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Article

Prey diet quality affects predation, oviposition and conversion rate of the predatory mite *Neoseiulus barkeri* (Acari: Phytoseiidae)

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Abstract

Biological characteristics of a predator can be affected by the nutritional history of its prey, e.g., change in the nutritional components of prey may influence the development and reproduction of its predator. In this study, the predation, reproduction and conversion rate of a native predatory mite, *Neoseiulus barkeri* were compared when fed with two colonies of the flour mite *Tyrophagus putrescentiae*, the one reared on common wheat bran (TPA) and the other on a mixture of wheat bran and yeast (TPB). The experiments were conducted under laboratory condition at 25°C, 80% relative humidity, and 16:8 light:dark photoperiod. The daily consumption of unmated and mated *N. barkeri* females fed with TPB was 1.30 and 1.93 times higher than those fed with TPA. The daily and cumulative fecundity of mated predator females fed with TPB were 1.55 and 2.47 times, and their eggs being 1.20 times as those of females fed with TPA. The daily consumption of unmated females gradually decreased with age. For both mated and unmated females fed with TPA or TPB, we observed a wave pattern curve of daily prey consumption. The mean difference between two wave peaks was 3.55±0.23 days. It seems a periodic trend instead of random vibration due to the relative consist differences between wave peaks and similar sizes of the peaks. About 82% and 74% overall consumption of mated females offered TPA and TPB were allocated to reproduction, with corresponding conversion rate being 0.39 and 0.28, respectively. Overall, adding yeast to the diet of *T. putrescentiae* led to increased fecundity of *N. barkeri* mainly through stimulating its predation.

Key words: yeast, *Tyrophagus putrescentiae*, *Neoseiulus barkeri*, conversion rate, prey quality

Introduction

Neoseiulus barkeri (Hughes) (Acari: Phytoseiidae) is a commercialized natural enemy of small piercing and sucking arthropods, such as thrips and spider mites (Hughes, 1948; Xin, 1988; Bonde, 1989; Fan & Petitt, 1994; Xu *et al*., 2013). Generally, this predator is mass-reared using the flour mite *Tyrophagus putrescentiae* (Schrank) (Acari: Acaridae), an alternative prey, which is often reared on wheat bran (Ramakers, 1983).

It is known that prey nutrient composition influences biological characteristics of predatory mites (Ferrero *et al*., 2013; Huang *et al*., 2013). Huang *et al*. (2013) reported that the addition of some protein sources, i.e., yeast, to the basic diet (wheat bran) of the prey *T. putrescentiae* leads to increased fecundity in the predatory mite *N. barkeri* and subsequently increase in mass-rearing efficiency. However, the authors did not explain how the presence of yeast in the prey diet indirectly affected the predator fecundity. We assumed two possible hypotheses. First, the predator *N. barkeri* consumes more *T. putrescentiae* with higher protein content as enriched by the presence of yeast. Previous studies showed that some general predators consume more prey with higher nutritional value than prey with lower nutrition value, when both preys are the same species of similar sizes

(Schmidt *et al.*, 2012). We assume similar situations to occur in generalist predatory mites. There are some evidences reporting preference of some phytoseiid species for different prey species with similar size within the same genus. For example, *Neoseiulus californicus* (McGregor) showed preference for eggs of *Panonychus citri* (McGregor) (Acari: Tetranychidae) to those of *P. ulmi* (Koch) during their development (Gotoh *et al.*, 2006).

Second, *T. putrescentiae* with high protein content provides more energy to *N. barkeri* than those with lower protein content. Mated females generally have a small proportion of energy (<30%) allocated to basal metabolism, while the majority is allocated to reproduction requirements (Nwilene & Nachman, 1996; Sabelis & Janssen, 1994). Fecundity of predatory mites differs when the same amount of different prey were consumed, which suggested different energetic supply from different prey (Janssen & Sabelis, 1992; Saber 2013).

In the present study, our objective was to investigate how the addition of yeast to the diet of the prey *T. putrescentiae* indirectly affects the predatory mite *N. barkeri*. We assessed the predation, fecundity, and conversion rate of this predator when fed with two colonies of *T. putrescentiae* with different nutritional histories: the one reared on a diet of common wheat bran and the other on a diet consisting of a mixture of common wheat bran and yeast, similar to that used in Huang *et al.* (2013). We also compared offspring egg size of the predator when fed the two types of prey, as a proxy for estimation of the effect of yeast on offspring size.

Materials and methods

Mite colony maintenance

Tyrophagus putrescentiae and *N. barkeri* were obtained from colonies maintained in the Lab of Predatory Mites, Institute of Plant Protection, Chinese Academy of Agricultural Sciences. The *T. putrescentiae* colony was initiated with individuals collected in wheat bran, and the *N. barkeri* colony was initiated from individuals collected in fruit orchards in Beijing. Both colonies have been kept in the laboratory for more than two years. *Tyrophagus putrescentiae* reared on sterilized wheat bran was used as prey for *N. barkeri*.

Two colonies of *T. putrescentiae* with different nutritional history were established: the one reared on a diet of wheat bran and the other reared on a diet consisting of a mixture of wheat bran and yeast For simplicity, these colonies will be referred to as TPA and TPB, respectively, along the text.

The wheat bran diet was prepared by mixing 130 g wheat bran (8–10% moisture content) with 20 ml distilled water, while the mixed diet was a mixture of 100 g wheat bran, 30 g yeast powder (ca. 200 µm dia.) obtained from yeast tablets, and 20 ml distilled water.

Each colonies of *T. putrescentiae* was maintained in plastic buckets (120 dia.×120 mm h.) with a 50×50 mm^2 hole drilled and sealed with 47 µm nylon fabric mesh on the lid, and was fed with the corresponding diet. Colonies were initiated at least 30 days prior to the commencement of the experiments.

The rearing unit for the predator consisted of a 75 mm-diameter black plastic film placed on a piece of filter paper (90 mm diameter), both placed on a water saturated sponge (90 mm diameter × 60 mm height) laying in a 140 mm ×120 mm (diameter x height) plastic buckets. Each bucket was filled with water at about ca. 40 mm of their height. The colonies were maintained at 25ºC, 80%RH, and 16:8 L:D. Both colonies were initiated 30 days before starting the experiments. The predatory mites produce 4–5 generations during 30 days and preliminary experiments showed that they have reached stable nutrition and metabolic levels.

Experimental procedures and data analysis

Effect of prey type on predation, oviposition, and conversion rate. Sixty female deutonymphs of the predator were selected from the base colony, and individually transferred to experimental units. The experimental units consisted of a 10 mm-diameter drilled on a Plexiglass (30×20×3 mm) board having one side sealed with 47 µm black nylon mesh, and the other side covered with a piece of glass. Upon reaching adulthood, half, i.e., 30 females, were allowed to mate for 24 hours, and the remaining half were maintained unmated. For each prey type, i.e., TPA and TPB, the predation, oviposition and conversion rate were recorded separately for both mated females and unmated females. Each day, 15 *T. putrescentiae* immature of similar body sizes were provided to each female and the prey remained from the previous day were removed. The *T. putrescentiae* immature used as the prey were checked under the microscope to make sure no yeast powder adhered.

Number of preys consumed per day per predator was recorded till the predator died. For mated females, the daily fecundity and oviposition period were also recorded. The actual numbers of replicates ranged from 16 to 29 (Table 1) for the four treatments, with females died accidently or mated females failed to oviposit excluded. Because *T. putrescentiae* were not reared individually from eggs, we were not able to select *T. putrescentiae* immatures of the same age. We assumed the bias introduced by prey age differences to be neglected when a large amount of prey was used.

The conversion rate (γ) from the prey biomass to the egg production was estimated using Eq. (1), as modified by Hayes (1988) from Beddington *et al.* (1976):

$$F = \gamma \times (I - M) \quad (1)$$

In phytoseiid mites, mating is a requirement for the oviposition to occur (Hoy 1982). In the formula above, F indicates fecundity, I indicates total prey biomass consumed estimated from mated females and M indicates prey biomass required for basal metabolism estimated from unmated females. We assumed therefore the difference (I-M) to represent the biomass required for the reproduction. The proportion of energy supply allocated to reproduction (P_R) is estimated using Eq. (2).

$$P_R = \frac{I - M}{I} \quad (2)$$

Effects of prey type on predator egg size. The eggs of predatory mite are ellipsoid. Lengths of the major (L_{max}) and minor (L_{min}) axis of 100 eggs were measured under microscope, and the average egg size (V) was estimated according to the formula of an ellipsoid (Eq. 3).

$$V = \frac{\pi}{6} L_{\min}{}^2 L_{\max} \quad (3)$$

Statistical analysis. Each response variable, including the daily predation, duration of preoviposition and oviposition periods, daily fecundity, and cumulative fecundity, was compared between the 2 treatments using independent two-sample t tests. Effects of diets and mating status on consumption rate were assessed using a 2-way ANOVA. All data were tested for homogeneity using Levene's test prior to their analysis. All analyses were performed using SPSS v.13.0 for Windows. Multiple comparisons for 2-way ANOVA were conducted with Turkey HSD test, with p<0.05 considered as statistically different. The Pearson's correlation coefficient of daily predation and fecundity was estimated using bivariate correlation analysis.

Results

Predation, oviposition, and conversion rate

The consumption rates of mated and unmated females of the predator *N. barkeri* are summarized in Table 1. Prey type significantly affected the predation of both mated and unmated females (df=1, 85; F=115.4; P<0.001). For each prey type, there was significant difference in the predation between mated and unmated females (df=1, 85; F=620.4; P<0.001). Mated females consumed more TPB than TPA, and their predation rate was about 4-fold and 6-fold higher, respectively, than that of unmated females. The addition of yeast to prey increased prey consumption to 1.30 and 1.93 folds for unmated and mated females, respectively. Estimated proportion of the energy allocated to the reproduction in the mated females was 73.8% for TPA and 82.3% for TPB.

TABLE 1. Predation rate of mated and unmated *Neoseiulus barkeri* females fed *Tyrophagus putrescentiae* reared on common wheat bran (TPA) or wheat bran mixed with yeast (TPB).

Prey	No. of consumed preys daily	
	Unmated	Mated (Mean±SE)
TPA	1.0±0.04Aa (19)	4.0±0.13Ab (16)
TPB	1.3±0.08Ba (29)	7.7±0.33Bb (21)

Means±SE followed by different uppercase letters in the same column were significantly different at p = 0.05, while those followed by different lowercase letters in the same row were significantly different at p = 0.05.

The fecundity of mated females is shown in Table 2. There were significant effects of prey type on the pre-oviposition and oviposition periods (t=3.1, p<0.01 and t=-9.7, p<0.01, respectively) as well as on the daily (t=-7.7, p<0.01) and cumulative (t=-23.0, p<0.01) fecundity. The pre-oviposition period was shorter and the oviposition period longer for females fed with TPB, with a daily and cumulative fecundity being 1.5 times and 2.0 times higher than females fed with TPA.

TABLE 2. Fecundity of *Neoseiulus barkeri* females fed *Tyrophagus putrescentiae* reared on common wheat bran (TPA) or wheat bran mixed with yeast (TPB).

Prey	Preoviposition (days)	Oviposition (days)	Daily fecundity (no. of eggs per day)	Total fecundity (no. of eggs)
TPA	4.0±0.30A	12.2±0.53A	1.1±0.03A	13.8±0.42A
TPB	3.0±0.18B	18.7±0.42B	1.7±0.07B	34.1±0.77B

Means±SE followed by different uppercase letters in the same column were significantly different at p = 0.05.

For both prey types, a significant correlation was observed between the predation and oviposition (TPA: R=0.920; TPB: R=0.918). On TPB, the daily consumption reached its first pick at the start of oviposition and the second pick at the date when the oviposition was the highest (ca. 11 days after the start of the experiments) (Figure 1a). Similar trend was observed for females fed with TPA reaching their highest consumption and oviposition rates approximately 13 days after the start of the experiments. Daily consumption of unmated females fed with either TPA or TPB, decreased with age (Figure 1b). For both mated and unmated females, we observed a wave pattern curve of daily prey consumption (Figures 1a,b). The mean difference between two wave peaks was 3.55±0.23 days. Similar vibrations were also observed for daily fecundity of mated females.

On each day, an average of 3.0 of TPAs was used to produce 1.1 eggs, and 6.4 of TPB to produce 1.7 eggs. Conversion rate from consumption to oviposition was significantly lower for females fed with TPB (0.28) than for those fed with TPA (0.39±0.01) (t=-11.0, p<0.01).

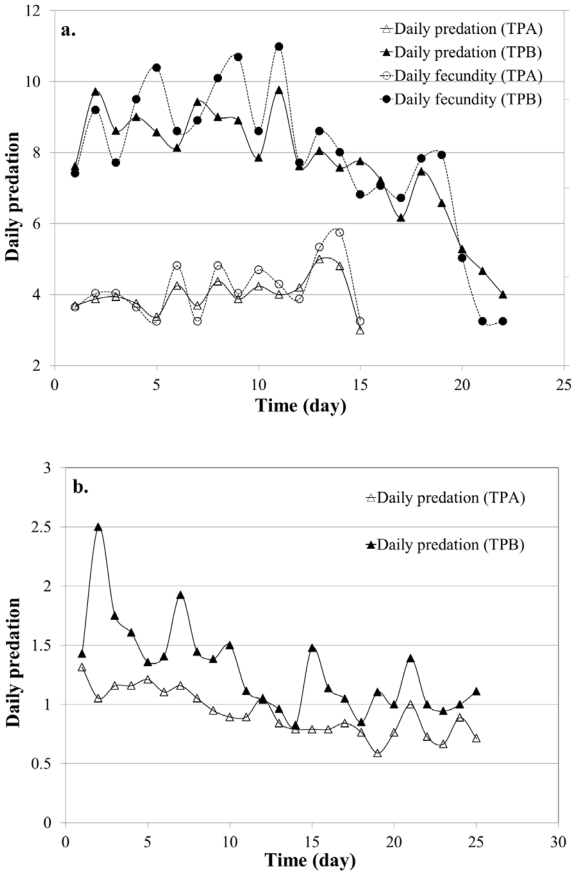

FIGURE 1. Predation and fecundity of mated (a) and unmated (b) females of *Neoseiulus barkeri* reared on (*Tyrophagus putrescentiae* fed with common wheat bran diet (TPA) and on *Tyrophagus putrescentiae* fed with a mixture of wheat bran diet and yeast (TPB).

Offspring egg sizes of N. barkeri.

The major and minor axis of the eggs produced by females fed with TPB were both 1.06 times longer, and the egg size was about 1.20 times larger than those produced by females fed with TPA (Table 3).

TABLE 3. Length and width of eggs produced by *Neoseiulus barkeri* females fed with common wheat bran (TPA) or wheat bran mixed with yeast (TPB).

Egg size (μm)	Prey	
	TPA	TPB
Length	171.1±1.13A	181.2±1.08B
Width	125.6±0.78A	133.5±0.77B

Means±SE followed by different uppercase letters in the same row were significantly different at p = 0.05.

Discussion

In the present study, we observed that the addition of yeast to the prey *T. putrescentiae* diet led to the increase in predation, reproduction, and conversion rate of the predatory mite *N. barkeri*. Mated females consumed a larger number of prey fed with TPB and allocated a large proportion of the energy to reproduction resulting in an increased fecundity and larger size of eggs produced.

Both mated and unmated *N. barkeri* females consumed more TPB, probably because of the increase in soluble protein level due to the addition of yeast (Huang *et al.* 2013). Previous studies have shown that insects consumed more food with higher protein content. For example, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) consumed its artificial diet 1.84 times as in the presence of yeast than in the absence of it (Plácido-Silva *et al.,* 2006). These authors indicated that insects preferred food resource with higher protein contents because protein is crucial for egg production and mating success. They also observed short periods of behavioral changes occurring in lab insect colonies fed diet with high protein content foods. Our findings suggest similar behavioral changes to occur in the predatory mite *N. barkeri* when yeast was added to its prey diet.

For both mated and unmated females fed with TPA or TPB, a wave pattern curve of daily prey consumption was observed. It seems a periodic trend instead of random vibration due to the relative consistent differences between wave peaks and the similar magnitude of the peaks. Because the mean timing difference between two wave peaks is 3.55d, we estimated the mean daily predation over every 4 days, and plotted it against timing: very smooth trends between predation and timing were observed (Figures 2a–b). Such periodic trend of predation for predatory mites has not been reported in previous studies. Due to the stable environment of rearing units used in the study, hunting difficulties should not be the major reason leading to differences of daily predation rate. But under natural circumstances, *N. barkeri* may consume excessively when highly nutrient food is available,

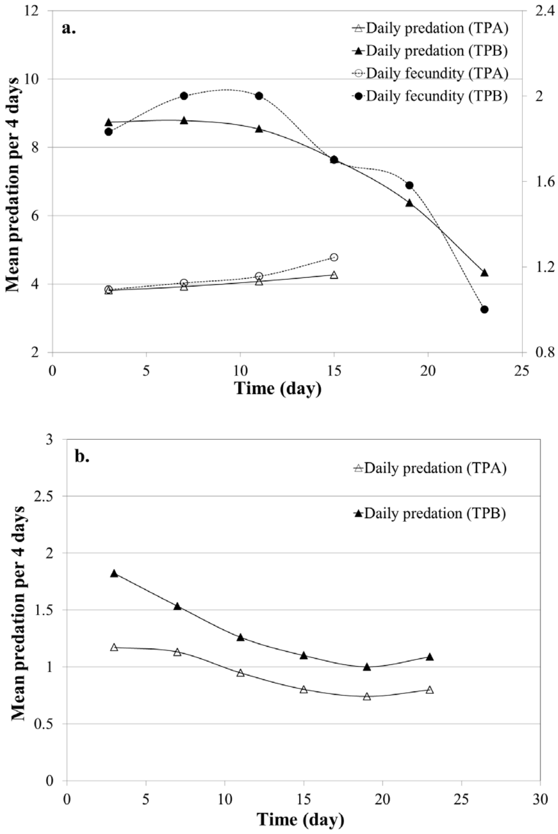

FIGURE 2. Mean predation and fecundity per 4 days of *Neoseiulus barkeri* reared on (*Tyrophagus putrescentiae* fed with common wheat bran diet (TPA) and on *Tyrophagus putrescentiae* fed with a mixture of wheat bran diet and yeast (TPB).

as many predators do, to deal with possible starving in the future. If so, will this characteristic still remain after long-time lab rearing? We speculate the length of the period is affected by nutrition metabolism level of *N. barkeri*; however, further evidences are required. It is also interesting to investigate whether periodical change of predation is common in the Phytoseiidae family or not.

The decrease in daily consumption with age, observed in unmated females fed with TPA and all females fed with TPB, could be explained by a low energy demand due to decrease in basal metabolism and reproduction requirements, similar to that reported in *Phytoseiulus persimilis* (Athias-Henriot) by Rasmy and Hussein (1995). However, this trend was not observed in mated females fed with TPA probably due to their shorter survival time, which in turn could be explained by the large fraction of their energy they allocated to the reproduction.

Previous studies showed that predatory mites convert energy supply into fecundity in a quite short period. Toyoshima and Amano (1998) indicated that it took six hours for the predatory mite *Phytoseiulus persimilis* to produce an egg when sufficient prey was provided. Gotoh and Tsuchiya (2009) observed that starved mated *N. californicus* females started to lay eggs immediately after reaching a spider mite colony. In our preliminary experiments, we found that starved mated *N. barkeri* females started to oviposit within 24 hours after prey was provided. Therefore, the daily predation appears as the most important factor that determines the oviposition rate, which is also supported by our results that showed high correlation between daily fecundity and consumption.

Castagnoli and Simoni (1999) and Saber (2013) also indicated that predatory mites showed lower converting efficiency when more prey was consumed. The addition of yeast to prey diet did not improve the conversion efficiency in *N. barkeri*, although an increase in prey consumption and in the proportion of energy allocated to the preproduction was observed. Perhaps the predator consumed more than the requirement for its metabolism and reproduction, which might also be a strategy to resist possible starvation in the future; this is consist with our assumption for explaining the periodic trend of daily consumption rates. Our finding that the addition of yeast to prey diet led to an increase in egg size produced by predator females is consistent with observations of Toyoshima and Amano (1998) for the predatory mite *P. persimilis*. Although offspring survival was not estimated quantitatively in the present study, no distinct increase in immature mortality has been observed for offspring produced by *N. barkeri* fed with TPB in preliminary mass rearing experiments. Huang *et al.* (2013) reported an increased proportion of female offspring of *N. barkeri* when yeast was added to prey diet. This suggests that the presence of yeast in the prey diet can promote the population growth of *N. barkeri*.

Results from the present study suggested that better nutrition conditions of the prey diet allowed *N. barkeri* to consume more preys and produce more offspring with larger sizes. It is also reasonable to assume good nutrition condition during immature stage leads to increased egg load, which further leads to increased predation to achieve sufficient energy for egg development. Yang *et al.* (2015) reported *N. barkeri* fed with TPB also showed higher consumption rate of its target pests, including spider mites and thrips, than the *N. barkeri* fed with TPA. Janssen and Sabelis (1992) compared the predation and fecundity of 36 species of predatory mites, and found species with high fecundity usually consumed more preys. Therefore, it is reasonable to assume that *N. barkeri* will perform better under natural environments with higher quality prey food. For example, further investigation might be conducted to explore whether *N. barkeri* could provide better biological control efficiency in highly fertilized field. Overall, the present study could serve as a guide line for mass-rearing of predatory mites used in biological control programs.

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