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RESEARCH

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Pre-emptive augmentative biological control of *Spodoptera frugiperda* in Europe using *Trichogramma* spp.

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Abstract

Background Fall armyworm, *Spodoptera frugiperda*, is a highly invasive pest of maize and other crops worldwide. It has recently been detected in Europe, and it is urgent to test and develop new sustainable control methods adapted to the European context and market. *Trichogramma* spp. are egg parasitoids that are sometimes used as biological control agent against *S. frugiperda* in other continents. However, a major issue using *Trichogramma* spp. against this pest is that females cannot reach all eggs in an egg mass, which is usually composed of one to three layers of eggs, often covered with scales and hair. Three European *Trichogramma* species were tested for their ability to parasitize egg masses with one to three layers and with or without hair and scale cover.

Methods *Trichogramma brassicae*, *T. dendrolimi* and *T. cacoeciae* were offered five types of *S. frugiperda* egg masses: one-layer without hair; one-layer with hair; two-layers without hair; two-layers with hair; three-layers with hair. For each treatment, an egg mass laid on paper was placed in a small vial saturated with females, to be sure that all reachable eggs would be parasitized. For each egg mass, the number of eggs in each layer was counted. Fifteen replicates were made per treatment, as well as 10 control vials without *Trichogramma* sp. Parasitism rates were calculated for each egg mass.

Results There were significant differences in the ability of parasitoid species to oviposit through hair and scales and to reach the lower egg layers. *Trichogramma dendrolimi* was the most efficient species and *T. cacoeciae* the least. Depending on the number of egg layers and hair and scales, parasitism rates by *T. brassicae*, *T. dendrolimi* and *T. cacoeciae* varied between 99 and 41%, 100 and 43% and 100 and 28%, respectively.

Conclusions The assays confirmed that *Trichogramma* females cannot easily oviposit through thick layers of hair and scales but overall parasitism rates were higher than found previously. Important variations between species were found, and more *Trichogramma* sp. and other local natural enemies should be tested pre-emptively before *S. frugiperda* has invaded Europe.

Keywords Egg parasitism, Fall armyworm, Proactive biological control, *Trichogramma brassicae*, *Trichogramma cacoeciae*, *Trichogramma dendrolimi*

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Background

The fall armyworm, *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae) is a very serious pest of maize and can occasionally damage many other crops such as sorghum, rice, soybean, cotton, etc. It is native to the Americas and was first found in West Africa in 2016, from where it quickly invaded most of the African continent, many Asian countries and Australia (Kenis et al. 2023). Pest distribution and climatic suitability models (du Plessis et al. 2018; Early et al. 2018) suggest that *S. frugiperda* will spread further north and establish in the extreme southern Europe. *Spodoptera frugiperda* migrates in summer to temperate regions in North America and Asia, causing damage to maize and other crops, and there is little doubt that, sooner or later, it will become a transient pest in most of Southern Europe as well (Babendreier et al. 2022). Indeed, in 2023, *S. frugiperda* adults and larvae were found in Cyprus and Greece and adults were also caught in traps in Romania (EPPO 2023).

In its native area, *S. frugiperda* is mostly controlled using GM crops and broad-spectrum insecticides. Various new management methods are presently being tested and developed in newly invaded areas in Africa and Asia (Kenis et al. 2023). However, in general, these methods have been developed for tropical and sub-tropical regions. If *S. frugiperda* were to arrive and become a pest of maize, and potentially some other crops in Europe, integrated pest management (IPM) should be developed that specifically focuses on migratory populations, taking into account the specificities of European countries such as the availability of biological control agents and the ban on GM crops (Babendreier et al. 2022). Preemptive, or proactive biological control, i.e. the study of biological control options before an invasive pest arrives in a region, is gaining importance worldwide (Avila et al. 2023; Hoddle 2024). In general, biological control can only be applied long after the arrival and spread of an invasive pest because the homologation of natural enemies for classical or augmentative biological control can take several years. Therefore, a pre-emptive approach assessing different biological control options in advance of the arrival of a pest would be desirable, in particular for rapidly spreading and devastating species such as *S. frugiperda*.

Trichogramma spp. (Hym.: Trichogrammatidae) are egg parasitoids that are commonly used as augmentative biological control agents against various pests worldwide (Smith 1996; Zang et al. 2021). They have been frequently tested and sometimes used against *S. frugiperda*, both in the native regions and in the invaded region (Li et al. 2023a; Navik et al. 2023). In the Americas, *T. pretiosum* Riley and *T. atopovirilia* Oatman & Platner have been tested against *S. frugiperda*, in particular in Brazil (e.g. Figueiredo et al. 2015; Parra 2010) and Mexico (Sanchez et al. 1999). Inundative releases of *T. pretiosum* sometimes resulted in egg mass parasitism of up to 79% with yield increase of 700 kg/ha per ha (Figueiredo et al. 2015). In recent years, several laboratory and field tests have also been conducted with *Trichogramma* spp. in China. For example, *T. chilonis* Ishii was released in maize fields in Shandong Province, resulting in parasitism rates of eggs and egg masses up to 73 and 86% respectively (Yang et al. 2019) while subsequent field trials with *T. chilonis*, *T. dendrolimi* Matsumura and *T. pretiosum* revealed egg parasitism rates between 11 and 31% (Yang et al. 2022). In Hainan Island, China, four *Trichogramma* species were released, with parasitism rates of *S. frugiperda* egg masses ranging from 61.5 to 87.5% (Jin et al. 2021). Despite high numbers of egg masses showing *Trichogramma* parasitism, it is usually considered that a full control of FAW with *Trichogramma* spp. alone is not easily achieved because many egg masses are composed of two or three layers of eggs and/or are covered by a variable amount of hair and scales, a strategy used by many Lepidoptera to protect their eggs from natural enemies and heavy rain (Hou et al. 2022). In general, *S. frugiperda* females tend to lay larger egg masses with more layers and more hair and scales when they are young (Li et al. 2023b) *Trichogramma* females are mostly able to access the upper layer of the egg masses and those at the edges and cannot easily oviposit through the hair and scales when these are too abundant (Beserra and Parra 2005; Dong et al. 2021; Hou et al. 2022; Liu et al. 2023). The ability to overcome this protection can vary between *Trichogramma* species. For example, *T. dendrolimi*, demonstrated higher biological control potential on *S. frugiperda* in the laboratory than three other *Trichogramma* species because it is better able to parasitize eggs through the hair and scales covering FAW egg masses (Sun et al. 2020). Other recent studies have shown differences in the ability of parasitizing *S. frugiperda* among *Trichogramma* species from China (Li et al. 2023a; Liu et al. 2023) and India (Navik et al. 2024).

While the suitability of *Trichogramma* spp. to parasitize *S. frugiperda* eggs has been extensively studied in Asia, such investigations have never been conducted in Europe. Neveretheless, if augmentative biological control methods must be developed against *S. frugiperda* in Europe, it is important to select among indigenous biological control agents and, if possible, among those that are already available. *Trichogramma* spp. are also used in Europe against crop pests, particularly *T. brassicae* Bezdenko against another important maize pest, *Ostrinia nubilalis* (Hübner) (Koch et al. 2019). It is commercially available for maize farmers and, thus, it is logical to consider this species as primary candidate for testing against *S. frugiperda* in Europe. *Trichogramma cacoeciae* Marchal is a species also commercially available in Europe against various pests, such as codling moth, *Cydia pomonella* (L.), in orchards and others in vegetables. *Trichogramma dendrolimi* is a Eurasian species that is also listed and available for augmentative biological control in Europe (e.g. Koch et al. 2019; Barantage et al. 2023; EPPO 2021). It is known as parasitoid of Noctuidae and showed promising results in China against *S. frugiperda* (Sun et al. 2020; Li et al. 2023b; Liu et al. 2023). Considering that the main issue of using *Trichogramma* spp. against *S. frugiperda* is their inability to parasitise the entire egg masses when these consist of more than one layer and/or are densely covered by hair and scales, we tested the parasitism rates of these three species in laboratory conditions on egg masses with 1–3 layers and with/without hair and scale cover.

Material and methods

Host and parasitoid rearing

Spodoptera frugiperda egg masses were obtained from a laboratory rearing maintained in the CABI quarantine laboratory in Delémont, Switzerland. The population belongs to the maize strain and was originally obtained from a laboratory colony maintained at the University of Neuchâtel, Switzerland. Adult *S. frugiperda* were reared in a bugdorm-1 rearing cage (30 × 30 × 30 cm), fed with drops of honey and water. Egg masses were laid on cellulose paper placed on the 4 walls of the cage and collected daily. Eggs were then placed in a Petri-dish (10 cm diameter) with a ventilated lid in which the larvae hatch. When they reached the third instar, they were placed in large (30 × 24 × 10 cm) plastic boxes with a ventilated lid, with cellulose paper at the bottom to allow pupation. Young larvae in the Petri-dishes were fed with the Beet Armyworm Diet of Frontier Scientific, Newark, NJ, USA, and the older larvae in plastic boxes were fed with a McMorran diet (Hervet et al. 2016). The pupae were placed in the adult cages for emergence.

A culture of *Trichogramma brassicae* was kindly provided by Andermatt Biocontrol whereas *T. dendrolimi* and *T. cacoeciae* were obtained from a permanent rearing at JKI, maintained by one of the authors (origin of strains, see Schäfer and Herz 2020). *Trichogramma brassicae* was first reared on *S. frugiperda* eggs obtained from the *S. frugiperda* rearing maintained in a quarantine laboratory at CABI. However, while this preliminary rearing has been very useful to calibrate future tests, it appeared that the method was highly unpractical because it required a high amount of eggs, and young larvae hatching after 3 days quickly fed upon parasitised eggs if the larvae were not removed very fast after hatching (– 12 h). Thus, it was decided to maintain the *T. brassicae* rearing, and later on the *T. dendrolimi* and *T. cacoeciae* rearing, on sterile eggs of the Mediterranean flour moth *Ephestia kuehniella* Zeller, first purchased from Bioline, France, and then from Agroline, Switzerland. *Ephestia kuehniella* eggs were glued on yellow pieces of cardboard before being offered to freshly emerged *Trichogramma* spp. adults. The whole rearing was maintained in an incubator at 25 ± 1 °C, 60 ± 10% relative humidity (RH) and with a photoperiod of 16:8 h. At this temperature, the development time is 9–10 days for *T. brassicae* and *T. dendrolomi*, and 10–11 days for *T. cacoeciae*. Thus, new carboard pieces with glued *E. kuehniella* were placed every 9–11 days in the vials, when old ones were removed.

Egg parasitism

Trichogramma brassicae, T. dendrolimi and *T. cacoeciae* were tested on 5 types of egg masses, varying in the number of egg layers and the presence or not of a cover of hair and scales (hereafter mentioned as "hair"): one-layer without hair; one-layer with hair; two-layers without hair; two-layers with hair; three-layers with hair. Egg masses with 3 layers from the laboratory rearing had always hair on them. The amount of hair was highly variable among egg masses. However, for the experiments, all egg masses without hair had all visible eggs directly accessible without hair (in some cases, egg masses were brushed to remove a small amount of hair). In contrast, all egg masses considered as "with hair" always had a rather large amount of hair, covering over 90% of the visible eggs, similar to egg masses shown with at least 180 µm of scales in Li et al. (2023c). Fifteen replicates per treatment were made and for each of the five treatments, 10 egg masses were used as control.

For control egg masses, the number of eggs was counted under a stereomicroscope, counting separately eggs visible from above from those supposedly hidden by the upper layers. Then, the larvae were counted after hatching (3 days at 25 °C). The number of larvae hatched divided by the number of counted eggs was considered as the expected percentage of emergence and integrated in the calculation of parasitism rates (see below).

For the tests themselves, egg masses of *S. frugiperda* were offered to *Trichogramma* spp. in a small vial (4 cm high, 1 cm diameter). Fresh egg masses laid in the previous night (< 24 h old) were placed with parasitised *E. kuehniella* eggs, aged of 9 days for *T. brassicae* and *T. dendrolimi*, and 10 days for *T. cacoeciae*, i.e. less than 1 day before adult emergence. About one *E. kuehniella* egg was provided for one *S. frugiperda* egg. Since parasitised *E. kuehniella* eggs contain one *Trichogramma* and most *S. frugiperda* eggs two or three (see below), and considering that *T. cacoeciae* is thelytokous and the sex ratio *for T. brassicae and T. dendrolimi* was about 50–50, we can consider that *S. frugiperda* eggs were saturated with *Trichogramma spp.* females. Although this saturation resulted in some *S. frugiperda* eggs collapsing because of an excess of superparasitism, this method was chosen to ensure that all reachable eggs were parasitised. For all egg masses placed in the vials, the number of eggs was counted under a stereomicroscope, counting separately eggs visible from above from those supposedly hidden by the upper layers.

The egg mass was exposed 2 days to *Trichogramma* spp. in the vial before being removed and placed in a small petri-dish (5 cm diameter). One day later, the hatching larvae (– 12 h old) were collected from the petri-dish and counted. Parasitism was calculated using the formula:

Parasitism rate = (1 – number of larvae hatched/number of expected larvae) X 100.

The number of expected larvae was calculated by multiplying the number of eggs counted by the proportion of counted eggs hatched in the control, measured separately for each treatment.

The sex-ratio and the number of *Trichogramma* adults developing per *S. frugiperda* egg of *T. brassicae* and *T. dendrolimi* in *S. frugiperda* eggs were calculated by selecting, for each species, 50 parasitised eggs, i.e. 10 eggs from 5 almost fully parasitised egg masses (but without collapsing eggs) and by counting and sexing all adults emerged per egg. The few eggs from where *Trichogramma* spp. did not hatch were excluded from the analysis. This experiment was not conducted with *T. cacoeciae*, which is known as a thelytokous species (Vavre et al. 2004). Only females were observed in our rearing.

Statistical analyses

Egg parasitism data were analyzed for the three factors '*Trichogramma* species', 'no. of egg layers' and 'hair coverage' based on general linear models, followed by Tukey's Honest Significant Difference post hoc tests for 'no. of egg layers' and '*Trichogramma* species' (n = 15 per treatment, n = 225 in total). In addition, differences in egg parasitism rates for the five treatments were tested, using general linear models and Tukey's HSD test, separately for the three species. Parasitism rates were arcsin-squareroot transformed to match assumptions, and all analyses were performed with SPSS 27.0.

Results

The control experiment showed that, in the five treatments, the number of larvae hatching from the egg masses represented between 90 and 98% of the number of eggs counted (Table 1). This is probably partly due to some non-hatched eggs (in particular in one layer egg masses without hair) and partly because very few eggs were eaten by hatching larvae. It may also indicate a slight error in egg counting under thick hair covers and hidden in the lower layers. These numbers represent the expected proportion of larvae hatched in the main experiments, on the basis of which parasitism rates were calculated.

Table 1 Proportion of larvae hatched per egg counted, for each category of egg mass

Egg mass category	No. of larvae hatched/ No. of eggs counted
One-layer, no hair	0.90 ± 0.02
One-layer, with hair	0.93 ± 0.02
Two-layer, no hair	0.95 ± 0.02
Two-layer, with hair	0.98 ± 0.02
Three-layer, with hair	0.93 ± 0.03

N = 10 ± SE

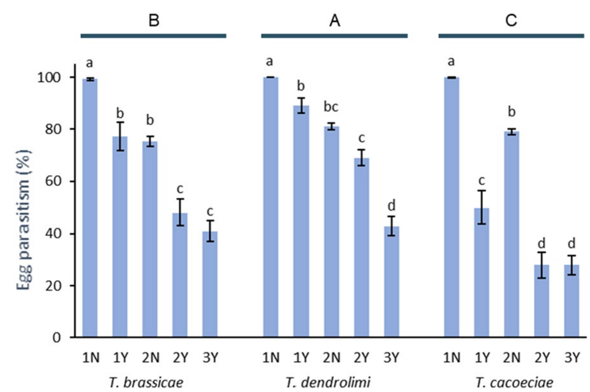

Fig. 1 Effect of the number of layers of egg masses and hair coverage on mean *S. frugiperda* egg parasitism (± SD) by *Trichogramma brassicae*, *T. dendrolimi* and T. *caoeciae,* for the five treatments: one-layer without hair (1N); one-layer with hair (1Y); two-layers without hair (2N); two-layers with hair (2Y); three-layers with hair (3Y). N = 15 for all treatments. Same small letters above bars within a *Trichogramma* species indicate non-significant differences at 0.05 level among treatments. Same capital letters above species indicate non-significant differences at 0.05 level among *Trichogramma* species

Parasitism rates by *T. brassicae*, *T. dendrolimi* and *T. cacoeciae* were 99.3, 100 and 99.9%, respectively, in one-layer egg masses with no hair and decreased to 41, 43 and 28% in the three-layers egg masses with hair (Fig. 1). All 75 egg masses counted together, parasitism rate was 67.7% for *T. brassicae*, 78.3% for *T. dendrolimi* and 56.9% for *T. cacoeciae*.

Highly significant differences in egg parasitism levels were found in the global analysis ($F_{12,210} = 196.0$, $p < 0.001$) and all three factors contributed to this. A highly significant difference was found between parasitism rates of egg masses with and without hair

($F_{1,210}=235.0$; $p<0.001$), thus the hair cover provides a certain level of protection of FAW egg masses. However, still a substantial proportion of eggs were successfully parasitized when egg masses were covered with hair (Fig. 1). Furthermore, egg masses with only one layer showed the highest and egg masses with three layers the lowest parasitism rates ($F_{2,210}=138.3$; $p<0.001$). Overall, significant differences were found in parasitism rates among the *Trichogramma* species ($F_{2,210}=19.34$; $p<0.001$) with *T. dendrolimi* showing the highest, and *T. cacoeciae* the lowest parasitism. This led to a highly significant interaction between the species tested and the factor hairs ($F_{1,210}=27.67$; $p<0.001$), indicating that *T. dendrolimi* is least affected by hair covering egg masses, and *T. cacoeciae* the most. Similarly, a highly significant interaction was found between the species tested and the number of egg layers ($F_{4,210}=4.85$; $p<0.001$), with *T. dendrolimi* being the species most capable of parasitizing hidden eggs and *T. cacoeciae* the least capable.

When looking at only the egg masses without hair, the differences in parasitism rate among species gets small, though still significant ($F_{2,84}=4.83$; $p=0.01$). The layers still affect parasitism highly significantly ($F_{1,84}=1271.7$; $p<0.001$) while the interaction between layers and species becomes insignificant ($F_{2,84}=0.71$; $p=0.50$). In contrast, when only analysing egg masses with hair, the difference in parasitism gets larger ($F_{2,126}=33.0$; $p<0.001$), with highly significant effects of the layers ($F_{2,126}=1271.7$; $p<0.001$) and a significant interaction between layers and species ($F_{4,126}=3.13$; $p=0.017$).

Figure 2 shows the rate of parasitism by *T. brassicae, T. dendrolimi and T. cacoeciae* against the proportion of hidden eggs in egg masses with two layers without hair (n = 15). For the three species, parasitism decreased with the proportion of hidden eggs. For the majority of the egg masses, parasitism was slightly higher than expected (more points are found above the red line than below). With *T. dendrolimi*, in all but one egg mass parasitism rate was higher than the proportion of exposed eggs (all points are found above or on the red line).

The number of adult *T. brassicae* and *T. dendrolimi* emerging per *S. frugiperda* varied between 1 and 4 and was on average 2.2 and 2.7, respectively. The sex ratio was 49% males in *T. brassicae* and 43% in *T. dendrolimi*.

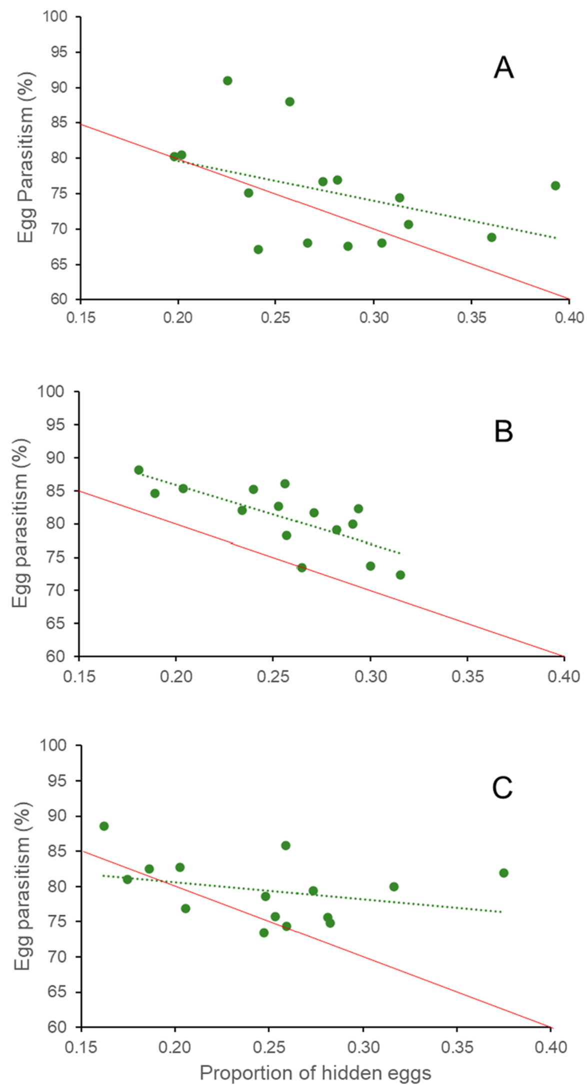

Fig. 2 Parasitism of *S. frugiperda* eggs by *T. brassicae* (**A**), *T. dendrolimi* (**B**) and *T. cacoeciae* (**C**) in the two-layers no hair treatment, in function of the proportion of hidden eggs. The blue dotted lines indicate the trend, and the red lines indicate the expected relation if all exposed eggs were attacked and if none of the non-exposed ones were attacked

Discussion

Our results confirm previous observations that *Trichogramma* females attack mainly the upper layer of the egg masses, or those exposed at the edges, and cannot easily oviposit through thick layers of hair and scales (Beserra and Parra 2005; Dong et al. 2021; Hou et al. 2022). A dense cover of hair and scales reduces the access of the *Trichogramma* spp. females to the eggs. In this experiment, not all egg masses with hair were covered exactly by the same amount of hair and scales, even though we selected only egg masses that were well covered (> 180 µm as described in Li et al. 2023c). It would be instructive to select different levels of hair and scale coverage and assess how *Trichogramma* spp. react to these differences, as done by Li et al. (2023c). Parasitism was higher in *T. dendrolimi* than in *T. brassicae* and *T. cacoeciae* for the treatments one-layer with hair and two-layers with hair, suggesting that *T. dendrolimi* can cope better with hair and scales than *T. brassicae* and particularly *T.*

cacoeciae. This confirms the results of Sun et al. (2020) who showed that *T. dendrolimi* reached higher parasitism rates than three other *Trichogramma* spp. Liu et al. (2023) also tested five *Trichogramma* spp. on *S. frugiperda* in China, including *T. dendrolimi* and *T. cacoeciae*, and found that *T. dendrolimi* was performing better than *T. cacoeciae*, but less well than *T. ostriniae* Pang et Chen. *Trichogramma dendrolimi* also had the longest ovipositor, which may explain its good performances (Liu et al. 2023).

Trichogramma spp. were unsuccessful in parasitising all eggs when the egg mass contained more than one layer, even without hair. This was expected based on similar reports from recent studies (e.g. Navik et al. 2024). When assessing the rate of parasitism in function of the proportion of hidden eggs in egg masses with two layers without hair, parasitism decreased with the proportion of hidden eggs. For the majority of the egg masses, parasitism was higher than expected. This is likely due to the fact that, when counting the exposed and hidden eggs, we assumed that the number of hidden eggs was similar to that of the top layer. At the edge of the egg masses, we counted as exposed eggs those that were at least half exposed. This may be slightly conservative because the eggs that are less than half exposed are probably still accessible to *Trichogramma* spp. females.

In the two arrhenotokous species tested in this study, the sex ratio was 49% males for *T. brassicae* and 43% for *T. dendrolimi*. A balanced or slightly female-biased sex ratio for the arrhenotokous species suggests that *S. frugiperda* is a highly suitable host for the two parasitoids. *Trichogramma cacoeciae* is a well-known thelytokous species in which thelytoky is genetically determined (Vavre et al. 2004; Russell and Stouthamer 2010).

If we assume that the type of egg masses provided in this study is similar to the overall egg mass distribution under natural conditions regarding layers and hair coverage, our data indicate that *Trichogramma* egg parasitoids can parasitize at maximum between 78% (*T. dendrolimi*) and 57% (*T. cacoeciae*). Unfortunately, the exact pattern of distribution of egg masses categories in the field is not well known, which prevents us from fully assessing the importance of the hair and scales cover and the number of layers on *Trichogramma* spp, parasitism in the field. The largest egg masses with grey hair cover are easier to spot when searching in field studies and, thus, it is a common thought that such egg masses dominate in the field. In our laboratory rearing, the first egg masses of females are mostly with 2–3 layers (rarely 4) covered with hair and scales. However, with time, females lay smaller egg masses with few or no hair and scales and, all together, these are more numerous than large egg masses fully covered with hair and scales. This has been thoroughly described by Hou et al. (2022), showing a decrease in scale thickness cover of egg masses and an increase in the proportion of egg masses having no cover with increasing age of females. Similar observations in laboratory rearing were made by Kasige et al. (2022).

The parasitism rates on *S. frugiperda* eggs found in the present study are higher than what has been reported previously from similar studies with *Trichogramma* spp. on *S. frugiperda*. This can be explained by the fact that other studies have used either single females or small groups of females (e.g. 10) (e.g. Hou et al. 2022, Navik et al. 2023). With the known fecundity of *Trichogramma* wasps and considering that *S. frugiperda* egg masses often have around 150 or more eggs and more than one parasitoid egg is usually laid per host egg, it may be expected that, in most cases, not all of the eggs that females could potentially parasitize, actually are parasitized. While this allows to compare different species, it does not allow to assess the maximum parasitism capacity of a species on *S. frugiperda* egg masses. We believe that the present study allows for the three tested *Trichogramma* species to conclude on the maximum number of eggs they are able to parasitize. These values can reach virtually 100% for single layer egg masses without hair and scales.

These experiments were conducted in 2021 and 2022 in an attempt to assess the potential of readily available augmentative biocontrol agent before the arrival of *S. frugiperda*, an approach called pre-emptive, or proactive biological control. This approach has recently been developed mostly for classical biological control to speed up preparedness for releases of biological control agents (Charles et al. 2019; Avila et al. 2023; Hoddle 2024). However, pre-emptive biological control can also be applied to augmentative biological control to fasten the homologation of macrobial and microbial biological control agents. In the case of *S. frugiperda*, other potential parasitoids, predators and pathogens should be tested for their potential application in Europe to prepare for registration of products against *S. frugiperda*, first targeting label extension of products existing for the control of other lepidopteran pests in Europe (Babendreier et al. 2022).

Conclusions

Trichogramma spp. are commonly used as biological control agents in Europe, including on maize pests. Thus, it would be important to study to what extend current releases (mostly *T. brassicae*) against European corn borer in Europe would be also efficient to control *S. frugiperda*. Our laboratory assays testing three European *Trichogramma* spp. on *S. frugiperda*, confirmed that the main issue is the fact that *T. brassicae*, *T. cacoeciae* and *T. dendrolimi* are not able to parasitize 100% of the eggs when egg masses are composed of two or three layers and when they are covered by a thick layer of hair and scales. Nevertheless, between 57 and 78% of eggs could be parasitized by *T. cacoeciae* and *T. dendrolimi*, respectively, which is more than previously thought. We further showed that there are differences between species in their capacity to parasitize hidden eggs. The comparison of European species should include more *Trichogramma* species and other parasitoids and predators so that field tests can be conducted in newly invaded areas with the most efficient species. Despite the handicap for *Trichogramma* arising from the egg laying habits of the pest, these egg parasitoids may play a role in controlling *S. frugiperda* in Europe and elsewhere, because of the exhaustive experience we have with them, their low costs, and the ease of use.

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Author contributions
Conceptualization: MK, DB, AH; Investigations and acquisition of data: MK, YZ, JF, JK; Provision of insects: AH; Analyses and interpretations: DB, MK; writing—original draft preparation: MK; writing—review and editing: DB, AH, YZ, JF, JK; funding acquisition: MK; supervision: MK and DB. All authors have read and agreed to the published version of the manuscript.

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Availability of data and materials
The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate
Not applicable.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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