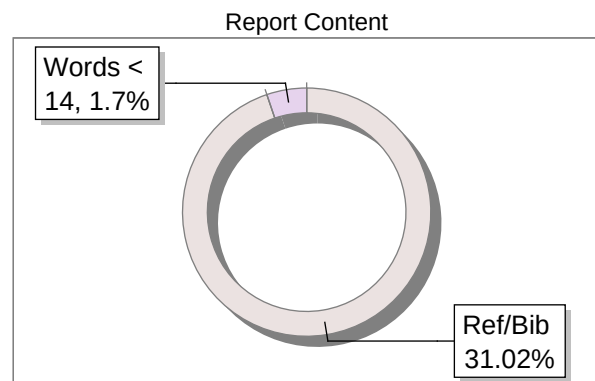
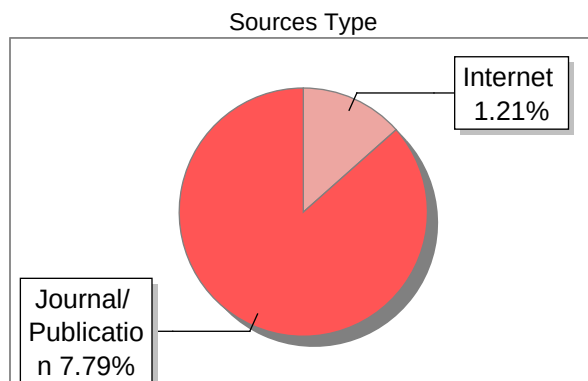


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Evaluation the parasitism efficiency of *Trichogramma evanescens* (Westwood) on *Spodoptera frugiperda* (J. E. Smith) eggs under the laboratory conditions.

Abstract

This study were carried-out to determine the potential parasitism of *Trichogramma evanescens* (Hymenoptera:Trichogrammatidae) on *Spodoptera frugiperda* (Lepidoptera: Noctuidae) egg masses under laboratory conditions. Egg masses of *S. frugiperda* and *Sitotroga cerealella* were exposed to *T. evanescens* females to detect its ability to parasitize Fall armyworm (FAW) egg masses and to evaluate the fitness components of emerged parasitoids after the parasitism. Parasitism percentage, percentage of adult's emergence, fecundity of emerged females, longevity and general productivity, females ratio in progeny were investigated for parasitoids females produced from parasitized FAW and *Sitotroga* eggs as control. Obtained results revealed that, females of *T. evanescens* accepted and succeeded in parasitizing all the offered FAW egg masses and the eggs of *Sitotroga* with the parasitism percentage of (96.69% and 93.25%) respectively, the parasitism resulted in the production of the highest percentage of emerged individuals (97.27% and 94.87%) from parasitized eggs of FAW and *Sitotroga*, respectively, with high ratio of produced females in progeny (71.94% and 69.83%) from parasitized FAW and *Sitotroga* eggs, respectively, those females had a fecundity of (54.88 and 53.71 eggs) from parasitized FAW and *Sitotroga* eggs, respectively, and lived the longest life days of (5.93 and 5.59) from parasitized FAW and *Sitotroga* eggs, respectively.

Keywords: *Trichogramma evanescens*, *Spodoptera frugiperda*, *Sitotroga cerealella*, Parasitism Potential, females ratio, General Productivity.

Introduction

Insect pests are the major constraint to food security, destroying the majority of the global crop production. Pest out breaks threatens the income of farmers, causes supply disrupt, which raises the market prices with the reduction of food accessibility. The out breaks occurred because of the insect pests and that could refer to many factors of which; nature and human activities (Prasanna *et al.* 2022 and Early *et al.* 2016). One of those pests is the fall armyworm *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae), it is a trans-boundary pest with high potential for fast dispersal and insecticides resistance, leading to threatens the most economic agricultural crops (Bueno, *et al.* 2023) causing unprecedented impacts and food security. That invasive pest originates from America, and then it invaded the world causing severe damage to maize crop (CABI 2021). For that, the integrated pest management which involves sustainable pest control strategies must be applied (Orr, 2003; Parsa *et al.* 2014). The fall armyworm outbreak causing severe damage to over 350 plant species mainly maize crop

resulting in great losses of the yield which promoting to increase use of chemical insecticides with its adverse impacts on human and the environment (Abro *et al.* 2021). Biological control is the safe and sustainable alternative way in the long term for the management of *S. frugiperda*, and *Trichogramma* parasitoid is the most important factor of the biological control strategy (Parra and Coelho 2019 and 2022), it has been used in augmentative biological control of the fall armyworm in many countries. Previously, Naviket *et al.* (2023) reviewed several works on different *Trichogramma* species except *T. evanescens* parasitizing Fall armyworm (FAW) egg masses as those of (Beserra *et al.* 2002 and Beserra and Parra 2005; Martinazzo *et al.* 2007; Camera *et al.* 2010; Diaz *et al.* 2012; Figueiredo *et al.* 2015; Junce *et al.* 2020; Jaraleno-Tenient *et al.* 2021; Jin *et al.* 2021; Martel *et al.* 2021; Sun *et al.* 2021; Zang *et al.* 2021; Kumar *et al.* 2022; Yang *et al.* 2022; Bueno *et al.* 2023; Dupatine *et al.* 2023; Li *et al.* 2023; Mohamed *et al.* 2023; Tang *et al.* 2023; and Sampio *et al.* 2024) who tested different species of *Trichogramma* parasitoids except *Trichogramma evanescens* for parasitizing FAW eggs and assessed the efficiency of resulting parasitoids, they reported that all the tested species of Trichgrammatidae could be incorporated in the biological control programs as a promising tool with other bio insecticides for suppressing FAW populations. In that sense, the present author thought that *T. evanescens* could act well in suppressing that pest, because, it is released locally and repeatedly against many lepidopteron pests invading different crops as cotton, tomato potatoes etc, (Siam, 2017). *Trichogramma evanescens* are gregarious wasps, sized 0.5 ml, its female searches the host eggs by its antennal drumming to detect its suitability and freshness to pierce and deposit its brood, which hatched into larva feeds and consumes the contents of the host egg, then it completes its developmental stages till the emergence of a new adult parasitoid. From that, the authors hypothesises that *T. evanescens* could acting well against FAW egg masses, they investigated an experiment to examined if the parasitism of FAW egg masses could occur or not, and the impacts of that parasitism on the emerged *Trichogramma* parasitoids fitness components. So, this work highlighted the potential parasitism of *T. evanescens* on FAW egg masses with no regard to the layers or thickness of the egg masses, and evaluated the impacts of that parasitism on the fitness components of resulting parasitoids (fecundity, females' ratio, longevity and the general productivity of resulting females aiming at incorporating that local parasitoid in the control programs of corn pests mainly the serious fall army worm *Spodoptera frugiperda*.

Materials and Methods

All the experiments of this work were accomplished at Fayoum laboratory of *Trichogramma* parasitoid mass rearing, Plant Protection Research Institute, Agricultural Research Center, Egypt. *Trichogramma. evanescens* was reared on *Sitotroga cerealella* Oliv eggs (< 24 hrs old) glued to a self-adhesive strip (2.5x8 cm). The strips carrying *S. cerealella* eggs were exposed to *T. evanescens* adults in glass jar (1 liter) covered with muslin cloth, held in position by rubber band. Colonies of *T. evanescens* were reared under the laboratory conditions of temperature (25±2°C) and 70±5% relative humidity.

Rearing of *Spodoptera frugiperda*

The colony of *S. frugiperda* was established before the experiment to assure enough supply of fresh Fall armyworm (FAW) egg masses free from any contamination and to avoid any natural parasitism, that rearing supply of FAW eggs masses were the base of the laboratory trials. The laboratory rearing of *S. frugiperda* was accomplished as following: Samples of infected corn plants from fields at Tamyah District, Fayoum Governorate were transported to laboratory fixation and separation of collected FAW larvae and pupae in separate jars, each group with the same age was put in separate jars. Feeding those larvae were accomplished daily with clean maize leaves planted in the laboratory till pupae stage which were placed in special cages waiting for the moths emergence and laying eggs on hard and folded papers put in the bottom of the cages with a piece of cotton dipped in sugar solution for the moths feeding and covered with a piece of cotton clothes. Daily collection for the laid egg masses were accomplished and put in clean jars till larvae hatching. The Newly hatched larvae fed daily till pupae development which in turn transformed to separate cages for moths emergence and laying eggs, the base of this experiment, as it was collected daily as fresh as possible to be treated by releasing *T. evanescens* on them. *S. frugiperda* was established under controlled room conditions ($25 \pm 2^{\circ}\text{C}$ and $55 \pm 5\%$ relative humidity).

Rearing *Sitotroga cerealella* (Oliv.)

The rearing process of *S. cerealella* rearing was done as a modification of those stated by Hassan (1995) when soft wheat was used as the rearing medium.

Mass production of *Trichogramma evanescens*(Westwood)

Trichogramma. evanescens was reared on main host ,*Sitotroga cerealella* Oliv eggs (< 24 hrs old) which glued to a self-adhesive strip (2.5x8 cm). The strips carrying *S. cerealella* eggs were exposed to *T. evanescens* adults in glass jar (2 liter), it was provided with cane sugar honey for nutrition and covered with muslin cloth, held in position by rubber band. Eggs sheet were renewed daily. Colonies of *T. evanescens* were reared under the laboratory conditions of temperature ($25 \pm 2^{\circ}\text{C}$) and $70 \pm 5\%$ relative humidity. Eggs cards were replaced daily to prevent the phenomenon of super-parasitism, and then the egg cards were maintained in clean jars.

Experimental Techniques

Parasitism experiments

The experiment was conducted to determine the parasitism potential of *T. evanescens* on *S. frugiperda* egg masses under laboratory conditions. Fresh *S. frugiperda* egg masses were collected and counted before releasing *Trichogramma evanescens*. This experiment comprises huge numbers of *S. frugiperda* egg masses which in turn was divided to seven groups each group comprises seven replicates; each replicate comprises 30 egg masses of *S. frugiperda* were put individually in separate jars. A similar group of fresh *Sitotroga cerealella* eggs < 24hrs were stickled on self-adhesive papers, then it was divided into pieces each comprising about 70

Sitotroga eggs, then those cards were put individually in glass vials (4X10cm). Newly hatched pairs of *T. evanescens* reared previously on *Sitotroga cerealella* eggs were released on the egg masses of FAW and on *Sitotroga* eggs per each replicate in the jars, each was provided with a droplet of sugar cane honey (Siam, *et al.* 2014) as a nutritive source for adult parasitoids feeding. Both the types of the tested eggs were removed daily to avoid super-parasitism and were put in separate jars with daily inspection to count blackened eggs (parasitized FAW and *Sitotroga* eggs) which recorded as fecundity and to remove any hatched *S. frugiperda* or *Sitotroga* larvae considering them as the un-parasitized eggs. After emergence, the percentage of emerged adults and female's ratio was determined. Females were checked daily to count longevity. Calculation for the general productivity was done according to Tshernyshev and Afonina (1995) as following:

$$\text{General Productivity} = \text{Rate of emergence} \times \text{Rate of females in progeny} \times \text{fecundity}$$

Data collection and Statistical analysis

Collected data from parental *T. evanescens* was analyzed with T-test at $P \leq 0.05$. The statistical analysis and figures were aided by the software Microsoft Excel

Results

Parasitism potential of FAW egg masses by *T. evanescens*

Regardless of the orientation or the number or thickness of *S. frugiperda* egg masses (Fig.1) *Trichogramma evanescens* females significantly parasitized *S. frugiperda* and *S. cerealella* eggs ($F = 6.907$). Regardless to the layers number or the thickness of the scales, *T. evanescens* females parasitized FAW egg masses with the mean of (96.69%) eggs; also, it parasitized (93.25%) *S. cerealella* eggs ($P \leq 0.01$) Also, the proportion of un-parasitized FAW egg masses was indicated by (3.31%) eggs, meanwhile the un-parasitized *S. cerealella* eggs averaged (6.75%) eggs, (Fig 2).

5

Fig. 1 T.evanscene parasitism on S.frugiperda egg masses regardless of the different orientation or layers thickness. A.S. frugiperda egg masses before parasitism. B S. frugiperda egg masses after parasitism.C.D.E.F.G.H.I.J. K., and L. Parasitized S. frugiperda egg masses of different orientations or layers thickness.

Fig. 2 Percentage of parasitism and un-parasitism of T.evanscens on S.frugiperda and S.cerealella eggs.

Influence of FAW eggs parasitism on the adult emergence

The efficiency of emerged individuals of T. evanescens from parasitized FAW and S. cerealella eggs was assessed based on the proportions of adult emergence. Percentage of the

0

50

100

% Parasitism % Unparasitism

FAW Eggs

Sitotroga Eggs

offspring (94.87%) emerged when parent female parasitized *Sitotroga* eggs. Regardless of the shape or the orientation of *S. frugiperda* egg masses and layers ($F= 0.014$), the maximum percentage was recorded with *Trichogramma* females parasitized *S. frugiperda* egg masses (97.27%), (Fig 3).

Influence of FAW egg parasitism on *T. evanescens* females fecundity

Fecundity of *T. evanescens* females had no significant difference between the two host eggs ($P \leq 0.01$). Females produced from parasitized FAW egg masses had fecundity of (54.88) eggs, while those emerged from parasitized *Sitotroga* eggs had fecundity with the average of (53.71) eggs. ($F= 1.125$), (Fig 3).

Influence of FAW egg parasitism on the *T. evanescens* longevity

Longevity of the parasite affected by the parasitism on armyworm

The mean life days Longevity of *T. evanescens* females produced from parasitized FAW egg masses and *S. cerealella* eggs were similar as it recorded (5.93 and 5.59) days, respectively ($F= 0.14$). There was no significant difference between the life span of the females parasitoid produced from the two hosts, (Fig 3).

Influence of FAW eggs parasitism on the produced female's ratio in progeny

The emerged progeny from both parasitized FAW and *Sitotroga* eggs were strongly female-biased ($F= 256.070$). The rate of *T. evanescens* females in progeny (71.94%) was produced from parent females 'parasitized FAW egg masses. Parent females produced the rate of females in progeny (69.83%) when they released on *Sitotroga* eggs, (Fig 3).

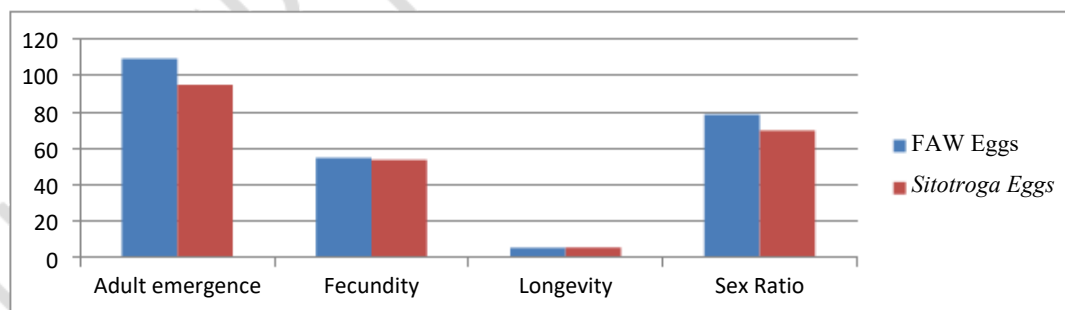


Fig. 3 Fitness components of resulted *T. evanescens* parasitoids from parasitized eggs of *S. frugiperda* and *S. cerealella*.

Influence of FAW egg masses parasitism on the general productivity (GP) of *T. evanescens*

The general productivity of *T. evanescens* was represented in Fig.4 *T. evanescens* females productivity was (37.095 females/ female) when they produced from parents parasitized FAW egg masses, while those resulted from parent females parasitized *S. cerealella* eggs recorded productivity of (35.584 females/ female), (Fig 4).

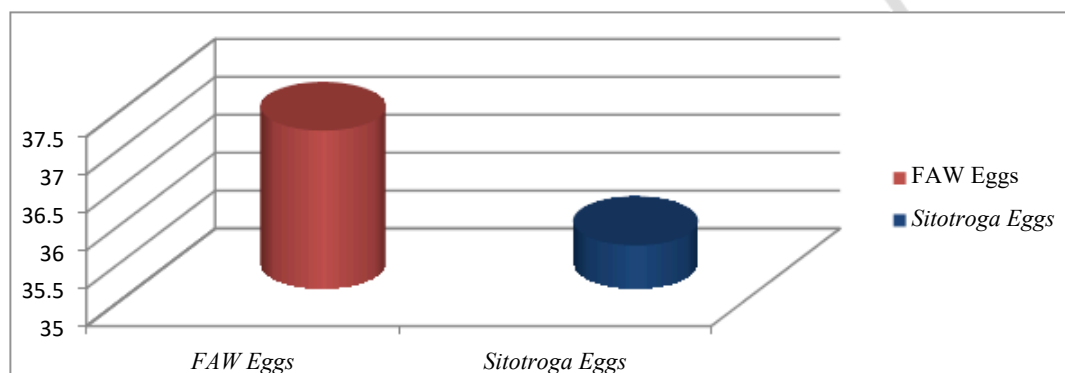


Fig. 4 General productivity of females produced from parents parasitized *S. frugiperda* and *S.cerealella* eggs

Discussion

In this study, *T. evanescens* which reared in the laboratory on the host *Sitotroga cerealella* eggs to release it on many lepidopteron eggs invading certain strategic and economic crops as cotton, tomato, etc., proved success in parasitizing the tested *Spodoptera frugiperda* egg masses as was revealed by the obtained results of this work, so, it could be considered as a promising hope for the control of the invasive *S. frugiperda*. Obtained results of this work revealed that *T. evanescens* which was tested against Fall armyworm (FAW) egg masses parasitized all the offered egg masses and produced high percentages of parasitoids adults with high rates of females in progeny of high fecundity and high general productivities calculated for the produced *T. evanescens* females. It must be mentioned that the present authors conducted many trials previously on the parasitism of FAW egg masses by releasing *T. evanescens* females whether those egg masses brought from fields or even reared under laboratory conditions, and in both cases the results were in favor of the parasitoid as it recorded high percentages of parasitism, in addition to the production of high quality parasitoids with high fitness components. Notably, parasitized FAW egg masses were utilized as an alternative host for *T. evanescens* to renew rearing process thus gaining strong parasitoids acting well when releasing them in the fields. When the produced parasitoids were re-reared and released on many economic and strategic crops as cotton in Fayoum Governorate, high potential parasitism of *T. evanescens* on many lepidopteron pests was recoded. The obtained results of this work revealed that, resulting

adults had high fitness components represented in strong female-biased which indicated by the average of 71.94%, and those females had high fecundity with the mean of 53.02 parasitized *Sitotroga* eggs and those females had the life span of 5.93 days, also, *T. evanescens* produced from parasitized FAW egg masses counted high general productivities averaged 37.09 females/female. Many authors declared that *Trichogrammatidae* had provided a prospect for augmentative biological control of *S. frugiperda* for the sustainability production of crops. Also, they reported that, natural parasitism of FAW egg masses by *Trichogramma* species averaged between different ecosystems and between different species of *Trichogramma* parasitoids, those species were; *T. pretiosum*; *T. atopovirilia* (Beserra *et al.* 2002; Beserra and Parra 2005; Bueno *et al.* 2010; Dequechet *et al.* 2013; Sisay *et al.* 2019; Jaraleno-Teniente *et al.* 2020; Junce *et al.* 2020; Jin *et al.* 2021; Li *et al.* 2023; Navik *et al.* 2019 and 2023). Other authors as Yang *et al.* (2022) evaluated the performance of three *Trichogramma* species as a bio control agent on *S. frugiperda* eggs, Mohamed *et al.* (2023) tried *Trichogramma toideabactrae* for the parasitism of *S. frugiperda* egg masses with different layers and scale thickness, they reported that, *T. bactrae* could be an efficient bio-control agent for controlling *S. frugiperda*. Sampio *et al.* (2024) evaluated *T. forsteri* as a newly discovered egg parasitoid for parasitism on *S. frugiperda* and *S. eridania* eggs and they reported that this species of *Trichogramma tidae* showed high potential against both pests. Navik *et al.* (2024) was trying to evaluate *T. chilonis*, *T. pretiosum* and *T. mwanzai* for the parasitism of *S. frugiperda* egg masses with different morphological structures, and then they found that morphological structure significantly affected the parasitism potential of *Trichogramma* species and they concluded that advised selecting *T. chilonis* for the parasitism of *S. frugiperda* egg masses. These results encouraged us to test *T. evanescens* that has been rare tested on FAW egg masses and It is worth mentioning that all the above works has been done on different species of the parasitoids except *T. evanescens*. On the other hand our work was compatible with El-Gepaly *et al.* (2024) who released *T. evanescens* on *S. frugiperda* egg masses and they reported that *T. evanescens* showed a good results promising for integrated pest management. In this response, we are looking forward that, this result obtained from our work will highlights on the potential parasitism of *T. evanescens* on FAW egg masses as one of the biological agents in integrated pest management system.

Conclusion

Results of this work revealed that the egg parasitoid *T. evanescens* had significantly parasitized all the offered FAW egg masses under laboratory conditions and the emerged parasitoids recorded high fitness components. So, it could be considered as a promising hope for suppressing *Spodoptera frugiperda* populations invading corn plantations after testing releasing it in the open fields in the next work. It could be used as an efficient bio-agent, thus preventing or reducing the hazard impacts of chemical insecticides.

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