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# Age and size at maturity in *Tyrophagus curvipenis* (Acari: Acaridae) when fed on three different diets

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#### **Abstract**

Tyrophagus curvipenis Fain & Fauvel, 1993 (Acari: Acaridae) has been reported from a very broad range of plant and animal hosts in various habitats in Australia, France, Portugal and New Zealand. However, except host/habitat records, there have been no reports of its life history and other aspects of biology. In this laboratory study conducted at 25°C, T. curvipenis were kept individually and fed three different diets. The time from egg to adult mite was recorded and size of resulting adult mites was measured. T. curvipenis completed development on dry yeast in 10 days, about half of time needed when fed rolled oats and 42% of time needed when fed wheat flour. The length and width of the prodorsal shield was used to determine adult size. Adult females fed yeast were 18% larger than those fed on oats and 25% larger than those fed on wheat flour. Adult males were 14% larger on yeast than on oats, and 27% larger on yeast than on wheat flour. Faster development and larger size at maturity on yeast are correlated with the higher protein content of yeast compared to that in oats and wheat. Males were smaller and developed faster than females on all three diets.

Key words: Food, development, size, Tyrophagus curvipenis

# Introduction

Animals that live in heterogeneous environments often display plastic phenotypes such as age and size at maturity (Pigliucci 2001). These developmental traits are highly variable and can affect overall species fitness and survival (Stearns 1992, Nylin & Gotthard 1998). Many life history traits are correlated; therefore, studying developmental traits, although taking much less time than constructing full life tables, may provide useful insights into the fitness consequences of these life history traits (Walzer & Schausberge 2011). Age and size at maturity are strongly affected by food limitation (Mikolajewski *et al.* 2005), and they may also be sex-specific. There is a paucity of studies on mites of this aspect of developmental plasticity (Walzer & Schausberger 2011). In this study, we examine the age and size at maturity of *Tyrophagus curvipenis* Fain & Fauvel, 1993 (Acari: Sarcoptiformes: Acaridae) when fed three different diets.

Species of *Tyrophagus* have a wide food range and habitats, and include some of the most economically important mites inhabiting stored food and other stored products (Hughes 1976, Fan & Zhang 2007). Several species of *Tyrophagus* are also known to cause damage to economic plants such as ornamental flowers and vegetables in greenhouses (Zhang 2003; Kasuga & Honda 2006). Studies of the life history and biology of this genus are numerous, but concern mainly a few common species. For example, *Tyrophagus putrescentiae*, a cosmopolitan species, is known to feed and reproduce on such food as fungi, pollen, garlic, seeds, cheese and various stored products, as well as

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living plants (Matsumoto 1962, Sinha & Mills 1968, Hughes 1976, Hafez & Tharwat 1989, Zdarkova 1991, Zhang 2003). The effects of different food on development and reproduction of *T. putrescentiae* have been studied; for example, the immature development time and female fecundity of *T. putrescentiae* fed on three different flours (soybean, wheat and maize) were significantly different (Sarwar *et al.* 2010). The effects of different food on body size have been less well studied. Liu *et al.* (2006) showed that food ingredient played an important role in the development of *T. putrescentiae* and affected the body length and width of the adult (Liu *et al.* 2006).

Tyrophagus curvipenis was described by Fain & Fauvel (1993) from mites feeding on algae covering the wooden structures of a greenhouse in Portugal. They also noted that the mites occasionally entered the flowers of orchids and may feed on pollen. In their monograph on Tyrophagus, Fan and Zhang (2007) reported this species in association with many plant and animal hosts in a very broad range of habitats in Australia, France, Portugal and New Zealand. Badieritakis et al. (2012, 2014) recently recorded this species in foliage and litter of Medicago species in Greece. However, virtually nothing else is known about the life history and biology of this species. In this study, we tested the effects of three different food on the age and size at maturity in T. curvipenis. The nutritional quality of the experimental food was compared to evaluate the effects of nutrient contents on mites. We also discussed the developmental plasticity of T. curvipennis fed on different food.

#### Material and methods

Mite culture: A colony of *T. curvipenis* was established from mites living on leaves of capsicum (Fig. 1) grown in a greenhouse in St. Johns, Auckland, New Zealand. The colony had been maintained in the laboratory of Landcare Research (Auckland) for more than two years. The mites were reared on dry yeast, *Saccharomyces cerevisiae* (produced by Goodman Fielder Limited, New Zealand, a common product used in bakery) sprinkled on a black plastic sheet (about 12 cm in diameter), which was placed on several layers of water-saturated paper tissue in a Petri dish (15 cm diameter). Water was added when needed to keep the paper tissue moist to prevent mites from escaping and to maintain a high humidity.



**FIGURE 1.** *Tyrophagus curvipenis* on capsicum leaf (female left, tritonymph lower right, with four eggs upper right). Photograph, Zhi-Qiang Zhang.

Experiment on influence of different diet on development: The experimental containers are modified from those used by Yu et al. (2002): the cell is of a cone shape with top diameter 6 mm and bottom diameter 3 mm in plexiglass slides (25 mm wide, 38 mm long, 3 mm thick). A pair of metal clips was used to hold two plexiglass slides of the same size (one top and one bottom) to keep the mites in the cell.

The three diets used in the experiment were dry yeast (the same as that used in source colony), rolled oats (*Avena sativa*; produced by Harraway and Sons Limited, New Zealand), wheat flour (*Triticum aestivum*, produced by Progressive Enterprises Limited, New Zealand). All tree items were purchased from a local supermarket. The component nutrients of the three diets are shown in Table 1.

**TABLE 1.** The main nutrient contents of three diets in the study. The data are from the packets.

Ingredients (% dry weight)	Dry yeast	Rolled oats	Wheat flour
Protein	46	13.5	11.0
Total fat (Saturated fat)	2.1 (0.8)	5.0 (1.0)	1.6 (0.2)
Carbohydrate (Sugars)	41.9 (17.7)	56.2 (1.0)	69.9 (0.5)
Sodium	0.065	0.009	0.004
Dietary fibre	0	9.2	3.2

Dry food of each of the three types was first mixed with water to a semi-liquid paste. A hair brush was then used to apply a small droplet (about 1 mm in diameter) to the bottom surface of each cell. The food was replaced in intervals of about 10 days when it was contaminated by mite faeces, by transerring the mite with a fine hair brush to a new cell with fresh food.

Fertilized females from the stock culture were individually transferred into a cell for egg laying. The eggs laid within a 24-hours-period were transferred into rearing cells, one per cell, for individual observation of immature mite development on each food. Fifty replicates were established for each food treatment. All the experiments were conducted in a room at  $25 \pm 1^{\circ}\text{C}$  and  $80 \pm 5$  %RH. The development of immature stages were observed at 24 h intervals until they reached adulthood. The presence of an exuvium was used as the criterion for successful development to the next stage. The duration of the egg, larval, protonymphal and tritonymphal stages were calculated.

When the mites developed into adults, they were mounted in a drop of Hoyer's medium. After drying the microscope slides in the oven for a week, the length and width of the prodorsal shield in each mite were measured under a compound microscope at 1000X magnification. All measurements are given in micrometers. Dorsal shield length is used as an indicator of body size in phytoseiid mites (Croft *et al.* 1999). Adult sex was determined under a microscope by using their secondary sexual characters (Fan & Zhang 2007).

Statistical analysis: To assess the effects of different food on the age and size of mites, data were analysed using ANOVA by generalized linear model (GLM) and means were separated by LSD at 5% level if treatment effects were significant. Survival of immature mites fed on different food was analysed using chi-square. Correlations between the total developmental times and the size of mites were performed using simple linear regression models. The data were analysed using DSP statistical software (Tang & Feng, 2007).

## **Results**

*Immature survival*: As expected, egg hatching success rates were similar in all diet treatments, averaging about 90% (Table 2;  $\chi^2 = 0.444$ ; df = 2; p = 0.8). Survival rates of larvae and protonymphs

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were 100% in all treatments. No deutonymphs were observed in this species. The survivorship of tritonymphs was 100% when fed on yeast, but were lower on two other diets (Table 2;  $\chi^2 = 5.79$ ; df = 2; p < 0.05). As a result, the overall survival rates from egg to adult were high and not significantly different among treatments (Table 2; last column;  $\chi^2 = 1.25$ ; df = 2; p = 0.536).

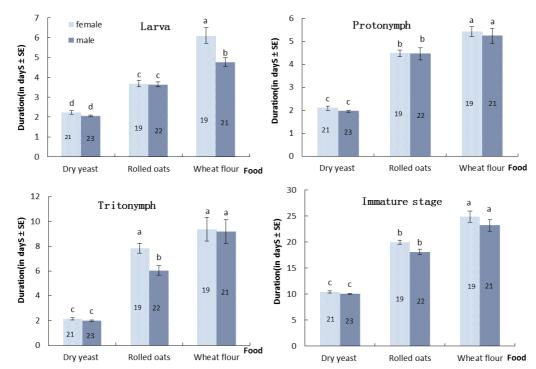
**TABLE 2.** Survival of immature *T. curvipenis* given three different diets.

Diet	n <sup>a</sup>	% survival in immature developmental stages (Means±SE)					
	11	Egg	Larva	Protonymph	Tritonymph	Immature stage	
Dry yeast	50	$88.0 \pm 4.6$	$100 \pm 0.0$	$100 \pm 0.0$	$100 \pm 0.0$	$88 \pm 4.6$	
Rolled oats	50	$90.0 \pm 4.3$	$100 \pm 0.0$	$100 \pm 0.0$	$91.1 \pm 4.3$	$82 \pm 5.5$	
Wheat flour	50	$92.0 \pm 3.9$	$100 \pm 0.0$	$100 \pm 0.0$	$86.9 \pm 5.0$	$80 \pm 5.7$	

<sup>&</sup>lt;sup>a</sup> Initial number of tested individuals.

*Larvae*: The development of larvae was fastest on protein-rich yeast (2.2 days for females; 2.0 days for males), intermediate on oats (3.7 days for females; 3.6 days for males), and the slowest on wheat flour (6.1 days for females; 4.7 days for males) ( $F_{2,124} = 147.33$ ; p < 0.001; Fig. 2 upper left). Males developed faster only when mites were fed on wheat flour ( $F_{1,124} = 11.29$ ; p = 0.001).

*Protonymphs*: The duration of protonymphal stage was affected by food ( $F_{2,124} = 143.15$ ; p < 0.001) but there was no significant difference between males and females on all three diets ( $F_{1,124} = 0.46$ ; p > 0.05): 2.1 days for females and 2.0 days for males on yeast, 4.5 days for females and 4.4 days for males on oats, and 5.4 days for females and 5.2 days for males on wheat flour (Fig. 2 upper right).



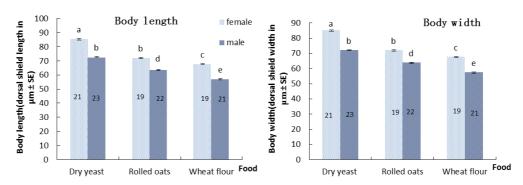
**FIGURE 2.** Average duration (in days  $\pm$ SE) of all immature stages of *T.cuvipensis* fed on different diets. Bars with the same letter are not significantly different (p > 0.05; LSD test). Number of replicates is indicated by the number in each bar. The mites that failed to develop to adults were not used in the final analysis.

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*Tritonymphs*: The food effect on tritonymphal development was significant ( $F_{2,124} = 81.34$ ; p < 0.001) but the gender effect was not ( $F_{1,124} = 2.13$ ; p > 0.05), with the exception for mites reared on rolled oats (with slightly faster developing males; Fig. 2 lower left): 2.1 days for females and 2.0 days for males on yeast, 7.8 days for females and 6.0 days for males on oats, and 9.3 days for females and 9.4 days for males on wheat flour.

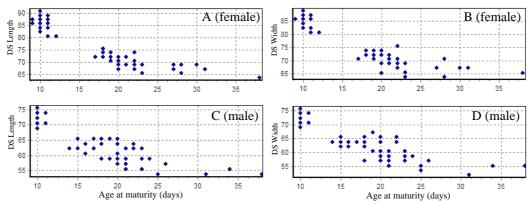
Age at maturity: Overall, the age at maturity was affected by food type ( $F_{2,124} = 209.84$ ; p < 0.001), with males developing slightly faster than females ( $F_{1,124} = 5.2447$ ; p = 0.0238): 10.4 days for females and 10.0 days for males on yeast, 20.0 days for females and 18.1 days for males on oats, and 24.9 days for females and 23.2 days for males on wheat flour (Fig. 2 lower right).

Size at maturity: Food type affected size at maturity, as measured by prodorsal prodorsal shield length and width ( $F_{2,124} = 654.11$ ;  $F_{2,124} = 656.19$ ; p < 0.001); females were always larger than males on the same food ( $F_{1,124} = 771.51$ ;  $F_{1,124} = 795.04$ ; p < 0.001): 85.4 × 85.0 µm for females and 72.5 × 72.0 µm for males on yeast, 72.0 × 72.0 µm for females and 63.5 × 63.8 µm for males on oats, and 67.7 × 67.5 µm for females and 56.9 × 57.3 µm for males on wheat flour (Fig. 3).



**FIGURE 3** Size at maturity (dorsal shield length and width in  $\mu$ m  $\pm$ SE) of *T. cuvipensis* fed on different diets. Bars with the same letter are not significantly different (p > 0.05; LSD test). Number of replicates is indicated by the number in each bar.

The smaller size of both adult females and males was correlated with the increased duration of development, and the correlations are highly significant, with correlation coefficients over 87% (Fig. 4).



**FIGURE 4.** Correlations between the total developmental times and the size of mites (the length and width of dorsal shield on the propodosoma or DS). A: correlation coefficient R = 0.9093, p < 0.001; B: R = 0.9038, p < 0.001; C: R = 0.8711, p < 0.001; D: R = 0.8788, p < 0.001.

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#### Discussion

Tyrophagus curvipenis was first reported to feed on algae and suspected of feeding on pollen of orchids (Fain & Fauvel 1993). Fan and Zhang (2007) reported that this species had been found in association with a broad range of plant and animal material in a variety of habitats. Recently, Badieritakis et al. (2012, 2014) recorded it from both the foliage and litter of Medicago species in Greece. These reports, however, are descriptive and observational in nature, although feeding of T. curvipenis on various plants, fungi and stored food is assumed, based on knowledge of related species of Tyrophagus. This study is the first experimental demonstration of T. curvipenis feeding and successful development on dry yeast, rolled oats and wheat flour. The effects of diets were reflected in the survival rates, age and size at maturity in T. curvipenis (Fig. 2 & Fig. 3; Table 2).

Dry yeast is a common food used for rearing *Tyrophagus* and other stored product mites in laboratory cultures. Its protein content is 3.4 times of that in rolled oats and 4.2 times of that in wheat flour used in this study, whereas its carbohydrate content is 75% of that in oats and 60% of that in wheat (Table 1). In this study, *T. curvipenis* completed development on yeast in 10 days at 25°C, or about half of time needed when fed oats and 42% of time needed when fed wheat flour at the same temperature (Fig. 2). By comparison, age at maturity of *T. putrescentiae* is 8 days on beer yeast powder at 25°C, or about 5 days on corn flour (Liu *et al.* 2006); the protein content of the yeast is 5.8 times of that in corn flour. In both *Tyrophagus* species, development was faster on a protein-rich diet. Yu *et el.* (2002) used yeast to rear *T. putrescentiae* and the time to reach maturity is 12 days at 25°C. Because the food (the same yeast) and other experimental conditions (rearing unit design, temperature, humidity) were similar in this study and Yu *et al.* (2002), it indicates that *T. curvipenis* has shorter development time on yeast than *T. putrescentiae*. However, *T. curvipenis* has longer development time on wheat than *T. putrescentiae*: average of 24 days in this study versus 12 days in Sarwar *et al.* (2010)—all at 25°C.

Size at maturity (in terms of prodorsal shield length and width) of *T. curvipenis* females fed yeast was 18% larger than those fed on oats and 25% larger than those fed on wheat flour (for males, it was 14% larger on yeast than on oats, and 27% larger on yeast than on wheat flour). Liu *et al.* (2006) showed in *T. putrescentiae* that females fed yeast were 28% larger in body length than those fed corn flour (for males it is 23% larger on yeast than on corn flour). Thus, in both species, size at maturity is greater on a protein-rich diet.

We also showed that there were significant inverse correlations between age and size at maturity in both males and females (Fig. 4). Because protein-richer food increased both the speed of development and size at maturity, this correlation was due to treatment effects as revealed by the ANOVA tests.

Most *Tyrophagus* species do not have deutonymphs (Zhang & Fan 2007). In this study, we did not observe deutonymphs in our laboratory colony, experiments and field samples of *T. curvipenis*.

While *T. putrescentiae* and many other species of *Tyrophagus* are more commonly found in stored food than on living plants, *T. curvipenis* is very common on living plants in New Zealand (Fan & Zhang 2007; also personal observation). Our laboratory culture was established with mites collected from leaves of capsicum (Fig. 1) and we have also observed this *T. curvipenis* on many other plant species. However, *T. curvipenis* have not been observed to damage plants. The presence of this species on leaves could, however, enhance the population of predatory mites such as *Neoseiulus cucumeris*, which is a species that has been mass-reared on *Tyrophagus* and other acarid mites (Zhang 2003). In the future, it will be interesting to examine the roles of *T. curvipenis* on plants, especially if they can serve as alternative prey for generalist phytoseiids that are commonly associated with them on plants in New Zealand.

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