capable of covering great distances, eradicating crops of grains, vegetables, and other vital food supplies, causing large crop losses, and jeopardizing the security of food in the affected areas. Numerous crops are targeted by the locusts, including grains that are basic diets for many populations, such as wheat, rice, and maize. Their feeding has the potential to bare entire fields, costing farmers money and raising food prices. Furthermore, outbreaks of locusts can harm pasturelands over time, decreasing the amount of feed available to cattle and negatively affecting agricultural livelihoods. In this study, we used the aggregation pheromones presented in the feces of aggregation locust culture to attract mature adult females to lay their eggs. Egg pods were then treated with a 50% locust feces extract solution to study its effects on egg hatching. The physiological analysis investigated that the total lipids, protein, and carbohydrates of the treated egg pods were significantly reduced compared to the control ones. This may be used as a good trap for controlling locust egg pods.

Introduction

Insects are known to have pheromone plasticity, which improves their capacity for survival, adaptation, and successful reproduction. Aggregation pheromones are well known for their unique role in causing noticeable individual aggregation, which in turn affects agriculture and human life (Guo *et al*., 2023). Pheromones, chemical messengers, significantly influence species behavior and physiology. In locusts, *Schistocerca gregaria* Forskål (Orthoptera: Acrididae), volatiles play a role in gregarious behavior and reproductive activity. Aggregated oviposition in females is caused by pheromone-based behavior, which interacts with environmental conditions. Pheromones also influence the timing of sexual maturity, with mature males emitting more volatiles, helping young males reach maturity faster. Locusts' gregarious behavior is a response to mechanical stimulation (Riaz and Hakeem, 2023). The study used fecal volatiles of *Locusta migratoria manilensis* L. (Orthoptera: Acrididae), an oriental migrating locust, to identify aggregation components. Adult men exhibited reduced behavioral reactions to synthetics of five individual chemicals compared to the mixture (Shi *et al.*, 2011). Desert locust *S. gregaria* exhibits phase polymorphism based on population density, affecting morphological, physiological, and behavioral traits. Recent discoveries suggest that volatiles in desert locusts play a role in their behavior and development. Females deposit volatiles in egg pods, potentially leading to aggregated oviposition when combined with external conditions. Mature males release volatiles, such as phenyl acetonitrile, which is believed to hasten the sexual maturity of juvenile males (Ferenz and Seidelmann, 2003). The desert locust, *S. gregaria*, will not deposit its egg pods on sand that has extracts from the locust's feces. This study examined the oviposition inhibitory (OI) impact on the migratory locust *L. migratoria*. More egg pods were deposited by *L. migratoria* females in control sand as compared to sand that included fecal extracts made from both *L. migratoria* and *S. gregaria*. However, compared to *S. gregaria* oviposition, *L. migratoria* oviposition was less responsive to fecal extracts. It appears that phase-specific metabolism is not involved in OI activity because the OI effect was shown in *L. migratoria* regardless of whether the fecal extracts were made from gregarious or solitaries locusts (Sugahara *et al*., 2021). Adults of desert locusts were exposed to a water extract of wild-collected frass mixed with sand, resulting in an oviposition inhibitory (OI) effect. This effect was observed when compared to sand alone, six plant species' leaves, and the frass created by the locusts. Frass extracts often had a higher OI impact than leaf extracts. The study also found that rescue grass also produced extracts of frass. The migratory and desert locust frass had a higher OI impact than the Bombay locust frass, and similar effects were observed in frost samples extracted using hot and cold water (Tanaka *et al*., 2019). The aim of this work is to investigate the effect of the aggregation pheromone of the young African locust, *L. migratoria* on the control of its egg hatching.

Materials and methods

1. Insects:

The African migratory locust *L. migratoria migratorioides*, and Egyptian locust *Anacridium aegyptium* (L.) (Orthoptera: Acrididae) were cultivated in a stock culture that lasted for more than three generations. The locusts were initially gathered from field-grown maize in the village of Abu Rawash, Giza, Egypt. At the Plant Protection Research Institute in Egypt, they were raised under gregarious phase circumstances within wooden cages measuring 40 x 40 x 30 cm. The number of adults in each cage was kept between 30 and 60. Under carefully regulated circumstances of 16:8 L:D, 30 ± 1°C, and 70% relative humidity, locusts were fed fresh maize leaves, *Zea mays* (Giza 94 variety) (Poales: Poaceae).

2.Bioassay:

In the experiment, locust frass was collected and dried for three days at 10–40% relative humidity. After drying, 30 g samples were soaked in 300 ml of water overnight at room temperature and then filtered following (Tanaka and Sugahara, 2017). We combined 1 kg of sterilized sand with 200 ml of the extract solution in 25 cm diameter pots. Each pot received five pairs of fully grown adult African locusts. After maturing adult females laid their egg pods, the sand was treated with a 50% concentration of extract. Juvenile African locust frass was spread on the pot's surface in amounts of 0, 10, or 30 g (Figure 1). This setup was repeated with the same components, substituting African locusts with five pairs of fully grown adult Egyptian locusts (Figure 2) for the differentiation between mature adult females of African and Egyptian locusts. Each treatment was replicated three times in five separate experiments.

3. Biochemical compositions analysis:

Ten egg pods from the African migrating locust control and ten egg pods from the frass treatment were collected over fourteen days. The egg pods were homogenized in distilled water and centrifuged in a Beckman GS-6R centrifuge at 3219 RCF for ten minutes at five degrees Celsius. Before the principal components were analyzed, the supernatant fluid was centrifuged, separated into tiny aliquots (0.5 ml), and stored at - 20°C. The formula for total protein founded in Bradford (1976). Singh and Sinha (1977) were utilized to evaluate total carbohydrates, while Knight *et al.* (1972) were utilized to examine total lipids. Every biochemical determination was conducted three times, using three copies of each treatment.

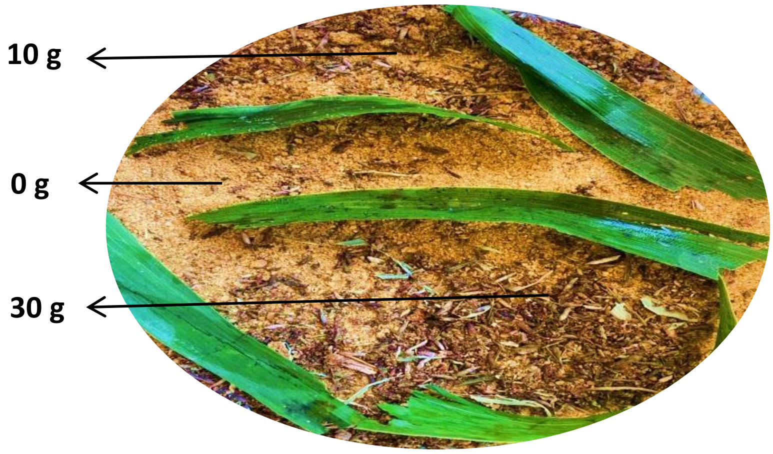

Figure (1): Different amounts of juvenile African locust frass were split onto the surface of the pot.

A

B

Figure (2): A. Mature adult female of African locust, *Lucsta migratoria.*
B. Mature adult female of Egyptian locust, *Anacridium aegyptium*

4. Statistical analysis:

The biochemical results of the treated and control eggs were compared statistically using the SPSS program, accounting for group differences (ANOVA). Duncan's multiple-range test was used to evaluate the significance of various treatments.

Results and discussion

1. Effect of the aggregation pheromone of juvenile African locust frass on locust egg-laying:

Juvenile African locust frass attracts mature female African locusts to lay their eggs at high concentrations (30 g) (Figure 3). However, when mature female Egyptian locusts were used instead, their behavior changed. Egyptian locust females avoided laying eggs at both low and high concentrations of African locust frass and lay their eggs only in the 0% frass concentration (Figure 4).

Figure (3): Mature adult females of African locusts lay their eggs at the 30 g of locust frass.

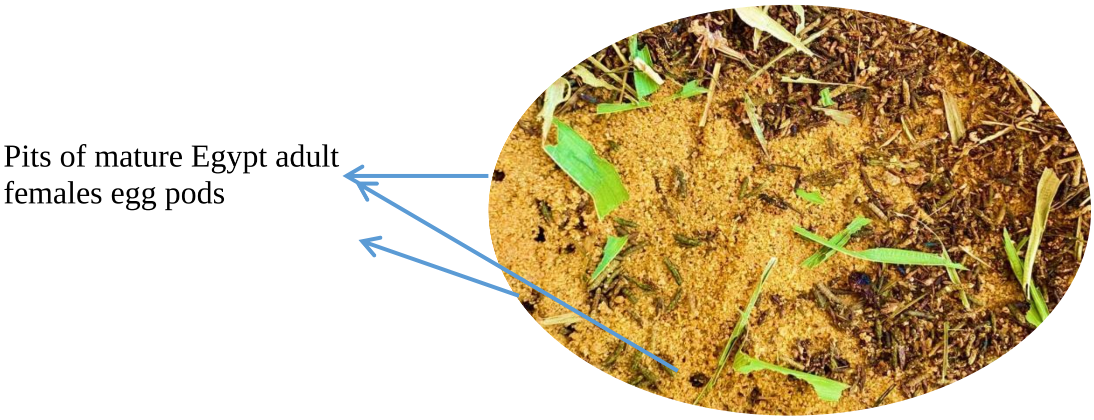

Figure (4): Mature adult females of Egypt locust avoid African locust frass and lay their eggs at 0 g of African locust frass.

2. Physiological effect of the juvenile African locust frass extract on egg hatching:

Applying a 50% juvenile African locust frass extract to the egg pods of *L. migratoria* resulted in no eggs hatching. Biochemical analysis of the treated egg pods presented in Table (1) reveals the effects of the 50 % frass solution on the African locust egg. It demonstrates that the total protein, lipids, and carbohydrate contents of the treated samples were significantly lower compared to the control samples. Statistical analysis confirms significant changes in total protein content after 5, 10, and 14 days of treatment (df = 1, F = 28.53, P = 0.0059; df = 1, F = 426.74, P < 0.001; and df = 1, F = 451.37, P < 0.001). Furthermore, significant changes in total carbohydrate contents were observed during the treatment (df = 1, F = 23.01, P = 0. 0087; and df = 1, F = 14.89, P = 0. 0182). However, no significant change was observed during the first five days of treatment (df = 1, F = 3.98, P = 0.1165). Also, no significant change was observed in total lipid content during the first five days (df = 1, F = 7.66, P = 0.0504), while exhibiting significant changes after 10 and 14 days of treatment (df = 1, F = 8.97, P = 0401; and df = 1, F = 50.38, P = 0.0021).

In summary, the statistical analysis of the frass solution's effects on the African locust egg indicates significant differences in total protein, carbohydrate, and lipid content throughout the treatment period, with varying patterns of change observed in each parameter during different times.

Table (1): Analysis of the total protein, carbohydrates, and lipid contents (mg /g body weight) in the *Locusta migratoria manilensis* egg control and treated with African locust frass solution.

Egg	Days	Total protein Mean ± S. E	Total carbs. Mean ± S. E	Total lipids Mean ± S. E
Control	5	31.61 ± 0.64[a]	9.11 ± 0.63[a]	10.41 ± 0.74 [a]
	10	48.5 ± 0.36[a]	8.42 ± 0.88 [a]	9.21 ± 1.21[a]
	14	55.75 ± 1.99[a]	6.16 ± 0.61[a]	7.46 ± 0.45[a]
Treated	5	24.84 ± 1.09[b]	7.70 ± 0.30[a]	7.84 ± 0.55[a]
	10	19.77 ± 1.34[b]	3.60 ± 0.47[b]	5.16 ± 0.58[b]
	14	11.34 ± 0.62 [b]	3.14 ± 0.48[b]	3.82 ± 0.23[b]

A comparison was made between the control and treated *Locusta migratoria manilensis* eggs. Bars (mean ± SE) with a different letter(s) are significantly different (P < 0.05).

Being the most common and perhaps most harmful locust species (Ma *et al*., 2012). Global agriculture is still seriously threatened by migrating locusts (Stige *et al*., 2007). A migrating locust's (*L. migratoria*) aggregation pheromone is 4-vinyl anisole (4VA), often referred to as 4-methoxystyrene. Regardless of their age or sexual orientation, 4VA attracts both gregarious and lonely locusts. On locust antennae, it selectively triggers reactions from basiconic sensilla. Field trapping studies confirmed that 4VA is appealing to both wild and experimental populations. These results pinpoint a pheromone associated with locust aggregation and offer guidance for creating innovative locust control tactics (Guo *et al*., 2020). The present investigations showed that mature adult female locusts attracted to the feces of the young once and laid their egg pods. This agreed with other studies, which found that aggregation pheromones have been detected in several species, most notably the desert locust *S. gregaria*. In addition to attracting other locusts, these pheromones aid in fostering group cohesiveness and behavior synchronization, all of which are essential for the creation of swarms (Hassanali *et al*., 2005 and Rai *et al*., 1997). These aggregation pheromones of *L. migratoria* are used as a trap in this research to control locust egg pods by the use of 50% of *L. migratoria* feces extract solution. The results showed that all treated egg pods did not hatch. This agreed with Tanaka *et al.* (2019); the impact of an oviposition inhibitor (OI) when given to adult desert locusts. Since the fraction samples produced OI effects that were equally significant when extracted using hot and cold water, it seemed doubtful that bacteria were involved in the extraction process. Hatching rates were dramatically decreased when sand and frass extracts from the previously described sources were used to incubate desert locust eggs. Within four days, embryos cultured in sand containing extracts from the frass of desert locusts that consumed rescue grass showed deformity. The application of a locust feces extract solution to locust eggs has been demonstrated to impact several facets of egg physiology, including levels of lipids, proteins, and carbohydrates; however, the precise biochemical alterations are not entirely elucidated in the current body of research. The general effects of fecal extracts on locust oviposition and egg quality were the subject of one investigation. It was said that the migrating locust's *L. migratoria* oviposition behavior and egg development were hindered by exposure to excrement extracts. This shows that fecal chemicals could interfere with the proper development of eggs and affect how nutrients are distributed within them, changing the amount of fat, protein, and carbohydrates (Sugahara *et al*., 2021).

References

Bradford, M. M. (1976): A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry, 72(1–2): 248–254. https://doi.org/10.1016/0003-2697(76)90527-3.

Ferenz, H. and Seidelmann, K. (2003): Pheromones to aggregation and reproduction in desert locusts. Physiological Entomology, 28(1): 11–18. https://doi.org/10.1046/j.1365-3032.2003.00318.x

Guo, X.; He, H.; Sun, J. and Kang, L. (2023): Plasticity of aggregation pheromones in insects. Current Opinion in Insect Science, 59, 101098. https://doi.org/10.1016/j.cois.2023.101098

Guo, X.; Yu, Q.; Chen, D.; Wei, J.; Yang, P.; Yu, J.; Wang, X. and Kang, L. (2020): 4-Vinylanisole is an aggregation pheromone in locusts. Nature, 584(7822): 584–588. https://doi.org/10.1038/s41586-020-2610-4

Hassanali, A.; Njagi, P. G. N. and Bashir, M. O. (2005): Chemical ecology of locusts and related acridids. Annual Review of Entomology, 50:223–245. https://doi.org/10.1146/annurev.ento.50.071803.130345

Knight, J. A.; Anderson, S. and Rawle, J. M. (1972): Chemical basis of the sulfo-phospho-vanillin reaction for estimating total serum lipids. Clinical Chemistry, 18(3): 199–202.

Ma, C.; Yang, P.; Jiang, F.; Chapuis, M.-P.; Shali, Y.; Sword, G. A. and Kang, L. (2012): Mitochondrial genomes reveal the global phylogeography and dispersal routes of the migratory locust. Molecular Ecology, 21(17): 4344–4358. https://doi.org/10.1111/j.1365-294X.2012.05684.x

Rai, M. M.; Hassanali, A.; Saini, R. K.; Odongo, H. and Kahoro, H. (1997): Identification of components of the oviposition aggregation pheromone of the gregarious desert locust, *Schistocerca gregaria* (Forskal). Journal of Insect Physiology, 43(1): 83–87. https://doi.org/10.1016/S0022-1910(96)00051-0

Riaz, U. and Hakeem, K. R. (Eds.) (2023): Locust outbreaks: Management and the world economy (First edition). Apple Academic Press.

Shi, W.-P.; Sun, H.-L.; Edward, N. and Yan, Y.-H. (2011): Fecal volatile components elicit aggregation in the oriental migratory locust, Locusta migratoria manilensis (Orthoptera: Acrididae): Fecal volatile components in the oriental migratory locust. Insect Science, 18(2): 166–174. https://doi.org/10.1111/j.1744-7917.2010.01341.x

Singh, N. B. and Sinha, R. N. (1977): Carbohydrate, lipid and protein in the developmental stages of *Sitophilus oryzae* and *S. granarius* (Coleoptera: Curculionidae). Annals of the Entomological Society of America, 70(1): 107–111. https://doi.org/10.1093/aesa/70.1.107

Stige, L. C.; Chan, K.-S.; Zhang, Z.; Frank, D. and Stenseth, N. C. (2007): Thousand-year-long Chinese time series reveals climatic forcing of decadal locust dynamics. Proceedings of the National Academy of Sciences, 104(41): 16188–16193. https://doi.org/10.1073/pnas.0706813104

Sugahara, R.; Hirota, K. and Sakakibara, S. (2021): Ovipositional inhibition effect of locust fecal extracts in the migratory locust, Locusta migratoria (Orthoptera: Acrididae). Applied Entomology and Zoology, 56(2): 199–205. https://doi.org/10.1007/s13355-021-00725-x

Tanaka, S.; Kotaki, T.; Nishide, Y.; Ben-Hamouda, A.; Abdellaoui, K.; Ebbe, M. A. B. and Ely, S. O. (2019): Effects of water extracts of frass from three locust species and various plants on oviposition and embryonic development in the desert locust, Schistocerca gregaria. Journal of Orthoptera Research, 28(2): 195–204. https://doi.org/10.3897/jor.28.34665

Tanaka, S. and Sugahara, R. (2017): Desert locusts *Schistocerca gregaria* (Acrididae: Orthoptera) do not lay eggs in old sand: Why? Applied Entomology and Zoology, 52(4): 635–642. https://doi.org/10.1007/s13355-017-0518-8