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Nesolynx thymus (Girault) as an Effective Biocontrol Agent of Uzi fly, *Exorista bombycis* (Louis)

Jasmeena Qadir[1*], Tajamul Islam[2], Somagaini Pavankumar[3], Ravi Kant[4] and Shalini Aryan[5]
[1]JRF, Division of Silkworm Crop Improvement,
College of Temperate Sericulture, Mirgund, SKUAST-Kashmir, India.
[2]Lecturer, Government Degree College Sopore, Jammu and Kashmir, India.
[3]State Department of Sericulture, Government of Andhra Pradesh, Lakkireddypalli Mandal, Andhra-Pradesh, India.
[4]Division of Sericulture,
Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, Chatha, India.
[5]Department of Agriculture, Mata Gujri College, Fatehgarh Sahib, Punjab, India.

(Corresponding author: Jasmeena Qadir)*
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ABSTRACT: The uzi fly, *Exorista bombycis* (Louis) is an endoparasitoid causing significant crop losses upto the tune of 20% in the rainy season followed by 11-15% loss in winter and 1.0 – 3.0% loss in summer seasons. There is high demand of minimizing the use of chemicals for pest control in sericulture as a little toxicity may deteriorate healthy silkworm rearing and ultimately raw silk production. *Nesolynx thymus* (Girault) is a serious endo-larval parasitoid and inundative pupal parasitoid of the order Hymenoptera: Eulophidae. It possess remarkable biological characteristics of biological control such as short life span, potential for high parasitization, high host searching ability, docile for mass production with good adaptability under laboratory as well as field conditions. The eco-friendly method of using N. *thymus* for control of Uzi fly promotes sustainable solution and balance in the environment. N. thymus is the pupal endoparsitoid of almost all the species of Uzi fly including *Blepheripa zebina, Exorista phillipinensis* as well. The industrial production of *N. thymus* parasitoid is completed in two processes which involves production of housefly (*Musca domestica*) pupae and production of N. thymus pupae on housefly pupae. Many behavioral responses culminate while oviposition of gravid female of *N. thymus* to parasitizes (host pupae), *Exorista bombycis*. The host factors *viz.*, pupae size, age and period of exposure to parasitoids influence the biology of parasitoids. The optimization of host factors influencing the parasitisation by parasitoid may enhance the efficacy of *N. thymus* and its mass multiplication and production in control of Uzi fly.

Keywords: Eco-friendly, *Exorista bombycis, Nesolynx thymus, Musca domestica*, parasitoid, uzi fly.

INTRODUCTION

India is the second largest producer of silk and produces all the five known silks (Padaki *et al*., 2015). Silkworm (*Bombyx mori* Linn.) is a domesticated insect and reared in mass numbers for the large scale production of mulberry silk (Giora *et al*., 2022). Many factors are responsible for severe or complete crop losses anually (Rahmathulla, 2012). Among them, the attack of insect and non-insect pests is of prime concern (Haripriya & Mamatha 2023). The mulberry silkworm is affected by a number of insect pests like Uzi fly (*Exorista bombycis* Louis), earwig, dermestes beetle and ants (Sakthival *et al.,* 2012; Belgumpe and Jadhav 2017). Recently it is learnt that, Uzi fly has also been introduced in various parts of Maharashtra (Jadhav and Sathe, 2016). The uzi fly, *Exorista bombycis* (Louis) is a larval endoparasitoid. It belongs to order diptera and family tachnidae (Singh *et al*., 2024). Many species of uzi fly are found to attack silkworms which include Japanese uzi fly (*Crossocosmia sericariae* Rondani), Hime uzi fly (*Ctenophorocera pavida* Meigen), Tasar uzi fly (*Blepharipa zebina* Walker) and the Indian uzi fly (*Exorista bombycis* Louis) (Sowmya and Ranjitha 2021). Among the pests, Uzi fly (Exorista bombycis) is the most serious pest of silkworm in Karnataka, Andhra Pradesh, Tamil Nadu and West Bengal. It has been reported that uzi fly cause silkworm crop loss upto the tune of 10 to 40 percent (Sakthivel *et al.,* 2012). It causes crop loss of more than 20% in the rainy season followed by 11-15% loss in winter and 1.0 – 3.0% loss in summer seasons (Bari *et al*., 2023). Uzi fly lays about 250-300 eggs on a single host of silkworm and hatches with an average hatching of 30-40 and within three days eggs hatch (Reddy *et al.*, 2015). It prefers 4th and 5th instar silkworm larva to lay eggs into their intersegmental regions. Upon hatching, maggots bore into the silkworm body. It feeds on silkworm's fluid and leaves a black scar on the insect body. They get nourishment until silkworm's pupal stage and the mature maggots emerge out of the cocoon shell and render the cocoons unreelable (Barsagade, 2017). If 4th stage silkworms are infested with Uzi fly, silkworms do not spin cocoons but when 5th instar silkworms are infested with uzi fly, silkworms spin cocoons but those cocoons are not suitable for silk reeling. The Uzi maggots pupate in the crevices and holes of rearing room (Choudhury *et al.*, 2014).

The use of natural enemy against the pest organism to reduce their densities is the effective method of pest control (DeBach *et al.*, 1971). Natural enemies include parasitoids, predators, entomopathogenic nematodes, pathogens, competing microorganisms without hyperparasites of plant pathogens, herbivores feeding on weeds and weed seeds, competitors for resources and organisms producing toxins, termed antibiosis or allelopathy as they directly or indirectly acts as the pest control with its management (Khandagale *et al.*, 2023). There are a number of hymenopteran parasitoids that can be used in nature to reduce the number of Uzi flies (Siddaiah and Danagoudra 2019; Baruah and Kalita 2020). Several parasitoids have been tested for parasitization against uzi fly pupae, including *Nesolynx thymus*, *Trichopria* sp., *Exoristobia philippiensis, Dirhinus* sp., *Brachymeri alugubris, Spalangi aendius, Pachycrepoideus veerannai*, and *Spilomicrus karnatakensis* (Nirmala and Veeranna 1998; Singh and Maheshwari 2002; Singh and Saratchandra 2003) (Table 1). In sericulture, chemical means of pest control may pose serious threat to silkworms, rearers and ecosystem as well (Singh and Saratchandra 2002). There is also high demand of minimizing the use of chemicals for pest control in sericulture as a little toxicity of it may deteriorate healthy silkworm rearing and ultimately raw silk production (Karthik and Rathinamoorthy 2017; Altman and Farrell 2022). The eco-friendly approach not only ensures a sustainable solution but also promotes the overall balance of the ecosystem.

Nesolynx thymus (Girault) is a serious endo-larval parasitoid and inundative pupal parasitoid of the Hymenoptera: Eulophidae. It possess remarkable biological characteristics of biological control such as short life span, potential for high parasitization, high host searching ability, docile for mass production with good adaptability under laboratory as well as field conditions. *N. thymus* has been regarded as potential biocontrol agent of *E. bombycis*. *N. thymus* have been found to control the uzi fly ((*Exorista philippinensis* and *Blepheripa zebina*) infesting muga silkworm (*Antheraea assamensis* Helfer) in sericulture (Choudhury *et al.*, 2014). It is considered as one of the important component of Integrated Pest Management (IPM) in sericulture by Central Sericultural Research and Training Institute (CSR & TI), Mysore (Hasan *et al.*, 2009; Gahukar, 2014). It is released on large scale for control the uzi fly infestation. Throughout this article, the significance of biological control agent, *Nesolynx thymus* and its mass production for the control of uzi fly is discussed.

Mass production of *N. thymus*. *N. thymus* a hymenopteran parasitoid was first discovered and utilized to control the Uzi fly, *E. bombycis* during 1985 (Aruna and Manjunath, 2010). Since then, this parasitoid has been used in India for respect to house fly *Musca domistica pupae*. It is observed that on E. bombycis maximum parasitization of *N. thymus* was recorded on first and second day old pupae (Belgumpe and Jadhav 2017). The industrial production of *N. thymus* parasitoid involves two different processes.

(a) Production of housefly pupae. The housefly pupae collected in cage are kept for adult emergence at at 25±2°C temperature, 75±5% RH duration (Kumar *et al.*, 2016). After 4-5 days, 80 % of adult flies will emerge and feeds on food containing sugar and milk powder in the ratio of 1:1. The cotton balls soaked in water is kept in a cage as reserved source of water. Temperature is increased to 28±2°C and 90±5% RH before introduction of the oviposition containers in 4-5 days old colony (Ortiz *et al.*, 2016). In oviposition containers, food container are kept which constitute mixture of 5-10g milk powder and 5g yeast powder to 100g of used larval medium which is wrapped in black moistened cloth. After 5-6 hours, 20,000 eggs are removed from oviposition containers using brush (Wilkes *et al.*, 1948; Pastor *et al.*, 2014).

The freshly prepared larval diet containing mixture of 1.2kg wheat bran and 15g dry yeast and water is added to final weight of 5.5 kg and it is kept on a surface. House-fly eggs, 20,000-25,000 are kept on surface of larval diet and covered with cloth. Eggs hatch within 24 hours and maggots start feeding on the diet (Kumar *et al.*, 2016). The maggots' colour changes into creamy white denoting initiation of pre-pupal stage. Transfer the pre-pupa gently on wire mesh (10 mesh size) on a perforated plastic tray (60cm × 30cm). The perforated plastic tray is kept on non-perforated plastic tray (60cm × 30cm). All the pre-pupa moves down through wire mesh and fall into the non-perforated tray kept below. Saw dust is spread in a thin layer to facilitate pupation. The color of pre-pupa turns into brownish color at pupal stage (Pastor *et al.*, 2014; Kumar *et al.*, 2016). The quantity of pupa by separating on wire mesh is determined by volume (one ml will contain about 30-35 pupae)

(b) Production of *N. thymus* on house fly pupae. The fresh house fly pupa (500ml; about 20,000) are spread in a single layer in 60 cm × 30 cm plastic tray. Smear 50% honey solution on both sides of a plastic strip (20 cm × 2.5 cm) and keep 2-3 such strips in each tray as food for parasitoid adults. Release 4-5 thousand, one-day-old *N. thymus* adults in the tray in the ratio of 1:4 (1 female parasite: 4 house fly pupae). About 350ml parasitized housefly pupae will serve the purpose. Cover the tray with thick white cloth and tightly fix it with elastic to prevent the escape of *N. thymus* (Kumar *et al.,* 2016) After 4 days, collect the parasitized pupa from the tray and keep in adult cage for emergence of house fly, if any Remove the empty shells by winnowing and collect parasitized pupae for supply to stake holders 7-10 days old parasitized pupae may be mixed in equal quantities so as to achieve emergence continuously for a week. After mixing, measure 50ml of house fly pupa parasitized by *N. thymus* and pack in 10cm x 10cm nylon net pouch. From each pouch, about 10,000 adults of the parasitoid emerge. These pouches can be easily transported to stakeholders by courier. 10-14 days old parasitized pupae can be refrigerated at 10±2°C for a maximum of one week in case of delay in supply, to prevent the quality damage to parasitoids (Senthoorraja *et al.,* 2020). *N. thymus* completes its life cycle in in 10-12 days of uzi fly host. Adult longevity is completed within 6-8 days. Adults mate within 8-10 hours of emergence (Belgumpe and Jadhav 2017). Males and females mate more than once. On parasitization, the larvae of *N. thymus* feed within the host pupae, develop rapidly and attain full growth (Kumar *et al.,* 1986).

Ovipositional behaviour of *Nesolynx thymus* on *Exorista bombycis* pupa. Behavioural manipulation approaches represent innovative and sustainable solutions for insect pest management in agriculture and ecosystem protection. These strategies leverage the intricate behaviours of insects, harnessing their natural instincts for the benefit of pest control (Chandana and Nadagouda 2023). Many behavioral responses culminate while oviposition of gravid female of *N. thymus* to parasitizes (host pupae), *Exorista bombycis* (Siddaiah and Danagoudra 2019). It involves series of behavioral responses before oviposition which includes host recognition, drumming, tapping and drilling. When a parasitoid (*N. thymus*) female is provided to host pupae, it walks in the oviposition container for host recognition. On recognizing the host, the parasitoid female mounted on the host, inspects the host by repeated contact using antennae. This process is known as drumming. After this, it is followed by tapping of the area by the abdominal tip that bents vertically downward using its sensory hairs. The parasitoid moves towards drummed area and coincides it with its abdominal tip. Drumming activity is little low frequent than tapping activity. When the process of drumming is completed, the parasitoid withdrew its ovipositor perpendicularly to the tapped area which is previously identified. At this time, the female parasitoid takes a firm grip on the host surface and became motionless in order to give sufficient reinforcement to the ovipositor (to hold it straight) for drilling .The process of drilling begins by clockwise and anticlockwise movement of the ovipositor and facilitates drilling. After drilling, entire ovipositor is inserted into the host to initiate oviposition on the pupa. At this time, both host and parasitoid remains immobile. After oviposition process, grooming process which involves cleaning of antennae and dorsal and lateral thoracic regions. The parasitoid feeds on the host body fluid (haemolymph) that oozes out from the feeding tube created in the hole drilled by the parasitoid on the host. Sometimes irregular behavioral response is observed which leads to host rejection (Siddaiah and Danagoudra 2019).

Host and parasitoid interactions. The host factors *viz.,* pupae size, age and period of exposure to parasitoids influence the biology of parasitoids. The parasitoid size is important quality trait influenced by host size in biological control of a pest (Aruna and Manjunth 2009). The mean parasitism by *N. thymus* is depicted by its host size. It is significantly higher (74 percent) in the large sized host pupae having a volume of 263.89 mm^3 of varying size with the mated females of N. thymus. Host age is the most important character that influences the mating behavior, reproductive traits and magnitude of parasitism in parastoids (Aruna and Manjunath 2010). Maximum progeny production in *N. thymus* was revealed on Two to four-day old puparia of *E. sorbillans* (Hasan *et al*., 2009). The nutritional quality of hosts decreases with age. The parasitoids show non-preference response to aged hosts. It was revealed that *Diodromus collaris* a pupal parasitoid of *Plutella xylostella* did not prefer old host pupae because of reduced nutritional quality which leads to cessation in development of the parasitoid. Hence the choice of host selection on the basis of age enhances survival advantage of hosts (Wang and Liu 2002). The host pupae of medium age classes were more preferred and more suitable for development (Pfannenstiel *et al.,* 1996). Senthoorraja *et al.* (2020) revealed higher level of parasitism (65%) by *N. thymus* to 24 - 48 hours old host pupae. The parasitism rate reveals significant decline with an increase in host age after 72 hours (Senthoorraja *et al.,* 2020).The period of exposure to parameters directly influences the rate of parasitism. Senthoorraja *et al.* (2020) revealed higher level of parasitism and with higher female progeny after 48 hours of exposure period. The reduction in parasitisation may be due to occurrence of superparasitism in exposure period beyond 48 hours. Higher level of superparasitism was reported when *T. howardi* was used against *Chilo partellus* (Baitha *et al.,* 2004). Therefore, it indicates that 24-48 h is a suitable exposure period to adopt during mass production of parasitoids. The optimization of host factors influencing the parasitisation by parasitoid may enhance the efficacy of *N. thymus* and its mass multification and production in control of Uzi fly.

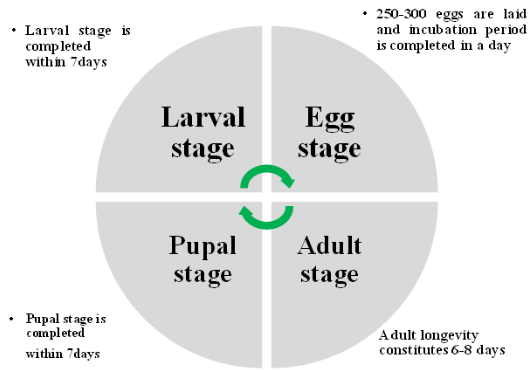
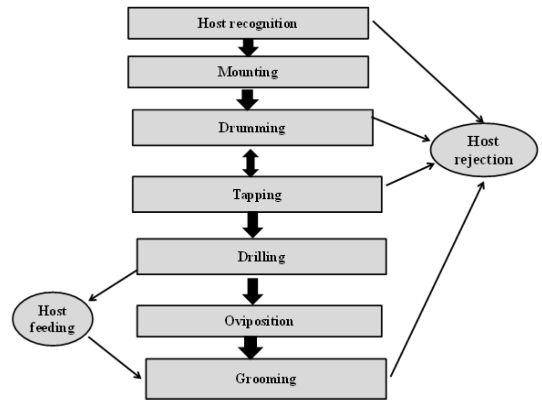


Fig. 1. Life cycle stages of *N. thymus.*



Scheme 1: Ovipositional behaviour of *Nesolynx thymus* on *Exorista bombycis* pupa (Siddaiah & Danagoudra 2019).

Table 1: Some important parasitoids against uzi fly pupae.

Sr. No.	Scientific name	Order	Family	Nature	Status
1.	*Nesolynx thymus*	Hymenoptera	Eulophidae	Ecto-pupal parasitoid	Gregarious
2.	*Trichopria* sp.	Hymenoptera	Diapriidae	Endo-larval pupal parasitoid	Gregarious
3.	*Exoristobia philippiensis*	Hymenoptera	Encyrtidae	Endo-larval pupal parasitoid	Gregarious
4.	*Dirhinus* sp	Hymenoptera	Chalcididae	Ecto-pupal parasitoid	Solitary
5.	*Brachymeri alugubris*	Hymenoptera	Chalcididae	Ecto-pupal parasitoid	Solitary
6.	*Spalangi aendius*	Hymenoptera	Ptermalidae	Endo-larval pupal parasitoid	Solitary
7.	*Pachycrepoideus veerannai*	Hymenoptera	Pteromalidae	Endo-larval pupal parasitoid	Gregarious
8.	*Spilomicrus karnatakensis*	Hymenoptera	Diapriidae	Ecto-pupal parasitoid	Solitary

Table 2: Parasitization rate of *N. thymus* to the host age of Uzi fly (Belgumpe & Jadhav 2017).

Sr. No.	Age of E. bombycis (day)	Parasitism by *N. thymus* (%)
1.	0	80
2.	1	75
3.	2	60
4.	3	45
5.	4	30
6.	5	20
7.	6	15
8.	7	8

CONCLUSIONS

The mass production of *Nesolynx thymus* requires little space and investment. The biological means of control are always eco-friendly, long lasting and solve the problem permanently. Hence, being cost-effective method with possession of no toxicity to silkworm and sericulture industry, it is highly recommended as effective means of pest control in sericulture for checking the Uzi infestation.

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