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Comparison of *Ephestia kuehniella* eggs sterilization methods for *Trichogramma* rearing

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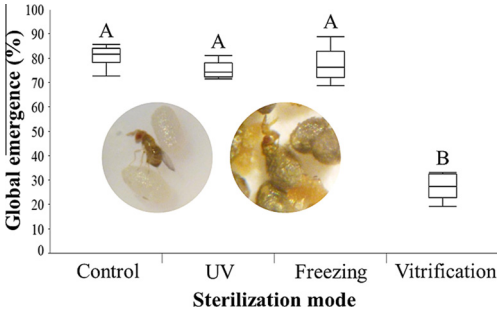
HIGHLIGHTS

- *Ephestia kuehniella* eggs sterilization methods were compared.
- UV irradiation, freezing at −15 °C and vitrification kill 100% of the embryos.
- UV and freezing were the most suitable sterilization methods for trichogramma rearing.

GRAPHICAL ABSTRACT



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ABSTRACT

Mass production is necessary to ensure the availability of biological control agents for the suppression of target pests. Many rearing hosts need to be sterilized to prevent development. Host egg sterilization also allows their storage for a longer period. *Ephestia kuehniella* eggs are frequently used as hosts for *Trichogramma* parasitoïds but they must be sterilized to prevent larvae from emerging and eating the unhatched parasitized eggs. Three sterilization methods were examined: UV irradiation, freezing at −15 °C and vitrification (liquid nitrogen submersion). The dosage and exposure duration to provide egg sterilization were determined and then the suitability of hosts sterilized by the different methods were compared. *E. kuehniella* eggs abortion was achieved after 15 min by UV irradiation, 4 h by freezing at −15 °C and 30 s by vitrification. Vitrification resulted in significantly lower parasitoids production with a global emergence rate of 28.7%, compared to UV irradiation (75.1%), freezing at −15 °C (77.4%) and control (80.9%). Host eggs sterilization method did not affect sex-ratio, occurrence of malformation in adults, and female walking speed. Fecundity was significantly reduced in the females emerging from UV irradiated (37.2 offsprings) and vitrified (36.9 offsprings) eggs, compared to control (43.1 offsprings).

1. Introduction

Dependable availability of biological control agents is essential for augmentative and inoculative biological control. For this reason, rearing of easily and cheaply obtained hosts/prey is fundamental to successful control programs. For *Trichogramma* production, factious host eggs used, are generally from stored grain moths like *Sitotroga cerealella* Olivier (Lepidoptera: Gelechiidae), *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) or *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae) (Singh, 1969). These species do not require laboriously maintained plants, have no diapause and females lay large quantities of eggs.

For parasitoids, reproduction depends on the suitability of the selected host for the parasitoid's development (Vinson and Iwantsch, 1980). For this reason, parasitoids are able to discriminate between different qualities of host. Different criteria are

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evaluated by the parasitoid after contacting a potential host, like host size, age, nutritional suitability and previous parasitism (Schmidt, 1994). Among *Trichogramma*, there is generally a preference for younger eggs over more developed ones (Calvin et al., 1997; Makee, 2005; Saour, 2004; Tunçbilek and Ayvaz, 2003). This is an important reason to sterilize host eggs early and so stop their development. Sterilization also prevents the subsequent cannibalism by hatched larvae on unhatched parasitized eggs (Mansour, 2010; Voegelé et al., 1974).

There are different means to sterilize host eggs. Cold, heat, ultraviolet and gamma irradiation, and chemicals have been used (Voegelé et al., 1974). Although UV irradiation has been recommended by Voegelé et al. (1974) for the treatment of *E. kuehniella* eggs used in *Trichogramma* rearing, this recommendation was based without consideration for the other techniques. Ayvaz et al. (2008) demonstrated that there was no significant difference between gamma-irradiated versus ultraviolet-irradiated *E. kuehniella* eggs in terms of the reproductive success of *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae). Singh (1969) found the freezing of the *C. cephalonica* eggs at −4 °C for 16–76 h an advantageous method for *Trichogramma australicum* Girault (Hymenoptera: Trichogrammatidae) rearing, citing its simplicity compared to UV irradiation. Finally, Huai-Yi (1988) obtained good parasitism and emergence rates of *Trichogramma dendrolimi* Matsumara and *Tribolium confusum* Viggiani (Hymenoptera: Trichogrammatidae) on tussah moth *Antheraea pernyi* Guerin-Meneville (Lepidoptera: Saturniidae) eggs stored in liquid nitrogen and thowed in water at 30 °C.

The mode of preventing host development differs with each technique. Since UV rays do not penetrate deeply, the effect of UV irradiation is different depending on the hardness and thickness of the chorion of the egg and/or stage of embryonic development (Hu et al., 1999). In *E. kuehniella*, eggs that are less than 24 h or more than 48 h are the most vulnerable to UV (Voegelé et al., 1974).

Cold sterilization has been tested at different temperatures (Greco and Stilinovic, 1998; Özder, 2002; Singh, 1969). Freezing begins only when molecule aggregation reaches a specific crystal size (Salt, 1961). The growth of the crystal decreases when the viscosity increases, resulting in the formation of several nuclei. If the conditions that formed the nuclei persist, the result is the formation of numerous very small crystals (Salt, 1961). When the cooling occurs very rapidly at extremely low temperatures without freezing, this is known as vitrification. In this case, numerous nuclei are formed but do not have the opportunity to grow (Salt, 1961). Depending on the temperature, sterilization can be done by freezing or by vitrification, using liquid nitrogen.

This study aims to compare for the first time the quality of *E. kuehniella* egg as host for *Trichogramma ostriniae* Pang and Chen (Hymenoptera: Trichogrammatidae) following their sterilization by UV irradiation, freezing at −15 °C and vitrification. Our hypothesis is that the sterilization by vitrification will allows the highest parasitism rate since the vitrification should not alter eggs quality.

2. Material and methods

2.1. Insect material

E. kuehniella eggs were obtained from Anatis Bioprotection Inc. (Quebec, Canada). *T. ostriniae* were obtained from IPM Laboratory Inc. (New York, USA), whose colony was started from individuals obtained from a Cornell University colony maintained since 1993 and initiated with specimens from the USDA APHIS Mission Biological Control Center, Mission, Texas, originally imported from Jilin Province in Northern China in 1990.

2.2. E. kuehniella eggs sterilization

Eggs less than 24 h old were used for the experiment. For UV irradiation, 2000 eggs were put in an UV irradiation cabinet at a distance of 10 cm from an 8 W lamp producing light at a 254 nm wave-length. About 200 eggs were retrieved every 5 min for 50 min. Subsequently eggs were maintained at 24 ± 1 °C, 60 ± 5% R.H., 16L: 8D photoperiod. Larval emergence was determined seven days later. Freezing sterilization was accomplished by placing 1600 eggs in a freezer at −15 °C. About 200 eggs were retrieved every hour over 8 h. These eggs were maintained as described above. To vitrify host, 800 eggs were put in 4 tubes and submerged in liquid nitrogen. One tube was retrieved every 15 s over 1 min, and these eggs were maintained as described above. For each sterilization method, the eggs were weighed before and after the sterilization and their appearance were compared to fresh eggs under optical microscope (×400).

2.3. Evaluation of the sterilized egg quality

To compare the quality of host eggs, we chose for each sterilization method, the minimum exposure time to reach 100% mortality of the host eggs. All experiments were done under laboratory controlled conditions (24 ± 1 °C, 60 ± 5% R.H., 16L: 8D photoperiod). For each sterilization method, 0.2 g (∼7400) sterilized eggs were glued on cardboard with non-toxic glue. The control treatment was prepared similarly, but with 0.2 g of unsterilized eggs. Eggs were placed in a glass jar with 0.02 g of parasitized *T. ostriniae* eggs about to emerge, ∼250 females. For each sterilization method, five replicates were done. Emerging parasitoids were allowed to parasitize eggs for 24 h, and then the eggs were removed.

Under these conditions, parasitized eggs began to blacken after 5 days and parasitoids began to emerge after 10 days. The parasitism rate (i.e. the number of black eggs over the total number of eggs) was calculated 7 days after the contact and the emergence 12 days after the contact by counting the number of eggs with an emergence hole. Global emergence was obtained by calculating the proportion of host eggs that yielded an adult parasitoid over the total number of eggs. The data for parasitism rate, emergence and global emergence were analyzed by a one-way ANOVA followed by an HSD Tukey test ($\alpha = 0.05$).

2.4. T. ostriniae quality

T. ostriniae quality was assessed in adults resulting from the parasitism and emergence experiments. The sex ratios and percentage of deformed adults were recorded for each sterilization method. Adult females used for quality comparisons were held in the presence of males for ∼24 h after hatching to allow time for mating. For each egg sterilization method, 20 females were used. They were first recorded with a Panasonic 12 mega pixels camera for 5 min walking on a one by one millimeter graph sheet. The walking speed was then calculated from the video for 10 continuous walking episodes of 10 s or more. After their walking speed evaluation, each female was put in a glass tube 4 cm high and 1½ cm of diameter, with 200 fresh *E. kuehniella* eggs glued on cardboard and a drop of 50% honey/water solution. The females were removed after 24 h and the number of black eggs was counted 7 days later. Data from quality comparison were analyzed by Fisher's protected LSD.

2.5. Female discrimination of vitrified eggs

Preliminary evidence suggested female parasitoids might discriminate against vitrified eggs. We exposed five vitrified and five untreated eggs to a 24 h old *T. ostriniae* female and noted the

number and type of host parasitized. We repeated this trial with 30 individual females. Data were analyzed by a Student *t*-test. All statistical analyzes were carried out using JMP 10 (SAS Institute Inc.).

3. Results

3.1. E. kuehniella eggs sterilization

For each sterilization method, we observed 100% mortality of the *E. kuehniella* eggs. The required time to obtain 100% mortality was different for each method (Fig. 1). With vitrification at 30 s being the fastest, followed by UV irradiation for 15 min and finally freezing at −15 °C for 4 h. Under an optical microscope (×400), there were no apparent differences in egg aspect among the control and the different sterilisation methods. However, the weight of the UV irradiated eggs was reduced by 3% compared to their pre-sterilization weight while no reduction of weight was noted for the freezing and vitrification methods.

3.2. T. ostriniae parasitism

E. kuehniella eggs sterilized by the different methods were accepted as hosts by *T. ostriniae* females. However, the parasitism rates differed significantly ($F_{(3,36)} = 133.86$; $P < 0.0001$). Eggs sterilized by vitrification yielded significantly a lower parasitism rate, 2.5 times lower than the unsterilized eggs (Fig. 2A). On the other hand, parasitism rates of *E. kuehniella* eggs sterilized by UV irradiation and freezing and unsterilized eggs did not significantly differ.

T. ostriniae adult emergence rates showed the same pattern as parasitism rates ($F_{(3,36)} = 95.63$; $P < 0.0001$). The emergence of *T. ostriniae* reared on eggs sterilized by vitrification was significantly lower than the other methods (Fig. 2B). The global emergence rates, the number of emerged parasitoids over the total number

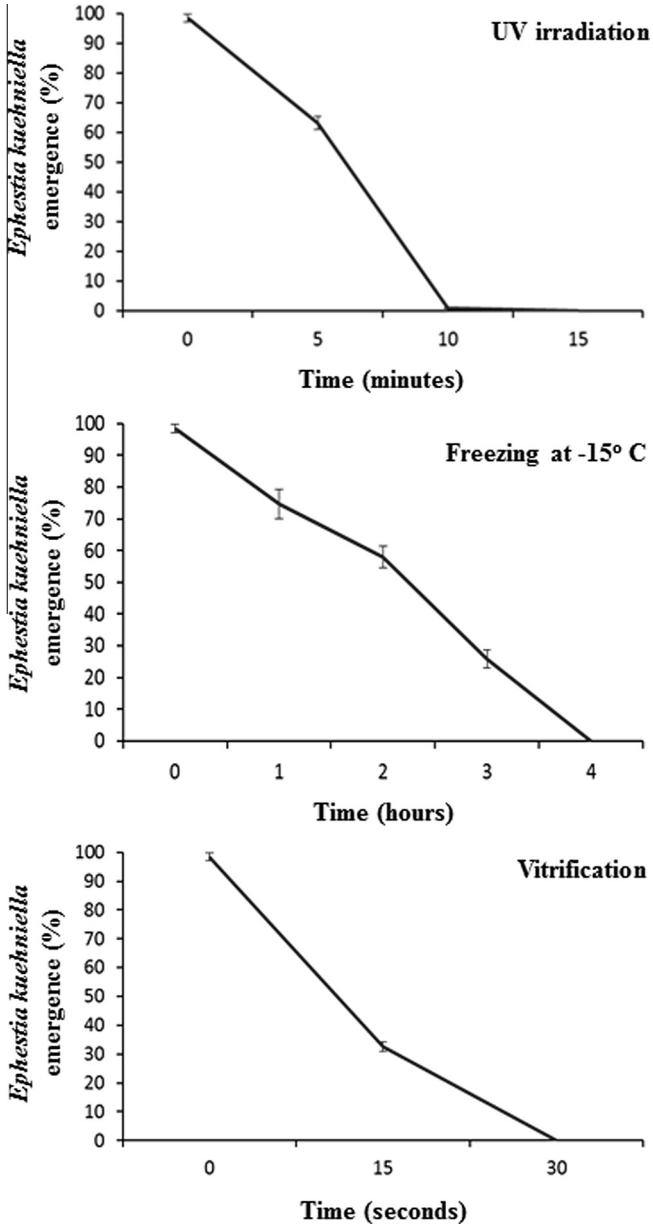

Fig. 1. Percentage of *E. kuehniella* larvae emergence after sterilization by UV irradiation, freezing at −15 °C and vitrification ($T°$: 24 ± 1 °C, R.H.: 60 ± 5%, 16L:8D).

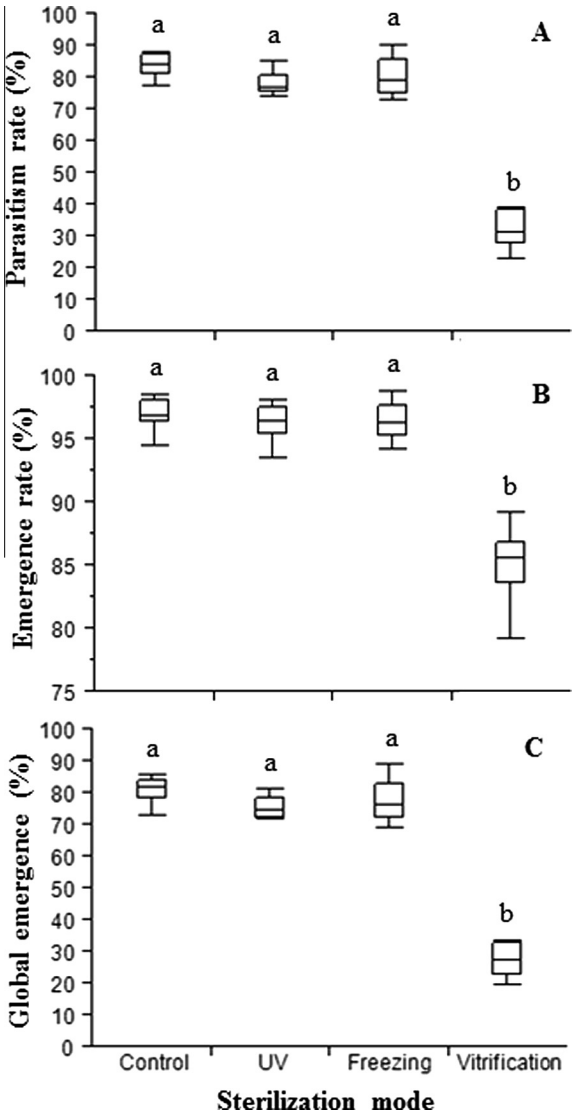

Fig. 2. *Trichogramma ostriniae* parasitism on *E. kuehniella* eggs according to different sterilization methods. (A) Parasitism rate (black eggs), (B) emergence rate (of parasited eggs), (C) global emergence, i.e. the number of emerged trichogramma over the total number of eggs submitted to parasitism. Boxes (graphique representation of five values: the smallest and largest observation and the lower, median and upper quartile) sharing the same letter are not significantly different (Tukey, $\alpha = 0.05$).

of eggs, were significantly different ($F_{(3,36)} = 160.77$; $P < 0.0001$) with vitrification yielding fewer adults than UV irradiated, frozen and unsterilized eggs (Fig. 2C).

3.3. T. ostriniae quality

There were no significant differences among adult parasitoids emerging from *E. kuehniella* eggs sterilized by different methods in terms of female proportion ($F_{(3,16)} = 0.01$; $P = 0.9981$), deformed adult percentage ($F_{(3,16)} = 1.74$; $P = 0.1982$), or female walking speed ($F_{(3,76)} = 1.04$; $P = 0.3817$). Female fecundity differed significantly among the different sterilization methods ($F_{(3,76)} = 3.13$; $P = 0.0304$). Females emerging from vitrified and UV irradiated eggs yielded fewer offsprings than control (Table 1).

3.4. Female discrimination of vitrified eggs

T. ostriniae females laid eggs in vitrified *E. kuehniella* eggs (96.0 ± 8.1) at the same rate as in unsterilized eggs (98.0 ± 6.1) ($t_{(58)} = -1.08$; $P = 0.1430$).

4. Discussion

An optimal sterilization method for *Trichogramma* rearing units should: (1) kill 100% of the factious host eggs, (2) provide high parasitism and parasitoids emergence rates, (3) yield parasitoids of high quality and (4) be the least expensive and time/equipment consuming.

Our results demonstrated that: (1) all the sterilization methods killed 100% of the factious host eggs, (2) host eggs sterilized by UV irradiation and freezing, but not vitrification, yielded high parasitism and emergence rates comparable to control, (3) all sterilization methods yielded high quality adults and (4) all three methods are easily available.

Contrary to our hypothesis, sterilization by vitrification generated the lowest parasitism and emergence rates, nearly 3 times lower than other sterilization methods. Liquid nitrogen has been used in some studies of different host eggs with various degrees of success but not always as a vitrification process. In most studies, liquid nitrogen was used with a cryoprotectant and the temperature was gradually decreased until the eggs were stored in liquid nitrogen. The quality of tussah moth, *A. pernyi*, eggs stored in liquid nitrogen was similar to fresh eggs for parasitism by *T. dendrolimi* and *T. confusum* (Huai-Yi, 1988). Although Hu and Xu (1988) considered the submersion in liquid nitrogen of the rice moth, *C. cephalonica*, eggs an effective method for *T. dendrolimi* production, the average parasitism rate was as low as 41.3%. However the percentage was higher (66.1%) for oak silkworm *Antheraea yamamai* Guérin-Méneville (Lepidoptera: Saturniidae). *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae) pupal production was reduced in *S. cerealella* vitrified eggs (Greco and Stilinovic, 1998). Although *T. ostriniae* production with *E. kuehniella* eggs sterilized by vitrification was lower, the adults had the same sex ratio, deformed adults percentage and walking speed as those from other sterilization methods. Female fecundity was lower than the control but according to Bigler et al. (1988) the most important variable to assess quality of *Trichogramma* is walking speed. Although quality criteria for *T. ostriniae* are not established, we can compare the present results with the criteria for another corn borer parasitoid *T. brassicae* Westwood (Hymenoptera: Trichogrammatidae). According to the IOBC Quality Control Guidelines (2002) female should produce at least 40 offspring in 7 days. The *T. ostriniae* emerging from vitrified eggs produced 36.9 offspring in 24 h.

The poor performance of the vitrified eggs as a suitable host raised the question whether females discriminate against these eggs, or whether the eggs laid by the females did not develop. Results of the discrimination trial showed that females did not discriminate against vitrified eggs, and therefore the low parasitism rate resulted from the inability of *T. ostriniae* parasitoids to develop after oviposition.

Two sterilization methods provided better results, UV irradiation and freezing at −15 °C. UV irradiation has been frequently used as the sterilization method of host eggs in *Trichogramma* studies (Hoffmann et al., 2001; Pavlik, 1993; Wang et al., 2004; Wright et al., 2001).

Based on previous studies, sterilization by freezing at −15 °C was a more uncertain method. Singh (1969) obtained suitable results with freezing at −4 °C of *C. cephalonica* eggs for *T. australicum* but Özder (2002) found that at −20 °C *E. kuehniella* eggs became quickly unsuitable for *T. evanescens* and *T. brassicae*. Greco and Stilinovic (1998) obtained no pupa of *T. pretiosum* from *S. cereallela* kept 5 days or more at −20 °C.

Since there was no significant difference between UV irradiation, freezing at −15 °C and control eggs in terms of parasitism rate, emergence rate and most of the quality control parameters, the choice of the sterilization method is a question of practicality. Freezers are more common equipment than UV lamp but UV sterilization takes less time.

There are other sterilization methods that we did not test. Heat was used by Bonnemaison (1972), and although it allowed *Trichogramma* development it also extended the length of development. Eggs sterilization can also be achieved by the sterilization of the host males, but that requires the separation of males and females, which is time consuming. Still another method is gamma radiation (Mansour, 2010). This technique however is limited by the availability of radioisotopes (Mastrangelo et al., 2010). For that reason, X-rays have been studied for its replacement in the sterile insect technique (SIT) (Mastrangelo et al., 2010).

Finally, the results obtained in this study compared eggs that were used immediately after sterilization. It remains to be determined how long the eggs, sterilized with each method, remain suitable for subsequent parasitism.

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Table 1
Quality control parameters of F1 generation *Trichogramma ostriniae* adults emerging from sterilized eggs according to sterilization methods (Mean ± SE).

	Female (%)	Deformed adults (%)	Female fecundity (24 h)	Walking speed (mm/s)
Control	63.8 ± 4.2a	4.8 ± 0.60a	43.1 ± 1.6a	4.9 ± 0.19a
UV	62.4 ± 3.7a	5.2 ± 0.56a	37.2 ± 2.2b	5.1 ± 0.18a
Freezing	62.4 ± 4.1a	5.2 ± 0.58a	41.8 ± 1.8ab	4.8 ± 0.23a
Vitrification	60.7 ± 3.0a	6.5 ± 0.37a	36.9 ± 1.4b	4.6 ± 0.15a

Numbers followed by the same letter in a column are not significantly different ($\alpha = 0.05$).

References

Ayvaz, A., Eyüp, K., Karabörklü, S., Tunçbilek, A.S., 2008. Effects of cold storage, rearing temperature, parasitoid age and irradiation on the performance of *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae). J. Stored Prod. Res. 44, 232–240.

Bigler, F., Bieri, M., Fritschy, A., Seidel, K., 1988. Variation in locomotion between laboratory strains of *Trichogramma maidis* and its impact on parasitism of eggs of *Ostrinia nubilalis* in the field. Entomol. Exp. Appl. 49, 283–290.

Bonnemaison, L., 1972. Diapause et superparasitisme chez *Trichogramma evanescens* Westwood (Hym. Trichogrammatidae). Bull. Soc. Entomol. Fr. 77, 122–132.

Calvin, D.D., Losev, J.E., Knapp, M.C., Poston, F.L., 1997. Oviposition and development of *Trichogramma pretiosum* (Hymenoptera: Trichogrammatidae) in three age classes of southwestern corn borer eggs. Environ. Enotmol. 26, 385–390.

Greco, C.F., Stilinovic, D., 1998. Parasitization performance of *Trichogramma* spp. (Hym., Trichogrammatidae) reared on eggs of *Sitotroga cerealella* Oliver (Lep., Gelechiidae), stored at freezing and subfreezing conditions. J. Appl. Entomol. 122, 311–314.

Hoffmann, M.P., Ode, P.R., Walker, D.L., Gardner, J., van Nouhuys, S., 2001. Performance of *Trichogramma ostriniae* (Hymenoptera: Trichogrammatidae) reared on factitious hosts, including the target host, *Ostrinia nubilalis* (Lepidoptera: Crambidae). Biol. Control 21, 1–10.

Hu Z., Xu Q., 1988. Studies on frozen storage of eggs of rice moth and oak silkworm, Trichogramma and other egg parasites. In: 2nd International Symposium Guangzhou, China, Paris: Les Colloques de L'INRA, 43, p. 644.

Hu, J.S., Gelman, D.B., Bell, R.A., 1999. Effects of selected physical and chemical treatments of Colorado potato beetle eggs on host acceptance and development of the parasitic wasp, *Edovum puttleri*. Entomol. Exp. Appl. 90, 237–245.

Huai-Yi, M., 1988. Studies on long-term storage of hosts for propagating Trichogramma, Trichogramma and other egg parasites. In: 2nd International Symposium Guangzhou, China, Paris: Les Colloques de L'INRA, 43, p. 644.

IOBC Quality Control Guidelines, 2002. http://users.ugent.be/~padclerc/AMRQC/guidelines.htm, page consulted September 18, 2013.

Makee, H., 2005. Factors influencing the parasitism of codling moth eggs by *Trichogramma cacoeciae* March. and *T. principium* Sug. et Sor. (Hymen. Trichogrammatidae). J. Pest Sci. 78, 31–39.

Mansour, M., 2010. Effects of gamma radiation on the Mediterranean flour moth, *Ephestia kuehniella*, eggs and acceptability of irradiated eggs by *Trichogramma cacoeciae* females. J. Pest Sci. 83, 243–249.

Mastrangelo, T., Parker, A.G., Jessup, A., Pereira, R., Orozco-Dávila, D., Islam, A., Dammalage, T., Walder, J.M.M., 2010. A new generation of X ray irradiators for insect sterilization. J. Econ. Entomol. 103, 85–94.

Özder, N., 2002. Parasitation performance of *Trichogramma cacoeciae*, *T. evanescens* and *T. brassicae* (Hym. Trichogrammatidae) reared on the embryos of *Ephestia kuehniella* Zell. (Lep. Pyralidae) killed by freezing. Great Lakes Entomol. 35, 107–111.

Pavlik, J., 1993. The size of the female and quality assessment of mass-reared *Trichogramma* spp. Entomol. Exp. Appl. 66, 171–177.

Salt, R.W., 1961. Principles of insect cold-hardiness. Annu. Rev. Entomol. 6, 55–74.

Saour, G., 2004. Efficacy assessment of some Trichogramma species (Hymenoptera: Trichogrammatidae) in controlling the potato tuber moth *Phthorimaea operculella* Zell. (Lepidoptera: Gelechiidae). J. Pest Sci. 77, 229–234.

Schmidt, J.M., 1994. Host recognition and acceptance by Trichogramma. In: Wajnberg, E., Hassan, S.A. (Eds.), Biological Control with Egg Parasitoids. CAB International, Oxon, U.K., pp. 165–200.

Singh, R.P., 1969. A simple technique for rendering host eggs inviable for the laboratory rearing of *Trichogramma* spp. Indian J. Entomol. 31, 83–84.

Tunçbilek, A.S., Ayvaz, A., 2003. Influences of host age, sex ratio, population density, and photoperiod on parasitism by *Trichogramma evanescens* Westw. (Hym., Trichogrammatidae). J. Pest Sci. 76, 176–180.

Vinson, S.B., Iwantsch, G.F., 1980. Host suitability for insect parasitoids. Annu. Rev. Entomol. 25, 397–419.

Voegelé, J., Daumal, J., Brun, P., Onillon, J., 1974. Action du traitement au froid et aux ultraviolets de l'œuf d'ephestia kuehniella (Pyralidae) sur le Taux de multiplication de Trichogramma evanescens et *T. brasiliensis* (Hymenoptera: Trichogrammatidae). Entomophaga 19, 341–348.

Wang, B., Ferro, D.N., Wu, J., Wang, S., 2004. Temperature-dependent development and oviposition behavior of *Trichogramma ostriniae* (Hymenoptera: Trichogrammatidae), a potential biological control agent for the European corn borer (Lepidoptera: Crambidae). Environ. Entomol. 33, 787–793.

Wright, M.G., Hoffmann, M.P., Chenus, S.A., Gardner, J., 2001. Dispersal behavior of *Trichogramma ostriniae* (Hymenoptera: Trichogrammatidae) in sweet corn fields: implications for augmentative releases against *Ostrinia nubilalis* (Lepidoptera: Crambidae). Biol. Control 22, 29–37.