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Jyoti Raama Bhardwaj
Department of Entomology, IGKV, Raipur, Chhattisgarh, India

Rajeev Gupta
Department of Entomology, IGKV, Raipur, Chhattisgarh, India

Akhilesh Kumar
Department of Entomology, IGKV, Raipur, Chhattisgarh, India

Sourabh Padmashali
Department of Entomology, IGKV, Raipur, Chhattisgarh, India

Corresponding Author:
Jyoti Raama Bhardwaj
Department of Entomology, IGKV, Raipur, Chhattisgarh, India

Effect of storage temperature on different biological parameters of *Trichogramma chilonis* survivability

Jyoti Raama Bhardwaj, Rajeev Gupta, Akhilesh Kumar and Sourabh Padmashali

Abstract
Developing efficient storage technique to produce quality parasitoids is essential for successful bio-control programs. In this study three different temperatures for six weeks were tested for best storage temperature and periods. Testing of survivability of *B. hebetor* and *T. chilonis* during storage at different temperatures revealed that in *T. chilonis* adult emergence was seen up to six weeks but at 5 ^{o}C and 95 to 97% RH almost 50% survivability was recorded up to five weeks. At 10 ^{o}C and 70 to 75% RH approx 50% survivability found up to four weeks and 15 ^{0}C and 85 to 89% RH upto 50% survivability was recorded only for two weeks.

Keywords: storage temperature, biological parameters, Trichogramma chilonis

Introduction
Trichogramma species are used throughout the world against various insect pests in biological control (Li, 1994). In India, 28 different species of *Trichogramma* and *Trichogrammatoidea* have been recorded. Among them, *T. chilonis* (Ishii) and *T. japonicum* (Ashmead) are the highly successful ones for pest management in tomato, sugarcane, cotton, and rice ecosystems, while *T. achaeae* Nagaraja and Nagarkatti has showed promising results against *Achaeae janata* (Linn.) (Noctuidae) on castor. *Trichogramma cordubensis* Vargas and Carbellois is an exotic species, which is used world-wide to control *Autographa gamma* (L.), *Chrysodeixis chalcites* (Esper), *Phlogophora meticulosa* (L.), *Peridroma saucia* (Hübner), *Xestia c-nigrum* (L.), and *Noctua pronuba* (L.) (Garcia *et al*. 1995; Lingappa and Hegde 2001; Singh *et al*. 2001; Lalitha and Ballal 2015) [2, 7, 10, 5]. Due to their worldwide success, these minute polyphagous endo-parasitoids are being commercially produced. Therefore, the commercial insectaries are challenged with a synchronous production rate during the seasonal high market demands. To address this problem, developing a suitable user-friendly storage protocol becomes mandatory. Cold storage is an appropriate way to extend the life of natural enemies such as insect parasitoids. Low temperature storage is a valuable method for increasing the shelf life of natural enemies such as insect parasitoids and predators.

Material Methods
T. chilonis culture was taken from Biocontrol laboratory Indira Gandhi Krishi Vishwavidyalaya, Raipur and this culture was again reared on universal host *C. cephalonica.* Freshly collected *Corcyra* eggs (0 to 4 h old) were UV irradiated. Hundred eggs were pasted on a thick paper card (2×1 cm) and all prepared sentinel cards were exposed to an old standard size tricho card which were about to emerge for 24 hour, inside sealing plastic envelope (20cm×15cm). Honey solution (5%) was used as food for parasitoids. After 24 hour of parasitization the parental Tricho card was removed. The plastic envelope containing parasitized cards were kept on BOD for 4 days till the developing embryos progressed into pupal stage, as indicated by darkening of the parasitized eggs. Then newly prepared tricho cards were stored in refrigerators at different temperatures. For investigating effect of cold storage temperature each treatments were taken out after one week, two week, three week, four and five week. Each card was transferred to a separate petridish and covered for observing adult emergence and adult longevity.

Result and Discussion

Effect of different storage temperature on parasitisation of *T. chilonis*

At 5 °C temperature and 95 to 97% RH

Long-term cold storage reduced the adult emergence percent and also longevity of parasitoids. The data revealed Table 4.11 that maximum per cent adult emerged were recorded at control 94.20% which is at par with first week of storage (93.40%) up to one percent reduction in percent adult emergence was recorded after first week of storage at (5 ºC and 95 to 97% RH).

Table 1: Effect of cold storage (5 ºC and 95 to 97% RH) at different periods on adult emergence and adult longevity of *T. chilonis*.

S. No.	Treatments (week of storage)	Survivability %	Adult longevity (Days)
1	One week	93.40 (75.15) ab	9.40 (3.06) ab
2	Two week	92.00 (73.61) b	9.40 (3.06) ab
3	Three week	83.20 (65.83) c	9.00 (2.99) ab
4	Four week	74.60 (59.75d	8.40 (2.89) b
5	Five week	54.00 (47.29) e	6.20 (2.48) c
6	Six week	32.20 (34.56) f	7.00 (2.64) c
7	Control (29 ºC-32 ^{0}C and RH 57-70%)	94.20 (76.13) a	10.00 (3.16) a
	SEm	0.04	0.07
	CD at 5%	1.75	0.21
	C. V. (%)	2.19	5.72

Significantly lowest adult emergence percent was observed in last week (sixth week) of storage (32.20%) up to 70% reduction in adult emergence was recorded after sixth week of storage. Adult emergence percent in second week, third week, fourth week and fifth week were significantly reduced weekly with 92.00, 83.20, 74.60 and 54.00 percent of adult emergence respectively. Fifty percent of pupal survivability recorded for five weeks. Adult longevity also affected significantly and highest longevity found in progeny emerged from control (10.00 days). Second highest days of adult longevity recorded in first and second week of storage (9.40 days). Least longevity found in adults emerged from trichocards which stored for fifth weeks (6.20 days). Third, fourth and fifth week of storage were significantly varied but not much with 9.00, 8.40 and 6.20 days respectively. Kumar *et al*. (2005) reported that *Trichogramma brasiliense* Ashmead can be stored for 10 days while *Trichogramme chilonis* Ishii and *T. pretiosum* can be stored for 20 days at 4 ± 0.5 °C without adversely affecting their emergence and parasitization efficiency. Similarly in this experiment 50 percent survivability recorded up to five week of storage, So *T. chilonis* can be stored at 5ºC upto five week effectively.

- **At 10 °C temperature and 67 to 72% RH**

The data revealed Table 4.12 that maximum per cent adult emerged were recorded at control (94.80%) which was comparable to first week of storage (93.00%). Only up to 1 percent of adult emergence was recorded less in first week of storage of tricho-cards in 10 ^{0}C. Significantly least percent of adult emergence recorded in a card which was stored for six weeks (25.20%). Fifty percent of pupal survivability recorded for four weeks.

Table 2: Effect of cold storage (10 ºC and 67 to 72% RH) at different periods on adult emergence and adult longevity of *T. chilonis*

S. No.	Treatments (week of storage)	Survivability %	Adult longevity (Days)
1	One week	93.00 (74.70)b	11.20 (3.33)a
2	Two week	83.40 (65.96)c	10.60 (3.25)ab
3	Three week	66.40 (54.58)d	10.20 (3.19)ab
4	Four week	55.80 (48.33)e	8.60 (2.93)bc
5	Five week	31.40 (34.07)f	7.60 (2.75)cd
6	Six week	25.20 (30.12)g	5.20 (2.27)d
7	Control (29 ºC-32 ^{0}C and RH 57-70%)	94.80 (76.85)a	9.60 (3.09)e
	SEm	0.81	0.07
	CD at 5%	1.63	0.21
	C. V. (%)	2.29	5.59

Percent of adult emergence in two week, three week, four week and fifth week were also significantly reducing weekly with 83.40, 66.40, 55.80 and 31.40 percent respectively.

- **At 15 ^{0}C temperature and 85 to 89% RH**

Significantly highest percent of adult emergence was recorded in control (96.00%). Similarly second highest adult emergence percent was found in first week of storage (95.00%). Least percent of adult emergence recorded in cards which stored for six weeks (21.26%). Percent of adult emergence in two week, three week, four week and fifth week were significantly varied with 79.00, 48.20, 36.40 and 21.40 percent respectively. Fifty percent of pupal survivability recorded for two weeks.

Table 3: Effect of cold storage (15 ^{0}C and 85 to 89% RH) at different periods on adult emergence and adult longevity of *T. chilonis*

S. No.	Treatments (week of storage)	Survivability %	Adult longevity (Days)
1	One week	95.00 (77.16)a	9.80 (3.12)a
2	Two week	79.00 (62.73)b	7.40 (2.71)b
3	Three week	47.60 (43.71)c	4.60 (2.14)c
4	Four week	37.60 (37.81)d	6.40 (2.52)d
5	Five week	27.40 (31.55)e	5.60 (2.23)d
6	Six week	21.26 (27.40)f	4.80 (2.18)d
7	Control (29 ^{o}C-32 ^{0}C and RH 57-70%)	96.00 (78.49)a	9.00 (2.99)a
	SEm	0.60	0.06
	CD at 5%	1.76	0.18
	C.V. (%)	2.65	5.62

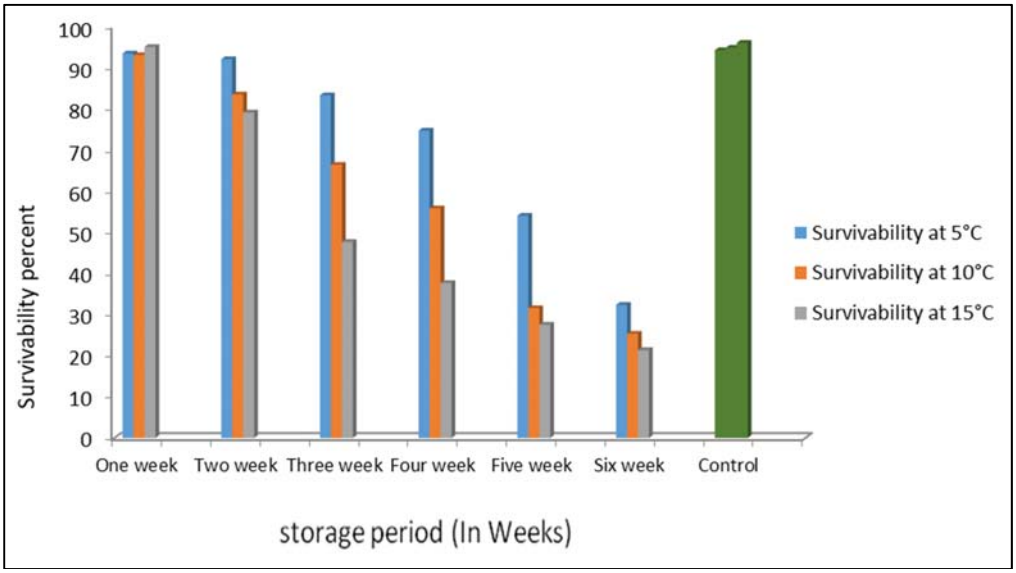

Fig 1: Effect of different storage temperature and storage periods on percentage of survivability of *T. chilonis*

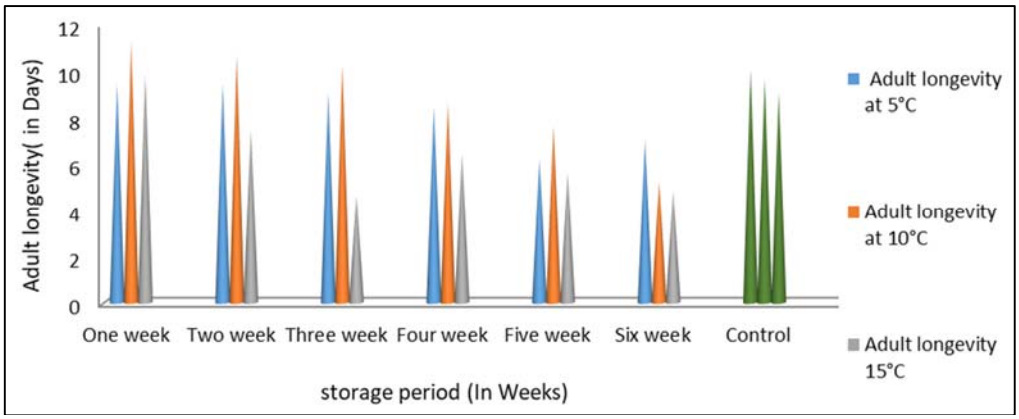

Fig 2: Effect of different storage temperature and storage periods on adult longevity of *T. chilonis*

Mehendale (2009) [8] found similar result of cold storage under refrigerated conditions at 15 ^{o}C temperature for varying periods on four days after parasitisation of trichocards by *T. chilonis*. He reported that per cent adult emergence was quite high at 5 days storage (89.00%) followed by 10 days (86.00%), 15 days (79.31%), 20 days (68.47%), 25 days (63.97%) and 30 days (63.80%). Thereafter, the adult emergence declined drastically. He further concluded that parasitoids trichocards could be slobbered effectively up to 30 days at 15 ^{o}C.

Adult emergence time is vital to realize the time needed for adult coming out after removing pupae from cold storage, particularly in order to synchronize adult emergence of parasitoid and availability of mature larvae of the pest in the field. Cold temperature prevented the embryonic growth of *Trichogramma nerudai* pupae (Tezze and Botto, 2004) [11].

Earlier work of Khosa and Brar (2000) [4] on storage study with *T. chilonis* strain collected from different places revealed that at 8 to 10 ^{0}C the mean per cent adult emergence was 96.2, 88.5, 85.1, 81.2, 73.2, 49.1, 33.6, 14.5 and 1.7 for a period of 1, 7, 15, 22, 29, 36, 43, 50 and 57 days, respectively. Similarly, Gupta and Bhardwaj (2002) [3] also reported that storage of *Trichogrammatoidea bactrae* Nagaraja at 15 ^{0}C for 10 days after 5 days of parasitisation was appropriate. However, Vishla *et al*. (2008) [12] reported that the parasitized eggs of *C. cephalonica* stored at 10 °C showed decreasing

trend of parasitoid emergence with the increase in storage period. Pathak *et al*. (2010) [9] studied the suitability of temperature for the storage of *T. chilonis*. They reported that among the treatment combinations, best treatment proved to be storage of cards at 15 °C for 30 days after 5 days of parasitisation, when highest parasitoid emergence was observed (72.4%).

Bradley *et al*. (2004) [1] investigated the effect of storage temperature (4 °C, 8 °C, and 10 °C) and time (1–8 wk) for *T. carverae*. They showed that lower temperatures (<10 °C) and 3 weeks storage had a negative impact on emergence and longevity.

Conclusion

T. chilonis at 5 ºC and 95 to 97% RH, it can be stored for five weeks with more than 50% survivability. Similarly at 10 ºC and 70 to 75% RH, it can be stored for four weeks with more than 50% survivability. At 15 ºC and 85 to 89% RH, more than fifty percent survivability recorded for two weeks of storage. Storage upto fifty percent survivability was only found economical.

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