



# Host-preference and parasitic capacity of new candidates of *Trichogramma* species (Hym.:Trichogrammatidae) against some stored product moths

Esmat Hegazi <sup>a,\*</sup>, Cornel Adler <sup>b</sup>, Wedad Khafagi <sup>c</sup>, Essam Agamy <sup>d</sup>

<sup>a</sup> Faculty of Agriculture, Alexandria University, Egypt

<sup>b</sup> Julius Kühn-Institute, Federal Research Centre for Cultivated Plants, Institute for Ecological Chemistry, Plant Analysis and Stored Product Protection, Königin-Luise-Str.19, 14195, Berlin, Germany

<sup>c</sup> Plant Protection Research Institute, Alexandria, Egypt

<sup>d</sup> Faculty of Agriculture, Cairo University, Egypt

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## ABSTRACT

The host-preference and parasitic capacity of some local *Trichogramma* spp. (*T. bourarachae*, *T. cordubensis*, *T. euproctidis* and *T. cacociae*) towards four species of stored product moth eggs were investigated in order to select new candidate species for use in mass rearing and biological control against moths in storages. The efficacy of the *Trichogramma* spp. was compared with *T. evanescens*, the common wasp used commercially for biological control. Experiments were carried out by offering a single parasitoid female to eggs of the Indianmeal moth *Plodia interpunctella* (Hubner), the Mediterranean flour moth *Ephestia kuehniella* Zeller, the warehouse moth *E. elutella* (Hubner), and the almond moth, *Cadra cautella* (Walker) in choice and no-choice assays. Two different choice experiments were used to certify the same conclusion in both methods. The bioassay for host-preference of *Trichogramma* was carried out by offering a single early female wasp the choice between equal numbers of host eggs on square cards “Petri dish tests” and/or strip cards “strip card tests”. In Petri dish tests, *E.kuehniella* was a highly acceptable host species for *T. bourarachae*, *T. euproctidis*, and *T. cacociae* wasps while *E.elutella* and *C.cautella* eggs were more acceptable for *T. evanescens* and *T.cordubensis*, respectively. In strip card tests, *E.kuehniella* eggs were highly acceptable for *T.bourarachae*, *T.cacociae* and *T.evanescens*. Eggs of *E.elutella* and *C.cautella* were more acceptable for *T.euproctidis* and *T. cordubensis*, respectively. Significant differences were found among the parasitic capacity of the tested *Trichogramma* spp.: *T. borarachae* showed a good parasitic potential against *S.cerealella* and *E. kuehniella*; *T.evanescens* and *T. cacociae* against *S.cerealella*; *T. cordubensis* against *S.cerealella* and *P. interpunctella* and *T. euproctidis* against *P. interpunctella*. All tested wasps had a propensity to superparasitize the host eggs. *T. cordubensis*, *T. euproctidis* and *T. borarachae* showed promise for further investigation into selecting new biological control agents against some stored product lepidopterous pests.

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## 1. Introduction

Most stored product insects are either beetles or moths. They develop large populations, consume food commodities, and contaminate the goods with feces, hairs, shed skins, webbing, cadavers, and toxins (Grethe et al., 2011; Niedermayer and Steidle, 2013). During storage, large quantities of Cereal crops, accounting

for 5–10% in temperate and 20–30% or more in the tropical regions, are lost annually due to different insect pests (Nenaah, 2014). Moth pests are important hazards to the storage of a wide variety of products. The most important moth species are the Indian-mealmoth, *Plodia interpunctella* (Hübner), the Mediterranean flour moth, *Ephestia kuehniella* Zeller, the warehouse moth, *E. elutella* (Hübner), and the almond moth, *Cadra cautella* (Walker) (Flinn and Schöller, 2012).

The control of stored product insects relies heavily on the use of synthetic insecticides such as organophosphates, pyrethroids and

\* Corresponding author.

E-mail address: [eshgazi@hotmail.com](mailto:eshgazi@hotmail.com) (E. Hegazi).

fumigants (mainly phosphine and methyl bromide) (Islam et al., 2010). However, new insecticide regulations resulting from the Food Quality and Protection Act as well as consumer concern over pesticide residues have limited the availability of insecticides for use in stored products (Arthur and Rogers, 2003; Opit et al., 2012). Intensive use of insecticides leads to resistant behavior and environmental pollution with negative side effects on human health (Skevas et al., 2014). For such reasons many researchers have been searching for environmentally friendly and low toxicity new natural products to provide alternatives (Gantner and Najda, 2013). Eco-friendly strategies to prevent such insect attacks on final packaged products are therefore highly required, e.g., essential oils (extracted from aromatic plants) could represent an alternative to chemical treatments due to their repellent properties (Piesik and Wenda-Piesik, 2015; Wenda-Piesik et al., 2016; Wenda-Piesik et al., 2017). Wenda-Piesik et al. (2018), reported that the blend of volatiles and their concentration can induce the insect behavior.

Biological control using egg parasitoids presents an another attractive alternative to insecticides for reducing infestation and damage from stored product insects in retail and warehouse environments. *Trichogramma* spp. have been suggested as potential natural enemies for a variety of stored product moths in bulk peanut storage (Flinn and Schöller, 2012; Schöller et al., 2006; Grieshop et al., 2006a & b; Stengård Hansen, 2005; Brower and Press, 1990), bulk wheat storage (Grieshop et al., 2007; Schöller et al., 1994, 1996), and bakeries (Prozell and Schöller, 1998; Steidle et al., 2001). Parasitoid searching capacity, host preference and dispersion are some traits that have been also evaluated as criteria for strain selection (Hassan, 1994), but these methods require more elaborate procedures.

Four naturally occurring *Trichogramma* species were collected for the first time in Egypt from two representative olive growing areas in arid area and semi-arid area. *T. bourarachae* was collected exclusively from Burg El-Arab farm, situated 60 km west Alexandria, near the coast (semi-arid area). Three species namely *T. cordubensis*, *T. euproctidis* and *T. cacociae* were isolated from arid areas (170 km south of Alexandria) (Herz et al., 2007). All these wasps were also bred from naturally parasitized host eggs during favorable and even at unfavorable temperature conditions of June–August. The presence of warm weather wasp strains may suggest the existence of well-adapted wasp species or strains which may be appropriate candidates for the control of stored product pests. The strains had also been collected in late winter and summer, thus demonstrating activity also during less favorable weather conditions, raising again the possibility of using these egg parasitoids as an innovative biological control agent in stored products. Studies of Flinn and Hagstrum (2001), have shown that the appropriate application of natural enemies does not lead to contamination of food with insect wasps, either because raw materials for production of food are cleaned prior to processing or because the pests are controlled outside packaged food. The objective of this investigation was to study the parasitoid efficacy of *T. bourarachae*, *T. cordubensis*, *T. euproctidis*, *T. cacociae* and *T. evanescens* (Hymenoptera: Trichogrammatidae) on some moth pests.

## 2. Materials and methods

*Trichogramma* wasps used in the study came from rearing colonies originating from field-collected material in Egypt. Species identity of *Trichogramma* specimen was confirmed using morphological and molecular biological methods (Herz et al., 2007) according to Pinto (1999) and Silva et al. (1999). *Trichogramma* populations (*T. bourarachae*, *T. cordubensis*, *T. euproctidis*, *T. cacociae*, *T. evanescens*) were maintained in the Laboratory of Entomology,

Alexandria University with fresh eggs of the alternative host *Sitotroga cerealella* Olivier, supplied by the International Company of Bioagriculture, Giza, Egypt. *S. cerealella* was used also as an experimental host. The quality of the mass rearing *Trichogramma* strains was maintained by regularly introducing individuals taken from the field into the rearing process (Bigler, 1994).

The experiments were conducted at the JKI Institute for Ecological Chemistry, Plant Analysis and Stored Product Protection für, Berlin, Germany. Host-species preference between the eggs of four stored product insects (*P. interpunctella*; *Ephestia kuehniella*; *E. elutella*; and *C. cautella*) was developed from studies of Hassan (1989) and Hassan and Guo (1991). Two different host –choice experiments with different arena (Fig. 1) were performed, to certify the same conclusion with two methods. Both experiments were conducted with individual early *Trichogramma* females, ( $\leq 2$  h old) ( $n = 10/\text{trail} \times 6$  replicates/species). In the 1st experiment (square card method), eggs of each moth species were glued on the ends of the four corners of the note card (35 × 35 mm) with non-toxic carpenter's glue (Elmer's® glue, Elmer's Products Inc., Columbus, Ohio). The note cards were placed into plastic petri dishes (55 × 15 mm) lined with filter paper (Ahlstrom®, Mt. Holly Springs, Pennsylvania). Petri dishes were sealed with parafilm® (Pechiney- Plastic Packaging, Menasha, Wisconsin) and arranged on plastic trays lined with moist tissue paper to increase relative humidity to 60–80% RH. In the 2nd experiment (strip card methods), the single early *Trichogramma* females were released in glass tubes (70 × 01 mm) containing each an egg cardstrip (50 × 5 mm) with four egg patches of the four host species. Each vial was tightly covered by parafilm and then perforated with a fine-tipped needle (000 micro pin). In both experiments, host eggs were randomly glued on fixed locations on the egg cards. The preference experiments were conducted by offering equal densities ( $70 \pm 5$  eggs/species) of four host species and equally apart to the single *Trichogramma* female. Larvae that hatched from the unparasitized eggs were daily removed.

To study the parasitic capacity of the test wasps on eggs of four moth species, the wasp species were evaluated by exposing each of them for the 1st time to eggs of the stored product test insects. From each species, (10 wasps × 6 repl.) freshly emerged females, were individualized in glass tubes (70 × 10 mm) containing a cardboard (20 × 5 mm) with host eggs and a drop of honey for feed. The host eggs were prepared by applying a fixed area from index card with water solution of non-toxic glue, and then spreading eggs ( $250 \pm 10$  eggs) on the surface. This number of host eggs available per female wasp was high enough to avoid superparasitism (Wajnberg et al., 1989). Each egg card was labelled with female number and parasitism date. For each egg card, the number of parasitized hosts was determined by counting the host eggs that turned black, using a dissecting

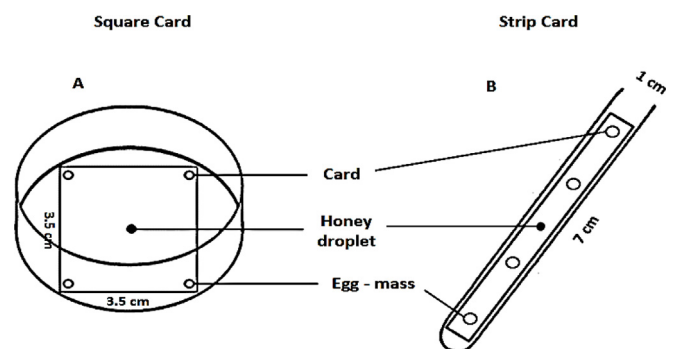


Fig. 1. Square (A) and strip (B) host egg-mass cards to test host preference by *Trichogramma* wasps.

microscope at a magnification of 25X. The parasitized eggs were detected by the dark colour of the chorion (Voegelé et al., 1974). Egg chorions of insects infested by *Trichogramma* turn black when parasitoids are in the pre-pupal stage. This is caused by black deposits that form a cocoon-shaped cuticle around the body of the larva and inner surface of the chorion of the host egg (Saakian- Baranova, 1991). Females that died in the honey were eliminated from the statistical analysis and replaced by new ones.

In all experiments, all females originated from rearings held at 25 °C on *S. cerealella* eggs. Individual females ( $\leq 1$  h old) were captured in the glass tub through the open end of a glass vial by allowing them to walk towards the light. The captured wasp was then examined using a binocular. Honey droplets were put in the centre of each note card, egg card strip or end of the cardboard to provide a supplemental source of food to released wasps as a lack of food was reported to adversely affect oogenesis (Van Lenteren et al., 1987). Host eggs offered to female wasps in the experiments were fresh, 0–24 h old, which is the generally preferred age of hosts in *Trichogramma* (Pak et al., 1986). Parasitism was allowed during the first 48 hours of female wasp life and parasitoid females were removed under stereoscopic microscope after this period. Tubes containing the card with parasitized eggs were maintained in climatized chambers. Host eggs with hole made by *Trichogramma* adults were counted in the morning and in the afternoon, at approximately the same time (hour), to evaluate emergence rate of these parasitoids. The emerged parasitoids per host species were evaluated by counting their numbers.

After completion of emergence remaining unemerged eggs and emerged one with exit holes were dissected to determine the number of undeveloped parasitoids. The growth chambers maintained at  $25 \pm 1$  °C, 16:8 h L:D (480–590 lux) and 60–80% RH were used for incubation of experimental units in all experiments.

### 2.1. Statistical analysis

Data on host preference and/or parasitization activity were subjected to one-way analysis of variance (ANOVA) and significant differences were identified using Duncan's multiple range test. Percentages were transformed to arcsine values before ANOVA. All the analyses were carried out at the 5% significance level.

### 3. Results

Results of the host-preference of four local *Trichogramma* spp. found in arid and semi-zones in Egypt, as well as the commercially available *T. evanescens* Westwood, originating from sugarcane fields in Egypt, towards eggs of four species of stored product moth pests by square and strip card methods are shown in Figs. 2–6. Individual wasp females of each of *T. bourarachae*, *T. cordubensis*, *T. euproctidis*, *T. cacoeciae*, or *T. evanescens*, readily parasitized and emerged from egg-mass of *C. cautella*, *Ephestia elutella*, *E. kuehniella*, or *P. interpunctella*. In some cases, the wasp female oviposited in only egg mass of one host species on the egg card, while in other cases oviposited in egg-masses of two host species and in few cases moved and oviposited in eggs of 3 host species.

Assessment of host preference by means of statistical comparison using individual *T. bourarachae* females for four host species is included in Fig. 2 based on the criteria of host acceptance 'parasitization activity', distinct differences were found among the four host species (for square card method:  $F = 108.3$ ,  $df = 3,20$ ,  $L.S.D. = 5.9$ ,  $P < 0.05$ ; for strip card method:  $F = 72.9$ ,  $df = 3,20$ ,  $L.S.D. = 5.91$ ,  $P < 0.05$ ). In square card method, parasitization rates were 18%, 16%, 60% and 22% for *T. bourarachae* on *C. cautella*, *Ephestia elutella*, *E. kuehniella*, and *P. interpunctella*, respectively. While parasitization rate in the strip card method on the same hosts were 16%, 12%, 50% and 28%, respectively. In both methods, *T. bourarachae* showed high oviposition frequency on *E. kuehniella* and lowest on *E. elutella*.

Results of square and strip card methods on parasitization of the four stored product moths by *T. cordubensis* are shown in Fig. 3. The parasitoid wasp showed a significant ( $P < 0.05$ ) preference for *C. cautella* eggs in both square card method ( $F = 25.2$ ,  $df = 3,20$ ,  $L.S.D. = 5.1$ ) and strip card method ( $F = 23.4$ ,  $df = 3,20$ ,  $L.S.D. = 7.8$ ). In the square card method, parasitization rates were 42%, 28%, 22% and 26% on *C. cautella*, *Ephestia elutella*, *E. kuehniella*, and *P. interpunctella* eggs, respectively while parasitization rates in the strip card method tests were 60%, 52%, 32% and 38% on eggs of the same host species, respectively.

Subsequent trials focused on parasitization activity (No. of parasitized eggs/female) of *T. euproctidis* individual females on host eggs of *C. cautella*, *E. elutella*, *E. kuehniella*, and *P. interpunctella* and showed different preference trends (Fig. 4). Parasitization activity varied

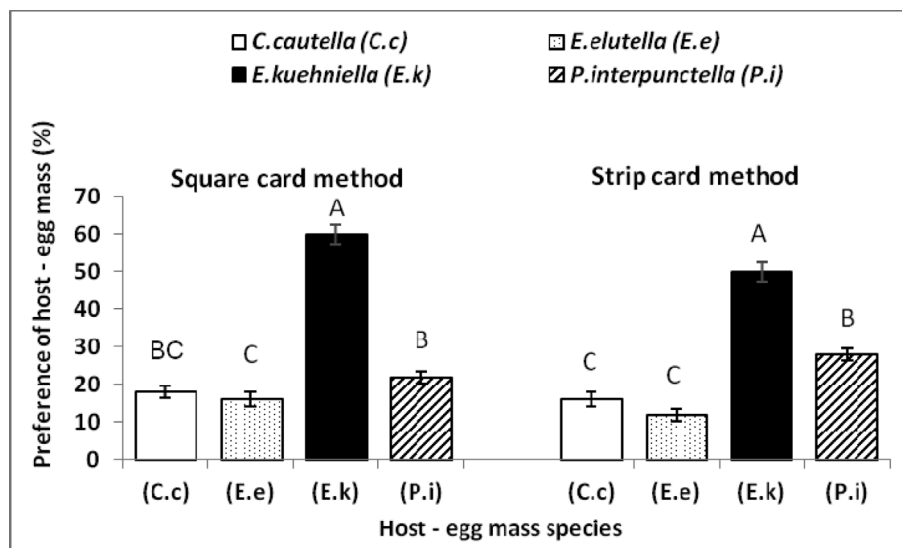


Fig. 2. Host-species preference ( $\% \pm SE$ ) in choice tests by individual females of *T. bourarachae* between egg masses of four lepidoptenous stored product insect species using square and strip card methods. For each set, bars with the same uppercase letter are not significantly different ( $P < 0.05$ ).

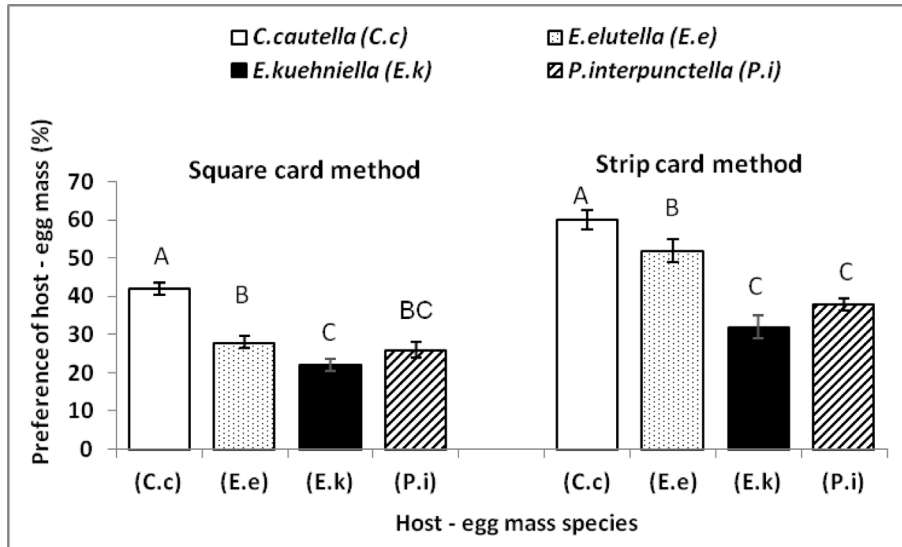


Fig. 3. Host-species preference (%  $\pm$  SE) in choice tests by individual females of *T. cordubensis* between egg masses of four lepidoptenous stored product insect species using square and strip card methods. For each set, bars with the same uppercase letter are not significantly different ( $P < 0.05$ ).

significantly among eggs of these hosts in both square card method ( $F = 49.96$ ,  $df = 3,20$ ,  $L.S.D. = 5.97$ ,  $P < 0.05$ ) or strip card method ( $F = 78.5$ ,  $df = 3,20$ ,  $L.S.D. = 6.38$ ,  $P < 0.05$ ) tests. In square card method the parasitoid wasp showed significant ( $P < 0.05$ ) preference to *E. kuehniella* eggs (44%) versus to 10%, 26% and 20% for *C. cautella*, *E. elutella* and *P. interpunctella* eggs, respectively, while in the strip card method, *T. euproctidis* showed significant preference ( $P < 0.05$ ) to *E. elutella* eggs (60%) followed by 44%, 36% and 14% among eggs of *C. cautella*, *E. kuehniella*, and *P. interpunctella*, respectively.

In the choice test either by square card ( $F = 43.8$ ,  $df = 3,20$ ,  $L.S.D. = 8.0$ ,  $P < 0.05$ ) or strip card ( $F = 61.1$ ,  $df = 3,20$ ,  $L.S.D. = 7.23$ ,  $P < 0.05$ ) methods, most individual female wasps of *T. cacoeciae* preferred the eggs of *E. kuehniella*. The wasps stayed and searched longer for the *E. kuehniella* eggs (Fig. 5). Consequently, more progeny emerged from *E. kuehniella* eggs than from *C. cautella*, *E. elutella*, and *P. interpunctella* eggs. In square card method tests, parasitization rates were 40%, 40%, 70% and 28% on *C. cautella*,

*E. elutella*, *E. kuehniella*, and *P. interpunctella* eggs, respectively, while parasitization rates in strip card method tests were 36%, 28%, 64% and 20% on eggs of the same host species, respectively.

Results of laboratory tests on host preference by the commercially available wasp (*T. evanescens*) against moth's pests of the stored products are shown in Fig. 6. Parasitization rates varied significantly among the eggs of *C. cautella*, *E. elutella*, *E. kuehniella*; and *P. interpunctella*, in both square card ( $F = 27.0$ ,  $df = 3,20$ ,  $L.S.D. = 7.0$ ,  $P < 0.05$ ) or strip card ( $F = 8.6$ ,  $df = 3,20$ ,  $L.S.D. = 7.3$ ,  $P < 0.05$ ) methods. In the former test, the highest percentage of parasitized host eggs presented to *T. evanescens* was in *E. elutella* hosts (56%) versus 40% 32% and 28% in *C. cautella*, *E. kuehniella*; and *P. interpunctella* eggs, respectively. Significant different parasitization behavior was recorded in the second test, where parasitization activity recorded on host eggs of *C. cautella*, *E. elutella*, *E. kuehniella* and *P. interpunctella* was 34%, 30%, 46% and 31%, respectively.

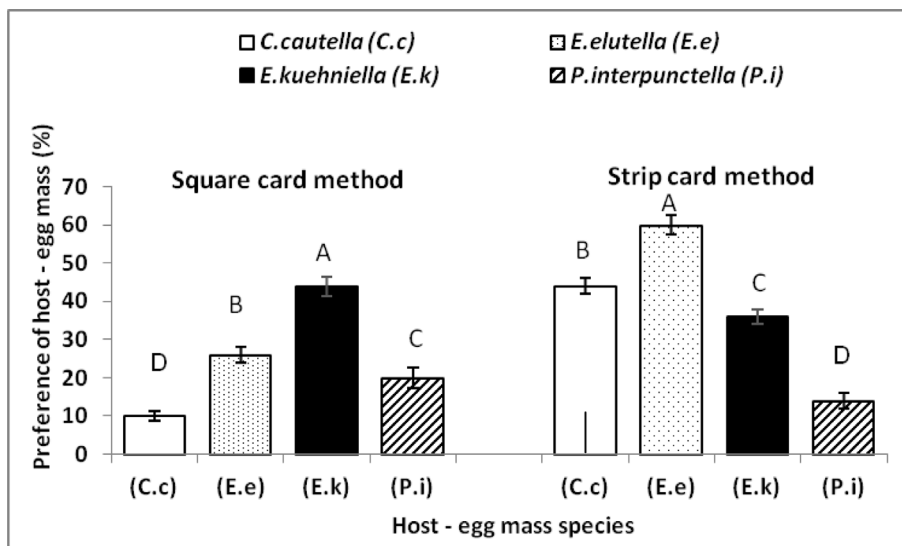
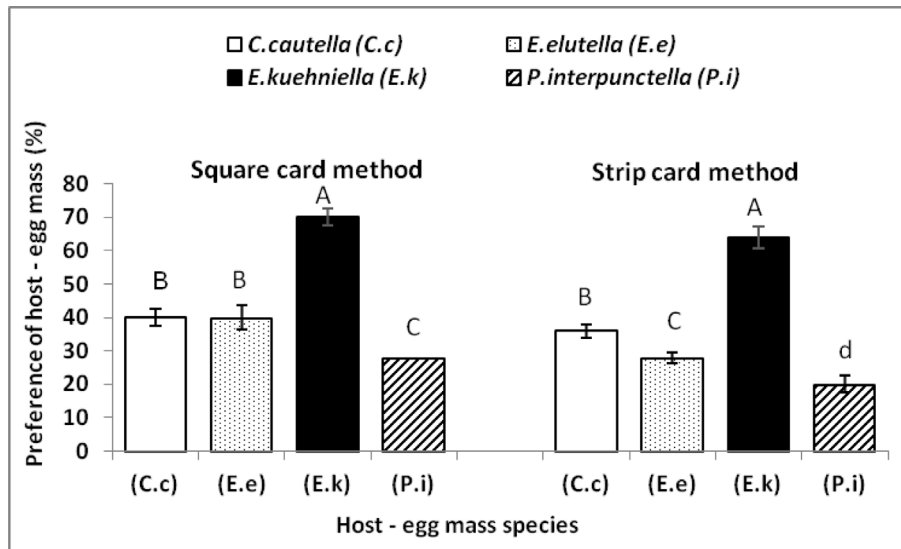


Fig. 4. Host-species preference (%  $\pm$  SE) in choice tests by individual females of *T. euproctidis* between egg masses of four lepidoptenous stored product insect species using square and strip card methods. For each set, bars with the same uppercase letter are not significantly different ( $P < 0.05$ ).



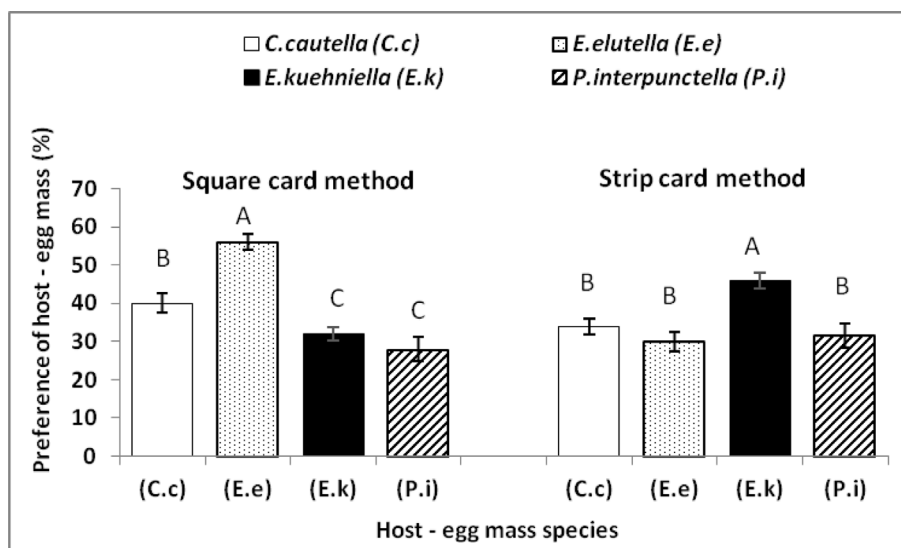


**Fig. 5.** Host-species preference (%  $\pm$  SE) in choice tests by individual females of *T. cacociae* between egg masses of four lepidoptenous stored product insect species using square and strip card methods. For each set, bars with the same uppercase letter are not significantly different ( $P < 0.05$ ).

In order to select effective *Trichogramma* species to control the stored product pests, unlimited number of host eggs was offered to a single wasp of *T. bourarachae*, *T. cordubensis*, *T. euproctidis*, *T. cacociae*, or *T. evanescens* to evaluate their parasitic capacity on eggs of moth species. Table 1 shows comparative analysis of the parasitic capacity/day of individual females of the five *Trichogramma* species provided host eggs for the first 48 h of female's life. For the parasitic capacity of *T. bororachae*, significant differences were found among the test egg moths ( $F = 15.74$ ;  $d.f. = 4,45$ ;  $P < 0.05$ ). The mean number of parasitic capacity was  $43.2 \pm 1.2$  eggs of *S. cerealella* and  $39.3 \pm 0.9$  eggs of *E. kuehniella* versus less significant number of eggs of *P. interpunctella*, *E. elutella* and *C. cautella* moths. The parasitoid wasp showed no significant ( $P < 0.05$ ) preference for *S. cerealella* and *E. kuehniella* eggs. The number of parasitized eggs by *T. cordubensis* significantly varied among eggs of moth species ( $F = 112.2$ ;  $df = 4,45$ ;  $P < 0.05$ ). The wasps parasitized the highest number of *S. cerealella* hosts followed by *P. interpunctella* eggs. Eggs of *E. elutella* and *C. cautella* moths

were less accepted. However, no significant preference was observed between eggs of *E. elutella* and *C. cautella* moths. *T. euproctidis* had the highest parasitic capacity on *P. interpunctella* eggs ( $F = 43.7$ ;  $df = 4,45$ ;  $P < 0.05$ ) followed by *S. cerealella* and *E. kuehniella* eggs, while eggs of *E. elutella* and *C. cautella* moths were also less accepted for this wasp. No significant preference was observed between eggs of *E. elutella* and *C. cautella* moths. *T. cacociae* females parasitized a significantly lower number of *P. interpunctella* host eggs, but had the highest parasitic capacity on *S. cerealella* hosts ( $F = 93.3$ ;  $df = 4,45$ ;  $P < 0.05$ ). *T. evanescens* had high parasitic capacity on both *S. cerealella* and *E. kuehniella* eggs with significantly lower parasitic capacity on *E. elutella*. So the parasitic capacities of *T. euproctidis* and *T. bororachae* strains (Table 1) are relatively high comparing to those of other wasps.

In the no choice experiments, when the number of parasitized eggs was counted, as well as the number of offspring, it was observed that the number of emerged wasps exceeds the number of parasitized eggs (hatched or not hatched). Dissection of some of



**Fig. 6.** Host-species preference (%  $\pm$  SE) in choice tests by individual females of *T. evanescens* between egg masses of four lepidoptenous stored product insect species using square and strip card methods. For each set, bars with the same uppercase letter are not significantly different ( $P < 0.05$ ).

**Table 1**  
Average parasitic capacity/day of individual females (n = 10 × 3) of five *Trichogramma* species provided with 250 ± 10 host eggs.

<i>Trichogramma</i> species	Host - eggs				
	<i>S. cerealella</i>	<i>C. cautella</i>	<i>E. elutella</i>	<i>K. kuehniella</i>	<i>Plodia</i>
<i>T. borarachae</i>	43.2 ± 1.2 a	13.1 ± 1.5 c	30.7 ± 1.41 b	39.3 ± 0.9 a	33 ± 1.8 b
<i>T. evanescence</i>	39.3 ± 0.9 a	30.4 ± 0.8 c	21.5 ± 0.8 e	35.8 ± 0.8 b	27.5 ± 0.6 d
<i>T. cacociae</i>	37.7 ± 0.6 a	23.5 ± 0.9 d	21 ± 0.6 e	28.9 ± 0.6 b	26.9 ± 0.6 c
<i>T. cardubensis</i>	34.7 ± 1.2 a	16.8 ± 0.5 c	17.5 ± 0.62 c	32.2 ± 0.9 b	34.4 ± 0.9 ab
<i>T. euproctidis</i>	39.2 ± 1.6 b	25.3 ± 1.6 c	22.6 ± 0.8 c	35.8 ± 1.1 b	48.2 ± 2.4 a

Mean values followed by the same letters in the same row are not significantly different at the 5% level of F test.

not hatched black eggs revealed that the eggs suffered desiccation at early stage due to superparasitism. This resulted in insufficient food to support the surplus parasitic larvae, resulting in premature death of developing wasps. Also, all eggs showed wasp's exit hole(s) were counted and dissected. These eggs having exit holes and containing more developing live or dead parasitoids in the same egg were considered superparasitized and identified as apparently superparasitized host eggs (Table 2). These host eggs contained 1 to more than 2 parasitoid wasps in the same egg. Superparasitism was observed by all test wasps. Dissection of sub-samples of non-emerged parasitized eggs revealed that all wasps tended to insert 1–3 eggs in *S.cerealella* eggs and 1–5 eggs in the other eggs of moth species. The results suggested (data not shown) that more than one parasitoid egg may be initially deposited per host egg. Larvae hatching from unparasitized eggs may feed on pest species as well as parasitized eggs, decreasing the number of emerging wasps.

#### 4. Discussion

Biological control using egg wasps of *Trichogramma* spp. presents an attractive alternative to chemical control for reducing damage caused by stored product moths, in retail and warehouse environments. It is generally known that most *Trichogramma* species exhibit a strong preference for certain hosts (Flinn and Schöller, 2012; Hassan and Guo, 1991). Host preference and host suitability in *Trichogramma* has now been fully recognized as important agents with regards to biological control. The research workers have shown that these characters can easily be tested in simple laboratory experiments (Lenteren et al., 1982). Two different methods were used to assess the host preference of early females (had a higher fitness than late females) of *Trichogramma* species (Hassan and Guo, 1991). In the first method, the behavior of a single parasitoid female released to parasitize the host eggs was assessed at the end of the experiment (Hassan, 1989; Wuhrer and Hassan, 2009). The other is based on the continuous and direct observation of the egg parasitoid to record its activities (Dijken et al., 1986). In both methods, counting the number of *Trichogramma* developing in the host eggs (parasitism) show the preference of the wasp for ovipositing and indicated the ability of the parasitoid to develop in these eggs (i.e. host suitability).

During the years 2002–2004, *T. bourarachae*, *T.cordubensis*, *T.euproctidis* and *T.cacociae* were captured in Egypt from two representative olive growing areas in arid and semi-arid areas (Herz et al., 2007). Since then, these species were under study at our laboratory with the goal of their future utilization for biological control of agricultural pests existing in Egypt. The host-selection behavior of these *Trichogramma* species, toward eggs of the Indian mealmoth, *P. interpunctella*, the Mediterranean flour moth, *E. kuehniella*, the warehouse moth, *E. elutella*, and the almond moth, *C. cautella*, to select new candidate species for inundative releases against pest species in dry product stores in future was investigated. The bioassay for host-preference of *Trichogramma* was carried out by offering a single early wasp female the choice between equal numbers of host eggs on square cards "Petri dish method" and/or strip cards "strip card method" (modified method of Hassan and Guo, 1991). For Petri dish method, *E. kuehniella* was a highly acceptable host species for *T.cacociae*, *T.bourarachae* and *T.euproctidis* wasps. While *E.elutella* and *C.cautella* eggs were more acceptable for *T. evanescens* and *T. cordubensis*, respectively. In strip card method, *E. kuehniella* eggs were highly acceptable for *T. cacociae*, *T. bourarachae* and *T. evanescens*. Eggs of *E. elutella* and *C. cautella* were more acceptable for *T.euproctidis* and *T.cordubensis*, respectively.

Superparasitism is a form of parasitism in which more than one parasite egg may be inserted into the same host egg. Superparasitism as a behavior of the parasitoids is expressed when numbers of hosts are relatively low (Papaj et al., 1989). According to this interpretation a female wasp oviposits in an already parasitized host because of non-availability of unparasitized hosts in sufficient number for all of her eggs (Mangel and Clark, 1988). Most superparasitism cases have been described in support of this interpretation (Roitberg et al., 1992; van Alphen et al., 1992). The incidence of superparasitism by test wasps was observed in both the local wasps as well as *T. evanescens*. However, it was more prevalent in *C. cautella*, *E. elutella*, *E. kuehniella*, *P. interpunctella* eggs than in *S.cerealella* eggs. The average number of parasitized eggs per female/d ranged from 13.1 ± 1.5 *C. cautella* eggs by *T. borarachae* to 48.2 ± 2.4 *P. interpunctella* eggs by *T. euproctidis*. So using 250 ± 10 host eggs/female seem to be unlimited and unparasitized hosts are quite available. The occurrence of superparasitized eggs suggesting that the wasp species had a

**Table 2**  
Apparently superparasitized host eggs (%) among hosts that showed wasp-exit hole(s) in no choice experiments with individual *Trichogramma* females exposed to 250 ± 10 host eggs.

<i>Trichogramma</i> species	Superparasitized host eggs with exit hole (s) (%)				
	<i>C.cautella</i>	<i>E.elutella</i>	<i>E.kuehniella</i>	<i>P.interpunctella</i>	<i>S.cerealella</i>
<i>T.bourarachae</i>	7.6 (431) <sup>a</sup>	4.1 (321)	7.1 (414)	6.5 (515)	2.1 (390)
<i>T.cordubensis</i>	5.8 (448)	4.9 (484)	6.6 (329)	5.2 (413)	2.9 (445)
<i>T.cacociae</i>	8.2 (419)	5.2 (359)	3.9 (229)	6.3 (321)	3.1 (498)
<i>T.euproctidis</i>	9.7 (421)	8.2 (225)	6.6 (373)	5.4 (392)	1.9 (491)
<i>T.evanescens</i>	4.2 (272)	3.9 (273)	5.8 (269)	4.5 (399)	1.8 (509)

<sup>a</sup> Numbers in parentheses refer to number of dissected host eggs with exit hole (s).

propensity to lay more than egg in the same host eggs. On the contrary, Garcia and Tavares (1995) reported that *T. cordubensis* females have a high discriminative capacity towards previous parasitized *E. kuhniella* eggs when the host eggs are in excess. Results of the present work suggest that the test wasps failed to discriminate parasitized hosts eggs among a large number of non-parasitized eggs, thus superparasitism occurred. Also, both of Petri dish and strip cards methods may underestimate the actual parasitization capacity due to self-superparasitism and mortality in black eggs that suffered desiccation during the early stages. Natural enemies are applied commercially against stored-product moths in Central Europe. In this contribution, we show promising for selecting new *Trichogramma* spp. against some stored product lepidopterous pests. *T. euproctidis* appears to be a new candidate which have a possible effective control agent of test insect populations in stores maintained without insecticides, because the parasitization rate of egg-masses was significantly higher among the tested moth species. The study showed that isolating egg wasps from arid and semiarid areas have a great potential that may be exploited along with other strategies in the management of some stored product moths.

### Author agreement

No conflict of interest. We all unanimously agreed to publish in the journal.

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