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Tyrophagus putrescentiae (Schrank) (Astigmata: Acaridae) as natural enemy for wood boring pest, *Psiloptera fastuosa* F. (Coleoptera: Buprestidae) in tropical tasar

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ABSTRACT: In tasar silkworm culture the stem-boring jewel beetle *Psiloptera fastuosa* Fabr. (Buprestidae: Coleoptera) is considered as a major pest of tasar plant (*Terminalia arjuna*, Combretaceae) cultivation. The grubs of *P. fastuosa* often damage the *Arjuna* stem by causing dieback. *Tyrophagus putrescentiae* Schrank (Acari: Acaridae) infested buprestid eggs up to 15% and caused egg mortality up to 9%. The mite predation on the buprestid beetle is reported for the first time. The mite seeps the newly-laid egg-fluids causing the egg mortality suggesting that tasar plant stem-boring pest (*P. fastuosa*) can be partially controlled by the mite as a natural enemy.

KEY WORDS: Tasar culture, mite-predation, buprestid eggs, biological control

INTRODUCTION

Tropical tasar silkworm culture is one of the major agricultural industries in central Indian provinces providing business and employment. Among non-mulberry silks, tasar silk is considered the second largest cash-crop from the tropics. *Antheraea mylitta* (Lepidoptera: Saturniidae) produces of this natural-protein-rich silk (Kundu *et al*., 2012). The tasar silkworms usually consume leaves from two plant species from Combretaceae family viz., *Terminalia arjuna* (Arjuna) and *Terminalia tomentosa* (Asan). Therefore, *Terminalia* species are considered as primary food plants for tasar silkworms (Ojha *et al*., 2009; Manabendra and Minu, 2013). Nearly 10-15 million hectares of land are being used for tasar plant cultivation (*T. tomentosa*) from central Indian provinces, but due to systematic denudation and destruction of forest, large areas of naturally-grown Tasar plantation (*T. tomentosa*) have been lost in forest fringes during the last couple of years (Sinha and Srivastava, 2002). To compensate the loss of naturally-grown *T. tomentosa*, large scale plantation of fast-growing *T. arjuna* plants were done in the affected zones. But extensive monoculture of such raised plantations has also made these plants vulnerable to various pest attacks (Singh and Saratchandra, 2002 and Singh *et al*., 2004) and among them buprestid insects are predominant.

The newly-grown Arjuna plants (*T. arjuna*) are mainly infested by a metallic green folivorous stem-boring insect, *Psiloptera fastuosa* (Fabricius,1775)

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(Coleoptera: Buprestidae) (Singh *et al.,* 1987, 1989; Mandal, 2007). The grubs of *P. fastuosa* often girdle inside the stem of young Arjuna and restrict translocation of plant nutrients and retard plant growth and development. As a result, the infested plants die (Dhar *et al*., 1989; Mandal and Singh, 1990; Tirkey *et al*., 2019). The stem-boring coleopteran is considered as a major pest for primary tasar food plant, *T. arjuna* (Reddy *et al.,* 1996). To control this pest, chemical pesticides though applied in some occasions but due to cryptobiotic nature of *P. fastuosa* grub, no effective measures have yet been successful. Therefore, the scope of biological control of this notorious pest is important. The absence of information regarding biotic check for buprestid beetles and its grubs by natural enemies makes it more challenging to monitor and control in changed habitats. Published literature also lacks in information on its natural enemies except for a few instances where the importance of buprestid natural enemies has only been reported so far (Mandal, 2007). Therefore, we conducted our investigation aiming to find out existence of any biological control agent for *P. fastuosa* and second, we paid attention to examine how egg mortality of *P. fastuosa* varied upon the parasitic/predatory intervention of that controlling agent.

MATERIALS AND METHODS

Field Study

Buprestid egg-masses were randomly collected from young Arjuna plants (7±2 years, mean ± SD) from four tasar plantation sites of Pali, Chhattisgarh, India (22.37° N, 82.32° E) during the breeding season of the beetles (September to October) for a period of three consecutive years (2016 to 2019). Study sites were maintained and managed by Central Tasar Research & Training Institute (CTRTI), Ranchi, Government of India. Average aerial distance between two sites was 5±2 km and each site was surrounded by mixed deciduous degraded forest vegetation. During field collection environmental temperature (25±3°C) and relative humidity (70±5% RH) were moderate without any incidence of rainfall. Beetle infested plants were spotted by observing yellowish scares on Arjuna shoot. The spotted plants were ribbon-marked for the year and selected for egg-mass collection. Throughout the survey, 1025 egg-masses were collected by digging the bark of the plant (0.2-1.2 cm) for three consecutive years (year I; n=278; year II; n=351; year III; n=396). Excavated egg-masses were transported to the laboratory and numbers of eggs per egg-mass were counted accordingly.

Microscopic study

Out of 1025 egg masses, a portion (n=205) of egg-mass was transferred to FAA (formaldehyde-acetic acid-alcohol) medium (Talbot and White, 2013) for taxonomic identification and the remaining portion (n=820) was kept in moist aerated test tubes so that the eggs were remained alive until further experiment. Mouths of test tubes were tightened with distilled-water-soaked-cotton-balls to keep it humid. Therefore, 205 randomly selected egg-masses were considered for mite identification (year I: 52; year II: 72, and year III: 81) under microscope (Leica, Wild M8). During this process of identification, if any mite was noticed, they were isolated and counted year-wise and thereafter identified taxonomically. The remaining portion of egg-masses (n=820) were separated again into two groups, a) non-infested egg-mass, where egg-masses were free from any mite attack, and b) infested egg-mass, where presence of any mite was recorded. During this separation, some adult alive mites (both males and females) were isolated from infested egg-masses in moistened test tubes.

Egg mortality assay of *P. fastuosa* by mite attack

Non-infested egg-masses were used for egg mortality assay. Non-infested egg-mass was kept in glass Petri-dish (8 mm diameter) and covered by a fine cotton mesh to prevent any contamination. Two groups of experimental sets (Set A and Set B) were prepared from these egg-masses. In Set A, 20 Petri-dishes were prepared and in each of which only one non-infested egg-mass was placed (control). Similarly, in Set B, another 20 replicates were prepared with non-infested egg-masses, but in each of which 3 to 4 adult mites were inoculated (treatment). The number of eggs per egg-mass was counted for each replica. Both the 'control' (n=20) and 'treatment' (n=20) Petri-dishes were kept at BOD incubator (25±3°C and 70±5% RH) for 21 days. During this period, each Petri-dish was observed daily to notice any nymphal emergence. When the eggs hatched out from a replica, either from 'control' or 'treatment', the number of emerged nymphs were counted accordingly. Similar to a few earlier reports we also noticed that mites generally seep the egg-fluids and as a result the egg dies (Moser, 1975; Brust and House, 1988; Canevari *et al*., 2012). After 21 days, total numbers of emerged hatchlings were pooled together. Numbers of non-hatched eggs from both sets (control and treatment) were also counted separately. Egg mortality (%) was calculated based on non-hatched eggs by the formula: number of non-hatched eggs / number of eggs taken for experiment × 100 (Hughes, 1959; Colloff, 1987). Egg mortalities were analysed and compared between 'control' and 'treatment' sets using SPSS software (ver. 25).

RESULTS AND DISCUSSION

During the field investigation buprestid eggs were found infested with mites and it was identified as *Tyrophagus putrescentiae* Schrank (Acari: Acaridae). It showed predation of *P. fastuosa* egg-masses in several occasions. Infestation incidence of *T. putrescentiae* on buprestid eggs has never been reported before. However, *T. putrescentiae* was earlier reported as a common store-grain-product pest (Eaton and Kells, 2011; Freitag and Kells, 2013) and usually attacked coccid eggs (Collins, 2006; 2012).

Buprestid beetles used to lay eggs inside the barks of *Terminalia* plants from late September to late October, but the mite finds its way and attacked buprestid eggs inside the bark. Like a few previous records it was observed that *T. putrescentiae,* the mould mite oviposit its eggs on Buprestid egg masses; usually seeps the newly-laid egg-fluids and as a result the eggs die (Moser, 1975; Brust and House, 1988; Canevari *et al*., 2012). Moreover, the mite passed rest of its life-stages (larva to adult) on the beetle's egg-mass by consuming the eggs maintaining their ovivorous feeding habit (Balazy and Kielczewski, 1965). Among the studied 205 buprestid egg-masses from three successive years, an average of nearly 12% egg-mass (range, 9-15%) was noticed predated by the mite (Table 1). Egg mortality of *P. fastuosa* was recorded 53±14.14% (mean±SD) for non-infested eggs (in control sets), but due to mite-predation egg mortality of *P. fastuosa* significantly increased to 62±15.87% (mean±SD) (ANOVA, $F_{1,\ 38}$ =7.02; p=0.012) (Fig. 1).

Several natural enemy complexes have been reported so far by numerous authors as controlling agents for several buprestid beetles across the world (Carlson and Knight, 1969; Loerch and Cameron, 1983, Bauer *et al*., 2005; Sallé, 2016; Wang *et al*., 2016; Zang *et al*., 2017; Abell *et al*., 2020). Mortality studies of buprestid eggs were mainly caused by parasitoids, however, pathogens and

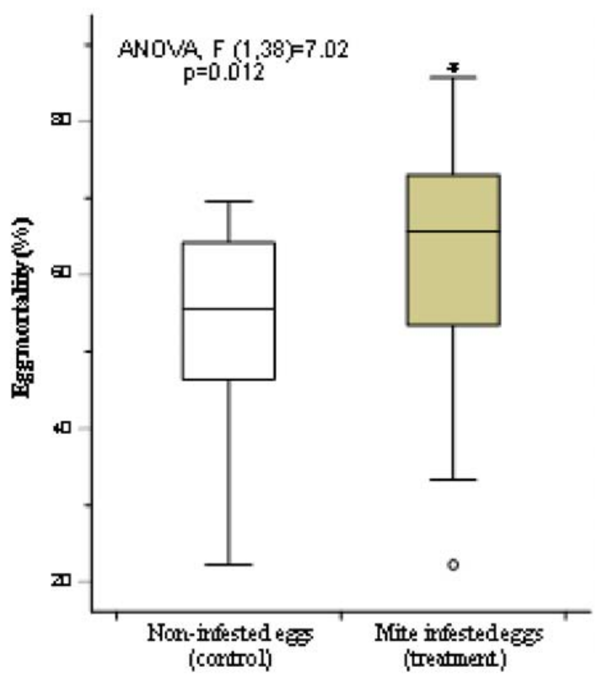

Fig. 1. Egg mortality % of *P. fastuosa* due to mite infestation (treatment) compared to non-infested eggs (control) at laboratory conditions. Each box represents 20 mortality assays (total assay, n=40). * indicates ANOVA (one way) result was statistically significant (p<0.05), transects in interquartile range indicate median value scaled in Y-axis, the circle indicates potential outlier

Table 1. Occurrence frequency of eggs per egg-mass of beetle (*P. fastuosa*) and mite (*T. putrescentiae*) infestation frequency to beetle's eggs

	P. fastuosa		***T. putrescentiae***	
	Egg mass observed/ Collected egg mass	Eggs counted/ Observed egg mass	Mites observed/ Counted eggs	% of eggs infested by mite
Year I	52/278	339/52	51/339	15.04
Year II	72/351	491/72	44/491	8.96
Year III	81/396	634/81	74/634	11.67
Total/Mean ± SD	205/1025	1464/205	169/1464	11.54±3.05

predators were also described in a few occasions (Oliveira *et al*., 2003). Most of the buprestid egg parasitoids were reported from hymenopteran insects from families Encyrtidae, Braconidae and Ichneumonidae. For example, natural enemies of Willow wood-borer, *Agrilus fleischeri* (Coleoptera: Buprestidae) were the parasitic non-stinging wasp, *Oobius* sp. (Hymenoptera: Encyrtinae) (Zang *et al*., 2017), where natural enemies for emerald ash borer, *Agrilus planipennis* were reported 3 species from braconids, 1 species from chalcid and an eupelmid parasitic wasp (Bauer et al, 2005). Another survey describes a complex of natural enemies including eight hymenopteran insects damage buprestid eggs (Zhang *et al*., 2003; Abell *et al*., 2020). Besides these, ichneumonid wasps were also reported on several occasions as potential parasitoids for buprestid beetles. At least 2 ichneumonid species along with 2 braconids and 1 chalcid established parasitoid on the sap-borer, *Trachypteris picta* (Kenis and Hilszczanski 2004). Solians (1974) has described several species of parasitoids (braconid and ichneumonid) for the Oak-borer buprestid, *Coraebus florentinus* larval instars. However, there was no report of any acarine parasite or predator record on buprestid eggs.

In most of the instances egg mortality of buprestid beetles varied approximately from 10 to 50%, though in some instances much lower value was observed. Variation of buprestid egg-mortality was described as an outcome of several reasons like varied geographic occurrence of the beetle, changes of sampling season, variation of forest plantation etc. (Bauer *et al*., 2005; Wang *et al*., 2016; Sallé, 2016; Zang *et al*., 2017; Abell *et al*., 2020). Mite infestations to a few coleopteran beetles, except buprestid, were reported. For example, a large number of *Caloglyphus* mites parasitized on Cranberry white grub, *Phyllophaga anxia* (Scarabaeidae) (Jarvis, 1964); the ovivorous mite, *Tarsonemoides gaebleri* fed spruce bark beetles, *Ips typographus* (Scolytidae) (Balazy and Kielczewski, 1965); the water mite, *Eylais* sp parasitized a variety of aquatic coleopterans beetles like *Dineutus nigrior* (Fairn *et al*., 2008) etc. Mites from several genera were reported as controlling agent for several coleopteran borers like *Sternochetus lapathi* (Curculionidae), *Pseudopityophthorus minutissium* (Scolytidae), and *Aphanisticus cochinchinae seminulum* (Buprestidae) (Berry and Bretz, 1966; Hall *et al*., 2005). The straw itch mite, *Pyemotes tritici* (Acari: Pyemotidae) was discovered parasitizing the gold spotted oak borers, *Agrilus auroguttatus* in the USA, and *Agrilus coxalis* in Mexico (Loghmani *et al*., 2014). Parasitizing mites, *Pyemotes* (Acarina: Pyemotidae) parasitoid on jewel beetle, *Ovalisia festiva*, and maintained a biological relationship (Ruseva *et al*., 2020).

The mite caused significant additional mortality to beetle's eggs up to 9% (mite-infested egg mortality: 62%) than its natural death (53%). Our result agrees with the observation of Canevari *et al*. (2012) where the authors noticed predation of *T. putrescentiae* on tobacco pest larvae, *Lasioderma serricorne* (Fabricius, 1792) (Coleoptera: Anobiidae) varied from 54 to 78%, depending on the larval stage.

Under laboratory trials the mite, *T. putrescentiae* showed a significant predatory activity to the beetle's eggs and therefore, the mite can be treated as a natural enemy for tasar plant stem boring pest, *P. fastuosa*.

Mites belonging to genus *Tyrophagus* have though been reported as parasitizing agent (Brust and House, 1988; Kumar, 1997) or predator for some insect groups such as beetles (Papadopoulou, 2006; Canevari *et al*., 2012), flies (Serpa *et al*., 2004), and bees (Maggi 2011; Texeira *et al*., 2014), but surprisingly no report has yet been available on *Tyrophagus* attack to any buprestid beetle. *Tyrophagus putrescentiae* (Schrank, 1781) is reported for the first time as an egg predator to buprestid beetle, *P. fastuosa*.

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