Biological Characteristics and Effect of Temperature on the Development and Reproduction of *Harpactor fuscipes* (Hemiptera: Reduviidae) reared on *Spodoptera litura* (Lepidoptera: Noctuidae) Larvae

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ABSTRACT

Harpactor fuscipes (Fabricius) is a potential biological control agent for *Myzus persicae*(Hemiptera: Aphididae), *Heliothis assulta* (Lepidoptera: Noctuidae) and *Spodoptera litura* (Fabricius) in tobacco fields. However, few studies have recorded the biological characteristics and effects of temperature on the development of this natural predator. We observed the biological characteristics and the effect of temperature on the development of immature and adult *H. fuscipes* predators at 15, 20, 25, 30, and 35°C with 60% relative humidity and a 16:8 L:D photoperiod on a diet of *S. litura* larvae. The results revealed two generations of *H. fuscipes* in Nanxiong Guangdong, including egg, 5 instars nymphal and adult, *H. fuscipes* live through the winter as adults. The optimum temperature for the development and reproduction of *H. fuscipes* was around 30°C. Temperatures in the range of 15-35°C had no effect on oviposition period, but the adults could not lay eggs normally at 15°C.

Our findings indicate that the biological traits and the differences in development and reproduction of *H. fuscipes* at different temperatures could be of importance for its mass rearing and use in the biological control of *M. persicae*, *H. assulta* and *S. litura* in tobacco fields.

Key words: Harpactor fuscipes, Myzus persicae, Heliothis assulta, Spodoptera litura, biological characteristics, developmental durations, biological control.

INTRODUCTION

Tobacco is one of the most important economic crops in China. Chemical pesticides are the main method of protecting tobacco from major pests, including *Myzus persicae* (Sulzer), *Heliothis assulta* (Guenee) and *Spodoptera litura* (Fab.). However, the overuse of pesticides may lead to increased pesticide residues in tobacco, which can impair tobacco quality and smokers' health. Pesticides use may also lead to resistance and killing of natural predators, thereby resulting in pest resurgence.

To better protect tobacco from its pests, we investigated these pests and their natural predators in tobacco fields in Guangdong province, China. We found that

Harpactor fuscipes Fabricius (Hemiptera: Reduviidae), which prey on the larvae of *S. litura* (Zhou *et al.*, 2007), *M. persicae* and *H. assulta* (DENG *et al.*, 2012), is the dominant natural predator of tobacco pests. The predatory function of *H. fuscipes* has been studied (DENG *et al.*, 2012), but life information is available about other biological characteristics and reproductive habits.

To protect and utilize this natural predator, in this study, we recorded the biological characteristics of *H. fuscipes*. Furthermore, to provide a basis of artificial breeding and biological control in tobacco fields, we investigated the development and reproduction of *H. fuscipes* at different temperatures.

MATERIALS AND METHODS

Insects

Adult *H. fuscipes* and larval *S. litura* were collected from the tobacco fields of Nanxiong Scientific Research Institute in Guangdong province, China (2009-2012). *Spodoptera. litura* larvae were fed tobacco and the normal growing 2nd, 3rd instar larvae of the 2nd generation were selected as prey for *H. fuscipes*.

Biological characteristics of H. fuscipes

In April, 10 pairs of *H. fuscipes* were cultured on tobacco plants covered by a cage $(120 \times 120 \times 140 \text{ cm}^3)$. One pair of *H. fuscipes* was cultured per cage. *S. litura* larvae were provided to feed the *H. fuscipes* until the next April. Observations of life cycle and morphological characteristics were conducted for 3 years and the life cycle and morphological characteristics were recorded.

Predation number by different insect states

We determined the predated numbers of *M. persicae*, and the 2nd instar larvae of *S. litura* and *H. assulta* by *H. fuscipes* in 12 cm glass culture dishes. Ten prey insects were placed in each dish; once consumed, more prey insects were supplied. Different life stages of *H. fuscipes* were put into each dish and the number of living prey insects were counted after 24 hours. All experiments were repeated 5 times.

Effect of temperature on *H. fuscipes* development and reproduction

Effect of temperature on *H. fuscipes* development and viability

Twelve-hour post-oviposition egg masses were put in the 12 cm culture dishes and cultured at 15°C, 20°C, 25°C, 30°C and 35°C in illumination boxes. The relative humidity was $60 \pm 15\%$ and the light:dark condition was 16 h: 8 h. Five egg masses were included in each experiment and hatching events were recorded every 12 hours. All experiments were repeated 3 times.

All of the nymphs from the same day were collected and cultured in 8 cm dishes and fed early instar *S. litura* larvae. One fresh tobacco leaf was provided for the *S. litura* larvae in the dish. The 8 cm dishes were put into illumination boxes at different

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temperatures and under the same humidity and light conditions as described above. Each experiment included 20 nymphs, and the developmental events were recorded every 12 hours. All experiments were repeated 3 times.

Effect of temperature on *H. fuscipes* lifespan and fertility

One pair of adult *H. fuscipes* was cultured in the flask and fed early instar *S. litura* larvae. The culture flasks were kept under different temperatures as described above. The adults' growth and ovipositions were recorded every day until they died. All experiments were repeated 3 times.

Statistical analysis

Variance analysis was performed using SPSS (version 12.0;); the average values were analyzed using Tukey's testy and values of P < 0.05 were considered statistically significant. The cumulative survival rates of the nymphs were transformed by arcsine square root transformation before the variance analysis was performed.

RESULTS AND DISCUSSION

Biological characteristics and life cycle

Results show that *H. fuscipes* has 2 generations annually. Males overwintered from middle or late November until early or middle March. *The* First generation eggs were laid in middle or late March, and the nymphs emerged in early April and the adults in late May. The eggs of the 2nd generation were laid in middle or late July and the adults emerged in late September. The adults entered into a diapause overwintering stage in November and became active when the temperature rose in late February or early March.

Adults (Fig. 1.) were 12.5-15.5 mm in length and had a red body color. The pronotum can be divided into anterior and posterior lobes. The front angle of the anterior lobe has a conical shape; the anterior part of the posterior lobe is black and the posterior part is red; the basal part of the scutellum is black; the anterior patagium is dark brown; the fore-, mid- and hind legs are black; and the medial and lateral femora have irregular yellowish-brown spots.

The eggs (Fig. 2.) are mostly laid on the back surfaces of the tobacco leaves. In the earlier stages, the egg masses are light yellow and translucent. The eggs are cylindrical and have a close arrangement and each egg mass contained 25-56 eggs. Eggs were 1.55-1.65 mm long and 0.31-0.34 mm in diameter. A circular egg cover is visible on each individual egg. In the late stage, the egg masses turn reddish brown. The nymphs broke the egg cover as they hatched.

The larvae (Figs. 3.) could be segregated into 5 instars. Upon hatching, the larval bodies are translucent or light yellow. Body size is the major difference among instars. The wing buds appear in the 4th instar larvae, but the mesothorax buds were shorter than those of the metathorax. The wing buds of the 5th instar larvae were longer and larger than those of the 4th instar larvae, and the mesothorax buds were significantly longer than those of the metathorax.

Predation numbers by each H. fuscipes stage

Harpactor. fuscipes can prey upon *M. persicae*, *H. assulta and S. litura* in tobacco fields. The numbers of *M. persicae* larvae predated by different instar, larval and adult stageswere significant different from those of *H. assulta and S. litura* (P < 0.05) (Table 1). However, the predation numbers did not differ significantly between *H. assulta and S. litura*. During the *H. fuscipes* nymphal stages, the prey numbers of increase as instar stage increases. However, the prey numbers for the 4th instar nymphs are greater than those of the 5th instar nymphs.

Impact of different temperatures on H. fuscipes growth and reproduction

The effect of different temperatures on the development of *H. fuscipes* on each instar stage is shown in Table 2. *Harpactor fuscipes* can normally develop to nymph states at temperatures of 15-35°C, but the duration of development of each state decreased as temperature increased. The longest duration of one generation, 56.3 d, was observed at 15°C; 42.1 d was seen at 25°C, 34.8 d was seen at 35°C and the shortest duration, 33.2 d, was seen at 30°C.

The effect of different temperatures on the survival rate of *H. fuscipes* nymphs at each instar is shown in Table 3. The survival rate of the nymphs and adults increased as the temperature increased from 15°C to 30°C. The survival rate of the 5th instar nymphs was only 33.33% at 15°C, but increased to 91.6 % at 30°C. Survival rates of the nymphs at different instar stages decreased when the temperature increased to 35°C, e.g., the survival rate of the 5th instar nymphs decreased to 61.6 %. The highest adult yield rate, 85.6 %, was observed at 30°C, while the lowest yield, 18.5 %, was observed at 15°C.

Impact of different temperatures on H. fuscipes life span and reproduction

As shown in Table 4, no eggs were obtained at 15° C; From 20- 35° C, no significant difference was observed between the pre-oviposition period and the oviposition period. The highest egg number, 122.4 eggs, was achieved at 30° C. The highest hatching rate, 88.2 %, was also observed at 30° C, whereas the lowest rate, 61.7%, was seen at 20°C. The longest life span of the adults was observed at 15° C (males, 45.12 d; females, 51.25 d). Average life spans of the males and females at 35° C were 23.74 and 30.41 days, respectively, a significant lower from those at other temperatures. Table 1. Predacious ability of *H. fuscipes* on three kinds of pests.

11 francisco life etc	Prey species				
H. fuscipes life stage	Myzus persicae	Heliothis assulta	Spodoptera litura		
1 st instar	2.5±0.10 a	1.7±0.07 a	1.8±0.07 a		
2 nd instar	6.7±0.23 a 4.3±0.14 a		4.6±0.16 a		
3 rd instar	14.2±0.38 a 7.5±0.25 b		7.8±0.25 b		
4 th instar	18.5±0.56 a	10.2±0.32 b	10.5±0.32 b		
5 th i nstar	15.6±0.42 a	8.1±0.24 b	8.7±0.31 b		
Adult female	22.8±0.87 a	14.5±0.75 b	14.9±0.75 b		

Note: Mean±SE, Values in the same line that are marked with the same letters were not significantly different at P < 0.05 using Duncan's multiple range test.

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Temp.	Duration of development (d)							
	1 st instar	2 nd instar	3 rd instar	4 th instar	5 th instar	Dev. durations		
15 °C	9.9±0.37 c	10.6±1.14 b	11.9±0.54 b	12.2±0.73 b	13.3±0.26 b	56.3±1.67c		
20 °C	6.4±0.67 b	8.5±0.33 b	8.7±0.26 a	9.2±0.18 a	11.5±0.35 b	43.6±2.51 b		
25 °C	5.7±0.28 a	6.8±0.43 a	6.7±0.35 a	7.1±0.67 a	10.8±0.26 b	42.1±1.54 b		
30 °C	3.9±0.26 a	4.3±0.54 a	6.1±0.72 a	7.6±0.43 a	7.1±0.75 a	33.2±2.37 a		
35 °C	4.2±1.13 a	4.5±0.41 a	6.5±0.62 a	6.9±0.50 a	7.7±0.54 a	34.8±1.72 a		

Table 2. Effect of different temperatures on developmental duration of H.fuscipes nymphs by instar

Note: Mean±SE , Values in the same column with same letters are not significantly different at P < 0.05 using the DMRT

Table 3. Effect of different temperatures on survival rate of H. fuscipes on S. litura larvae

Temp.	Survival rate (%)					Adult yield
	1 st instar	2 nd instar	3 rd instar	4 th instar	5 th instar	(%)
15 ℃	51.6 ±5.67 d	41.8 ±4.85 d	40.6 ±4.85 d	36.6 ±3.26 d	33.3 ±2.87 d	18.5 ±0.75 c
20 °C	88.4 ±8.32 b	85.1 ±7.55 b	85.0 ±7.21 b	81.6 ±6.59 b	78.3 ±7.53 b	65.3 ±6.38 b
25 °C	93.8 ±8.78 a	93.3 ±8.37 a	93.3 ±8.37 a	91.6 ±7.78 a	90.0 ±7.41 a	81.3 ±7.15 a
30 °C	96.6 ±9.65 a	96.6 ±9.15 a	95.0 ±8.75 a	93.3 ±8.41 a	91.6 ±7.78 a	85.6 ±7.47 a
35 ℃	75.1 ±7.43 c	74.2 ±7.26 c	71.6 ±6.94 c	63.3 ±5.56 c	61.6 ±6.22 c	58.3 ±5.19 b

Note: Mean±SE , Values in the same column with same letters are not significantly different at P< 0.05 using the DMRT

Table 4. Effect of different temperatures on *H. fuscipes* adult fecundity and longevity.

Temp.	Oviposition					Adult life span	
	Pre-oviposition(d)	Oviposition (d)	Eggs/female number	Hatching(%) rate (%)	Female (d)	Male (d)	
15 °C	-	-	-	-	51.2±7.13 a	45.1 ±3.15 a	
20 °C	6.4 ±0.05 a	24.2 ±2.15 a	94.5 ±8.49 b	61.7 ±5.87 b	43.3±5.33 a	35.3 ±2.73 b	
25 °C	5.1 ±0.04 a	22.5 ±2.17 a	112.1 ±10.14 a	85.8 ±8.45 a	38.4±5.64 b	32.6 ±2.56 b	
30 °C	5.2 ±0.04 a	21.7 ±2.05 a	122.4 ±10.76a	88.2 ±8.36 a	36.5±3.77 b	39.4 ±2.79 a	
35 °C	4.7 ±0.04 a	21.5 ±2.05 a	88.6 ±7.61 b	67.3 ±6.04 b	30.4±4.58 c	23.7 ±2.16 c	

Note: Mean±SE , Values in the same column with same letters are not significantly different at P < 0.05 using the DMRT.

DISCUSSION

Biological characteristics

The biological characteristics of predators play an important role in their predation (Broufas *et al.*, 2007). Their body size determines their predatory capability (Pereira *et al.*, 2009). In our study, the body size of *H. fuscipes* was relatively large (length, 12.5-15.5 mm; abdominal width, 3.5-4.8 mm), which indicates that *H. fuscipes* can prey upon the higher-stage instar larvae of *S. litura* and *H. assulta* in the tobacco field (Fig. 4, 5).

Predacious ability of H. fuscipes on different pests

A predator's prey requirements are determined by the different instar stages and the prey types (Pappas *et al.*, 2011). Shahayaraj and Sathiamoorthi (2002) reported that the prey number for *Corcyra cephalonica* by *Rhynocoris marginaturs* increased with the instar stages. In addition, the predation by nymphs was significantly higher than by adults. In our study, the Predacious ability of *H. fuscipes* on the 1st to 4th instar nymphs increased with the instar. However, the prey numbers of 5th instar nymphs decreased compared with that of the 4th instar nymphs. We also found that the prey number through the entire nymph stages were significant higher than for the adults. In similarly, the predation of *M. persicae* by *H. fuscipes* was much higher than for *H. assulta and S. litura*. This is perhaps because lower body size of aphid in comparison with caterpillars.

Effect of temperature on development and reproduction

Temperature is one of the most important factors that influences the development and predation of predators (Parajulee *et al.*, 2006). Pakyari and Enkegaard (2012) explored the effect of temperature on the development of *Harmonia axyridis* and found that its developmental duration decreased as temperature increased. Pakyari *et al.* reported that the predation number of the 2 spotted mite by *Scolothrips longicornis* showed a positive correlation with temperature from 15°C to 35°C. In our study, *H. fuscipes* could survive at 15-35°C and the developmental duration of each instar stage decreased as temperature increased. The longest development duration of nymphs and adults was observed at 15°C, but no eggs were laid at this temperature. In contrast, the shortest duration was observed at 30°C and most eggs were laid at this temperature. Our conclusion is consistent with those of Kolokytha *et al.* (2011) and Aline (Pomari *et al.*, 2012), which showed that the life span and development duration of predators have a negative correlation with temperature.

H. fuscipes are mainly distributed in the southern tobacco fields in Guangdong, Guangxi, Yunnan and Fujian Province (Wu *et al.*, 2003). Based on our observations, *H. fuscipes*, including adults and nymphs, is a polyphagous predaceous species with a high predation capacity (predation number), Good oviposition and survival rates permit breed in the laboratory. Potential use of *H. fuscipes* as a biocontrol agent of pests in tobacco fields will require further investigation.

Conflict of interest

The authors declare that they have no conflict of interest.



Fig. 1. An adult Harpactor fuscipes.



Fig. 2. A Harpactor fuscipes egg mass.



Fig. 3. *Harpactor fuscipes;* a) 1st instar nymph, b) 2nd instar nymph, c) 3rd instar nymph, d) 4th instar nymph, e) 5th instar nymph.



Fig. 4. Behaivor of H.fuscipes preying on H. assulta.



Fig. 5. Behaivor of H.fuscipes preying on S. litura.

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