

# *Cephalonomia tarsalis* (Hymenoptera: Bethyilidae) for the control of the sawtoothed grain beetle, either alone or in combination with the predatory mite *Blattisocius tarsalis*

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## ABSTRACT

The sawtoothed grain beetle *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) is an important pest of stored cereals. Due to the phasing out of many active compounds registered as insecticides and the occurrence of insect resistance to the remaining active compounds, it is crucial to develop alternative pest control strategies, such as the use of natural enemies. In this study, we evaluated the ability of *Cephalonomia tarsalis* (Ashmead) (Hymenoptera: Bethyilidae), a specialised larval ectoparasitoid, to locate and parasitize *O. surinamensis* in vertical PVC pipes 20 cm in diameter filled with 7–26 kg of paddy rice. The parasitoid was able to move through the rice and reduce pest emergence up to 60% at depths of 40, 100, and 150 cm, even when larvae were offered simultaneously at all three depths. Since most of the pest populations are found in the upper layer of 1 m of the grain piles, this result suggests that the parasitoid would be able to locate and parasitize the pest within the stored paddy rice piles. We also evaluated the possibility of complementing pest reduction by the parasitoid with the aid of the egg-predatory mite *Blattisocius tarsalis* (Berlese) (Mesostigmata: Ascidae). When each natural enemy was offered its target pest instar for one week, both were able to reduce the pest similarly by 52–65%. When they were released in 2 kg of paddy rice with 10 *O. surinamensis* females for 10 weeks, the parasitoid achieved higher pest reduction (96%) than the predatory mite (42%). The pest reduction obtained by combining both natural enemies did not improve the efficacy of the parasitoid, suggesting that the parasitoid alone can be an effective alternative to maintain the beetle populations under control.

## 1. Introduction

The sawtoothed grain beetle *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) is one of the major pests in stored grain products and causes significant quantitative and qualitative losses (Castañé and Riudavets, 2015; Castañé et al., 2020; Duarte et al., 2021; Morrison et al., 2023). *Oryzaephilus surinamensis* feeds externally on grain kernels broken or damaged by other pests, increasing weight loss, grain damage and contamination. It exhibits high mobility and can be found at least 1 m deep within grain piles (Navarro et al., 1981; Armitage et al., 1983). Due to its relatively small size compared to other stored-product insect species, it is difficult to detect and control, as it can reside in the cracks and crevices of storage facilities (Arthur, 2001; Hill, 2003; Mahroof and Hagstrum, 2012).

Conventional insecticides remain the primary strategy for managing insect pests in stored products, despite the appearance of resistance in insect populations caused by the repeated use of a limited number of permitted active compounds and the negative impacts on the

environment and human health (Kavallieratos et al., 2015; Vélez et al., 2017; Feroz et al., 2020). Consequently, there is a need to develop and enhance sustainable alternative pest control methods, such as biological control. This approach, which uses natural enemies to regulate pest populations effectively, has proven to be economically feasible and successful (Hervet and Morrison, 2021).

*Cephalonomia tarsalis* (Ashmead) (Hymenoptera: Bethyilidae) is an ectoparasitoid specific to *Oryzaephilus* spp. that parasitises both larvae and pupae (Howard et al., 1998). Like many other parasitoids, *C. tarsalis* employs chemical cues to locate the host. Subsequently, it paralyzes

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several larvae, often feeding and hiding them in distinct locations. This behaviour enhances the potential effectiveness of the parasitoid, as paralyzed larvae do not recover (Powell, 1938; Howard et al., 1998). *Cephalonomia tarsalis* has been regarded by some studies as an economically unimportant biocontrol agent of *O. surinamensis* due to its low parasitic rate (Powell, 1938; Howard et al., 1998; Eliopoulos et al., 2002). However, other studies have shown promising results when released the parasitoid alone (Eliopoulos et al., 2017; Eliopoulos, 2019) or together with other natural enemies, such as parasitic protozoans (Lord, 2006) or mites (Žďárková et al., 2003). Combining parasitoids with other natural enemies that target distinct developmental stages of the pest can improve the suppression of the pest population.

*Blattisocius tarsalis* (Berlese) (Mesostigmata: Ascidae) is a predatory polyphagous egg mite commonly found in warehouses and grain storage facilities associated with a wide range of Acari, Psocoptera, Lepidoptera, and Coleoptera species (Haines, 1974, 1981a, 1981b; Nielsen, 1999; Riudavets et al., 2002; Stejskal et al., 2006; Thind and Ford, 2006; Gallego et al., 2020). Iturralde-García et al. (2020a) found that the combination of this mite with the larval parasitoid *Anisopteromalus calandrae* (Howard) (Hymenoptera: Pteromalidae) improved the control of the bruchid weevil *Acanthoscelides obtectus* (Say) (Coleoptera: Chrysomelidae) compared to the use of each biological agent alone. In the case of *C. tarsalis*, few studies have focused on its suitability and practical application as a biological control agent in large storage facilities.

This study aimed to determine if the parasitoid *C. tarsalis* could be a potential candidate for managing *O. surinamensis* populations when developed in paddy rice and whether combining it with the predatory mite *B. tarsalis* would improve pest reduction. We hypothesised that the parasitoid could effectively reduce sawtoothed grain beetle populations and that its effectiveness would be enhanced by combining it with the predatory mite *B. tarsalis*. Therefore, we evaluated the ability of the parasitoid to penetrate and parasitise the host at different depths of paddy rice. We also evaluated the efficacy of both natural enemies in reducing the pest in different arenas.

## 2. Materials and methods

All experiments were performed, and the stock colonies were maintained under environmentally controlled conditions ( $28 \pm 2^\circ\text{C}$ ,  $70 \pm 2\%$  RH, 16:8 L:D). The stock colonies of *O. surinamensis*, *C. tarsalis* and *B. tarsalis* were initiated with individuals collected more than three years ago in warehouses of different grain commodities in northeast Spain. *Oryzaephilus surinamensis* was reared in our laboratory on a diet containing a mix of whole wheat flour and brown rice. *Cephalonomia tarsalis* was reared in our laboratory on the fourth instar larvae of *O. surinamensis*. The predatory mite *B. tarsalis* was supplied by Agrobio SL (Almería, Spain). Females of *C. tarsalis* were identified by the lower number of antennal segments in comparison with males (Powell, 1938), and females of the mite were recognized by their larger body than males (Gu et al., 2022).

### 2.1. Searching ability of the parasitoid at different depths of paddy rice

In this experiment, we aimed to evaluate the ability of the parasitoid to locate and suppress the host at different depths in a vertical column of paddy rice. Polyvinyl chloride (PVC) pipes (20 cm in diameter) were filled to the top with paddy rice and arranged vertically. A small glass jar (8 cm high x 6.5 cm internal diameter) was placed at the bottom of each pipe. The jar contained a thin layer of whole wheat flour (10 g) and 15 g of paddy rice infested with the fourth instar larvae of *O. surinamensis*. The jar was open but covered with a  $1 \times 1.5$  mm mesh screen that enabled the parasitoid to pass through but prevented the larvae from escaping. A paper strip with honey was placed on the grain's surface to nourish the parasitoids. Three pairs of *C. tarsalis* adults (0–7 days old) were released on the surface of the grain, after which the top of the pipes

was sealed with fabric mesh. After one week, paddy rice in PVC pipes was poured off, and the glass jars were recovered and isolated in ventilated plastic containers to allow the hosts and parasitoids to reach the adult stage. Afterwards, the number of *O. surinamensis* adults and parasitoids was recorded.

Three treatments consisted of PVC pipes with lengths of 40, 100 and 150 cm, containing 6.8, 17.0, and 25.5 kg of paddy rice, respectively. In a fourth treatment, three jars containing each 5g of infested rice, that is each with 1/3 of the amount used in the other three treatments, were placed at depths of 40, 100 and 150 cm each in the same PVC pipes. This was done to offer an equal number of larvae as in the previous three treatments. Six replicates were conducted for each treatment. A control treatment was also included outside the pipes, consisting of the same-sized jars with 25 g of diet infested with *O. surinamensis* 4th instar larvae.

### 2.2. Effectiveness of parasitoid and predatory mites in a small setting

In this experiment, we aimed to evaluate separately the capacity of parasitoids and predatory mites to reduce a given number of pests when offered the target instar in a short period and on a small scale. A total of 45 eggs of *O. surinamensis* were placed in glass jars (13.5 cm high x 9 cm internal diameter) containing 0.8 g of whole wheat flour and 8 g of paddy rice. Next, in the parasitoid treatment, 300 g of paddy rice was added to each glass jar. After three weeks, the time needed for the beetle larvae to develop, three parasitoid females were released, and sugary water was added for their nourishment.

In the treatment of predatory mites, three females were released together with *O. surinamensis* eggs. Four days later, jars were filled with *O. surinamensis* rearing diet (120 g brown rice and 180 g whole wheat flour) to ensure the development of non-predated eggs.

The emergence of adult beetles and parasitoids was recorded beginning in the fourth week. Control treatments with *O. surinamensis* eggs but without any releases of parasitoids or predatory mites were also included. Ten replicates of each treatment were performed.

### 2.3. Effectiveness of the parasitoid and predatory mite in a medium setting

In this experiment, we aimed to evaluate the ability of each natural enemy to reduce the pest population separately and in combination over a more extended period and on a larger scale. Ten adult females of *O. surinamensis* were released on 2 L ventilated plexiglass cylinders containing 2 kg of paddy rice and 0.02 kg of rice flour. In the predatory mite treatment, four releases of 10 females each were done in weeks two, four, six and eight. In the parasitoid treatment, 10 females were released in week three, when the first *O. surinamensis* eggs laid were estimated to have reached third to fourth larval instar. In the combination of both natural enemies treatments, 10 female parasitoids were released in week three, and 10 predatory mite females per release were introduced in weeks two, four, six and eight. The number of adult beetles and parasitoids that emerged was recorded in week 10. A control treatment with 10 female *O. surinamensis* but without any release of parasitoids or predatory mites was also included. Ten replicates of each treatment were performed.

### 2.4. Data analysis

Parasitoid-induced mortality (PIM), which includes host feeding, unsuccessful parasitism and/or super parasitism, was computed as the difference between total host mortality and effective parasitism (number of adult parasitoids emerged). When testing the parasitoid searching ability in the PVC pipe experiment, a generalized linear model (GLM) with a quasi-binomial error distribution was applied for pest reduction related to control treatment, effective parasitism, and PIM. A GLM fitted in a quasi-Poisson distribution was used for the number of emerged parasitoids. Post hoc comparisons were analyzed using the Tukey test.

In the experiment on the efficacy of each natural enemy in a small

setting, *O. surinamensis* reduction related to control treatment among natural enemies was compared using a GLM with a quasi-binomial error distribution. Additionally, differences in the number of pest adults that emerged in the control among natural enemies were assessed using a GLM fitted to a Poisson distribution.

When testing natural enemies individually and in combination in 2 L containers, pest reduction related to the control treatment and effective parasitism was analyzed through a GLM with a quasi-binomial error distribution and post hoc comparisons using a Tukey test. In this trial, the sex ratio was calculated from 15 randomly selected emerged parasitoids. Significant deviation from 1:1 female-to-male in emerged parasitoids was determined in each experiment using a Student's *t*-test. All the statistical analyses were conducted with R Studio software (RStudio Team, 2023) and a nominal significance of 5% ( $P < 0.05$ ).

### 3. Results

#### 3.1. Searching ability of the parasitoid at different paddy rice depths

The parasitoid could locate and parasitise *O. surinamensis* larvae at all tested depths, indicating that adult parasitoids had no difficulty finding hosts in a 1.5 m layer of paddy rice. In addition, the reductions in pest adult emergence when compared with the control treatments were similar regardless of whether the hosts were offered at any specific depth or if they were offered a spread simultaneously across the three pipe depths ( $F_{3,20} = 1.26$ ;  $P = 0.314$ ) (Fig. 1 A).

Moreover, when hosts were offered simultaneously at three different depths, the reduction in host emergence was similar at the three levels ( $F_{2,15} = 2.35$ ;  $P = 0.794$ ) (Fig. 1 B). When examining the causes of this reduction, no differences in effective parasitism were observed, either when offering hosts at a specific depth or when offered a spread across the pipe ( $F_{3,20} = 2.47$ ;  $P = 0.092$ ). Additionally, there were no differences among the three depths when offering the host simultaneously ( $F_{2,15} = 0.11$ ;  $P = 0.900$ ) (Fig. 1 B). Effective parasitism ranged from 8.7% to 23.5%. Similarly, PIM, which ranged from 20.21 to 54.8%, was not significantly different at any of the three depths tested ( $F_{3,20} = 1.03$ ;  $P = 0.399$ ), nor did it differ among the three depths when offered simultaneously ( $F_{2,15} = 0.18$ ;  $P = 0.838$ ) (Fig. 1). Compared with effective parasitism, PIM was the primary cause of host reduction (Fig. 1).

Table 1 shows the number of adult parasitoids and the percentage of females that emerged in each treatment. As expected, based on host

**Table 1**

The number (mean  $\pm$  SE) of *C. tarsalis* adults that emerged and the percentage (mean  $\pm$  SE) of *C. tarsalis* females that emerged when host larvae were offered at three depths (40, 100, and 150 cm) or simultaneously (mixed) at the three depths tested in a vertical PVC pipe of paddy rice.

Treatment	No. <i>C. tarsalis</i>	% <i>C. tarsalis</i> females	Student's <i>t</i> -test	
			<i>t</i>	<i>P</i>
40 cm	13.0 $\pm$ 6.3	74.7 $\pm$ 9.0 <sup>a</sup>	2.80	<0.01
100 cm	13.5 $\pm$ 6.5	64.6 $\pm$ 9.7	1.50	0.195
150 cm	20.3 $\pm$ 6.9	57.4 $\pm$ 6.0	1.19	0.289
Mixed depths	29.2 $\pm$ 11.4	50.4 $\pm$ 5.7	1.25	0.265
Mixed – 40 cm	11.2 $\pm$ 4.1	66.8 $\pm$ 3.2 <sup>a</sup>	5.25	<0.001
Mixed – 100 cm	8.0 $\pm$ 5.7	56.5 $\pm$ 6.5	1.00	0.423
Mixed – 150 cm	10.0 $\pm$ 3.7	61.3 $\pm$ 9.9	1.64	0.162

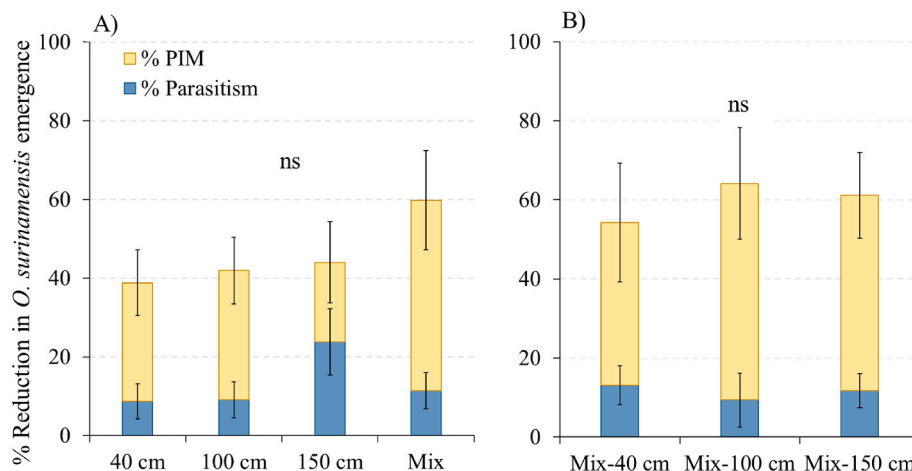
<sup>a</sup> Denotes a significant deviation from 1:1 female-to-male (Student's *t*-test).

reduction results, parasitoid offspring were similar among the three depths and the mixed treatment ( $F_{3, 20} = 0.55$ ;  $P = 0.657$ ). Parasitoid progeny presented a bias towards females at 40 cm depth, either when host larvae were offered only at this depth or when offered simultaneously with the other two depths (Table 1). The average number of hosts that emerged from the four control treatments was  $125.5 \pm 12.3$ .

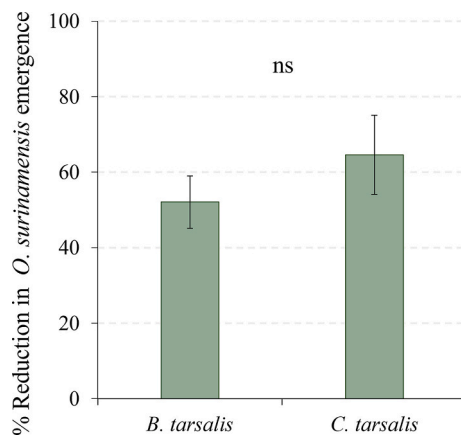
#### 3.2. Effectiveness of the parasitoid and predatory mite in a small setting

When testing predatory mite females (1:10 predator:prey ratio), *O. surinamensis* reduction in relation to its respective control was  $52.1 \pm 6.9\%$  (Fig. 2). In the control treatment, a mean of  $29.0 \pm 1.6$  beetles emerged from the initial 45 eggs, indicating a preimaginal mortality rate of  $35.6 \pm 3.5\%$ .

Regarding the parasitoid (1:9 parasitoid:host ratio), pest reduction in relation to its respective control was  $64.5 \pm 10.4\%$  (Fig. 2). Of this reduction,  $20.2 \pm 4.6\%$  was due to successful parasitism, and  $44.7 \pm 7.3\%$  was due to PIM. The average number of emerged parasitoids was  $9.1 \pm 2.1$ , and the sex ratio was not biased ( $t = -0.85$ ;  $P = 0.420$ ). A mean of  $26.6 \pm 1.4$  *O. surinamensis* adults emerged in the control treatment out of the 45 eggs introduced, indicating a preimaginal mortality of  $41 \pm 3.1\%$ . The percentage of *O. surinamensis* reduction ( $F_{1,18} = 1.56$ ;  $P = 0.227$ ) (Fig. 2) and the number of adults that emerged in the control ( $F_{1,18} = 1.04$ ;  $P = 0.309$ ) were similar for both natural enemies.



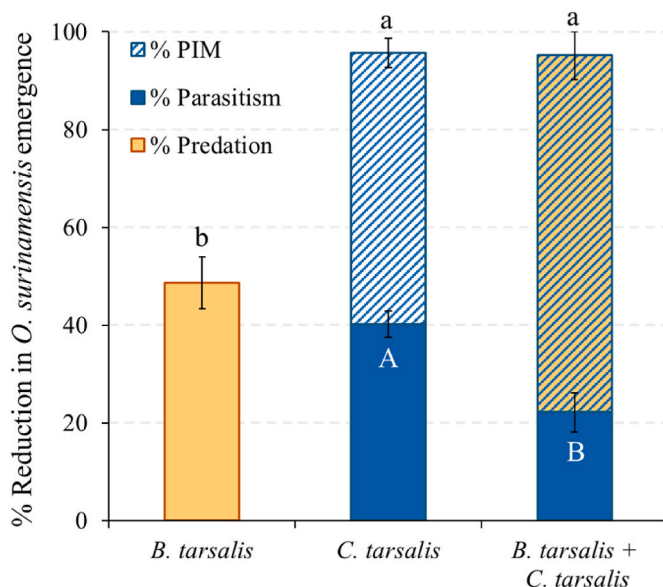
**Fig. 1.** Percentage (mean  $\pm$  SE) reduction in the emergence of *O. surinamensis* in comparison with the control treatment disaggregated in the percentage of effective parasitism (blue) and the percentage of PIM (yellow). Host larvae were offered at three depths (40, 100, and 150 cm) alone or simultaneously (mixed) at the three levels in a vertical PVC pipe filled with paddy rice. A) Comparison between the three depths separately and the total of the mixed treatments. B) Comparison of the three different depths in the mixed treatment. There were no significant (ns) differences in the percentage reduction of *O. surinamensis* emergence, the percentage of parasitism, and the percentage of PIM observed in the treatments.



**Fig. 2.** Percentage (mean ± SE) reduction in adult host emergence after introducing 45 eggs of the pest in glass jars with wheat flour and paddy rice. Two treatments were considered: the release of three females of the predatory mite *B. tarsi* with *O. surinamensis* eggs and three females of the parasitoid *C. tarsi* with *O. surinamensis* third–fourth instar larvae. No significant (ns) differences in the percentage reduction of *O. surinamensis* emergence were observed among the natural enemies (Tukey's test,  $P < 0.05$ ).

### 3.3. Effectiveness of the parasitoid and predatory mite at a medium scale

Releasing the predatory mite, the parasitoid, or both together had a significant negative impact on *O. surinamensis* adult emergence ( $F_{2,27} = 76.13$ ;  $P < 0.001$ ) (Fig. 3). Pest:natural enemy ratios tested were 1:4, 1:1 or 1:4 + 1:1, respectively. When the predatory mite was released, a  $48.7 \pm 5.9\%$  reduction of *O. surinamensis* adult emergence was observed (Fig. 3). Nevertheless, the parasitoid was more effective, reducing the pest by  $95.7 \pm 1.0\%$  (Fig. 3). The combination of the predatory mite and the parasitoid resulted in a  $95.2 \pm 1.1\%$  reduction in the pest, similar to that obtained with the parasitoid alone (Fig. 3).



**Fig. 3.** Percentage (mean ± SE) reduction in adult host emergence after 10 weeks compared with a control treatment when 10 females of the parasitoid and/or 40 females of the predatory mite were released. Arenas consisted of containers with 2 kg of paddy rice, where 10 females of *O. surinamensis* were initially released. Host reduction is disaggregated into predation, effective parasitism, and PIM. Treatments with different lowercase letters differ significantly in the reduction of *O. surinamensis* emergence; those with different uppercase letters differ significantly in effective parasitism (Tukey's test,  $P < 0.05$ ).

Effective parasitism, that is, the parasitoid progeny that reached the adult stage, was significantly higher when the parasitoid was released alone ( $42.0 \pm 2.8\%$ ) than when it was combined with the predatory mite ( $23.3 \pm 4.2\%$ ) ( $F_{2,27} = 76.13$ ;  $P < 0.001$ ) (Fig. 3). In both treatments, a bias towards female production was observed in the parasitoid progeny (*C. tarsi*  $t = 7.47$ ;  $P < 0.01$ , *B. tarsi* + *C. tarsi*  $t = 7.62$ ;  $P < 0.001$ ). The primary factors contributing to the reduction of the pest population were high PIM in the case of the parasitoid treatment and high predation activity of the mites, in addition to the PIM produced by the parasitoid in the treatment with both natural enemies tested together (Fig. 3). The mean number of *O. surinamensis* adults that emerged in the control treatment was  $719.7 \pm 87.6$ .

## 4. Discussion

Parasitoid females were able to penetrate and locate host larvae up to a depth of 150 cm in a vertical column of paddy rice. They had the ability to move freely through the paddy rice kernels in a layer of that thickness since they were able to reduce *O. surinamensis* emergence up to 50% and produced an equal number of offspring at each of the three depths, even when hosts were offered simultaneously at the three depths. Similar results were obtained with another parasitoid, *A. calandrae*, when the host *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) was offered inside the PVC pipes filled with paddy rice at the same three depths tested in the present study (Riudavets et al., 2021). Also, *A. calandrae* and another pteromalid parasitoid, *Lariophagus distinguendus* (Förster), were able to similarly parasitise and reduce the host population, *Callosobruchus chinensis* (L.) (Coleoptera: Chrysomelidae) larvae, at the three depths tested in PVC pipes filled with chickpeas (Iturralde-García et al., 2020b). *Laelius pedatus* (Say) (Hymenoptera: Bethyridae) were also able to penetrate 90 cm deep into wheat grains, successfully parasitizing *Trogoderma granarium* Everts (Coleoptera, Dermestidae) larvae (Al-Kirshi et al., 1997). However, *Holepyris sylvanidis* (Brèthes) (Hymenoptera: Bethyridae) was unable to penetrate 8 cm of fine grist while succeed into coarse grist, indicating that particle size of the substrate affects parasitoid movement (Lorenz et al., 2010).

Therefore, it seems that the small interstitial spaces left among rice grains in that layer thickness are not an obstacle to the movement, location, and host parasitisation by these parasitoids. Since most pests attacking grains perform their activity in the top layer of 1–1.5 m of grain mass (Žďárková et al., 1982; Anukiruthika et al., 2021), the limit for the effectiveness of parasitoids in controlling rice pests must rely on factors beyond the thickness of the grain layer, such as the dispersion of host odours.

Like most parasitoids, *C. tarsi* locates its host by detecting the chemical cues emitted by the grain and target instar activity. Once females locate the host habitat, they locate the larvae by the odours emanating from their faeces and by detecting the host's movement upon contact (Collatz and Steidle, 2008). Traces left by the host larvae when moving on the substrate have a low persistence (Collatz and Steidle, 2008) and are not used by the parasitoid *C. tarsi* for host finding.

Females paralyze the host larvae by stinging them and rubbing their abdomens on them to camouflage them from other females. Once the larvae are paralyzed, the female uses shelters to hide several larvae before oviposition (Howard et al., 1998). In the pipe trial, host larvae were aggregated at one or at three points, which do not reflect their natural distribution when infesting grain, in which host larvae had a more random distribution (Jian et al., 2012). Therefore, in our study, locating the host was supposedly easier than under natural conditions because host odor was concentrated in specific areas.

Both natural enemies were able to similarly reduce the host population by 52–64% when their instar target (eggs or fourth instar larvae) was offered for a short period (one week), as in the small-scale experiment, indicating that both may be comparably effective control agents of *O. surinamensis*. Pre-imaginal mortality of *O. surinamensis* observed in



this experiment, although high, was similar to that observed in other studies (Beckett and Evans, 1994; Collins et al., 1989). Eggs and first instar larvae are so delicate that laboratory handling had probably reduced their survival significantly. Egg consumption by the predatory mite observed in this study was comparable to rates reported for his species with other coleopterans commonly found in storage facilities (Riudavets et al., 2002), confirming this species' broad polyphagia. This is an essential and desirable characteristic of a natural enemy for release in storage facilities since grains and other commodities are attacked by several beetles and other pest species simultaneously.

Also, both natural enemies showed high efficacy when tested in a more realistic setup, such as an intermediate scale with 2 kg paddy rice in which 10 *O. surinamensis* females were introduced and maintained for 10 weeks. The parasitoid proved highly effective in reducing the emergence of *O. surinamensis* by up to 95% when released at a 1:1 parasitoid-to-host ratio. More than 40% of this reduction was caused by effective parasitism, while most of the reduction (54%) was due to PIM, which also occurred in the PVC pipes and the small setup experiments. Host feeding, a behaviour associated with PIM, has been widely described in bethylids (Amante et al., 2017b). For example, oviposition behaviour of *C. stephanoderis* was dependent on *Hypothenemus hampei* (Curculionidae, Scolytinae) density and became independent as host numbers increased (Lauziere et al., 1999): as host density increased, more hosts died from feeding and paralysis than from parasitism. Other bethylids such as *Plastanoxus westwoodi* (Kieffer) and *H. sylvanidis*, attacking the beetle *Cryptolestes pusillus* (Schoenherr) (Coleoptera: Cucujidae) and *Tribolium confusum* (du Val) (Coleoptera: Tenebrionidae), respectively, have also showed high killing rates of their hosts, both through host feeding and oviposition (Ahmed and Khatun, 1996; Rahman and Islam, 2006; Amante et al., 2017a).

Host feeding is a common strategy among parasitoids (Heimpel and Collier, 1996) and is considered essential for both somatic maintenance and reproductive development. All known host-feeding parasitoids are synovigenic (possessing ovaries in which eggs mature throughout their adult life) (Heimpel and Collier, 1996). It is likely that eggs present upon adult emergence are not completely developed and require the female to feed on the host to mature them. In addition, interference among conspecific females could explain the elevated PIM values since it affects their foraging and reproductive behaviour. Similar efficacy results in reducing host emergence and % of PIM in this reduction were obtained with other parasitoids of stored-product pests, as in the case of *A. calandrea* with *Sitophilus* spp. as host (Solá et al., 2020).

In this experiment, the predatory mite reduced the pest population by nearly half, a lower proportion than that caused by the parasitoid. This predatory mite feeds exclusively on eggs, while the parasitoid oviposits in fourth instar larvae or pupae and can also feed on any larval instar. We completed four releases of the predatory mite every two weeks. However, females of *O. surinamensis* laid eggs continuously, and some eggs laid between successive releases were free from predation. The longevity of these mites at 28 °C probably does not reach two weeks (Çobanoğlu et al., 2007). Possibly, if we had done more frequent releases of the predatory mite, the effectiveness of this natural enemy might have increased. In the 10 weeks of the study, two generations of the pest occurred (Eliopoulos, 2019), while four generations of the parasitoid (Eliopoulos, 2019) and 12 of the predatory mite took place (Çobanoğlu et al., 2007).

In contrast to the findings of Žďárková et al. (2003), in which the combination of *C. tarsalis* and the predatory mite *Cheyletus eruditus* (Schrank) led to an improvement in reducing *O. surinamensis*, the presence of *B. tarsalis* in our study did not enhance the effectiveness of the parasitoid alone. This could be attributed to the already high efficacy of the parasitoid, leaving little room for further improvement. A similar high effectiveness of the larval parasitoid *A. calandrea* was observed with *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) and *S. zeamais* (Solá et al., 2020).

In conclusion, *C. tarsalis* has been shown to be a promising natural

enemy for the control of *O. surinamensis* at the rates tested in this study. Further studies should improve our knowledge of its behaviour to determine how to use it in more realistic scenarios. *Blattisocius tarsalis* has also shown that it is a promising control agent of interest for controlling this pest and other coleopterans that are pests of stored products.

## CRedit authorship contribution statement

**Lidia del Arco:** Data curation, Investigation, Methodology, Writing – review & editing. **Jordi Riudavets:** Conceptualization, Formal analysis, Methodology, Writing – review & editing, Funding acquisition. **Cristina Castañé:** Conceptualization, Formal analysis, Funding acquisition, Methodology, Writing – review & editing.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

Data will be made available on request.

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## References

- Ahmed, K.N., Khatun, M., 1996. The biology of pilachna beetle, *Epilachna septima* Dieke (Coleoptera: Coccinellidae: Epilachninae) infesting bittergourd in Bangladesh. *Bangladesh J. Sci. Ind. Res.* 31, 147–152.
- Al-Kirshi, A.G., Reichmuth, Ch, Bochow, H., 1997. Potential of the larval parasitoid *Laelius pedatus* (Say) (Hymenoptera, Bethyidae) for the control of the khapra beetle *Trogoderma granarium* Everts (Coleoptera, Dermestidae) in grain. *Mitt. Dts. Ges. Allg. Angew. Entomol.* 11, 367–372.
- Amante, M., Schöller, M., Hardy, I.C., Russo, A., 2017a. Reproductive biology of *Holepyris sylvanidis* (Hymenoptera: Bethyidae). *Biol. Control* 106, 1–8.
- Amante, M., Schöller, M., Suma, P., Russo, A., 2017b. Bethyids attacking stored-product pests: an overview. *Entomol. Exp. Appl.* 163, 251–264. <https://doi.org/10.1111/eea.12587>.
- Anukiruthika, T., Jian, F., Jayas, D.S., 2021. Movement and behavioral response of stored product insects under stored grain environments - a review. *J. Stored Prod. Res.* <https://doi.org/10.1016/j.jspr.2020.101752>.
- Armitage, D.M., Day, P.G., Lewis, G.J., 1983. The effect of aeration on the vertical distribution of grain beetles (Coleoptera) in wheat. *Bull. Entomol. Res.* 73, 233–237. <https://doi.org/10.1017/S0007485300008828>.
- Arthur, F.H., 2001. Immediate and delayed mortality of *Oryzaephilus surinamensis* (L.) exposed on wheat treated with diatomaceous earth: effects of temperature, relative humidity, and exposure interval. *J. Stored Prod. Res.* 37, 13–21. [https://doi.org/10.1016/S0022-474X\(99\)00058-2](https://doi.org/10.1016/S0022-474X(99)00058-2).
- Beckett, S.J., Evans, D.E., 1994. The demography of *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae) on kibbled wheat. *J. Stored Prod. Res.* 30, 121–137. [https://doi.org/10.1016/0022-474X\(94\)90190-2](https://doi.org/10.1016/0022-474X(94)90190-2).
- Castañé, C., Riudavets, J., 2015. Sampling arthropod pests and natural enemies in stored barley. *J. Stored Prod. Res.* 64, 54–61. <https://doi.org/10.1016/j.jspr.2015.08.005>.
- Castañé, C., Agustí, N., Estal, P. del, Riudavets, J., 2020. Survey of *Trogoderma* spp. in Spanish mills and warehouses. *J. Stored Prod. Res.* 88, 101661 <https://doi.org/10.1016/j.jspr.2020.101661>.
- Çobanoğlu, S., Bayram, Ş., Denizhan, E., Sağlam, H., 2007. The effect of different temperatures and food densities on development of *Blattisocius tarsalis* (Berlese) (Acari: Ascidae) reared on Mediterranean flour moth *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae). *Türkiye Entomoloji Dergisi* 31 (1), 7–20.
- Collatz, J., Steidle, J.L.M., 2008. Hunting for moving hosts: *Cephalonomia tarsalis*, a parasitoid of free-living grain beetles. *Basic Appl. Ecol.* 9, 452–457. <https://doi.org/10.1016/J.bae.2007.04.002>.

- Collins, P.J., Mulder, J.C., Wilson, D., 1989. Variation in life history parameters of *Oryzaephilus surinamensis* (L). (Coleoptera: Silvanidae). J. Stored Prod. Res. 25, 193–199. [https://doi.org/10.1016/0022-474X\(89\)90024-6](https://doi.org/10.1016/0022-474X(89)90024-6).
- Duarte, S., Barros, G., Carvalho, L., Guerreiro, O., Mourato, M., Carvalho, M.O., 2021. Early-warning detection protocol of khapra beetle (*Trogoderma granarium* Everts) and other insect pests associated with stored grains in Portugal - preliminary results. In: Jayas, D.S., Jian, F. (Eds.), Proceedings 11<sup>th</sup> International Conference on Controlled Atmosphere and Fumigation in Stored Products (CAF2020), CAF Permanent Committee Secretariat. Winnipeg, Canada, pp. 186–192.
- Eliopoulos, P.A., Athanassiou, C.G., Buchelos, C.H., 2002. Occurrence of hymenopterous parasitoids of stored product pests in Greece. IOBC-WPRS Bull. 25 (3), 127–140.
- Eliopoulos, P.A., 2019. Life table parameters of the parasitoid *Cephalonomia tarsalis* (Hymenoptera: Bethyilidae) and its host the sawtoothed grain beetle *Oryzaephilus surinamensis* (Coleoptera: Silvanidae). J. Plant Protect. Res. 59, 544–551. <https://doi.org/10.24425/jppr.2019.131269>.
- Eliopoulos, P.A., Kapranas, A., Givropoulou, E.G., Hardy, I.C.W., 2017. Reproductive efficiency of the bethylid wasp *Cephalonomia tarsalis*: the influences of spatial structure and host density. Bull. Entomol. Res. 107, 139–147. <https://doi.org/10.1017/S0007485316000651>.
- Feroz, A., Shakoori, F.R., Riaz, T., Shakoori, A.R., 2020. Development of resistance in stored grain pest, *Trogoderma granarium* (Everts) against deltamethrin and its effective control by synergistic toxicity of bifenthrin and chlorpyrifos. J. Stored Prod. Res. 88, 101673. <https://doi.org/10.1016/j.jspr.2020.101673>.
- Gallego, J.R., Caicedo, O., Gamez, M., Hernandez, J., Cabello, T., 2020. Selection of predatory mites for the biological control of potato tuber moth in stored potatoes. Insects 11, 196. <https://doi.org/10.3390/insects11030196>.
- Gu, X., Li, G.Y., Zhang, Z.Q., 2022. Stage and sex-dependent responses of immature predatory mites (*Blattisocius dentriticus*) to predation risk from cannibalistic conspecifics. J. Stored Prod. Res. 99, 102027. <https://doi.org/10.1016/J.JSPR.2022.102027>.
- Haines, C.P., 1981a. Laboratory studies on the role of an egg predator, *Blattisocius tarsalis* (Berlese) (Acari: Ascidae), in relation to the natural control of *Ephestia cautella* (Walker) (Lepidoptera: Pyralidae) in Warehouses. Bull. Entomol. Res. 71, 557–574. <https://doi.org/10.1017/S0007485300010075>.
- Haines, C.P., 1981b. Insects and Arachnids from Stored Products: a Report on Specimens Received by the Tropical Stored Products Centre 1973-77, L54. Rep. trop. Prod. Inst., pp. 1–73.
- Haines, C.P., 1974. Insects and arachnids from stored products: a report on specimens received by the Tropical Stored Products Centre 1972-1973. Rep. trop. Prod. Inst. L 39, 22.
- Heimpel, G.E., Collier, T.R., 1996. The evolution of host-feeding behaviour in insect parasitoids. Biol. Rev. 71, 373–400.
- Hill, D.S., 2003. Pests of Storage Foodstuffs and Their Control. Kluwer Academic Publishers, New York.
- Hervet, V.A.D., Morrison III, W.R., 2021. Prospects for use of biological control of insect and mites for the food industry in North America. Agronomy 11, 1969. <https://doi.org/10.3390/agronomy11101969>.
- Howard, R.W., Charlton, M., Charlton, R.E., 1998. Host-finding, host-recognition, and host-acceptance behavior of *Cephalonomia tarsalis* (Hymenoptera: Bethyilidae). Ann. Entomol. Soc. Am. 91, 879–889. <https://doi.org/10.1093/aesa/91.6.879>.
- Iturralde-García, R.D., Castañé, C., Wong-Corral, F.J., Riudavets, J., 2020a. Biological control of *Acanthoscelides obtectus* and *Zabrotes subfasciatus* in stored dried beans. BioControl 65, 693–701. <https://doi.org/10.1007/s10526-020-10048-5>.
- Iturralde-García, R.D., Riudavets, J., Castañé, C., 2020b. Biological control of *Callosobruchus chinensis* (Coleoptera: Chrysomelidae) in stored chickpeas through the release of natural enemies. Biol. Control 149, 104322. <https://doi.org/10.1016/j.biocontrol.2020.104322>.
- Jian, F., Larson, R., Jayas, D.S., White, N.D.G., 2012. Three dimensional temporal and spatial distribution of adult *Oryzaephilus surinamensis* and *Sitophilus oryzae* in stored wheat under different temperatures, moisture contents, and adult densities. J. Stored Prod. Res. 49, 155–165. <https://doi.org/10.1016/j.jspr.2012.01.001>.
- Kavallieratos, N.G., Athanassiou, C.G., Arthur, F.H., 2015. Efficacy of deltamethrin against stored-product beetles at short exposure intervals or on a partially treated rice mass. J. Econ. Entomol. 108, 1416–1421. <https://doi.org/10.1093/jee/tov060>.
- Lauziere, I., Pérez-Lachaud, G., Brodeur, J., 1999. Influence of host density on the reproductive strategy of *Cephalonomia stephanoderis*, a parasitoid of the coffee berry borer. Entomol. Exp. Appl. 92, 21–28. <https://doi.org/10.1046/j.1570-7458.1999.00520.x>.
- Lord, J.C., 2006. Interaction of *Mattesia oryzaephili* (Neogregarinorida: Lipotrophidae) with *Cephalonomia* spp. (hymenoptera: Bethyilidae) and their hosts *Cryptolestes ferrugineus* (Coleoptera: Laemophloeidae) and *Oryzaephilus surinamensis* (Coleoptera: Silvanidae). Biol. Control 37, 167–172. <https://doi.org/10.1016/j.biocontrol.2006.01.016>.
- Lorenz, S., Adler, C., Reichmuth, C., 2010. Penetration ability of *Holepyris sylvanidis* into the feeding substrate of its host *Tribolium confusum*. Julius-Kühn-Archiv 425, 721.
- Mahroof, R.M., Hagstrum, D.W., 2012. Biology, behavior, and ecology of insects in processed commodities. In: Hagstrum, D.W., Phillips, T.W., Cuperus, G. (Eds.), Stored Product Protection. Kansas State University, Manhattan, KS, pp. 33–44.
- Morrison, W.R., Agraftioti, P., Domingue, M.J., Scheff, D.S., Lampiri, E., Gourgouta, M., Baliota, G.V., Sakka, M., Myers, S.W., Athanassiou, C.G., 2023. Comparison of different traps and attractants in 3 food processing facilities in Greece on the capture of stored product insects. J. Econ. Entomol. <https://doi.org/10.1093/jee/toad107>.
- Navarro, S., Amos, T.G., Williams, P., 1981. The effect of oxygen and carbon dioxide gradients on the vertical dispersion of grain insects in wheat. J. Stored Prod. Res. 17, 101–107. [https://doi.org/10.1016/0022-474X\(81\)90008-4](https://doi.org/10.1016/0022-474X(81)90008-4).
- Nielsen, S., 1999. The impact of temperature on activity and consumption rate of moth eggs by *Blattisocius tarsalis* (Acari: Ascidae). Exp. Appl. Acarol. 23, 149–157. <https://doi.org/10.1023/A:1006054627912>.
- Powell, D., 1938. The biology of *Cephalonomia tarsalis* (Ash.), a vespid wasp (Bethyilidae: hymenoptera) parasitic on the sawtoothed grain beetle. Ann. Entomol. Soc. Am. 31, 44–49. <https://doi.org/10.1093/aesa/31.1.44>.
- Rahman, M.M., Islam, W., 2006. Influence of temperature on *Platanoxys westwoodi* (Kieffer) (Hymenoptera: Bethyilidae), an ectoparasitoid of red flat grain beetle *Cryptolestes pusillus* (Schon.) (Coleoptera: Cucujidae). Pak. entomol. 28, 39–44.
- Riudavets, J., Maya, M., Montserrat, M., 2002. Predation by *Blattisocius tarsalis* (Acari: Ascidae) on stored product pests. IOBC-WPRS Bull. 25 (3), 121–126.
- Riudavets, J., Martínez-Ferrer, M.T., Campos-Rivela, J.M., Agustí, N., Castañé, C., 2021. Releases of the parasitoid *Anisopteromalus calandrae* (Hymenoptera: Pteromalidae) can control *Sitophilus zeamais* (Coleoptera: Curculionidae) in big bags of paddy rice. Biol. Control 163, 104752. <https://doi.org/10.1016/j.biocontrol.2021.104752>.
- RStudio Team, 2023. RStudio: Integrated Development for R. RStudio, Version 2023.06, 0+421.
- Solá, M., Riudavets, J., Castañé, C., 2020. Control of *Rhyzopertha dominica* and *Sitophilus zeamais* in stored rice with different release rates of the larval parasitoid *Anisopteromalus calandrae*. Entomol. Gen. 40, 323–330. <https://doi.org/10.1127/entomologia/2020/0899>.
- Stejskal, V., Kosina, P., Kanyomeka, L., 2006. Arthropod pests and their natural enemies in stored crops in northern Namibia. J. Pest. Sci. 79, 51e55. <https://doi.org/10.1007/s10340-005-0109-2>.
- Thind, B.B., Ford, H.L., 2006. Laboratory studies on the use of two new arenas to evaluate the impact of the predatory mites *Blattisocius tarsalis* and *Cheyletus eruditus* on residual populations of the stored product mite *Acarus siro*. Exp. Appl. Acarol. 38, 167–180. <https://doi.org/10.1007/s10493-005-5829-8>.
- Vélez, M., Barbosa, W.F., Quintero, J., Chediak, M., Guedes, R.N.C., 2017. Deltamethrin and spinosad-mediated survival, activity, and avoidance of the grain weevils *Sitophilus granarius* and *S. zeamais*. J. Stored Prod. Res. 74, 56–65. <https://doi.org/10.1016/j.jspr.2017.10.002>.
- Žďárková, E., Lukáš, J., Horák, P., 2003. Compatibility of *Cheyletus eruditus* (Schränk) (Acari: Cheyletidae) and *Cephalonomia tarsalis* (Ashmead) (hymenoptera: Bethyilidae) in biological control of stored grain pests. Plant Protect. Sci. 39, 29–34. <https://doi.org/10.17221/3824-PPS>.
- Žďárková, E., Verner, P.H., Novosad, J., 1982. Dispersion and distribution of mites and beetles in stored grain. J. Stored Prod. Res. 19, 73–80. [https://doi.org/10.1016/0022-474X\(83\)90029-2](https://doi.org/10.1016/0022-474X(83)90029-2).