Series: ✓ *Biology*
✓ *Horticulture*
✓ *Food products processing technology*
✓ *Environmental engineering*

ANNALS OF THE UNIVERSITY OF CRAIOVA

Vol. XXIX (LXV) – 2024

THE EFFECTIVENESS OF FUMIGATION WITH ALUMINIUM FOSFINE AGAINST TRIBOLIUM CONFUSUM AND TRIBOLIUM CASTANEUM POPULATION AT A WHEAT MILL IN BUCHAREST AREA IN ROMANIA

Spiliotis Spyros-Angelos[1*], Mitrea Ion[2]
[1]*Doctoral School of Plant and Animal Resources Engineering, Faculty of Horticulture, University of Craiova, 13 A.I. Cuza Street, 200585, Craiova, Romania
[2]University of Craiova, Faculty of Horticulture, 13 A.I. Cuza Street, 200585 Craiova, Romania
**Correspondence author. E-mail:* aggelos212000@yahoo.com

Keywords: Tribolium Confusum, Tribolium Casteneum, Rezistance to fosfine

ABSTRACT
Stored grain pests, particularly Tribolium castaneum and Tribolium confusum, pose significant threats to grain storage facilities. This study examines their populations before and after phosphine fumigation in a wheat mill near Bucharest, Romania. Monitoring was conducted over four months using XLure Rusel MST pheromone traps. Results revealed a 97.14% reduction in total insect population, from 1,606 to 46 individuals. However, surviving populations were noted, indicating potential resistance to phosphine. This study underscores the persistence of pest populations and the necessity for continued monitoring and research on resistance mechanisms in real-world settings.

INTRODUCTION

Insect pests of stored cereal products represent a major threat to mills and storage facilities, causing significant economic losses. The costs of infestation control, including the implementation of integrated pest management strategies, are necessary to prevent larger losses, such as product recalls and reputational damage (Hagstrum and Athanassiou 2019).

The pest species affecting cereals include *Sitophilus granarius*, *Sitophilus oryzae*, *Tribolium castaneum*, and *Tribolium confusum*, known for their destructive impact on stored cereals. These insects spread rapidly and cause significant damage, affecting product quality and processing facilities (Hagstrum and Athanassiou 2019).

Tribolium castaneum and *Tribolium confusum* are frequently encountered in cereal processing facilities and storage units, where they find favorable conditions in wall and floor crevices. These pests significantly contribute to cereal degradation (Abdullahi, Mohammad, and Sule 2019).

Integrated pest management is widely used in mills and storage facilities, and structural fumigation with phosphine is a common method for controlling *Tribolium castaneum* infestations (Hema *et al.* 2020). Phosphine is a fumigant gas applied via aluminum or magnesium phosphide tablets or pellets, which release the gas upon contact with atmospheric moisture. The phosphine gas reacts with water vapor, forming lethal compounds for insects within the structures.

However, phosphine resistance has become an increasing concern. Phosphine acts by suppressing the oxidative metabolism of insects, as a result of differential encoding of enzymes such as dihydrolipoamide dehydrogenase (L-protein) and unsaturase (Schlipalius et al. 2012).

Recent studies have examined the resistance of *Tribolium castaneum*, one of the most widespread and harmful insect species in global cereal storage facilities. *Tribolium castaneum* was the first species declared to be highly resistant to phosphine, with fumigation failures becoming more frequent. The gene responsible for phosphine resistance is P450 CYP346, and insects possessing this gene have demonstrated a resistance level varying between 1.8 and 862.7 (Wang et al. 2020).

Global reports, including those from China, the USA, Australia, and Turkey, have confirmed phosphine resistance in populations of *Tribolium castaneum* and *Tribolium confusum* as well as in another 8 species of warehouse insects. (Huang *et al.* 2019; Opit *et al.* 2012; Chen *et al.* 2015; Kocak *et al.* 2015, Song XuHong *et.al.*). Furthermore, recent European studies have revealed a decrease in phosphine sensitivity in insects sampled from various European countries (Sakka and Athanassiou 2023; Aulicky, Stejskal, and Frydova 2019).

None of these studies, however, have been conducted under real field conditions in Romania, with prior research being limited to laboratory settings. The objective of this study is to gain a better understanding of the behavior of *Tribolium castaneum* and *Tribolium confusum* populations before and after a phosphine fumigation in a wheat mill located in the Bucharest area, Romania, using pheromone traps. Monitoring was conducted for 45 prior to and 45 days after the main fumigation.

This article aims to provide new data and valuable insights regarding phosphine resistance in *Tribolium castaneum* and *Tribolium confusum* in Romania, based on a real-world phosphine application.

MATERIALS AND METHODS

The study was conducted in a mill with a production capacity of 500 tons per day, located in the Bucharest area, Romania. Pheromone traps, XLure Rusel MST, specifically designed to capture crawling insects from storage areas, were installed using a specialized pheromone for *Tribolium castaneum* and *Tribolium confusum*. In total, 49 XLure Rusel MST pheromone traps were used and strategically placed throughout the mill. The traps were positioned near production machinery as well as on the fifth and sixth floors, where the lids of the cereal and flour silos are located.

Measurements and insect identification were carried out over a period of four months, with two months before and two months after the main phosphine fumigation. Insect measurements were conducted at 15-day intervals. The total insect population was analyzed, as removing insects from the trap's control surface is impossible without destroying them.

For the fumigation, 7 grams of aluminum phosphide per cubic meter were used, with a total of 422 kg of aluminum phosphide applied for a total volume of 60,359 cubic meters. The maximum phosphine concentration reached 1150 ppm after 60 hours. The fumigation lasted 96 hours. After recording the insect populations detected in the pheromone traps, a comparison was made between insect populations before and after the fumigation.

Figure 1. Fosfine treatment. Application of solid AlP (Original)

Figure 2. Maximum concentration of 1150 ppm reading with Dräger X-am 5000. (Original)

Figure 3. Placed feromone trap XLure MST (Original).

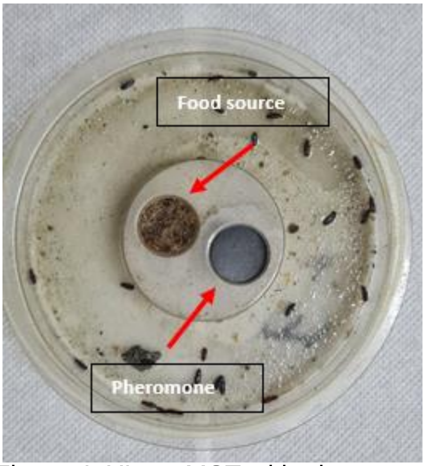

Figure 4. XLure MST with pheromone lure and food source. (Original)

Figure 5. XLure MST with pheromone lure and food source. (Original)

Figure 6. *Tribolium confusum* si *Tribolium castaneum* on XLure MST Feromone traps when collected.

RESULTS AND DISCUSSION

Counting insect populations using pheromone traps confirmed the presence of *Tribolium castaneum* and *Tribolium confusum*, both before and after the phosphine fumigation process. A significant reduction in the total population was observed, reaching 97.14%, with absolute numbers decreasing from 1,606 to 46 insects (Table 1).

The interval between the two counts, conducted before and after the central fumigation, was 15 days. This indicates the existence of insect populations that survived the phosphine treatment, considering that the minimum life cycle for the emergence of new adult insects is approximately 30 days.

Additionally, an increase in the insect population was observed 15 days after fumigation, closely aligning with the insect life cycle. It appears that the additional captures involved insects that survived the treatment. At 45 days post-fumigation, a rapid increase in the number of insects captured was observed; this can be attributed not only to surviving insects but also to those generated by the first generation of survivors.

Table 1

Total number, average and percentage difference of *Tribolium (castaneum and confusum)* observations before and after fumigation.

	T-45d	T-30d	T-15d	T=0	T+15d	T+30d	T+45d
Total nr.	1163	1397	**1606**	**FUMIGATION**	**46**	92	318
Avg.nr/trap	23,73	28,51	**33,46**		**0,98**	1,87	6,48
Dif. %	-	18,28%	13,91%		-97,14	100%	245%

In conclusion, although phosphine fumigation has a profound impact on insect populations, resulting in a substantial reduction, it appears that some insect populations exhibit resistance to the treatment. This resistance may stem from genetic factors or from exposure to lower concentrations of phosphine in hard-toreach areas covered with flour residues, where phosphine penetration is difficult. This warrants further investigation.

CONCLUSIONS

This study demonstrates the effectiveness of phosphine fumigation in significantly reducing populations of *Tribolium castaneum* and *Tribolium confusum* in a wheat mill setting. However, the observation of surviving insects post-treatment highlights the potential development of resistance to phosphine. These findings emphasize the importance of ongoing surveillance and research into pest management strategies. To mitigate economic losses in grain storage, it is crucial to integrate alternative control methods and adapt management practices in response to the evolving resistance patterns of stored product pests. Further investigations are needed to better understand the mechanisms of resistance and to ensure the long-term efficacy of fumigation treatments.

REFERENCES

Abdullahi, *et.al.* 2019. Biology, host range and management of red flour beetle Tribolium Castaneum (Herbst)(Coleoptera: Tenebrionidae): a review. Taraba Journal of Agricultural Research 7.1 (2019): 48-64. DOI: N/A

Aulický, R. *et. al.* 2015. Validation of hydrogen cyanide fumigation in flourmills to control the confused flour beetle. Czech Academy of Agricultural Sciences 33 (2): 174-179. DOI: https://doi.org/10.1016/j.jspr.2019.02.003

Chen, Z, *et. al.*. 2015. Diag-nostic molecular markers for phosphine resistance in US popula-tions of Tribolium Castaneum and Rhyzopertha Dominica. Genes 10 (3). DOI: https://doi.org/10.1371/journal.pone.0121343.

Donahaye, E.J. 2020. Current status of non-residual control methods against stored product pests. Crop Protection 19 (8-10): 571-576. DOI: https://doi.org/10.1016/S0261-2194(00)00074-0.

Emery, RN, MK Nayak, and JC Holloway. 2011. Lessons learned from phosphine resistance monitoring in Australia Stewart Postharvest 7 (3): 1-8. DOI: http://dx.doi.org/10.2212/spr.2011.3.8

Hagstrum. D. and Athanassiou C. 2019. Improving Stored Product Insect Pest Management: From Theory to Practice. Insects 10(10):332. Doi: http://dx.doi.org/10.3390/insects10100332

Hamel, Darka, Vlatka Rozman, and Anita Liška. 2020. Storage of cereals in warehouses with or without pesticides. Insects 11.12: 846.

Hema, L., et. al.. 2020. IOT Based Real-Time Control and Monitoring System for Food Grain Procurement and Storage. OP Conf. Ser.: Mater. Sci. Eng. 993 012079. https://doi.org/10.1088/1757-899X/993/1/012079

Huang, Y. *et. al.* 2019. Susceptibility of Tribolium Castaneum to phosphine in China and functions of cytochrome P450s in phosphine resistance, Journal of Pest Science 92 (3) : 1239-1248. https://doi.org/10.1007/s10340-019-01088-7.

Kocak, E, *et. al.*. 2015. Determin-ing phosphine resistance in rust red flour beetle, Tribolium casta-neum (Herbst.) (Coleoptera: Tenebrionidae) populations fromTurkey. Turk Aust J Entomol 39 (2): 129-136. https://doi.org/http://dx.doi.org/10.16970/ted.17464.

Opit, P., *et. al.* 2012. Phosphine Resistance in Tribolium Castaneum and Rhyzopertha dominica From Stored Wheat in Oklahoma. Journal of Economic Entomology 105 (4): 1107-1114. https://doi.org/10.1603/EC12064

Schlipalius, DL, *et. al.* 2012. A Core Metabolic Enzyme Mediates Resistance to Phosphine Gas American Association for the Advancement of Science 338 (6108) : 807-810. https://doi.org/10.1126/science.1224951

Song, XuHong, *et.al.* 2011. Phosphine resistance in Rhyzopertha dominica (Fabricius)(Coleoptera: Bostrichidae) from different geographical populations in China. African Journal of Biotechnology, 10(72) 16367-16373. http://dx.doi.org /10. 5897/AJB11.1101

Wang, Kangxu, et al. 2020. Identification and functional analysis of cytochrome P450 CYP346 family genes associated with phosphine resistance in Tribolium Castaneum. Pesticide Biochemistry and Physiology 168, https://doi.org/10. 1016/j.pestbp.2020.104622