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PREDATORY EFFICACY OF STORED GRAIN MITE (*TYROPHAGUS PUTRESCENTIAE* (SCHRANK) (ASTIGMATA: ACARIDAE) AGAINST COTTON INSECT PESTS UNDER LABORATORY CONDITIONS

Faiz Ullah Faiz[1], Zahid Mahmood Sarwar[1] Nazeer Ahmed[2], Pravej Alam[3], Abdulrahman A. Almadiy[4], Sadeq K. Alhag[5,6], Mona Mohammed Al Dawsari[3], Ferjeni Zouidi[5], Kamlesh Himmatrao Mali[7]**

1 Department of Entomology, FAST, Bahauddin Zakariya University Multan. 60800, Pakistan. 2 Department of Agriculture, University of Swabi, Anbar-23561, Swabi-Khyber Pakhtunkhwa-Pakistan. drnazeerento@gmail.com (N.A)

3 Department of Biology, College of Science and Humanities; Prince Sattam bin Abdulaziz University, Saudi Arabia.

4 Department of Biology, Faculty of Arts and Sciences, Najran University, 1988 Najran, Saudi Arabia

5 Biology Department, College of Science and Arts, King Khalid University, Muhayl Asser, Saudi Arabia.

6 Biology Department, College of Science, Ibb University, Ibb, Yemen.

7 Zoology department, Comrade Godavari Shamrao Parulekar College of Arts Commerce and Science, Talasari- India.

Correspondence: zmsarwar@bzu.edu.pk

Abstract:

Common pest control approaches include the use of synthetic pesticides and the resistance of many insects to chemical insecticides, coupled with potential environmental health hazards gave birth to a search for biological control as substitutes to synthetic insecticides. There are many organisms in agro-ecosystems that may have a value, negative or positive, to crop production. Cotton crop is damaged by different insect pests like *Phenacoccus solenopsis*, *Dysdercus koenigii*, *Oxycarenus hyalinipennis*, *Amrasca biguttula*, *Bemisia tabaci*, and *Aphis gossypii*. These pests deteriorate the quality and quantity of the crop ultimately reducing the yield. These pests have also conquered the resistance against different insecticides like bifenthrin, deltamethrin, lufenuron, and chlorpyrifos. There is a need to study different control methods which are environment friendly and safer for food like biological control. Stored grain mite *Tyrophagus putrescentiae* (Oder: Astigmata) is a cosmopolitan, polyphagous, and also omnivorous species belonging to the family Acaridae. It has an efficient predatory potential in the IPM program. In the present study, the predatory efficacy of *Tyrophagus Putrescentiae* (Schrank) against some of the sucking insect pests of cotton i.e., *P. solenopsis, D. koenigii,* and *O. hyalinipennis* and the effect of predator density and various time intervals under laboratory conditions was evaluated and determined. The

experiment was designed under a complete randomized block design (CRD) according to factor factorial design. Maximum predation was observed in all stages of red cotton bug followed by the cotton mealy bug and dusky cotton bug. Similarly, a high predation rate was recorded in the adult stage of all insect pests followed by nymphal and egg stages. Mortality (%) increased with the passage of time and maximum mortality was observed after 48 hours in eggs and 72 hours in nymphal and adult stages of all insects in all treatments. It is concluded that *T. putrescentiae* is a good biological control agent sucking insect pests.

Keywords: Red cotton bug; Cotton mealy bug; Dusky cotton bug; predatory potential; predator density

Introduction

Mankind is consistently in race with insect pests for available natural resources and most significantly food products and food crops. Insect pests and vectors of diseases are main intimidations to increased production of agricultural and to the health and well-being of human and his livestock. Food and agriculture department's estimates that food demand will dual by 2050 and to overcome this demand, grains yields especially developing countries will have to increase by 40% and an additional 100-200 million hectare of land may be required (FAO, 2008). Cotton is a most important fiber and oilseed crop in the world. This kharif crop is a major source of foreign exchange incomes. It contributes up to 8.2 percent in agricultural value added and roughly 2 percent in Gross Domestic Product (GDP) (Ehsan et al. 2008).

The factors either climatic or synthetic like irrigation, land preparation, plant protection measures, seed, and qualitative variables like farming experience, age, education and insect pests affect cotton productivity. Among these factors, the most important are the insect pests of the cotton because they reduce seed-cotton yield and hamper fiber quality (Bakhsh et al. 2005). The outbreak of sucking and chewing insect pests is the major problem of the cotton crop and almost 93 species of insect pests have been reported in Pakistan that cause significant loss in cotton crop (Yunus & Yousuf, 1979). They deteriorate the quality of crops and cause 30% to 40% losses (Shah, 2014). After the adoption of Bt cotton in Pakistan, the lepidopteron pests especially bollworms are almost controlled, but the attack of sucking pests increases rapidly. Cotton main sucking pests like Cotton mealy bug, *Phenacoccus solenopsis* (Tinsley), (Homoptera: Pseudococcidae), red cotton bug, *Dysdercus koenigii* (Fabricus) (Hemiptera: Pyrrhocoridae), and dusky cotton bug, *Oxycarenus hyalinipennis* (Costa) (Hemiptera: Lygaeidae) causes quantitative and qualitative losses. Insecticides are being used in large quantities to control sucking pests (Arshad et al. 2009). But the lack of farmer knowledge and injudicious use of insecticides with similar modes of action results in the development of resistance to insect pests. Resistance in cotton bugs against different insecticides like bifenthrin, deltamethrin, lufenuron and chlorpyrifos have been reported (Shah, 2014; Naqqash et al. 2014; Ullah et al. 2016; Venkatesan et al. 2016; Hassan et al. 2020).

Dysdercus koenigii has developed a strong resistance against neonicotinoids **(**Saeed et al. 2018**),** and *O. hyalinipennis* also has high resistance against various tested insecticides including bifenthrin, profenofos, chlorpyrifos, methomyl, cypermethrin, emamectin benzoate, deltamethrin, triazophos, fipronil, acetamiprid, spinosad, spirotetramat, imidacloprid, and Nitenpyram (Ullah et al. 2017). The best alternative of these issues is biological control method. This method has to be adopted to avoid the frequent use of insecticides, resistance risk, and safeguard for the environment (Timprasert et al. 2014).

In biological control method, acarine bio-control agent is a well-documented to use against arthropod pests. In stored food commodities, stored grains, and the upper surface of soils, Family Acaridae is abundantly present (Mullen & Durden, 2009). Stored grain mite, *Tyrophagus putrescentiae* (Schrank) (Astigmata: Acaridae) species belong to the family Acaridae (El-Atta et al. 2016), Slow-moving colorless body and long setae (Gerson et al. 2008). The predatory efficacy of cigarette beetle larvae, *Tyrophagus putrescentiae* has been described against different stages of many insects, larval stage of cigarette beetle (Fabricius) (Coleoptera: Anobiidae) (Papadopoulou, 2006). Southern corn rootworm eggs (Brust & House, 1988), adult of *Aedes aegypti* (Linnaeus) (Diptera: Culicidae), and *Aedes albopictus* (Skuse) (Diptera: Culicidae) (Serpa et al. 2004).

In the present study, the predatory efficacy and intraspecific competition of *T. putrescentiae* has been evaluated on all life stages of three important sucking insect pests i.e., *P. solenopsis*, *D. koenigii*, and *O. hyalinipennis* of the cotton crop on various time intervals However, in this research, an eco-friendly way to control the different insect pests of important crops by using their natural enemies were used. The predatory properties of the predator elucidated in this study can provide a basis for a biological control program of three bugs using their specialist predator as *T. putrescentiae*. It is also probable technique that the availability of local insect pests control strategy providing economical substitutes to improved one like food security and safety especially in Pakistan.

Materials and Methods

Collection and rearing of *D. koenigii*, *P. solenopsis*, and *O. hyalinipennis*

Nymphs and adults of three bugs were collected individually in a different plastic jars (2×2×4 inches) from cotton and China rose (*Hibiscus rosasinensis* Linnaeus) plants from the field area of Bahauddin Zakariya University (BZU), Multan (30.1978° N, 71.4697° E), Pakistan. After collection, insects were brought to the rearing laboratory and shifted in glass cages (6×6×10 inches), covered with fine mesh cloth. Soaked fuzzy seeds of cotton variety (NIAB-2008) was used as a food for *D. koenigii*. The cotton seeds were changed on daily basis. Soaked cotton swab was provided as water source. A layer of dry sterilized soil (2 inches) was spread inside the jar for egg-laying for *D. koenigii* (Jaleel et al. 2013). China rose twigs with leaves was provided as food for the rearing of *O. hyalinipennis* and *P. solenopsis*. Twigs were replaced after two days (Saddiq et al. 2014). Opened bolls of cotton, fresh twigs of *H. rosasinensis* and soaked fuzzy seeds of

cotton were also given as food to *O. hyalinipennis*. Feed was changed twice a week (Ullah et al. 2016). Experiment was performed under laboratory conditions (Temperature 27 ± 2ºC, Relative humidity 60 ± 5%, and light: dark photoperiod 14:10 h). In all experiments insect uniform in age were used.

Collection of *Tyrophagus putrescentiae*

Samples of damage -stored grains were collected from government godown and other local grain markets of district Multan. *Tyrophagus putrescentiae* mites were separated from grains processed through Barlese funnel extraction method (Neethirajan et al. 2007).

Identification of *Tyrophagus putrescentiae*

Tyrophagus putrescentiae samples were identified up to species level with the help of published materials. These mite samples were also identified from the Center for Agriculture and Bioscience International (CABI) organization, Rawalpindi, Pakistan.

Rearing of *Tyrophagus putrescentiae*

Tyrophagus putrescentiae were reared on uncontaminated and sterilized wheat germ and wheat porridge. Rearing was done in plastic jars (10×10×25 cm), covered with lids under laboratory conditions (Temperature 27 ± 2ºC, Relative humidity 60 ± 5% and light: dark photoperiod 14:10 h). *Tyrophagus putrescentiae* were reared up to two generations for getting homogeneous progeny. F3 Generation was used for experiment [31].

Predation on egg stage

The experiment was conducted under laboratory conditions. Six treatments with three replications were used to determine the predatory efficacy of mite on the eggs of *D. koenigii, P. solenopsis* and *O. hyalinipennis*. In glass Petri plates of two-inch in size, eggs were put on filter paper. Fifteen numbers of eggs were used in all treatments. While numbers of mites were increased in 1st, 2nd, 3rd, 4th, and 5th treatments having 1, 2, 3, 4, and 5 mites, respectively. Moreover, 6th treatment was considered as control. Mortality was observed after 24 hours and 48 hours as eggs hatched in short period of time.

Predation on nymphs and adult stages

Predatory efficacy of mite on adult stage was evaluated by using similar method as described above as predation on egg stage.

Statistical analysis

The effect of predator density and time intervals on the predatory efficacy of *T. putrescentiae against* all life stages of *P. solenopsis*, *D. koenigii,* and *O. hyalinipennis* were determined through one-way analysis of variance (ANOVA) at ($P< 0.05$) by using statistical software SAS V9.0.

Further, the correlation coefficient was calculated by using statistical software SPSS V20.0 (IBM Co., NY).

Results

Predatory potential of *T. putrescentiae* on *D. koenigii*

The results of the current study showed that significant predation of *T. putrescentiae* was observed after 24 hours on all developmental stages (eggs, nymphs, and adults) of *D. koenigii*. Maximum predation 80% was recorded on adult stage followed by nymph and egg stages. The predation % increased with the increase of predator density and maximum mortality % was recorded in treatment 5 with five predatory mites (Fig. 1). The same trend of predation was recorded after 48 and 72 hours, respectively (Fig. 2, 3).

The correlation between mortality % of eggs, nymphs, and adults of *D. koenigii* and the numbers of predators was recorded significantly positive. The correlation between eggs mortality % and time intervals was recorded significantly negative while the correlation with nymph and adult mortality % was recorded positive (Table 1).

The significant positive correlation was found between the mortality % of all stages (egg, nymph, and adult) of *P. solenopsis* and the predator density. The correlation between egg mortality % and time intervals was recorded significantly negative while the correlation with nymph and adult mortality % was recorded positive with time intervals (Table 2).

The morality % of egg, nymph, and adult of *O. hyalinipennis* was positively correlated with the predator density. The correlation between egg mortality % and time intervals was recorded significantly negative while the correlation with nymph and adult mortality % was recorded positive (Table 3).

Predatory potential of *T. putrescentiae* on *P. solenopsis*

The results revealed that maximum predation of *T. putrescentiae* was recorded after 24 hours on all stages (eggs, nymphs, and adults) of *P. solenopsis*. Maximum predation was recorded on adult stage followed by nymph and egg stages. Moreover, maximum mortality was recorded in the treatment having 5 numbers of predators (Fig. 4). The same trend of predation was recorded after 48 and 72 hours, respectively (Fig, 5, 6).

Predatory potential of *T. putrescentiae* on *O. hyalinipennis*

Tyrophagus putrescentiae exhibited maximum predation after 24 hours on all developmental stages (eggs, nymphs, and adults) of *O. hyalinipennis*. Maximum predation was recorded on adult stage followed by nymph and egg stages. The highest mortality % was observed in the treatment having 5 numbers of predators (Fig. 7). After 48 and 72 hours the same trend of predation was recorded with relatively high mortality % (Fig. 8, 9).

Discussion

Biological control method is an economic and effective method for agricultural pest management. Biological control is pollution-free and environmentally safe method. Biological control method abridged ecologically hazardous chemicals.

In the present study *T. putrescentiae* showed maximum predation against *D. koenigii followed by P. solenopsis and O. hyalinipennis.* Predation was the highest on adult stage of *D. koenigii*, *P. solenopsis and O. hyalinipennis followed by nymphs and eggs.Tyrophagus putrescentiae* has been reported as an important biological control agent (Serpa et al. 2004; Papadopoulou et al. 2006). It feeds voraciously on the insects, soil nematodes, and fungus (Bilgrami & Tahseen, 1992). The predatory potential of *T. putrescentiae* was determined against all life stages of cotton sucking insect pests *i.e., D. koenigii*, *O. hyalinipennis,* and *P. solenopsis*. It was observed that *T. putrescentiae* showed valuable predation on the eggs of cotton bugs. Similar research was conducted by Carly and Roselyne (2021), who proved that *T. putrescentiae* actively fed on the eggs of western flower thrips *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) (Nguyen et al. 2019).

In the present research, the percentage of egg consumption of *O. hyalinipennis*, *P. solenopsis*, *and D. koenigii* by *T. putrescentiae* increased as time increased but the egg mortality % showed negative correlation with time intervals this might be due to the eggs hatch within 72 hours and the mortality was not recorded after 72 hours. Similar research proved that the predatory potential of *T. putrescentiae* increased overtime against the eggs of European cyst nematode, *Heterodera avenae* Wollenweber (Walia & Mathur, 1995). Results also showed that utilization of Southern corn rootworm eggs by predatory mite increased with time (Brust & House, 1988).

The results showed that mean mortality % against all life stages of *O. hyalinipennis*, *P. solenopsis*, *and D. koenigii* had significant positive correlation to the numbers of the predatory mite. Maximum mortality was observed in the treatment 5 having more (5) numbers of predators. The findings of Brust and House (1988), correlated with current results that mortality % of Southern corn rootworm *Diabrotica undecimpunctata* howardi Barber was increased as numbers of predators was increased. *Tyrophagus putrescentiae* was tested for predatory potential against nymphal stages of *D. koenigii*, *P. solenopsis*, *and O. hyalinipennis*. Maximum mortality was observed in T5 against immatures of all insects. Previous studies proved that bio-control potential of *T. putrescentiae* against the immature of *L. serricorne* and 78% mortality was recorded in immatures (Canevari et al., 2012).

This study examined that the predation occurred at the nymphal stage of *O. hyalinipennis* by *T. putrescentiae* was lower as compared to the predation rate against the nymphs of *D. koenigii and P. solenopsis*. This might be due to the fast-moving ability of the nymphal instars of *O. hyalinipennis.* Similar result was recorded in previous study that fast-moving ability of cigarette beetle, *Lasioderma serricorne (Fabricius)* grubs resulted in low predation by *T. putrescentiae* as

the mortality was observed 54, 68, and 78% after the fourth, fifth, and sixth days, respectively (Canevari et al., 2012).

Tyrophagus putrescentiae caused the highest damage to *D. koenigii*, *P. solenopsis*, and *O. hyalinipennis* at nymphal and adult stages. Feeding was done mostly from the thorax, abdomen, and legs due to this behavior maximum mortality was recorded on the 3rd day. It was recorded that *T. putrescentiae* attack on abdomen of *L. serricorne* for feeding (apadopoulou, 2006). Another study described that immature and adults of *T. putrescentiae* consumed the thorax, abdomen, and legs of *A. aegypti* and *A. albopictus* adults (Serpa et al. 2004). In the present research, it was evaluated that due to the large size of host insects like the adult stage of *D. koenigii*, *P. solenopsis*, and *O. hyalinipennis*, mite takes more time for feeding or killing them. If the time period increases mite can feed the adult completely. Similar results were reported that longer periods were required by mites to consume larger prey as compared to smaller ones (Bilgrami, 1994). It was recorded that *T. putrescentiae* slowly feed on the adults of *L. serricorne* (Canevari et al. 2012). Another study showed that the adult mites having a size of 400 to 500 µm completely consumed nematode in 1 to 2 minutes but the immature stage of mite having size < 400 µm take more than 10 minutes for complete feeding (Walter et al. 1986).

In this study it was observed that *P. solenopsis* secreted sugary materials in its surroundings and predatory mite attracted towered these secretions. This behavior of mite revealed that *T. putrescentiae* has capabilities to detect prey from the secretions. In previous study it was recorded that predatory nematode species *Mononchoides forlidens Schuurmans Stekhoven* and *Mononchoides longicaudatus (Khera)* stimulated through prey secretions and fed on the prey (Bilgrami, 1990).

Conclusions

The control of population of cotton sucking pests by *T. prutescentiae* is a very innovatory and ecofriendly. Then this predator could be used in pest management programs for the other harmful pests.

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Table Legends

Table 1 Correlation coefficient (person R) of mortality % of egg, nymph, and adults of *D. koenigii* with predator density and time intervals.

Mortality %	**Predator density**	**P value**
Egg	0.345*	0.020
Nymph	0.945**	0.000
Adult	0.957**	0.000
Mortality %	Time	P value
Egg	-0. 740**	0.00
Nymph	0.179	0.240
Adult	0.131	0.389

Table 2 Correlation coefficient (person R) of mortality % of egg, nymph, and adults of *P. solenopsis* with predator density and time intervals.

Mortality %	Predator density	P value
Egg	0.368*	0.013
Nymph	0.949**	0.000

Adult	0.956**	0.000
Mortality %	Time	P value
Egg	-0.724**	0.000
Nymph	0.155	0.309
Adult	0.133	0.384

*= Significant, **= Highly Significant (P < 0.05).

Table 3 Correlation coefficient (person R) of mortality % of egg, nymph, and adults of *O. hyalinipennis* with predator density and time.

Mortality %	Predator density	P value
Egg	0.367*	0.013
Nymph	0.846**	0.000
Adult	0.857**	0.000
Mortality %	Time	P value
Egg	-0.702**	0.000
Nymph	0.446**	0.002
Adult	0.437**	0.003

*= Significant, **= Highly Significant (P < 0.05).

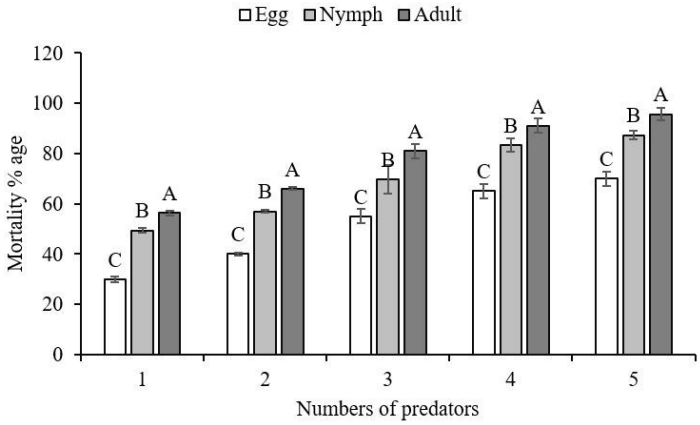
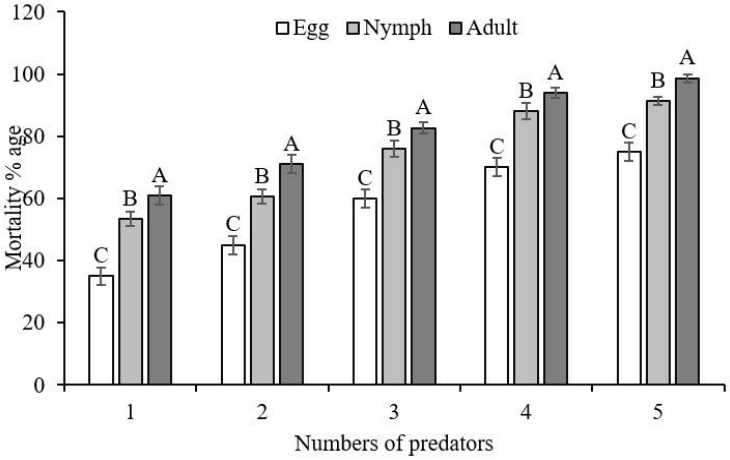


Figure Legends

Figure 1 Mortality % of *T. putrescentiae* after 24 hours on all developmental stages (eggs, nymphs, and adults) of *D. koenigii* on different predator densities.



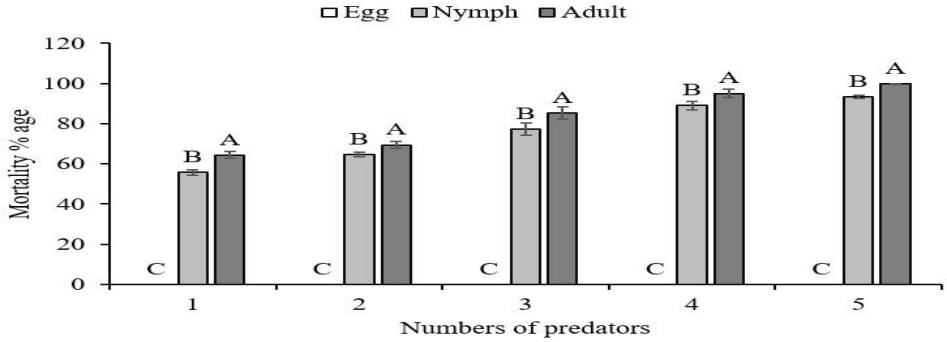
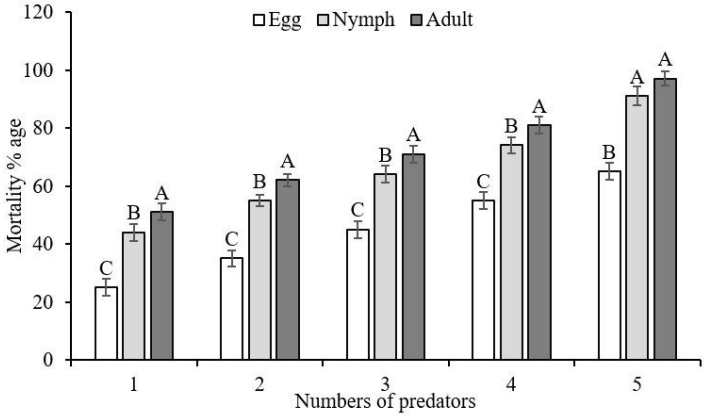


Figure 2 Mortality % of *T. putrescentiae* after 48 hours on all developmental stages (eggs, nymphs, and adults) of *D. koenigii* on different predator densities.

Figure 3 Mortality % of *T. putrescentiae* after 72 hours on nymphs and adults of *D. koenigii* on different predator densities.



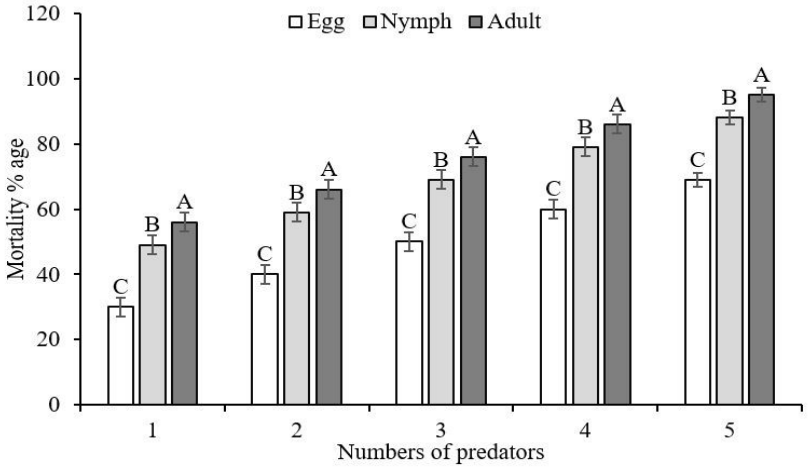


Figure 4 Mortality % of *T. putrescentiae* after 24 hours on all developmental stages (eggs, nymphs, and adults) of *P. solenopsis* on different predator densities.

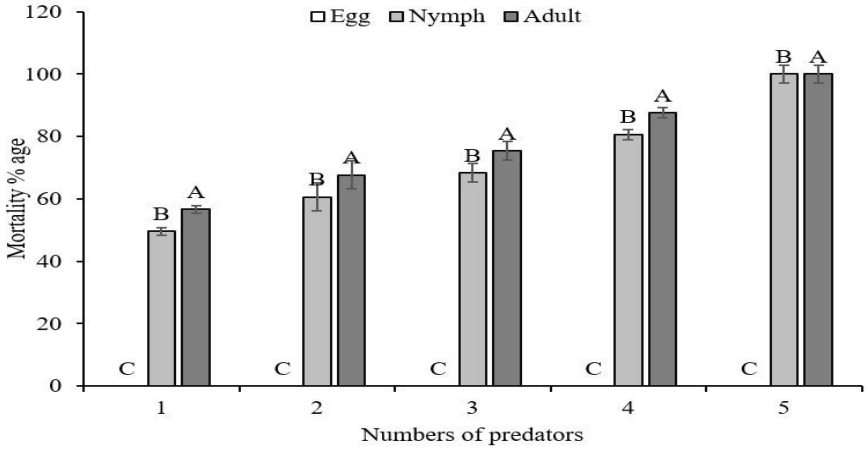


Figure 5 Mortality % of *T. putrescentiae* after 48 hours on all developmental stages (eggs, nymphs, and adults) of *P. solenopsis* on different predator densities.

Figure 6 Mortality % of *T. putrescentiae* after 72 hours on nymphs and adults of *P. solenopsis* on different predator densities.

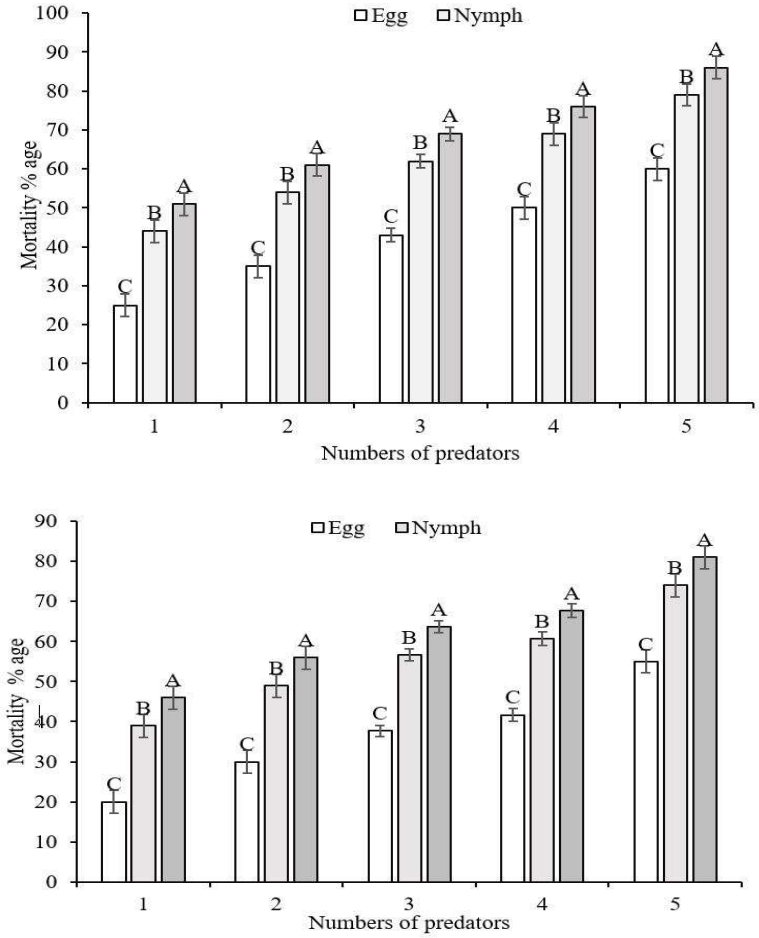


Figure 7 Mortality % of *T. putrescentiae* after 24 hours on all developmental stages (eggs, nymphs, and adults) of *O. hyalinipennis* on different predator densities.

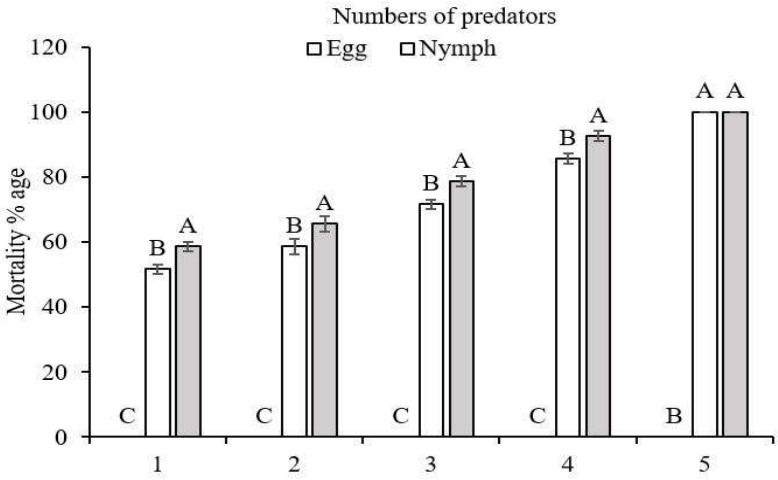


Figure 8 Mortality % of *T. putrescentiae* after 48 hours on all developmental stages (eggs, nymphs, and adults) of *O. hyalinipennis* on different predator densities.

Figure 9 Mortality % of *T. putrescentiae* after 72 hours on nymphs and adults of *O. hyalinipennis* on different predator densities.

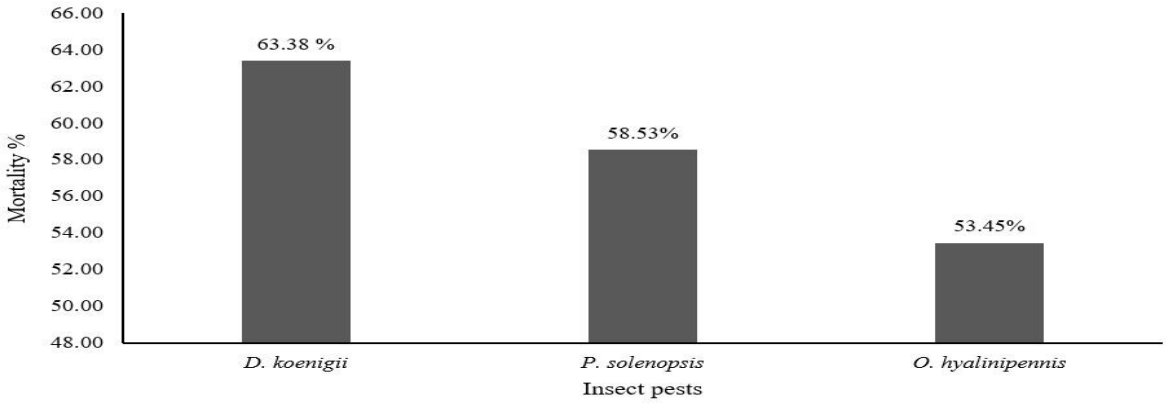


Figure 10. Mean mortality % of *D. koenigii, P. solenopsis and O. hyalinipennis*.