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Article

Effects of temperature on development and reproduction of *Euseius nicholsi* (Ehara & Lee)

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Abstract

The development, survival, and reproduction of *Euseius nicholsi* (Ehara & Lee) (Acari: Phytoselidae), an important and rapacious predatory mite that feeds on *Tetranychus cinnabarinus* (Boisduval), were evaluated at six constant temperatures between 19°C and 32°C. The temperature range 22°C to 28°C was found to be optimal for development and reproduction of the mites. Both were significantly impaired at temperatures higher than 28°C. At 30°C and 32°C, survival rates were significantly reduced. At all temperatures, the egg incubation period was the longest developmental stage, accounting for 40% to 45% of total development time. The sex ratios (female:male) of the offspring at the six temperatures (19, 22, 25, 28, 30 and 32°C) were 1.46:1, 1.35:1, 1.32:1, 1.52:1, 1.65:1 and 1.71:1, respectively. The lowest sex ratio occurred at 25°C, suggesting that this was the natural temperature for this species. At 25°C and 28°C, the population doubling times of *E. nicholsi* were 4.27 d and 3.13 d, respectively, indicating that these were appropriate temperatures for population growth. We conclude that the optimal temperature range for the development of *E. nicholsi* was 22°C to 28°C and that the most suitable temperature for both development and reproduction of the mites was 25°C.

Keywords: *Euseius nicholsi*, development, reproduction, life table, temperature

Introduction

With increasing population pressures on food and natural resources, sustainable development is becoming a global concern. Particularly in relation to the control of agricultural pests, the so-called 'three Rs' (Resistance, Resurgence and Residue) are causing serious negative effects on people's lives. Consequently, biological control methods are increasingly accepted as having an important contribution to agricultural pest control. *Euseius nicholsi* (Ehara & Lee), which is mainly distributed in southern China and Thailand, is an important natural enemy of sap-sucking mite pests. As it is polyphagous (type IV according to McMurtry *et al.* 2013), *E. nicholsi* is potentially a highly valuable control agent for several species of pest mites, including *Tetranychus cinnabarinus* (Boisduval), *T. urticae* (Koch), *Panonychus citri* (McGregor), *Eotetranychus kankitus* (Ehara), *Polyphagotarsonemus latus* (Banks), *P. ulmi* (Koch), and *Acaphylla theae* (Watt) (Zheng & Jin 2009; Ma & Guo 2011). Arthropod populations are affected by many environmental factors, among which temperature has particularly important effects on growth, reproduction, survival and behavior (Ding 1980). To date, research on *E. nicholsi* has mainly focused on development and reproduction at different temperatures when feeding on citrus mites. However, the development and reproduction of this mite feeding on mulberry pests, and its possible role in controlling *T. cinnabarinus*, have not been reported. The aim of the current study was to investigate the effects of temperatures on the development and reproduction of *E. nicholsi* on mulberry leaves. These data will have an important

bearing on reproduction, release and protection of *E. nicholsi* in the field. More precisely, the work reported here was designed with the following objectives: 1) determination of the effects of temperature on the developmental rates of immature and adult mites; 2) determination of the lower developmental temperature and the required thermal conditions using linear and logistic models; 3) estimation of its biological characteristics, including survivorship, preoviposition time, oviposition rate, and total oviposition; and 4) establishment of life tables for this species at different temperatures.

Materials and Methods

Mite sources. Mites were collected from the wild mulberry trees of Southwest University (Beibei, Chongqing, China) and reared in the laboratory on a diet of *T. cinnabarinus* fed on *Vigna unguiculata* (L.) Walp. The mites were maintained at 22–28°C, in 8:16 (L:D) photoperiod growth chambers at a relative humidity of 75–90%. Fresh mulberry leaves were changed every 3–4 d and sufficient prey was supplied each day.

Experimental methods. Incubation and development times of *E. nicholsi* were observed at six constant temperatures: 19°C, 22°C, 25°C, 28°C, 30°C and 32°C. Sixty eggs were observed at each temperature. Eggs that were spawned within a 12-h period were recorded as the same stage. Each mite was individually fed on *T. cinnabarinus* after hatching. The developmental durations, deaths, numbers of males and females at each stage (larva, nymph and adult) at each temperature were checked under the microscope and recorded every 8 h after hatching. Males and females of *E. nicholsi* were mated after eclosion and the duration of preoviposition and spawning, the latest time of spawning, daily oviposition rate and longevity were observed and recorded.

Analytical approaches. The regression relationship between growth rate and temperature was fitted to linear and logistic equations (Wang *et al*. 1982).

Linear equation: $V(T)=(1/K)*T-C/K$. In the model, $V(T)$ represents growth rate at temperature T, K represents the effective accumulated temperature, and C represents the developmental threshold temperature.

Logistic equation: $V(T)=K/[1+\exp(a-bT)]$. In the model, $V(T)$ represents growth rate at temperature T, K represents the highest developmental rate, and a and b are model coefficients.

The experimental data were used to generate life tables at different temperatures. The statistical analysis software SPSS and Excel were used for analysis and graphing of data. The columns of the life tables have the following meanings: x is time interval in units per day; l_x is the survival rate of any one individual during the time x, that is the survival probability in a specific time; and m_x is the average number of female offspring produced by each female mite; this value was calculated from the observed female oviposition rate and the sex ratio at different experimental temperatures.

Results

Development times of *E. nicholsi* at different temperatures

The development times of eggs ranged from 1.65 d at 30°C to 3.81 d at 19°C; for larva the range was 0.65 d at 28°C to 1.67 at 19°C; and for nymphs the range was 1.43 d at 30°C to 2.45 d at 19°C (Table 1). Within the range of 19–28°C, the development times were significantly shorter at higher temperatures ($P<0.05$). At 28°C and 30°C, the development times were rather similar but at the highest temperature (32°C) there were small increases relative to 28°C (Fig. 1). Thus the fastest development of eggs, larvae, and nymphs occurred at 30°C, 28°C and 30°C, respectively.

Considering the whole generation, the development times were not significantly different at 28°C (3.94 d), 30°C (3.80 d) and 32°C (4.03 d). At each temperature, incubation of the egg was the longest stage, accounting for 40–45% of the total development time; larval development was the shortest stage, accounting for 15–21% of development; and the nymphal stage occupied about 30% of the whole developmental period.

TABLE 1. Development times (days) of *E. nicholsi* at six constant temperatures.

Temperature (°C)	n	Egg	Larva	Nymph	The whole generation
19	60	3.81±0.140a	1.67±0.159a	2.45±0.186a	8.94±0.121a
22	60	2.82±0.102b	1.27±0.189b	2.41±0.235a	6.52±0.223b
25	60	2.06±0.098c	0.93±0.241c	1.95±0.176b	4.97±0.154c
28	60	1.81±0.115d	0.65±0.263e	1.48±0.331c	3.94±0.273d
30	60	1.65±0.241e	0.71±0.155e	1.43±0.192d	3.80±0.166d
32	60	1.70±0.132d	0.82±0.232d	1.50±0.225cd	4.03±0.167d

Note: n represents the number of mites. Values are means ± SD. Different letter labels in the same column indicate significant differences (P<0.05, Fisher's LSD test).

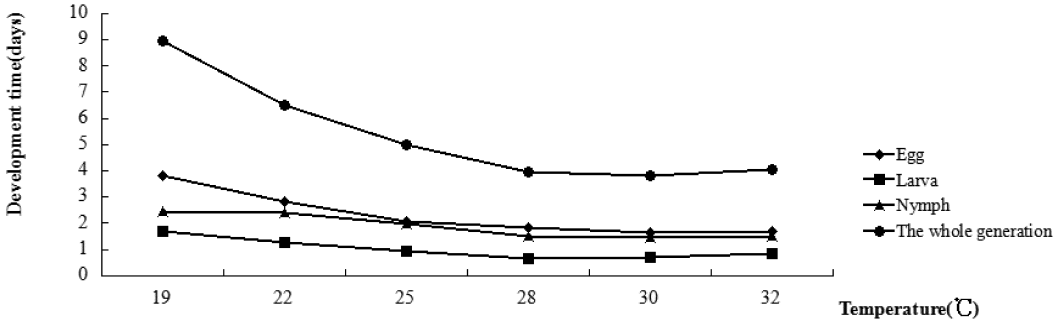

FIGURE 1. The relationship between development time and temperature in *E. nicholsi* .

The regression relationship between developmental rate and temperature

The development times shown in Table 1 were transformed to developmental rates, V=1/D, and the developmental rates for each stage of *E. nicholsi* at different temperatures were fitted by linear and logistic equations. This analysis indicated that the maximum growth rates of each stage were different. The maximum growth rate for larvae was 1.430 and the minimum growth rate for the whole generation was 0.276.

The developmental rates of each stage were positively correlated with temperature. The rates increased rapidly with the temperature between 19°C and 28°C and more slowly from 28°C to 32°C. Both models described the increases in developmental rates with temperature but the logistic model had a greater correlation coefficient and showed that the rates increased more slowly at the higher temperatures of 28°C, 30°C and 32°C. Therefore, we consider the logistic equation to more accurately indicate the relationship between developmental rates and temperature.

Effects of the different temperatures on the survival rates of various stages of *E. nicholsi*

The survival rates of the eggs (Table 2) increased progressively with temperature from 19°C (93.87%), to 22°C (96%), to 25°C (98%) but decreased at higher temperatures (85% at 28°C, 82.67% at 30°C and 59% at 32°C). Thus, the highest survival rate of eggs occurred at 25°C and the lowest survival rate occurred at 32°C. Similarly, the survival rates of larvae and nymphs increased from 19°C to 25°C (larvae: 86.32% at 19°C, 92.51% at 22°C and 95.85% at 25°C; nymphs: 89.25% at 19°C, 95.43% at 22°C and 98.02% at 25°C) but decreased from 28°C to 32°C (larvae: 91.3% at 28°C, 75.3% at 30°C and 60.41% at 32°C; nymphs: 90.15% at 28°C, 80.32% at 30°C and 50.88% at 32°C). Therefore, at 25°C, the survival rates of eggs, larvae and nymphs were all highest (98.00%, 95.85% and 98.02%, respectively). The next highest survival values were observed at 22°C (96%, 92.51% and 95.43%, respectively).

TABLE 2. Survival rates (%) of developmental stages of *E. nicholsi* at different temperatures.

Stage	Temperature (°C)					
	19	22	25	28	30	32
Egg	93.87	96.00	98.00	85.00	82.67	59.00
Larva	86.32	92.51	95.85	91.30	75.30	60.41
Nymph	89.25	95.43	98.02	90.15	80.32	50.88

The influence of temperature on life table parameters of *E. nicholsi*

Life tables of *E. nicholsi* were constructed for the experimental population at the six temperatures (Table 3). Net reproduction rate (R_o) reached a maximum value at 25°C, (9.90) and was next highest at 28°C (7.87). The intrinsic rate of natural increase (r_m) and the finite increase rate (λ) reached maximum values at 28°C (0.22, and 1.33, respectively) and the next highest values were at 25°C (0.16 and 1.25, respectively). At higher temperatures (>28°C), the three parameters of R_o, r_m and λ gradually declined. The shortest population doubling time (t) was 3.13 d at 28°C followed by 4.27 d at 25°C. The highest average generation time (T) was 14.14 d at 22°C and the lowest value was 7.62 d at 32°C.

TABLE 3. Life table parameters of the experimental population of *E. nicholsi* at different temperatures.

Life table parameters	Temperature (°C)					
	19	22	25	28	30	32
Net reproduction rate, R_o	4.64	4.78	9.90	7.87	2.99	1.05
Intrinsic rate of natural increase, r_m	0.11	0.12	0.16	0.22	0.13	0.01
Finite increase rate, λ	1.12	1.13	1.25	1.33	1.18	1.01
Population doubling time, t	5.71	5.37	4.27	3.13	5.54	10.05
Average generation time, T	14.00	12.82	14.14	9.31	8.75	7.62
Longevity (days)	28.48	26.71	29.78	20.78	15.05	14.36
Preovipositing period (days)	3.02	2.04	1.87	2.31	3.26	3.85
Ovipositing period (days)	19.14	22.76	23.33	15.42	12.90	9.31
Total fecundity	14.43	20.77	24.72	12.36	6.43	3.52
Fecundity rate	0.75	0.91	1.06	0.80	0.50	0.38
Sex ratio (♀:♂)	1.46：1	1.35：1	1.32：1	1.52：1	1.65：1	1.71：1

The highest fecundity of female *E. nicholsi* was 1.06 at 25°C, followed by 0.91 at 22°C and 0.80 at 28°C. Fecundity decreased sharply at 30°C (0.50) and 32°C (0.38) to only 47.17% and 35.85% of the value at 25°C. The mean duration of the life of adult mites reached 29.78 d at 25°C. However, the average lifetimes were only 15.05 d at 30°C and 14.36 at 32°C. The average ovipositing period peaked at 23.33 d at 25°C, 2.51 times the ovipositing period at 32°C. Despite differences in the number of eggs laid and the spawning time, the most suitable temperature for oviposition by *E. nicholsi* was 25°C. The sex ratios of the offspring indicated that the lowest percentage of female offspring was 56.90% at 25°C and the highest percentage of female offspring was 63.10% at 32°C.

Effects of temperature on survival rates and reproduction of *E. nicholsi* females

At high temperatures (>30°C) (Fig. 2), the slopes of the survival curves were greater than at lower temperatures, indicating that female mites died more quickly at high temperatures. At 22°C and 25°C, there were four peaks of egg-laying but there was a single oviposition peak at each of the other temperatures. At 22°C, the four peaks corresponded to oviposition rates of 0.88 eggs/d on day 13, 0.74 eggs/d on day 16, 0.76 eggs/d on day 20 and 0.92 eggs/d on day 23. At 25°C, the four peaks of egg-laying were 1.36 eggs/d on day 11, 1.14 eggs/d on day 19, 0.62 eggs/d on day 22, and 0.7 eggs/day on day 25. The maximum rates of oviposition of females (and the corresponding survival rates) were 0.95/d at 19°C (survival: 75%), 0.92/d at 22°C (survival: 11%), 1.36/d at 25°C (survival: 86%), 2.01/d at 28°C (survival: 73%), 0.95/d at 30°C (survival: 59%), and 0.53/d at 32°C (survival: 47%) and occurred on days 12, 23, 11, 9, 9, and 8, respectively. Interestingly, at higher temperatures (30°C and 32°C), the survival rate dropped very quickly initially.

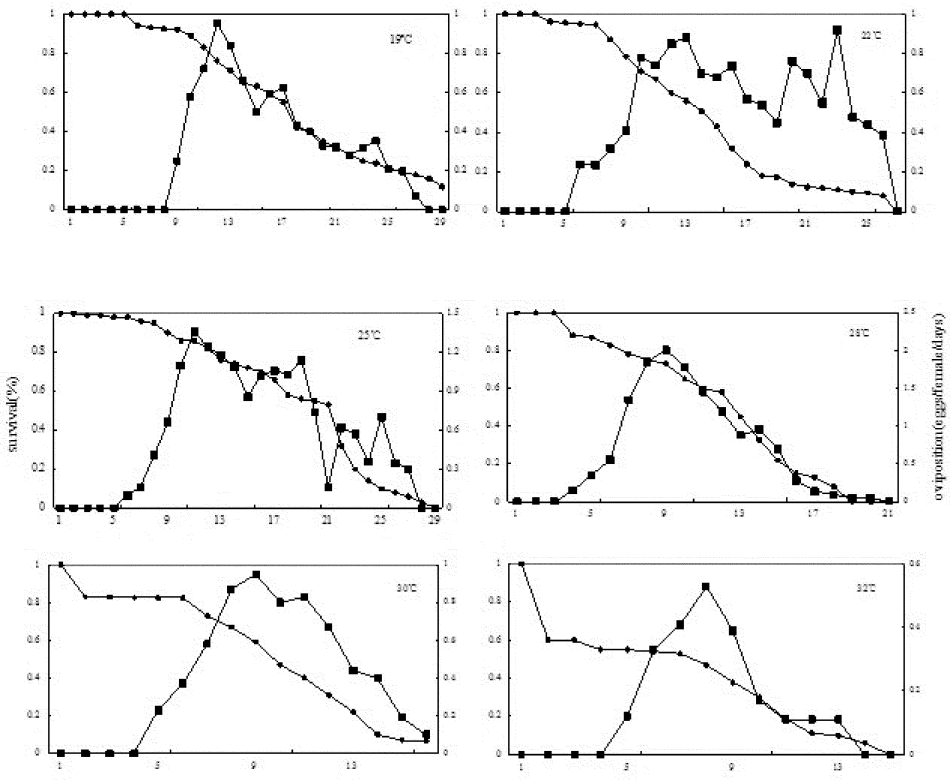

FIGURE 2. The time course of survival, and fecundity rates, of *E. nicholsi* at different temperatures. The dots represent survival rates and the squares represent the fecundity.

Discussion

It has been well documented that the development time of *E. nicholsi* is greatly affected by temperature (Zhi *et al*. 1992). In this experiment, the whole generation times at 19°C, 22°C, 25°C, 28°C, 30°C and 32°C were 8.94 d, 6.52 d, 4.97 d, 3.94 d, 3.80 d and 4.03 d, respectively. Raising the temperature shortened the development time and also caused immature mites to develop more quickly into mature mites. In practical applications, temperature could be controlled at about 28°C to reduce the development time of *E. nicholsi*. The development times observed in the present study of *E. nicholsi* at the same temperatures were significantly shorter than those reported in an earlier study of *E. nicholsi*, which observed development times at constant temperatures of 19°C, 22°C, 25°C, 28°C, and 31°C of 12.61 d, 7.87 d, 7.03 d, 5.22 d and 5.23 d, respectively (Zhi *et al*. 1992). Possibly, this reflects our selection of *T. cinnabarinus* as food in our study, which had a higher nutritional value than the castor pollen (from flowers of *Ricinus communis*) used by Zhi *et al.* It is noteworthy that the egg stage of *E. nicholsi* was the longest developmental stage. Therefore, it would be necessary to avoid applying lethal pesticide during the egg stage of *E. nicholsi* for its efficient use as a control agent.

Our data indicated that there were four peaks of egg-laying at 22°C and 25°C but single peaks at other temperatures. The optimal temperature range for development of *E. nicholsi* was 22°C to 28°C. The peaks of egg-laying eggs and average heights of the peaks indicated that the most appropriate temperature for egg production by female mites was 25°C.

The survival rates showed that the deaths of *E. nicholsi* feeding on *T. cinnabarinus* were progressive. This differs from the general law of insect death. With increasing temperature, the mortality rates of the mites also increased. The highest death rates appeared at higher temperatures (30°C and 32°C) and mortality gradually decreased as the temperature decreased. Possibly the *E. nicholsi* deaths were related to the manufacture of webs by *T. cinnabarinus*.

The population doubling time was shortest at 28°C (3.13 d), followed by 4.27 d at 25°C and 5.37 d at 22°C. These data indicate that temperatures of 25°C to 28°C are most appropriate for growth of the population. The intrinsic rates of natural increase (r_m) of *E. nicholsi* were 0.11, 0.12, 0.16, 0.22, 0.13, and 0.01, respectively, at the six constant temperatures. A previous study reported population doubling times of *T. cinnabarinus* of 2.86 d, 2.38 d, 1.91 d at 25°C, 30°C and 35°C, respectively, and corresponding r_m values of 0.23, 0.24, and 0.34 (Wu, 1988). Therefore, the reproductive potential of *T. cinnabarinus* is stronger than that of its natural enemy, *E. nicholsi*. To control *T. cinnabarinus*, it would therefore be necessary to release a large number of *E. nicholsi* and to supplement them with additional mites at intervals after the first release. We conclude that *E. nicholsi* mites would be most effective in the early stages of biological control of *T. cinnabarinus*.

It was reported that 25°C is the most favorable temperature for reproduction of *E. nicholsi* (Zhi, 1992) and this is consistent with our data. In the current study, at six constant temperatures (19°C, 22°C, 25°C, 28°C, 30°C and 32°C), the offspring sex ratios (female: male) were 1.46:1, 1.35:1, 1.32:1, 1.52;1, 1.65:1 and 1.71:1, respectively. The lowest value was 1.32:1 at 25°C. Therefore, at higher or lower temperatures less favorable for reproduction, female mites would produce more female offspring, which would be beneficial for population survival and reproduction. This may be a general adaptive strategy by *E. nicholsi* to maintain the population in harsh environmental conditions.

Previous studies have shown that humidity, light and other ecological factors greatly influence mite populations, as well as temperature (Xin 1988; Wu & Ding 1985). Our data demonstrated that the optimal temperature range for populations of *E. nicholsi* was 22°C to 28°C. However, the effects of temperature on mite populations need to be further investigated at the population level. Furthermore, the relationship between changes in multiple ecological factors and population

dynamics should be investigated under conditions more appropriate to natural populations (Wu *et al*. 1988; Shipp & Gillespie 1993). Thus, the relationships among humidity, light, other ecological factors and the population dynamics of *E. nicholsi* require further investigation.

In conclusion, our results demonstrated that the optimal temperature range for the development of *E. nicholsi* was from 22°C to 28°C, while the most suitable temperature for mite development and reproduction was 25°C. However, the development and reproduction of *E. nicholsi* would be significantly impaired at temperatures above 28°C. Our results provide basic biological information on *E. nicholsi* that will facilitate its propagation and application in biological control systems.

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