

**Effect of cold storage on egg hatching in two reduviid predators *Rhynocoris marginatus* FAB. and *R. fuscipes* FAB. (Hemiptera Reduviidae)**

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**Summary**

The effect of cold storage on egg hatching in *Rhynocoris marginatus* FAB. and *R. fuscipes* FAB. (Hemiptera : Reduviidae) was determined at two different temperatures, i.e., 14 and 20°C, for different periods of storage namely 1, 2, 4, 8 and 12 days. For both reduviids, no hatching was observed at 14°C. Eggs of *R. fuscipes* were not suitable for storage at 20°C either, but *R. marginatus* eggs could be stored for upto 4 days without any nymphal mortality and minimum abnormalities in nymphs. However, storage for 8 days or more affected *R. marginatus* egg hatching and nymphal survival considerably. In a control 99% hatching was observed for *R. fuscipes* whereas it was 90% for *R. marginatus*. Cold storage at 20°C extended the incubation period in both reduviids.

**Keywords :** *Rhynocoris marginatus*, *R. fuscipes*, biocontrol agent, incubation, abnormality.

**Introduction**

An adequate storage of the natural enemies of pests is essential to face problems related to production, planning and the unpredictability of demand. Cold storage can be a useful technique to ensure the availability of beneficial insects for further research or field release without maintaining a continuous rearing. Furthermore, tolerance to cold may be considered as a desirable attribute for shipment procedures (VAN LENTEREN & WOETS, 1988). Information about the effect of cold storage is available for eggs of a pentatomid predator, *Podisus maculiventris* SAY (GORYSHIN & TUGANOVA, 1989); pupae of a parasite *Trichogrammatoidea armigera* NAGARAJA (BAITHA & RAM, 1997); eggs and adults of *Podisus maculiventris* Say and *P. sagitta* FAB. (DE CLERCQ &

DEGHEELE, 1992, 1993) and eggs of *Chrysoperla carnea* STEPHENS. (GEETHA & SWAMIAPPAN, 1996). The reduviids *Rhynocoris marginatus* FAB. and *R. fuscipes* FAB. are important polyphagous predators with a wide distribution throughout India (IMMS, 1965; NAYAR *et al.*, 1976; SINGH, 1985; PAWAR *et al.*, 1986; AMBROSE, 1995; BABU *et al.*, 1995 and SAHAYARAJ, 1995). They are predominantly found in the agricultural ecosystems. Previous papers have presented information on the laboratory rearing and biology of these predatory reduviids (KUMARASWAMI, 1991; AMBROSE & KUMARASWAMI, 1993; SAHAYARAJ, 1995). Determination of embryonic developmental rate of *R. marginatus* at different temperature regimes is a pre-requisite for the preparation of a programming schedule which effectively incorporates storage of *R. marginatus* eggs and prediction of its emergence in the field. In the present study, the effect of cold storage on the hatching of eggs of *R. marginatus* and *R. fuscipes* was evaluated.

### Materials and Methods

#### Insects

A laboratory colony of *R. marginatus* and *R. fuscipes* was initiated with nymphs and adults obtained from groundnut fields and other agricultural ecosystems in and around the Trichy district, Tamil Nadu, India, 8.IV.1998. These predatory bugs were maintained in the laboratory (temperature  $32.5 \pm 0.5^\circ\text{C}$ , r.h. 85% and photoperiod 12±1hrs) on third and fourth instar larvae of the tobacco armyworm, *Spodoptera litura* Fab. (Lepidoptera : Noctuidae) in transparent plastic vials (650 ml volume).

#### Storage of eggs

First batches of newly laid eggs of *R. marginatus* and *R. fuscipes* were collected from paper towelling of stock rearing containers and were placed on filter paper in small plastic vials (30 ml volume) with a perforated lid. The cold treatment was given to the eggs of both predators for different periods viz. 1, 2, 4, 8 and 12 days. Thirty eggs were subjected to each treatment and replicated six times. The vials were kept in the refrigerator at two different temperatures, i.e. at 14 and  $20^\circ\text{C}$ . Control eggs were maintained by keeping eggs at room temperature ( $32 \pm 1^\circ\text{C}$ ). At the end of the treatment periods, vials from each treatment were taken out of the refrigerator and kept at room temperature until hatching. The vials were observed daily and the number of eggs hatched was recorded. Incubation period and percentage egg hatching were calculated. Nymphal mortality up to 4 days after hatching, number of abnormal nymphs, and nature of abnormality in the nymphs were also recorded.

#### Statistical Analysis

For comparing the effect of storage on the incubation and hatching, an ANOVA was conducted using differences between the number of nymphs hatched from the treatment and control. Duncan's Multiple Range Test (DMRT) was used to separate treatment means (DANIEL, 1987).

## Result

No hatching was observed either predator when the eggs were placed at 14°C.

*R. marginatus*

The results presented in table 1 clearly indicated that *R. marginatus* eggs withstand up to 12 days of storage at 20°C, but their hatching was affected when duration of storage period increased (df=5,17,  $p<0.05$ ,  $F=12.89$ ). The hatching rates of eggs for 1, 2, 4, 8 and 12 days 89, etc and 59.33%, respectively. The hatching was similar for 1 to 4 days period of storage. Nymphal mortality was found from the fourth day of storage. A nymphal mortality occurred within 4 days after hatching. Abnormal nymphs were found in storage of 8 and 12 days. The abnormalities observed were curved legs and thorax as well as fused antennae. Hatching and incubation period were significantly different in the controls. In tested eggs, the incubation period increased as the duration of cold storage increased (Table 1) (df=5,17,  $p<0.05$ ,  $F=653.65$ ).

Table 1. Values ( $\bar{x}\pm SE$ ) for incubation period (in days), hatching (%), nymphal mortality (%), and abnormal nymphs (%) produced at cold storage (20°C) of *R. marginatus* and *R. fuscipes* eggs for different storage periods.

Parameters	Species	Control	Storage period (in days)*				
			1	2	4	8	12
Incubation period	<i>R. marginatus</i>	8.5±0.032 <sup>a</sup>	10.3±0.04 <sup>b</sup>	11.7±0.042 <sup>bc</sup>	12.7±0.042 <sup>cd</sup>	18.0±0 <sup>bc</sup>	19.7±0.042 <sup>abcd</sup>
	<i>R. fuscipes</i>	5.0±0	7.0±0	-	-	-	-
Hatching	<i>R. marginatus</i>	90.4±0.073 <sup>a</sup>	88.9±0.07 <sup>ac</sup>	88.6±0.01 <sup>ab</sup>	88.4±0.08 <sup>ac</sup>	87.8±0.054 <sup>abcd</sup>	59.3±0.29 <sup>abcde</sup>
	<i>R. fuscipes</i>	98.9±0.08	45.0±0.03	- 0%	- 0%	- 0%	- 0%
Nymphal mortality	<i>R. marginatus</i>	-	-	-	3.2±0.071	26.0±0.07	41.6±0.384
	<i>R. fuscipes</i>	-	-	-	-	-	-
Number of abnormal nymphs	<i>R. marginatus</i>	-	-	-	-	3.8±0.104	26.3±0.463
	<i>R. fuscipes</i>	-	-	-	-	-	-

(-) No results observed.

\* Mean value followed by different letters within a row are significantly different ( $p<0.05$ ) using Duncan's Multiple Range Test (DMRT).

*R. fuscipes*

When stored at 20°C for a day, 45% eggs hatched, which was nearly 45% of those hatched the in control (99% per cent). The incubation period was also less (5 days) in control than in experimental set (7 days). Abnormalities were not found among the hatched nymphs.

### Discussion

Low temperature has a negative effect on the incubation period of the eggs of both studied predators. Similarly, reduction in the incubation period at higher temperature was observed in another reduviid predator, *Allaeocranum biannulipes* (MONTR et SIGN) (TAWFIK & AWADALLAH, 1983) and also in a hymenopteran parasitoid, *Tetrastichus howardi* (OLLIFF) (KISHORE *et al.*, 1994). Furthermore, the incubation of reduviid eggs requires a certain amount of moisture, prolonged dryness curtails the egg development (VENNISON & AMBROSE, 1990). *R. marginatus* eggs were observed to have tolerance to cold conditions in comparison with those of *R. fuscipes*, and this was clearly understood through hatching of these predators. The rate of hatching was observed in *R. marginatus* eggs for all the tested cold storage periods. BAKTHAVATSALAM *et al.* (1995) noticed that the storage of eggs of *Chrysoperla carnea* STEPHENS (Neuroptera : Chrysopidae) at 10°C for two weeks did not reduce the hatching. In *R. fuscipes*, hatching was not observed beyond 1 day of cold storage. The hatching percentages of *R. marginatus* eggs at 1, 2 and 4 days of storage were similar and this was only 2% lesser than that of the control. However, the prolonged storage of eggs strongly reduced the hatching and viability of nymphs. From this observation it was clear that prolongation of the period of chilling leads to the death of the embryos and non-hatching of the eggs. Furthermore, the gradual decrease in hatching might be due to adverse effects of the cold on developing embryo. A similar kind of susceptibility was observed in the coccinellids *Coccinella septempunctata* (SETHI & ATWAL, 1964) and *C. repanda* Thumb. (SAHARIA, 1981), the reduviid *Pristhesancus plagipennis* WALKER (JAMES, 1992), the pentatomid *Podisus sagitta* FAB. (DE CLERCQ & DEGHEELE, 1993) and in the neuropteran predator *Chrysoperla carnea* STEPHENS (BAKTHAVATSALAM *et al.*, 1995). Furthermore, the age of the egg as well as the period of refrigeration are the two major factors governing hatching (SINGH, 1969). In this study, abnormal nymphs were produced in *R. marginatus* when eggs were stored for 8 days and beyond that period. This indicated that the development of the embryo was not completely stopped during the storage period, but rather slowed down. Moreover, 4 days storage is the optimal for most of the metabolic processes. When this storage period increases, either it may kill the developing embryo or cause many abnormalities in the body. All the abnormal nymphs died within 4 days after hatching. Immature development was observed in a coccinellid predator at 11.7°C and their survival was only of 42% (NARANJO *et al.*, 1990). Adult emergence was not affected up to 6 days of storage of the pupae of a parasitoid, *Trichogrammatoidea armigera* Nagaraja (BAITHA & RAM, 1997). KUZNETZOVA (1970), MORRISON (1985) and OSMAN & SELMAN (1993) reported that the eggs of the neuropteran predator, *Chrysoperla carnea* STEPHENS can be stored up to 21 days at 8-10°C. Similar predator eggs can be stored for more days (20-35 days) but at a lower temperature (5°C) (ADASHKEVICH *et al.*, 1972). The possibility of storing reduviid eggs in the refrigerator for a long period and the subsequent use of the insects as a biocontrol agent would be a great benefit to

the biological control programmes. It can be feasible if this predator is used when mass production is required.

From this study it is concluded that *R. fuscipes* eggs are not suitable for at 14 and 20°C cold storage and *R. marginatus* eggs can be stored for 4 days with no nymphal mortality and minimum abnormalities in nymphs at 20°C. Further studies on the effects of cold storage of eggs of these and other reduviids on biology, biocontrol potential, predatory behaviour, and physical fitness are necessary.

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