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## Storage conditions' effects on two *Bemisia tabaci* parasitoids' (*Eretmocerus mundus* and *Encarsia sophia*) emergence

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### Abstract

The storage conditions of pests' natural enemies are prominent factors that delineate the biological control technique applicability and its effectiveness. In this work we evaluated two indigenous *Bemisia tabaci* parasitoids *Eretmocerus mundus* and *Encarsia Sophia* emergences following short -terms storage at temperature ranging from 0 to 12 °C. To do so, we used *E. mundus* and *E. Sophia* pre-pupae that were submitted to one combination of the following conditions: constant temperature of 0, 2, 4, 6, 8, 10 or 12 °C and duration of 5, 10, 15, 20, 25 or 30 days. Our results showed that the two wasps' emergence rate significantly increases when temperature is hissed, but it drops down when storage duration is longer. The optimal emergence rate were observed when storage conditions were set at 10 – 12 °C for 5 to 10 days, both for *E. mundus* (from  $93 \pm 4.83$  to  $98 \pm 4.22\%$ ) and *E. Sophia* (from  $81 \pm 9.94$  to  $95 \pm 7.07\%$ ). The evaluation of time lapses to emergence arrest and its maximal value permitted to define the chronology of 2<sup>nd</sup> release of parasitoids that might be applied 7 days after the first one, in the geothermal greenhouses. It is concluded that 10 to 12 °C temperature and duration of 5 to 10 days are optimal conditions to conserve the *E. mundus* and *E. Sophia* pre-pupae, in order to allow a maximal rate of their emergence. Further studies are in need to better explore these findings to ameliorate the two wasps' use in managing *B. tabaci* pest.

**Keywords:** Storage conditions, wasps, whiteflies' pests, managements

### 1. Introduction

*Bemisia tabaci* (Gennadius) species (Hemiptera: Aleyrodidae) are a serious polyphagous ravaging whitefly causing great damages in more than 700 different species of crops, vegetables and ornamental plants [1]. It affects both fields and greenhouse protected agricultures. These insects cause either direct plants' injuries through their feeding process (leaves piercing, sap sucking and secreting honeydew) [2], or indirectly via vectoring viruses. *B. tabaci* is known to transmit over 100 phytopathogenic viruses, including *Tomato yellow leaf curl virus* (TYLCV) [3]. In south Tunisia, *B. tabaci* was detected under geothermal greenhouse, and causes severe damages, especially for crops and vegetables where it finds favorable conditions (high air humidity and temperature) to pullulate. Since 1940s, *B. tabaci* geographical distribution has enlarged to invade all the terrestrial globe [4, 5]. Thus, worldwide efforts are concentrated to manage this pest. Recently, *B. tabaci* control using its natural enemies, as an alternate to chemical pesticides that become less effective because of the greater resistance developed by these whiteflies, is sought as the adequate, healthy and environmentally safe technique [6, 7]. Multiple parasitic, parasitoid or predator insects and acaries' species are already used in such objectives. However, the synchronization of the biological agents to adequately meet the favorable *B. tabaci* developmental stage that is indispensable remains a limiting factor for the effectiveness of the biological control and the used insects- commercialization. Two approaches were well developed to comply with this requirement. The first focused on the conservation of the biological agents' instars at low temperature (simulating the natural hibernation process); and the second is to use indigenous enemies of the pest. In this context, this work aimed to evaluate the effect of the storage's temperature and duration on two indigenous parasitoids of *B. tabaci*: *Eretmocerus mundus* (Mercer) and *Encarsia Sophia* (Girault & Dodd) (Hymenoptera: Aphelinidae) pre-pupae development to adult flies.

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## 2. Materials and methods

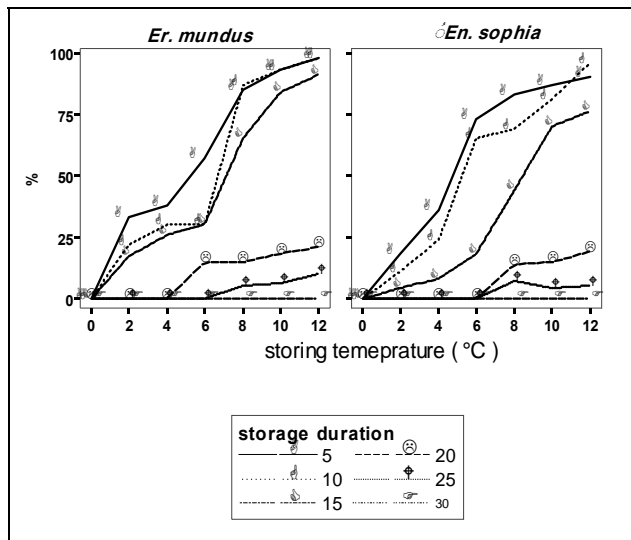
The two indigenous parasitoid wasps (*E. mundus* and *E. Sophia*) were reared on *B. tabaci* infesting the ornamental plant *Lantana camara* (at the experimental site of the Institute of Arid Regions of Kebili – Tunisia). 100 pre-pupae of each parasitoid were daily collected and mounted on (3x5 cm<sup>2</sup>) cardboard filled with date- honey (a carbohydrate-rich solution extracted from dates), in a ratio of 10 individual per one cardboard. Both, *E. Sophia* and *E. mundus* were similarly exposed to different storage conditions (temperature (°C) and durations (days)), in order to determine their influence on pre-pupal development. The cardboards were stored under seven constant temperatures (0, 2, 4, 6, 8, 10 and 12 °C) and in combination with six continuous durations (5, 10, 15, 20, 25 and 30 days), in dark. 10 cardboards supporting pre-pupae were stored at each condition's combination. Immediately, after storage cardboards were placed in an emergence- room at 27 °C (the medium temperature of the geothermal greenhouses), a photoperiod of 16/8 Light/Dark and 50-70% air relative humidity. Throughout the experiment, there was no pre-conditioning for temperature or storage duration. Daily adult- flies' emergence was recorded until its arrest, and the daily cumulative rate of emergence was calculated. Pre-pupae's death was considered when pupae did not hatch at the end of the experimental period (10 days after the release).

### 2.1 Statistical analysis

The obtained results were analyzed using SPSS.17 for windows. Pearson correlations were checked to outline the variation of pre-pupal development within storage's temperature and duration. ANOVA- test was used to compare results between the different storage conditions.

## 3. Results

### 3.1. Storage's duration and temperature effect on *B. tabaci*'s parasitoids' emergence rate:



**Fig 1:** the 10<sup>th</sup> day – cumulative emergence's rate of *E. mundus* and *E. Sophia* stored under different conditions.

Our results showed that the total emergence's rate significantly increase when the storage's temperature is augmented, at all the storing durations both for *E. mundus* and *E. Sophia*, as checked by Pearson's simple correlations. In addition, ANOVA analysis showed significant variability of the total cumulative rate of emergence (at the 10<sup>th</sup> day after the release) dependently on storage durations, both for *E. mundus* and *E.*

*Sophia* ( $F = 6.791$  and  $6.599$ , respectively at  $p \leq 0.01$ ). Among these durations, 5, 10 and 15 days allowed up to 75% of pre-pupae development into flying insects, at 12 °C. Other storage's duration permitted low emergence rate that was null at 30 days. Furthermore, no eclosion was observed when pre-pupae were conserved at 0 °C. Because of that only temperatures equal or more than 2 °C and storage durations strictly lesser than 20 days were considered for the later analyses. Both 0 °C and 30 days duration of storage are considered as lethal, for the two studied indigenous wasps. The following analyses were then focused on favorable conditions of storage (durations of 5, 10 and 15 days; and temperature 2 to 12 °C) allowing convincing emergence rates. The total cumulative emergence rate ranged from  $93 \pm 8.23$  to  $98 \pm 4.22$  for *E. mundus*; and from  $87 \pm 6.75$  to  $90 \pm 6.66$ , for *E. Sophia* when pre-pupae were conserved for 5 days; respectively at 10 and 12 °C. This rate varied between from  $93 \pm 4.83$  to  $98 \pm 4.22$  for *E. mundus*; and from  $81 \pm 9.94$  to  $95 \pm 7.07$ , for *E. Sophia* pupae when storage lasted for 10 days; for the same temperature interval (figure 1).

### 3.2 Storage's duration and temperature effect on parasitoids' latency to maximal emergence and its arrest:

The estimated delay from the experimental release at 27 °C to adults' maximal emergence was the lowest at 12 °C both for the two studied wasps when they are conserved for two weeks ( $1.1 \pm 0.31$  days and  $1.95 \pm 0.16$  respectively for *E. mundus* and *E. Sophia*). Similarly the lowest lapse of time to emergence arrest was also observed at the same condition ( $3.40 \pm 2.83$  and  $3.60 \pm 0.70$  for *E. mundus* and *E. Sophia*, respectively). The *E. mundus*'s maximal emergence rate was observed  $2.05 \pm 0.76$  days and  $1.80 \pm 0.42$  days after the release of pupae that were stored during 5 days at 10 and 12 °C, respectively. For *E. Sophia*, at the same storage duration, the delay to maximal emergence was equal to  $3.15 \pm 0.67$  and  $2.95 \pm 0.68$  respectively for 10 and 12 °C. For 10 days conservation at 10 and 12 °C this lapse of time ranged from  $2.40 \pm 0.51$  and  $3.15 \pm 0.57$  days, for both the two wasps (table 1).

## 4. Discussion

*B. tabaci* was sought with great concern, since the late 20<sup>th</sup> century when it invaded several regions of the World and caused severe damages in agriculture yielding. Yet, numerous researches are focusing on managing this pest using its natural enemies as a safe - alternate to chemical pesticides [8]. More than 500 different parasitoids of *B. tabaci* are described, among them *Eretmocerus* and *Encarsia* genera showed important effectiveness in controlling the ravager [9, 10]. An efficacious application of biological control requires an adequate chronological synchronization between the pest and its enemy's developmental stages. To comply with this optimal condition, cold storage of pupae (simulating the natural hibernation of the insect) and recruitment of indigenous species are envisaged. In the present study, the imaginal emergence rate of both *E. mundus* and *E. Sophia*, two Tunisian indigenous parasitoids of *B. tabaci*, was evaluated under various storing temperatures and durations. Conservation of pre-pupal instars of the two parasitoids at 0 °C or for durations longer than weeks (15 days) gave emergence rate lesser than 25%. This rate significantly increases when storing temperatures are augmented (from 2 to 12 °C), particularly for 5 and 10 days durations. Similar results have been observed using various species from *Eretmocerus* and *Encarsia* genera [11-13]. Our results pointed that temperatures of

10 to 12 °C and 5 to 10 days durations constitute desirable conditions for storing both *E. mundus* and *E. Sophia*, that allowed over 80% of imaginal emergence. It is spotted that at these conditions the metamorphosis physiological process is not affected, as it does using the other studied conditions. In

effect, several researches demonstrated that metamorphosis' process and pupae's survival are greatly affected by both the temperature and storing durations [14-16]. For example, the lower thermal threshold for development of *E. Formosa* pupae has been estimated at 13 °C [14].

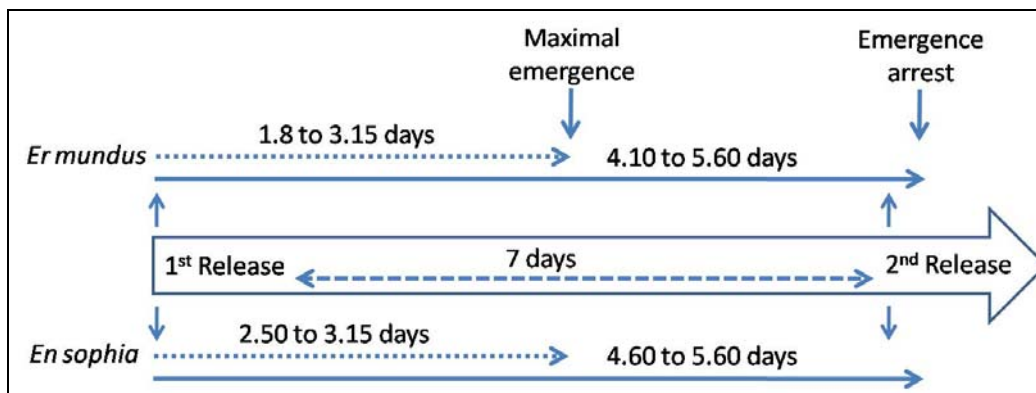


Fig 2: perspective parasitoids releasing chronology, after storage at 10- 12 °C for 5- 10 days.

The evaluation of the time- lapses to the maximal rate of emergence and its arrest, are sought as prominent factors during the application of the biological control techniques. It might define the chronological features of the first and subsequent wasps' release in the field or greenhouses. According to our findings, after storage at the chosen conditions of *E. mundus* and *E. Sophia* pre- pupae, the second release should be applied 7 days after the first one, in order to allow a continual and maximal adults' population coexistence in the geothermal greenhouses.

**5. Concluding remarks**

Storing biological agents is an indispensable tool to ameliorate the biological control, in terms of quality and quantity. In this context, our findings pointed toward the choice of temperature of 10 to 12 °C and 5 to 10 days duration for an optimal conservation of both the two parasitoids of *B. tabaci*: *E. mundus* and *E. Sophia*. The estimated delays to maximal flies' emergence and its arrest might permit the adjustment of the chronology of their application process, to maximize the active population against the pest. To better demarcate these findings, further studies are envisaged to evaluate post- emergence events (parasitoids survival and their population dynamic, and effectiveness against the pest).

Table 1: Time-lapse's variation to maximal emergence and its arrest after release of *E. mundus* and *E. Sophia* stored pre-pupae.

T ( °C)	Delay to (days)	5 days	10 days	15 days	
<i>E. mundus</i>	2	Maximal emergence	6.55 ± 0.83 (b,c,d,e,f)	6.43 ± 0.69 (b,c,d,e,f)	6.75 ± 0.67 (b,c,d,e,f)
		Emergence arrest	8.60 ± 0.28 (c,d,e,f)	8.50 ± 2.83 (c,d,e,f)	8.50 ± 2.83 (c,d,e,f)
	4	Maximal emergence	5.59 ± 1.23 (a,c,d,e,f)	5.62 ± 0.72 (a,c,d,e,f)	5.58 ± 0.69 (a,c,d,e,f)
		Emergence arrest	8.30 ± 2.83 (c,d,e,f)	8.50 ± 2.83 (c,d,e,f)	8.50 ± 2.83 (c,d,e,f)
	6	Maximal emergence	1.70 ± 1.33 (a,b)	2.30 ± 0.48 (a,b,e)	2.45 ± 0.49 (a,b,f)
		Emergence arrest	4.20 ± 2.83 (a,b)	4.10 ± 2.83 (a,b,d,e,f)	4.10 ± 2.83 (a,b,d,e,f)
	8	Maximal emergence	2.00 ± 0.78 (a,b,□)	2.65 ± 0.74 (a,b)	2.90 ± 0.87 (a,b,f,*)
		Emergence arrest	4.70 ± 2.83 (a,b)	5.40 ± 2.83 (a,b,c)	5.30 ± 2.83 (a,b,c,f)
	10	Maximal emergence	2.05 ± 0.76 (a,b,#,□)	3.15 ± 0.57 (a,b,c,f,*)	2.90 ± 0.73 (a,b,f,*)
		Emergence arrest	4.60 ± 2.83 (a,b,#,□)	5.60 ± 2.83 (a,b,c*)	5.40 ± 2.83 (a,b,c,f,*)
	12	Maximal emergence	1.80 ± 0.42 (a,b,#,□)	2.40 ± 0.51 (a,b,e,*,□)	1.10 ± 0.31 (a,b,c,d,e,*,#)
		Emergence arrest	4.10 ± 2.83 (a,b,#)	5.10 ± 2.83 (a,b,c,*,□)	3.40 ± 2.83 (a,b,c,d,e,#)
<i>E. Sophia</i>	2	Maximal emergence	6.50 ± 0.90 (d,e,f)	6.27 ± 0.90 (b,c,d,e,f,□)	7.66 ± 0.57 (b,c,d,e,f,#)
		Emergence arrest	8.33 ± 1.12 (d,e,f)	7.56 ± 1.24 (b,d,e,f)	8.05 ± 1.16 (c,d,e,f)
	4	Maximal emergence	6.44 ± 1.04 (d,e,f)	5.90 ± 1.02 (d,e,f)	6.40 ± 1.56 (a,c,d,e,f)
		Emergence arrest	8.40 ± 0.70 (d,e,f)	8.33 ± 0.50 (a,d,e,f)	8.30 ± 0.95 (c,d,e,f)
	6	Maximal emergence	6.27 ± 0.94 (d,e,f,#,□)	5.30 ± 0.79 (a,d,e,f,*,□)	4.10 ± 0.74 (a,b,f,*,#)
		Emergence arrest	8.20 ± 0.79 (d,e,f,□)	7.90 ± 0.74 (d,e,f, □)	5.40 ± 0.89 (a,b,f,*,#)
	8	Maximal emergence	4.55 ± 0.44 (a,b,c,e,f,#,□)	3.65 ± 0.41 (a,b,c,f,*,□)	3.90 ± 0.57 (a,b,f,*,#)
		Emergence arrest	7.20 ± 0.63 (a,b,c,e,f,#,□)	6.00 ± 0.67 (a,b,c,f,*)	5.80 ± 0.42 (a,b,c,f,*)
	10	Maximal emergence	3.15 ± 0.67 (a,b,c,d,f,□)	3.15 ± 0.41 (a,b,c,f,□)	3.70 ± 0.67 (a,b,f,*,#)
		Emergence arrest	5.30 ± 0.42 (a,b,c,d,f)	5.60 ± 0.70 (a,b,c,f)	5.50 ± 0.57 (a,b,f)
	12	Maximal emergence	2.95 ± 0.68 (a,b,c,d,e, #,□)	2.50 ± 0.47 (a,b,c,d,e,*,□)	1.95 ± 0.16 (a,b,c,d,e,*,#)
		Emergence arrest	4.60 ± 0.52 (a,b,c,d,e,□)	4.80 ± 0.63 (a,b,c,d,e,□)	3.60 ± 0.70 (a,b,c,d,e,*,#)

Significant differences at 0.05 are represented as follows: \*, # and □ represent significant differences from 5, 10 and 15 days, respectively; and a, b, c, d, e and f show significant difference from 2, 4, 6, 8, 10 and 12 °C, respectively.

**6. References**

1. McKensie CL, Anderson PK, Villareal N. An extensive survey of *Bemisia tabaci* (Homoptera: Aleyrodidae) in agricultural ecosystems in Florida. Florida Entomol. 2004; 87(5):403-407.
2. Oliveira MRV, Henneberry TJ, Anderson P. History, current status, and collaborative research projects for *Bemisia tabaci*. Crop Protection. 2001; 20:709-723.
3. Jones D. Plant viruses transmitted by whiteflies. Eur J Plant Pathol. 2003; 109:197-221.
4. Boykin LM, Shatters RG, Rosell RC, McKenzie CL, Bagnall RA, De Barro P. Global relationships of *Bemisia tabaci* (Hemiptera: Aleyrodidae) revealed using Bayesian analysis of mitochondrial COI DNA sequences. Mol. Phylogen. Evol. 2007; 44:1306-1319.
5. Gerling D. Status of *Bemisia tabaci* in the Mediterranean countries: opportunities for biological control. Biological Control. 1996; 6:11-22.
6. Omer AD, Tabashink BE, Johnson MW, Costa HS, De Ulman. Genetic and environmental influences on susceptibility to acephate in sweet potato whitefly (Homoptera: Aleyrodidae). J Econ Entomol. 1993; 86(4):625-659.
7. Horowitz AR, Ishaya I. Chemical Control of *Bemisia* Management and Application. In *Bemisia* Taxonomy, biology, Damage, Control and Management, Gerling, D. and R. T. Mayer (Eds.). Andover, Intercept, 1996, 537-556.
8. Lopez SN, Botto E. Effect of cold storage on some biological parameters of *Eretmocerus corni* and *Encarsia formosa* (Hymenoptera: Aphelinidae). Biological Control. 2005; 33:123-130.
9. Gerling D. The overwintering mode of *Bemisia tabaci* and its parasitoids in Israel. Phytoparasitica. 1984; 12:109-119.
10. Lopez-Avila A. Two new of *Encarsia* (Hymenoptera: Aphelinidae) from Pakistan, associated with the cotton whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae). Bull Entomol Res. 1987; 77(3):425-430.
11. Ballal C, Singh S, Jalali S, Kumar P. Cold tolerance of cocoons of *Allorhogas pyralophagus* [Hymenoptera: Braconidae]. Bio Control. 1989; 34:463-468.
12. Jalali SK, Singh SP. Differential response of four *Trichogramma* species to low temperatures for short term storage. Bio Control. 1992; 37:159-165.
13. Bueno R, Van Cleave HW. The effect of cold storage on the emergence of *Aphelinus perpallidus* (Hymenoptera: Aphelinidae): a parasitoid of *Monellia caryella* (Homoptera: Aphididae). Southwestern Entomologist. 1997; 22:39-51.
14. Lacey LA, Millar L, Kirk AA, Perring TM. Effect of storage temperature and duration on survival of eggs and nymphs of *Bemisia argentifolii* (Homoptera: Aleyrodidae) and pupae of the whitefly parasitoid *Encarsia formosa* (Hymenoptera: Aphelinidae). Annals of the Entomological Society of America. 1999; 92:430-434.
15. Easwaramoorthy S, Kurup NK, Santhalakshmi G, Shanmugasundaram M. Effect of low temperature storage on the variability of puparia of *Sturmiopsis inferens* Townsend (Diptera: Tachinidae): a larval parasitoid of sugarcane moth borers. Journal of Biological Control. 2000; 14:63-65.
16. Venkatesan T, Singh SP, Jalali SK. Effect of cold storage on cocoons of *Goniozus nephantidis* Muesebeck (Hymenoptera: Bethylinidae) stored for varying periods at different temperature regimes. Journal of Entomological Research. 2000; 24:43-47.