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Research Article Evaluation of six cucumber greenhouse cultivars for resistance to *Tetranychus turkestani* (Acari: Tetranychidae)

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Abstract: The strawberry spider mite (SSM), Tetranychus turkestani Ugarov & Nikolski, is an important pest of greenhouse cucumber in tropical and temperate regions of the world. The use of the host plant resistance is a basic method to reduce pesticide application in greenhouses. In this study, the biological responses of SSM to six greenhouse cucumber cultivars (Puia, Hedieh, Milad Ghadim, Milad Jadid, Khasib and Negin) were investigated. Non-choice tests were performed to evaluate the interaction between the host plant-mite. All tests of this study were carried out under laboratory conditions at 28 ± 1 °C, $60 \pm 5\%$ RH and 16:8 h (L: D) in a walk-in growth chamber. The results indicated that immature development time and adult longevity of SSM was significantly influenced by cucumber cultivars. The highest r, λ , and R_0 were 0.275 (day⁻¹), 1.316 (day⁻¹) and 36.180 (offspring/generation) all of which were recorded for the mites reared on the Hedieh cultivar. No significant difference was observed for these parameters among the other cultivars. Mean generation times of T. turkestani reared on Negin (14.020 day) and Milad Ghadim (13.57 day) cultivars were significantly longer than for the other cultivars. This mite had the shortest generation time on Puia cultivar (11.43 day). In conclusion, it seems that Hedieh and Milad Jadid cultivars are the more susceptible and resistant cultivars to the SSM than the other tested cultivars, respectively.

Keywords: host plant resistance, greenhouse cucumber, non-choice test, life table, strawberry spider mite

Introduction

The greenhouse cultivations are rising in recent years, at the same time the problems of pests are increasing as well. Cucumber (*Cucumis sativus* L.) is one of the most important and widely grown crops in greenhouse conditions (Rich *et al.*, 2013; Khaghani, 2009). The greenhouse crops are

affected by many pests that cause extensive yield losses (Zhang, 2003). Manufacturers use chemical pesticides to control the pests, and this process threatens the health of consumers, particularly because these crops are consumed as raw and fresh (Kosary and Kharazy Pakdel, 2006; Baniameri, 2003). Among the wide range of pests which attack to greenhouse crops, strawberry spider mite (SSM), Tetranychus turkestani Ugarov & Nikolski, is an economically important pest (Martinez-Ferrer al., 2006). et Mite population increases enormously in the constantly warm and moist conditions of

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greenhouses (Zhang, 2003). The pest further infests the underside of leaves, where it produces plenty of web and feeds in a piercing-sucking manner that damages plant cells and tissue. The feeding results in appearance of yellow chlorotic spots on leaves (Martinez-Ferrer *et al.*, 2006). One of the strategies for sustainable management of this pest would be the use of resistant host plants.

Host plant resistance can be a fundamental factor of a successful integrated pest management (IPM) system. Integration of resistant plants with other pest control methods results in conservation of beneficial natural enemies and decreased usage of pesticides (Lorenzen et al., 2001). In the first step, recognition of resistance sources in plants is consequential (Jyoti et al., 2001). Also, information about the type of resistance is essential for breeding programs (Stoner and Shelton, 1988). Plant resistance to a pest can be explained in three main assortments: antixenosis, antibiosis and tolerance, or some combinations of these mechanisms (Kogan and Ortman, 1978). Antibiosis is the most important because of its direct effect on the biology of a pest. Therefore. comparison of biological parameters of pest on different host plant cultivars can be used to select resistant cultivars to the pest (Ofomata et al., 2000; Li et al., 2004). Different host plant cultivars affect the life table parameters of SSM differently. Host plant resistance to T. turkestani was reported on some host plants such as melon (Mansour et al., 1987), raspberry (Wilde et al. 1991), strawberry (Gimenez-Ferrer et al. 1994), watermelon (Fadel et al., 1994), tomato (Saeidi, 2006), bean (Mohammadi et al., 2008), soybean (Sedaratian et al., 2011), cotton (Kabiri et al., 2012) and eggplant (Khanamani et al., 2012, 2013). The establishment of spider mites on different cultivars is affected by diverse factors. including secondary morphological metabolites. structures. nutritive value of leaf and leaf age.

Defensive chemical compounds or poorquality plants can make changes in life history and reproductive parameters of spider mites (Krips *et al.*, 1998; Agrawal, 2000; Gotoh and Gomi, 2003).

A life-table study provides two types of information including basic data and derived parameters (Jha et al., 2012; Huang and Chi, 2012). The age-stage specific growth rate. age-stage specific developmental rate, agestage specific survival rate and age-stage specific fecundity are the basic data that are calculated directly from the life history data of a cohort. These basic data are used to estimate derived parameters, such as the intrinsic rate of increase (r), finite rate of increase (λ), net reproductive rate (R_0) and mean generation time (T). The derived parameters are applicable only to a population with a stable age distribution, which is unlikely to occur under changing environmental conditions. According to basic data, an age-stage, two-sex life table offers a way to simulate population growth for a month without assuming a stable age distribution (Chi and Liu, 1985). A simulation based on an age-stage, two-sex life table represents the stage structures of both sexes in a pest population at any time (Chi, 1988, 1990; Tsai and Chi, 2007; Kavousi et al., 2009; Huang and Chi, 2012).

The aim of the present study is to determine the effects of six greenhouse cucumber cultivars on life table parameters of strawberry spider mite. The data obtained from these studies are used to understand the mechanism of population build-up of pest on different cucumber cultivars and to develop a comprehensive pest management program for cucumber in greenhouses.

Materials and Methods

Cultures

In this study, six greenhouse cucumber (*Cucumis sativus*) cultivars, namely Puia, Hedieh, Milad Ghadim, Milad Jadid, Khasib and Negin were used as host plants. The

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cultivars were selected by advisement of plant protection experts in Khuzestan province, Iran. Seeds were drenched 24 h. before planting. Then, seedlings were planted in pots (10 liters in size) that were filled with coco peat and perlite (1:1) in a hydroponic system. The pots were kept in a greenhouse at 28 ± 2 °C, with a light intensity of 13000 Lux and were fed by the nutrient solution (Rash) three times daily.

The initial populations of mites (*T. turkestani*) were collected from infested European bindweed, *Convolvulus arvensis* L., in the agricultural College campus of Shahid Chamran University, Ahvaz, Iran. Collected mites were separately reared on cucumber leaves of each experimental cultivar for three generations.

The mite rearing and experiments were done in a walk-in growth chamber at 28 ± 1 °C, $60 \pm 5\%$ RH and a photoperiod of 16:8 (L: D) h.

Experimental unit

The effect of cucumber cultivars on life parameters of the spider mite and non-choice test was assessed on leaf discs (2 cm diameter) placed on soaked cotton in a Petri-dish (3 cm diameter), in which a 1.5 cm diameter hole was drilled and covered with ruching for ventilation. To keep the leaves fresh, distilled water was daily added to cotton.

Non-choice test

This experiment was carried out in a completely randomized design with six treatments (cucumber cultivars), each with six replications. Five female adult mites were released on the leaf disc per unit. After 72 h., the damage score of the leaf discs, the number of eggs and the percentage of dead mites were recorded. The damage was assessed according to a damage score method as explained by Nihoul *et al.* (1992), Gimenes-Ferrer and Scheerens (1993), Smith (2005) and Seaidi (2006).

Life table

Female and male spider-mite cohorts were formed by placing a pair of SSM on a leaf disc. The mites were eliminated after 3 h. and only one egg was maintained on each leaf disc. Each greenhouse cucumber cultivar was tested with 40 replicates. Units were checked every 12 h. (at 7 am and 7 pm). After adult emergence, females were coupled with males taken from the colony of the mite on the same cultivar. Then the duration of egg, larva, protonymph and deutonymph, adult longevity, fecundity and mortality of the mite were recorded for each cultivar until the death of the last mite.

Life table parameters

The age-stage survival rate $(s_{xj};$ where x and j are age and stage, respectively), the age-stage fecundity (f_{xj}) , the age-specific survival rate (l_x) , and the age-specific fecundity (m_x) , as well as the population parameters, the intrinsic rate of increase (r), the finite rate of increase (λ) , the net reproductive rate (R_0) , and the mean generation time (T), were estimated, according to Chi (2015). The intrinsic rate of increase (r) was calculated by using the iterative bisection method from the Euler-Lotka formula with the age indexed from zero (Goodman, 1982).

$$\sum_{x=0}^{\infty} e^{-r(x+1)} l_x m_x = 1$$
 (1)

Where l_x and m_x were computed according to Chi and Liu (1985) as (k is the number of stages):

$$l_x = \sum_{j=1}^k s_{xj} \tag{2}$$

The mean generation time is the time length that a population needs to increase to R_0 -times of its size as the stable age distribution and the stable increase rate are reached. Therefore, the mean generation time is calculated by $T = \ln R_0/r$. The R_0 is estimated at:

$$R_{0} = \sum_{x=0}^{\infty} \sum_{j=1}^{\kappa} s_{xj} f_{xj}$$
(3)

Statistical analysis

Mean compression of the data was subjected to a one-way Analysis of Variance (ANOVA) using the Proc GLM procedure of SAS 9.2 using the LSD tests at P < 0.05 (SAS Institute Inc, 2006). The data analysis and population parameters (r, λ, R_0) and T) were calculated by using the TWOSEX-MS Chart program and charts were drawn with Sigma Plot 12.0 (Chi, 2015). The standard errors were estimated by using the bootstrap technique and the differences of life table bootstrap-values among the treatments were compared with paired bootstrap test by TWOSEX-MS Chart program (Efron, 1979; Huang and Chi, 2012, 2013).

Result

Non-choice test

The result of the non-choice tests showed a significant difference between the mean mortality percentages ($F_{5, 24} = 9$. 20; P = 0. 0001), mean egg numbers ($F_{5, 24} = 29.67$; P = 0. 0001) and mean damage scores ($F_{5, 24}$ = 22. 13; P = 0. 0001) of T. turkestani fed on different cucumber cultivars leaf disc after 72 h (Table 1). The highest mortality percentage was observed on the Milad Jadid cultivar (36 percent/72 h) and those of other cultivars had no significant difference. The SSM females laid the highest eggs on the Milad Ghadim cultivar during 72 h (119.60 eggs/72 h) and the lowest on the Milad Jadid cultivar (38.80 eggs/72 h). The highest and lowest damage scores were observed on the Hedieh and Milad Jadid cultivars leaf discs (4.40 and 0.80 damage score/leaf disc), respectively (Table 1). The following aspects of life history of SSM as

affected by mite-host plant interactions were studied in more details.

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Developmental time and adult longevity

Effects of different greenhouse cucumber cultivars on the developmental time and adult longevity are presented in table 2. Significant differences among some biological traits such as egg ($F_{5, 54} = 18.32$; P < 0.0001), larvae ($F_{5, 54} = 17.30$; P <0.0001), protonymphal ($F_{5}, F_{5}, 5_{4} = 12.45$; P < 0.0001) and deutonymphal (F_{5, 54} = 12.10; P < 0.0001) durations, total immature developmental time and adult longevity of SSM were observed on the different tested cultivars. The longest (3.52 days) and shortest (2.10 days) egg development were observed on Hedieh and Puia cultivars, respectively. The longer durations of larvae (2.28 days), protonymph (2.28 day) and deutonymph (2.00 days) of the SSM were observed on Puia cultivar.

Statistical comparison between the mean adult pre-oviposition periods (APOP) of T. turkestani on six cucumber cultivars showed that this mite has the longest APOP on Milad Jadid cultivar (1.32 days) and the shortest on Hedieh cultivar (0.62 day) ($F_{5.54}$ = 32.80; P < 0.0001). The shortest total pre-oviposition period (TPOP) (from egg to first oviposition) however was observed on Milad Jadid (9.06 days) and Puia (9.14 days) and the longest on Negin (9.5 days) and Hedie (9.41 days) cultivars ($F_{5, 54}$ = 3.44; P < 0.009). It seems that Negin and Hedieh cultivars lead to increase in duration of the SSM immature life stages. Also, the female adult longevity was shortest (12.77 days) on Milad Jadid and longest on Hedieh cultivar (16.26 days) ($F_{5, 54} = 63.67$; P <0.0001). On the whole, it seems that developmental time and adult longevity of this pest on Hedieh cultivar are longer than those of the other cultivars.

Table 1 Mean (\pm SE) percent mortality, number of eggs and damage score of *Tetranychus turkestani* fed on the six tested greenhouse cucumber cultivars in the non-choice tests under laboratory conditions.

Cultivars	Mortality (%) (in 72 h)	No. eggs (in 72 h)	Damage score on leaf disk (in 72 h)
Puia	$0.00 \pm 0.00b^*$	$45.20 \pm 2.94c$	$4.00 \pm 0.45 ab$
Hedieh	$8.00 \pm 4.91b$	$74.80 \pm 8.02b$	$4.40 \pm 0.25a$
Milad Jadid	$36.00 \pm 7.50a$	$38.80 \pm 7.43c$	$0.80 \pm 0.20d$
Milad Ghadim	$0.00 \pm 0.00b$	$119.60 \pm 6.04a$	$3.20 \pm 0.20b$
Khasib	$2.00\pm0.91b$	$75.20 \pm 3.08b$	$3.20 \pm 0.38b$
Negin	$4.00 \pm 1.40b$	$104.20 \pm 5.52a$	$2.00 \pm 0.00c$

* The means followed by the same letters in each column are not significantly different (LSD, P < 0.05).

Table 2 Mean (\pm SE) of *Tetranychus turkestani* development time and adult longevity reared on the six tested greenhouse cucumber cultivars in laboratory condition.

Development time (day)	Puia	Hedieh	Milad Jadid	Milad Ghadim	Khasib	Negin
Egg	$2.10 \pm 0.04d^*$	$3.52 \pm 0.76a$	$2.55\pm0.08c$	$2.95\pm0.03b$	$2.92\pm0.03b$	$2.93\pm0.03b$
Larval	$2.28\pm0.10a$	$1.77\pm0.10c$	$2.05\pm0.08b$	$2.10\pm0.06b$	$2.05\pm0.05b$	$2.08\pm0.04b$
Protonymph	$2.16\pm0.12a$	$1.60\pm0.08b$	$1.78\pm0.08b$	$1.63\pm0.08b$	$1.18\pm0.06c$	$1.77\pm0.09b$
Deutonymph	$2.00\pm0.12a$	$1.65\pm0.09b$	$1.48\pm0.55b$	$1.60\pm0.11b$	$1.59\pm0.09a$	$1.29\pm0.10a$
Total immature	$8.58 \pm 0.16 ab \\$	$8.55\pm0.73ab$	$7.82\pm0.09d$	$8.30\pm0.12b$	$8.14\pm0.07d$	$8.71\pm0.60a$
APOP**	$0.91\pm0.18c$	$0.62\pm0.13d$	$1.32\pm0.25a$	$1.00\pm0.14bc$	$1.12\pm0.10b$	$1.15\pm0.18b$
TPOP***	$9.04\pm0.23c$	$9.41 \pm 1.03 ab$	$9.05\pm0.25c$	$9.30\pm0.17abc$	$9.24 \pm 0.13 bc$	$9.50\pm0.17a$
Adult longevity (Male)	$10.50\pm0.5b$	$11.80\pm0.58a$	$10.00\pm0.58c$	$12.00 \pm 1.00 a$	$12.00\pm0.44a$	$11.83\pm0.7a$
Adult longevity (Female)	$13.17 \pm 0.40 \text{cd}$	$16.26 \pm 1.13a$	$12.77\pm0.57d$	$13.82\pm0.67b$	$13.57\pm0.48bc$	$13.91\pm0.63b$
Total longevity	$13.08\pm0.47cd$	$15.60\pm1.01a$	$12.49\pm0.52d$	$13.25\pm0.63bc$	13.14 ± 0.46 bcd	$13.76\pm0.56b$

Notes: * The means followed by the same letters in each row are not significantly different (LSD, P < 0.05). **APOP, adult pre-ovipositional period; *** TPOP, total pre-ovipositional period (from egg to first oviposition).

Survival and mortality rate

The result showed that there was a difference among mortality and survival rates of *T. turkestani* reared on the six cucumber cultivars, comparatively (Fig. 1). Mortality of this pest on the tested cucumber cultivars nearly occurred between 5-6 days, except that it started on the 7 day on Hedieh cultivar (Fig. 1). The mortality rate of this pest on cultivar Hedieh ascended with a quick slope between 7-22 days and was fixed from 22^{nd} day till closing date (47^{th} day). On the other cultivars, mortality started on 5^{th} or 6^{th} day and increased till closing date (47^{th} day) (Fig. 1). These results suggest that survival rate of *T. turkestani* fed on Hedieh cultivar was comparatively slowed down.

Population projection

The population growth rates of the SSM reared on the six greenhouse cucumber cultivars were projected. The resulting figures revealed the change in the age-stage structure of *T*. *turkestani* reared on the mentioned host plants. After the second peak, the trend of population growth of SSM on the Hedieh cultivar was equable in the each stage and then the population increased. However, population growth of this pest on the other cultivars was descending after the second peak (Fig. 2).

Life table parameters

The result showed that there was a significant difference between the intrinsic rate of increase (r) ($F_{4, 22} = 4.22$; P = 0.0012), the finite rate of increase (λ) ($F_{5, 54} = 4.43$; P =

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0.0008) and the net reproductive rate (R_0) (F_5 , $_{54} = 6.77$; P = 0.0001) of the SSM fed on the different tested cultivars (Table 3). The highest r, λ , and R_0 were 0.275 (day⁻¹), 1.316 (day⁻¹) and 36.18 (offspring/generation) recorded for the mites that were reared on the Hedieh cultivar. No significant difference was observed for these parameters among the other cultivars. Mean generation time of *T. turkestani* reared on Negin (14.020 day) and Milad Ghadim (13.570 day) cultivars was significantly longer than that of other cultivars($F_{5, 54} = 4.86$; P = 0.0003). This parameter was least for mites that reared on Puia cultivar (11.430 day) (Table 3).



Figure 1 Age-specific survival rate (solid white circle, left Y-axis) and mortality rate (solid black circle, right Y-axis) of *Tetranychus turkestani* reared on six greenhouse cucumber cultivars in laboratory conditions.

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Figure 2 Population projection of age-stage structure of *Tetranychus turkestani* reared on the six greenhouse cucumber cultivars during population growth.

 Table 3 Mean (± SE) population parameters of *Tetranychus turkestani* reared on six cucumber greenhouse cultivars.

Cucumber cultivars	Population parameter $(means \pm SE)^1$					
	Intrinsic rate of increase (day ⁻¹)	Finite rate of increase (day ⁻¹)	Net reproductive rate (offspring/generation)	Mean generation time (day)		
Puia	$0.183 \pm 0.026b^*$	$1.201 \pm 0.03b$	8.15 ± 1.85b	$11.43 \pm 0.41c$		
Hedieh	$0.275\pm0.26a$	$1.316 \pm 0.02a$	$36.18\pm5.19a$	$13.06\pm0.27ab$		
Milad Jadid	$0.207\pm0.02b$	$1.233\pm0.03b$	$15.050\pm4.05b$	$13.04\pm0.43ab$		
Milad Ghadim	$0.199\pm0.02b$	$1.221\pm0.02b$	$15.05\pm4.05b$	$13.57\pm0.42a$		
Khasib	$0.209\pm0.02b$	$1.232\pm0.02b$	$13.433\pm2.37b$	$12.40\pm0.23b$		
Negin	$0.184 \pm 0.02b$	$1.200\pm0.03b$	$13.333 \pm 3.09b$	$14.02 \pm 0.29a$		

¹The means followed by the same letters in each column are not significantly different (Paired bootstrap test, P < 0.05).

Discussion

The main purpose of this study was to evaluate resistance of six greenhouse cucumber cultivars to the strawberry spider mite. The pests have different survival. development and reproductive rates on the plant species and cultivars because plants differ greatly in suitability as hosts. Shorter development time and high reproduction of pests on host plants indicate greater suitability of those plants for colonization of pests (van Lenteren and Noldus 1990). Previous studies demonstrated that not only the quantity, but also the quality of food sources is a consequential factor for growth and development of all organisms (Eischen and Dietz, 1987; Hagley and Barber, 1992, Awmack and Leather, 2002; Blanco et al., 2006; Lee, 2007; Hwang et al., 2008). The results of this study confirmed the previous studies regarding effect of quality and quantity of host plant on the pest biology.

Egg development time of the SSM showed significant difference in the various tested cucumber cultivars. This means that embryonic development of eggs laid by mite females on the different cucumber cultivars was affected by the feeding of females before oviposition. However, Sedaratian *et al.* (2009) pointed out that egg incubation period of *T. urticae* was not significantly different on soybean genotypes. These noticeable differences might be related to differences in, for example the host plant and mite species.

Mean development time of *T. turkestani* varied significantly on the tested cucumber cultivars. According to previous studies, these differences could be due to variation in plant quality, particularly host plant nutrient levels and secondary compounds (Awmack and Leather, 2002; Blanco *et al.*, 2006; Lee, 2007; Hwang *et al.*, 2008).

Khanamani *et al.* (3013) and Kabiri *et al.* (2012) reported that developmental time of *T. urticae* fed on the resistant eggplant and cotton cultivars was longer than on susceptible cultivars. Uckan and Ergin (2002) suggested that increasing the life stage duration of pests

may be a major fitness for the pestfor survival of a generation when food is inadequate, because only a limited number of females have the potentiality to survive. In this study *T. turkestani* had a longer development time on Hedieh cultivar and hence it seems this cultivar may susceptible to the SSM while it was shorter on Milad Jadid cultivar which appears to be resistant. Different responses of this mite to cucumber, eggplant, cotton and or other host plants may be due to the different mite species or variant host plant.

The range of the value of r for the mites that fed on the six greenhouse cucumber cultivars varied from 0.183 to 0.275 (day⁻¹), which is nearly consistent with reported results by Helle and Sabelis (1985). Khanamani et al. (2013) reported that the intrinsic rate of increase of two spotted spider mite varied from 0.022 to 0.157 (day^{-1}) on different eggplant cultivars. T. urticae reared on soybean genotypes had arrange between 0.392 and 0.236 (day⁻¹) (Sedaratian et al., 2011). The declared values of R_0 on the different eggplant cultivars by Khanamani et al., (2013) varied from 1.425 to 11.585 (offspring/generation), but the range of parameter for the SSM in our study was wider (8.150 to 36.180 offspring/generation) and on soybean genotypes was 45.521 at 12.149 (offspring/generation) (Sedaratian et al., 2011). These results showed that the cucumber cultivars may be a more suitable host plant for the larva and nymph of the spider mite than eggplant cultivars but not better than soybean genotypes.

The population parameters (r, R_0 , λ and T) are the derived parameters. They are calculated based on the presumption the that environmental factors are constant and the population structure reaches a stable age-stage distribution as time approaches infinity (Huang and Chi, 2012). The intrinsic rate is a good parameter to reveal and compare the potential of insect populations under different treatments. Its practical application in pest management, however, is limited. The basic data, including developmental rate, survival rate. and fecundity, describe the life history and stage

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differentiation. These data can be used in population projection a stop predicts growth trends, as well as the stage structure of a population in the short-term or long-term future.

These laboratory results may be related to antibiotic or antixenotic effects (or their combinations) of host plants on both life history and fecundity of the SSM. This study did not assay the effect of chemical compounds or morphological structure of these cultivars on biological parameters of *T. turkestani*. Therefore further work is imperative to explain whether the differences observed in mentioned parameters in our study are related to the chemical compounds, the morphological structure of the plants or to both. As well as obtaining more applied results of field experiments should be considered in future.

In conclusion, the use of resistant and moderately resistant cultivars with biological and chemical control methods can help to success an IPM program. Different levels of resistance to *T. turkestani* were observed in Milad Jadid, Puia, Negin, Khasib and Milad Ghadim cultivars. Hedieh and Milad Jadid cultivars were the more susceptible and resistant cultivars to the SSM than other tested cultivars, respectively.

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ارزیابی مقاومت شش رقم خیار گلخانهای نسبت به Tetranychus turkestani (Acari: Tetranychidae)

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