

LEAF STRUCTURE EFFECTS ON THE LIFE CYCLE AND REPRODUCTION OF TWO TETRANYCHID SPECIES

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ABSTRACT

This study was conducted to investigate the effect of some physical and chemical host plant leaf properties such as thickness and hairness of cuticular layer and carbohydrates on the biology of the two tetranychid mites; *Tetranychus urticae* Koch and *Eutetranychus orientalis* (Klein). *Phaseolus vulgaris* L., *Gossypium barbadense* L. and *Solanum melongena* L. were considered to be the most suitable for *T. urticae* as it accelerated development (9.93, 11.00 and 11.37 days), prolonged adult female longevity (18.50, 15.27 and 12.24 days) and increased its fecundity (100.34, 70.40 and 63.34 eggs), respectively. *T. urticae* preference of the lower leaf surface may be due to its very thin cuticular layer which couldn't be measured, compared with that of the other plants (1.6 μ). Dense hair leaf surface was suitable for mite morphology due to its narrow body and long legs and cheliceral stylets.

On the contrary, *Citrus aurantium* (Macfad) and *Plumeria obtusa* L. were the most suitable hosts for *E. orientalis* as it accelerated development (11.02 and 11.48 days), prolonged female longevity (15.32 and 12.00 days) and increased its fecundity (25.40 and 22.34 eggs), respectively. Smooth leaf surface in *C. aurantium* and *P. obtusa* as well as thick cuticle covering the upper epidermis (3.1 μ) proved to be suitable for *E. orientalis* development and reproduction.

INTRODUCTION

The family Tetranychidae includes several mite species that infest a wide variety of field crops, vegetables, fruit trees, shrubs, ornamental plants and grasses. They suck the plant sap by inserting their mouth stylets through the leaf cuticle and the epidermis. Thus, the physical properties of leaves include factors of which the thickness and hairness of cuticular layer might affect the mite feeding process.

Plant resistance to pests involves passive and active mechanisms of resistance. Active resistance comprises mechanisms that are coded and regulated by the plant but are triggered by pathogens and / or insects (Sebastian *et al.* 2004). The outermost barrier of plants towards their environment includes the epicuticular wax crystalloids and the cuticle which are the first defence of plant against the pathogens (Barthlott and Neinhuis, 1997). The cuticle thickness in the adaxial and abaxial leaf surfaces was found to be a considerable physical barrier against pathogen or insect penetration (Lucas, 1998). Among leaf morphological characters, trichomes are one of the most important traits contributing to plant passive resistance against pests (Levin, 1973). Walter *et al.* (1992) found a relationship between both the morphology of the phytoseiids such as body size and leg length of

legs and leaf surface texture. The dense hair of leaf surface allows easy movement for mites with narrow body, relatively longer legs and longer and stronger dorsally directed seta.

Therefore, the objective of this study is to contribute to the effect of physical and chemical structures of host plant leaf on the life cycle and reproduction of the two main tetranychid mites, the two spotted mite *Tetranychus urticae* Koch and the citrus brown mite *Eutetranychus orientalis* (Klein).

Also chemical properties such as nitrogen, potassium, phosphorus, carbohydrates, reducing and non-reducing sugars affect tetranychid mite female life cycle, fecundity and population, Rodriguez (1958), Rasmy (1966) and Ibrahim (1980).

MATERIAL AND METHODS

Biology of the mites:

Two tetranychid mite species; *T. urticae* Koch and *E. orientalis* (Klein) were reared on different plants. Leaf discs of different surface textures were used as substrate. These leaf discs represented field crops, cotton (*Gossypium barbadense* L.); fruit trees, Bitter; (*Citrus aurantium* Macfad) apple (*Malus sylvestris* Mill) and peach, (*Prunus persica* Bats); grasses, bind weed (*Convolvulus arvensis* L.), vegetables; egg plant (*Solanum melongena* L.), kidney beans, (*Phaseolus vulgaris* L.) and castor oil bean (*Ricinus communis* L.) and ornamental plant temple tree (*Plumeria obtusa* L.).

Leaf discs of about one square inch of every plant were put on wet cotton pads in Petri dishes. Twenty five newly hatched larvae of every *T. urticae* and *E. orientalis* were singly transferred each to a leaf disc, and kept to develop till adulthood. Adult females were copulated with males after emergence and left for oviposition. Deposited eggs per female were recorded daily all over the oviposition period. Experiments were conducted at $25 \pm 2^\circ\text{C}$ and $70 \pm 10\%$ R.H. and observation were undertaken twice daily. Measurements of body length and width, length of legs and stylets, of *T. urticae* and *E. orientalis* females (at least 15 individuals) were given in micrometers (μm).

Anatomical studies:

1. Specimens of leaves of the studied plants were taken from the middle of the lamina including the midrib. Specimens were killed, fixed, dehydrated, embedded in paraffin wax (Sass, 1956), sectioned to $20\mu\text{m}$, stained, then mounted in Canada balsam on glass slides for examination (Nassar and El-Sahhar, 1998). Counts and measurements (μ) of the different tissues were taken and averages of 10 readings were calculated.
2. Prints of stomata of leaves were made, using an adhesive, to detect the type and numbers of stomata/ mm^2 in every tested plant. A peel was taken from a basal surface of the leaf to examine types of epidermal trichomes.

Chemical analysis:

To determine the total soluble sugars and reducing sugars, the leaves of the studied genera were collected and dried in a suitable oven adjusted at 70°C for at least 72 hours. Dry materials were ground to fine powder with an electric willy mill and used for chemical analysis. Total soluble sugars were determined according to Dubios *et al.* (1956) and reducing sugars according to Nickerson *et al.* (1975). Non-reducing sugars were mathematically calculated.

RESULT AND DISCUSSION

Phytophagous mites show variable degrees of host preference, therefore, this study was carried out to verify the influence of different host plants on *T. urticae* and *E. orientalis* biology at 25 ± 2°C and 70 ± 10% R.H. These two injurious mites differ in their location on the leaf as the former prefers the lower surface while the latter prefers the upper. However, the two phytophagous mites were reared on nine hosts including, field crops, fruit trees, vegetables, ornamentals and weeds *Gossypium barbadense* L., *Ricina communis* L., *Prunus persica* Bats, *Malus sylvestris* Mill, *Citrus aurantium* (Macfad), *Phaseolus vulgaris* L., *Solanum melongena* L., *Plumeria obtusa* L. and *Convolvulus arvensis* L.

The results showed variable responses throughout the developmental time (life cycle) of *T. urticae* with minimum duration on *P. vulgaris*, *G. barbadense* and *S. melongena* (9.93 ± 0.33, 11.00 ± 0.31 and 11.37 ± 0.25 days) and maximum on *P. obtusa* and *C. aurantium* (14.05 ± 0.28 and 14.55 ± 0.32 days) respectively (Table 1). In addition, female longevity varied according to host plant, as female survived for longest periods on *P. vulgaris* and *G. barbadense* L. (18.50 ± 0.52 and 15.27 ± 0.65 days) and for shortest periods on *C. arvensis* and *M. sylvestris* (9.99 ± 0.50 and 10.20 ± 0.71 days), respectively.

T. urticae female fecundity increased on the same aforementioned host plants (100.34 ± 4.35 and 70.40 ± 3.30 eggs) while decreased on *C. aurantium* and *P. obtusa* (27.40 ± 3.65 and 30.27 ± 3.71 eggs), respectively, (Table 1). Ibrahim (1980) reared *T. urticae* Koch (= *T. arabicus* Attiah) on the seven hosts, ullayg, pear, egg plant, cotton, sweet potato, kidney beans and edible fig, and showed variable life cycle durations with minimum duration on kidney beans and maximum on sweet potato.

Comparing between the structure of the lamina of studied plants, it was obvious that there were several differences among the cuticle thickness covering the lower leaf surface being so small to be difficult to measure in all studied plants, except in *C. aurantium*, *P. obtusa* and *C. arvensis*, which recorded the same measurement (1.6µ). On the other hand, the lower epidermis, thicknesses recorded (14.5 and 12.4µ) for *C. arvensis* and *P. obtusa*, respectively followed being by being nearly equal in measurement (10.1 to 10.5µ) for *G. barbadense* *Ph. vulgaris* and *P. persica* whereas *C. aurantium* had the lowest thickness (7.2µ) (Fig. 1), (Table 2).

Table (1): Duration of *Tetranychus urticae* female different stages and fecundity on nine host plants.

Stages	<i>Phaseolus vulgaris</i>	<i>Gossypium barbadense</i>	<i>Solanum melongena</i>	<i>Ricinus communis</i>	<i>Convolvulus arvensis</i>	<i>Prunus persica</i>	<i>Malus sylvestris</i>	<i>Plumeria obtusa</i>	<i>Citrus aurantium</i>
Egg	4.50 ± 0.20	5.05 ± 0.10	5.70 ± 0.10	5.83 ± 0.07	5.84 ± 0.12	5.40 ± 0.11	5.49 ± 0.07	5.90 ± 0.11	6.02 ± 0.14
Total immatures	5.43 ± 0.12	5.95 ± 0.21	5.67 ± 0.13	6.13 ± 0.12	6.34 ± 0.10	6.85 ± 0.21	7.75 ± 0.14	8.15 ± 0.16	8.53 ± 0.17
Life cycle	9.93 ± 0.33	11.00 ± 0.31	11.37 ± 0.25	11.96 ± 0.20	12.18 ± 0.23	12.25 ± 0.32	13.24 ± 0.21	14.05 ± 0.28	14.55 ± 0.32
Oviposition	15.25 ± 0.63	12.35 ± 0.60	10.12 ± 0.50	9.34 ± 0.42	8.62 ± 0.34	8.32 ± 0.65	8.24 ± 0.80	7.65 ± 1.02	7.50 ± 0.71
Longevity	18.50 ± 0.52	15.27 ± 0.65	12.24 ± 0.72	10.56 ± 0.61	9.99 ± 0.50	11.52 ± 0.90	10.20 ± 0.71	10.60 ± 0.53	11.22 ± 0.61
No. of eggs/♀	100.34 ± 4.35	70.40 ± 3.30	63.34 ± 5.21	54.40 ± 3.55	43.70 ± 3.40	39.40 ± 2.34	35.90 ± 2.35	30.27 ± 3.71	27.40 ± 4.55
Daily rate	6.58	5.70	6.26	5.82	5.07	4.74	4.36	3.96	3.65

Table (2): Measurements (µ) and counts of different tissues of foliage leaves of the studied plant genera (Averages of 10 readings).

Characters	<i>Gossypium barbadense</i>	<i>Ricinus communis</i>	<i>Phaseolus vulgaris</i>	<i>Solanum melongena</i>	<i>Citrus aurantium</i>	<i>Prunus persica</i>	<i>Malus sylvestris</i>	<i>Plumeria obtusa</i>	<i>Convolvulus arvensis</i>
Thickness (µ) of cuticle on the upper epidermis	3.1	1.6	1.6	1.6	3.1	3.1	1.6	3.1	3.1
Thickness (µ) of cuticle on the lower epidermis	-	-	-	-	1.6	-	-	1.6	1.6
a) Upper epidermis	14.7	14.7	14.7	16.3	10.3	14.7	16.7	15.5	16.5
b) Lower epidermis (µ)	10.1	7.8	10.5	7.8	7.2	10.1	8.5	12.4	14.5
Thickness of Lamina (µ)	207.2	197.2	143.4	166.1	278.6	156.1	246.6	287.4	396.7
Length of midrib region (µ)	733.4	678.2	1362.9	1108.5	739.9	488.9	571.1	1395.4	729.5
Width of midrib region (µ)	586.7	843.7	843.7	1168.2	936.8	519.2	656.8	1959.9	839.4
No. of palisade layers	1	1	2	1	2	2	3	2	2
No. of stomata/mm ²	26	upper. 28 lower. 34	40	40	44	22	20	2	12

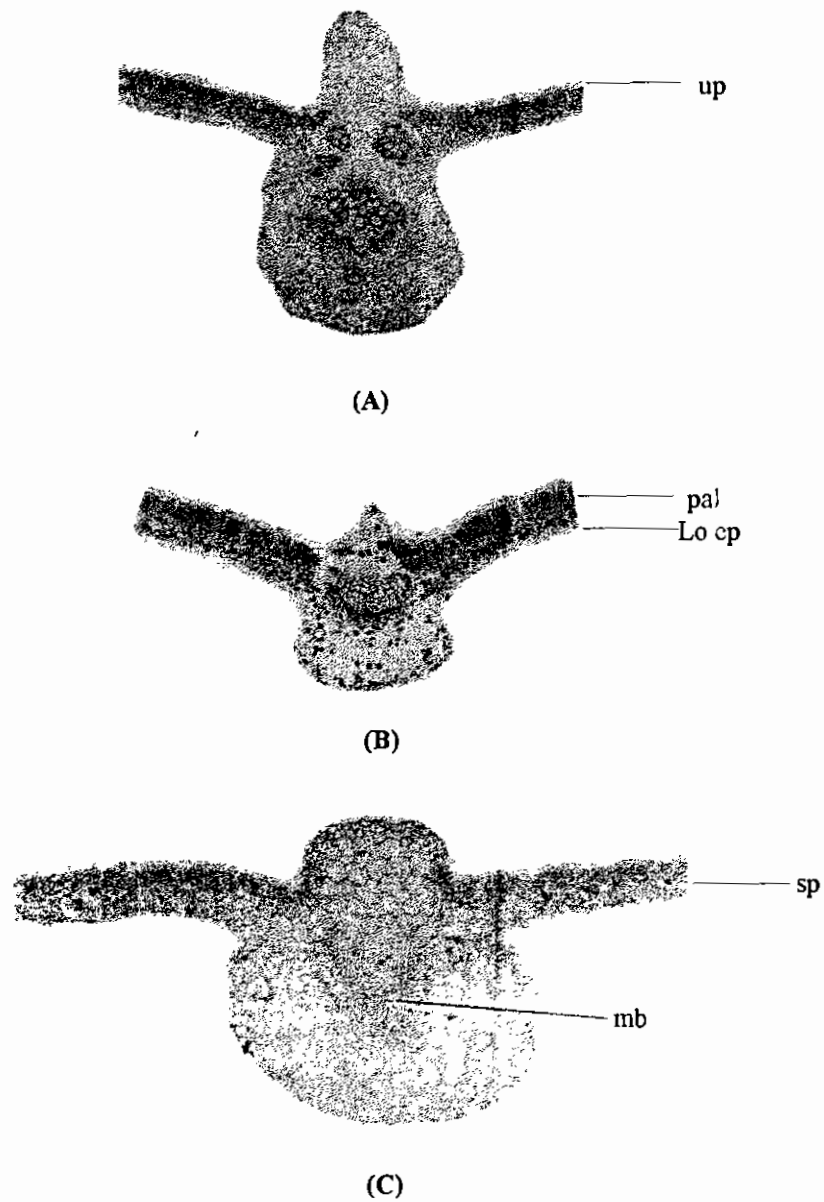
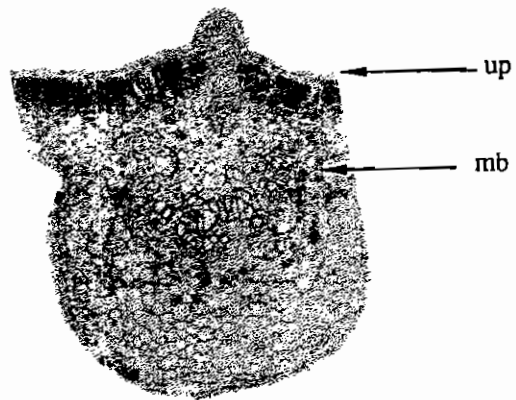
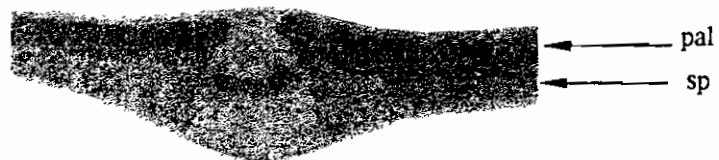


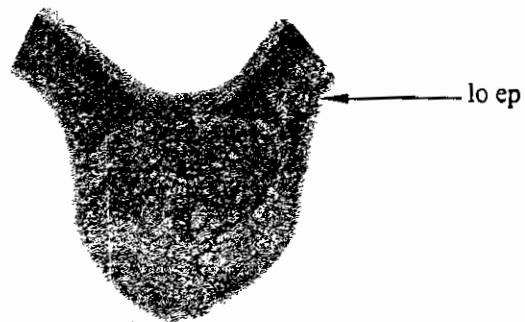
Fig. (1): Transections of the lamina of (A) *Phaseolus*, (B) *Gossypium*, (C) *Solanum* (X40).
Details: lo ep; lower epidermis; mb, midrib bundle; pal, palisade tissue; sp, spongy tissue; up, upper epidermis.



(D)



(E)

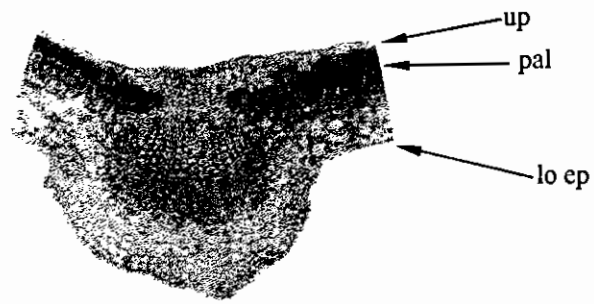


(F)

Fig. (1): Cont.

(D) *Ricinus* (X100), (E) *Convolvulus* (X40), (F) *Prunus* (X100).

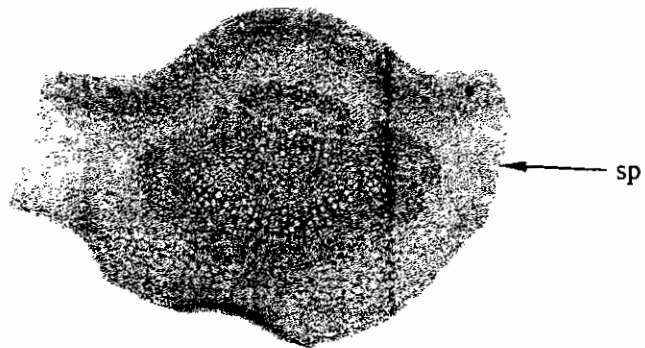
Details: lo ep, lower epidermis; mb, midrib bundle; sp, spongy tissue; up, upper epidermis.



(G)



(H)



(I)

Fig. (1): Cont.

Malus (X100), (H) *Plumeria* (X40), (I) *Citrus* (X100).

Details: lo ep, lower epidermis; mb, midrib bundle; sp, spongy tissue; up, upper epidermis.

As to the type of stomata present on the lower epidermis, it is obvious that, stomata types were anomocytic in the three genera, *Gossypium*, *Solanum* and *Prunus*; paracytic in *Ricinus* and *Plumeria*; anomotetracytic in *Citrus*; paracytic and anomocytic in *Phaseolus*; anisocytic and actinocytic in *Malus* and anomocytic and anisocytic in *Convolvulus*. (Fig. 2) and (Table 2).

Concerning trichomes upon the lower epidermis of the lamina, *Gossypium* recorded stellate hairs with four branches of one cell, *Phaseolus* showed different types of epidermal hairs; stellate hairs with 8 branches each consisted of 2 - 3 cells; glandular and non-glandular multicellular hairs, and unicellular hairs; *Solanum* shows stellate hairs with 7 to 8 branches of one for each branch. These leaves were found to be suitable for mite infestation as it positively affected mite biology. *Malus* had very long and slightly spiral unicellular hairs, (fig 3. d) and gave negative effect. On the other hand, no trichomes were observed on the lower epidermis of the other plant genera. (Fig. 3) and (Table 2).

Generally, *Ph. vulgaris*, *G. barbadense* L. and *S. melongena* proved to be the most suitable hosts of *T. urticae* as it accelerated developmental time, prolonged female longevity and increased female fecundity. Thus, it can be stated that one of the reasons that *T. urticae* prefers the lower leaf surface may be due to its very thin cuticular layer which could not be measured compared with that of the upper surface (1.6 – 3.1 μ), and to the hairs found on the lower surface in addition to escaping from direct sunlight and/or other chemical contents (nutrients or/and odour). However, morphological structure of *T. urticae* showed that cheliceral stylets measured (16.8 μ) body length (69.02) and width (52.72), and legs, (51.16, 42.88, 44.76 and 55.54 μ). Walter (1992) found a relationship occurred between both the morphology of the phyoseiids such as body size and leg length and leaf surface texture. The dense hair leaf surface allows easy movement for mites with narrow body and relatively long legs and longer and strong dorsally directed setae.

Concerning the citrus brown mite *E. orientalis*, it was found that *C. aurantium* and *P. obtusa* were the most suitable hosts as it accelerated developmental time (life cycle) (11.02 ± 0.24 and 11.48 ± 0.20 days) while *G. barbadense* and *S. melongena* prolonged developmental time (14.41 ± 0.53 and 14.93 ± 0.75 days), respectively. (Table 3).

Also female longevity varied according to host plant. It prolonged on *C. aurantium* (15.32 ± 1.60 days) while decreased to (9.07 ± 0.75 days) on *M. sylvestris*, respectively.

Moreover, female fecundity increased on *C. aurantium* and *P. obtusa* (25.40 ± 4.90 and 22.34 ± 3.73 eggs) while decreased on *G. barbadense* and *S. melongena* (9.40 ± 2.76 and 7.65 ± 2.40 eggs), respectively. On the other hand the cuticle thickness covering the upper epidermis was the highest in *C. aurantium*, *P. obtusa*, *P. persica* and *C. arvensis*, (3.1 μ) while reduced by 50% (1.6 μ) in the other plant leaves.

Also, upper epidermis thickness that gave the lowest measurement (10.3 μ) was recorded by *C. aurantium* (Table 2). In *Plumeria*, it was noted that the midrib bundle and the secondary ones were connected to the upper and lower epidermis with compact parenchymatous cells. *Citrus* has many oil ducts present underlying the upper epidermis and this odour may play a part in preferring the plant as well as leaf surface.

Table (3): Duration of *Eutetranychus orientalis* female different stages and fecundity on nine host plants.

Stages	<i>Citrus aurantium</i>	<i>Plumeria obtusa</i>	<i>Ricinus communis</i>	<i>Prunus persica</i>	<i>Convolvulus arvensis</i>	<i>Malus sylvestris</i>	<i>Phaseolus vulgaris</i>	<i>Gossypium barbadense</i>	<i>Solanum melongena</i>
Egg	5.04 ± 0.11	5.27 ± 0.10	5.72 ± 0.12	5.92 ± 0.11	6.24 ± 0.07	6.50 ± 0.10	6.50 ± 0.14	6.51 ± 0.10	6.29 ± 0.22
Total immatures	5.98 ± 0.13	6.21 ± 0.50	6.54 ± 0.12	6.78 ± 0.42	6.80 ± 0.51	7.02 ± 0.63	7.63 ± 0.63	7.90 ± 0.43	8.64 ± 0.53
Life cycle	11.02 ± 0.24	11.48 ± 0.20	12.26 ± 0.25	12.70 ± 0.53	13.04 ± 0.59	13.52 ± 0.73	14.13 ± 0.79	14.41 ± 0.53	14.93 ± 0.75
Oviposition	11.20 ± 0.63	10.10 ± 0.50	10.29 ± 0.53	9.75 ± 0.43	8.70 ± 0.60	8.53 ± 0.70	8.20 ± 0.98	7.07 ± 0.43	6.52 ± 0.63
Longevity	15.32 ± 1.60	12.00 ± 1.23	13.27 ± 1.22	10.94 ± 0.89	9.25 ± 0.80	9.07 ± 0.75	10.40 ± 1.05	9.50 ± 0.90	9.14 ± 1.02
Nc. of eggs/♀	25.40 ± 4.90	22.34 ± 3.73	21.50 ± 4.73	19.25 ± 3.75	17.34 ± 2.50	14.00 ± 2.70	12.50 ± 1.73	9.40 ± 2.76	7.65 ± 2.40
Daily rate	2.27	2.21	2.08	1.97	1.99	1.64	1.52	1.33	1.17

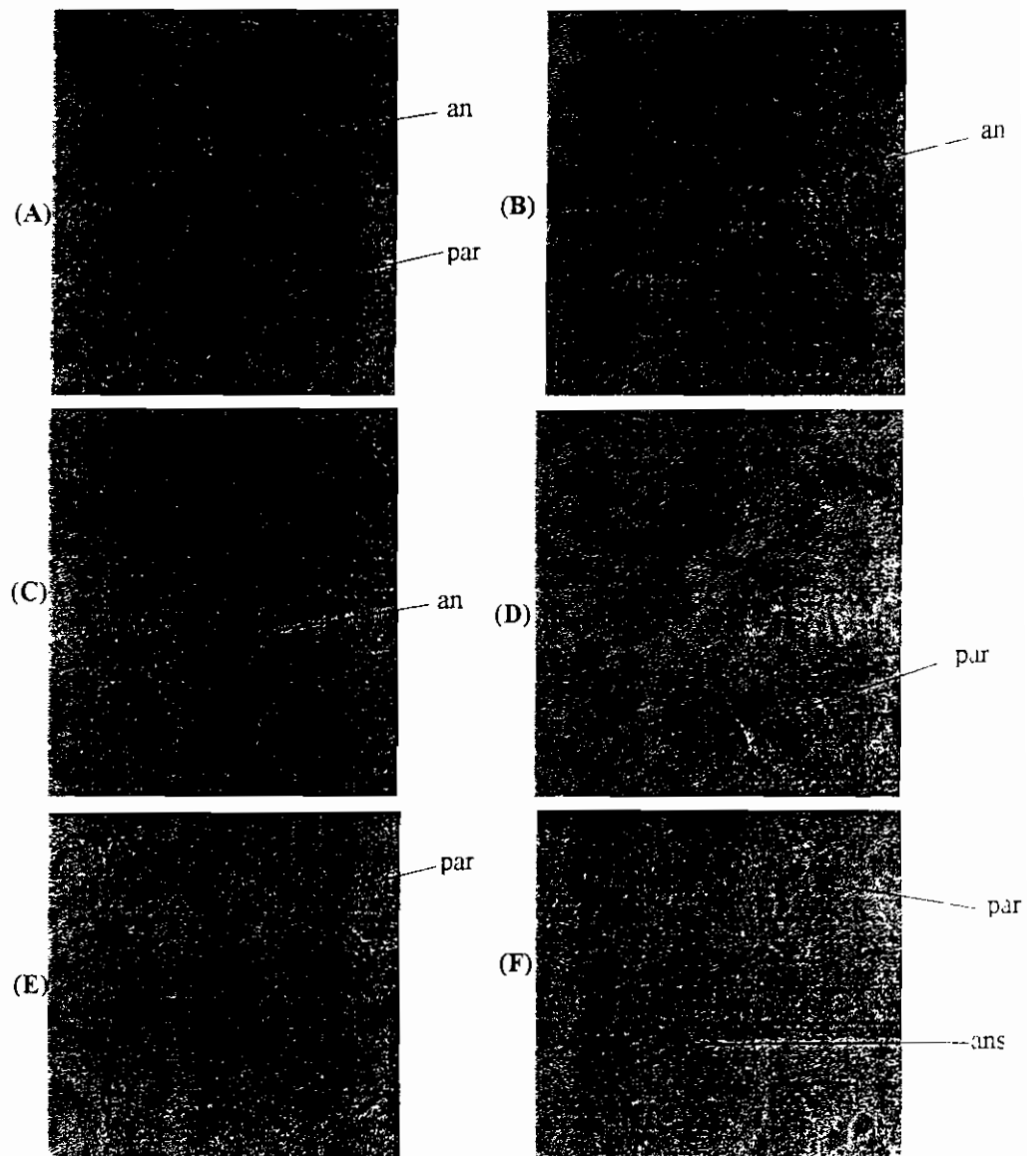


Fig. (2): Types of stomata of the lower epidermis of the leaves; (A) *Phaseolus*; (B) *Gossypium*; (C) *Solanum*; (D and E) upper and lower epidermis of *Ricinus*; and (F) *Convolvulus* (X 400).
Details: an, anomocytic type; ans, anisocytic type and par, paracytic type.

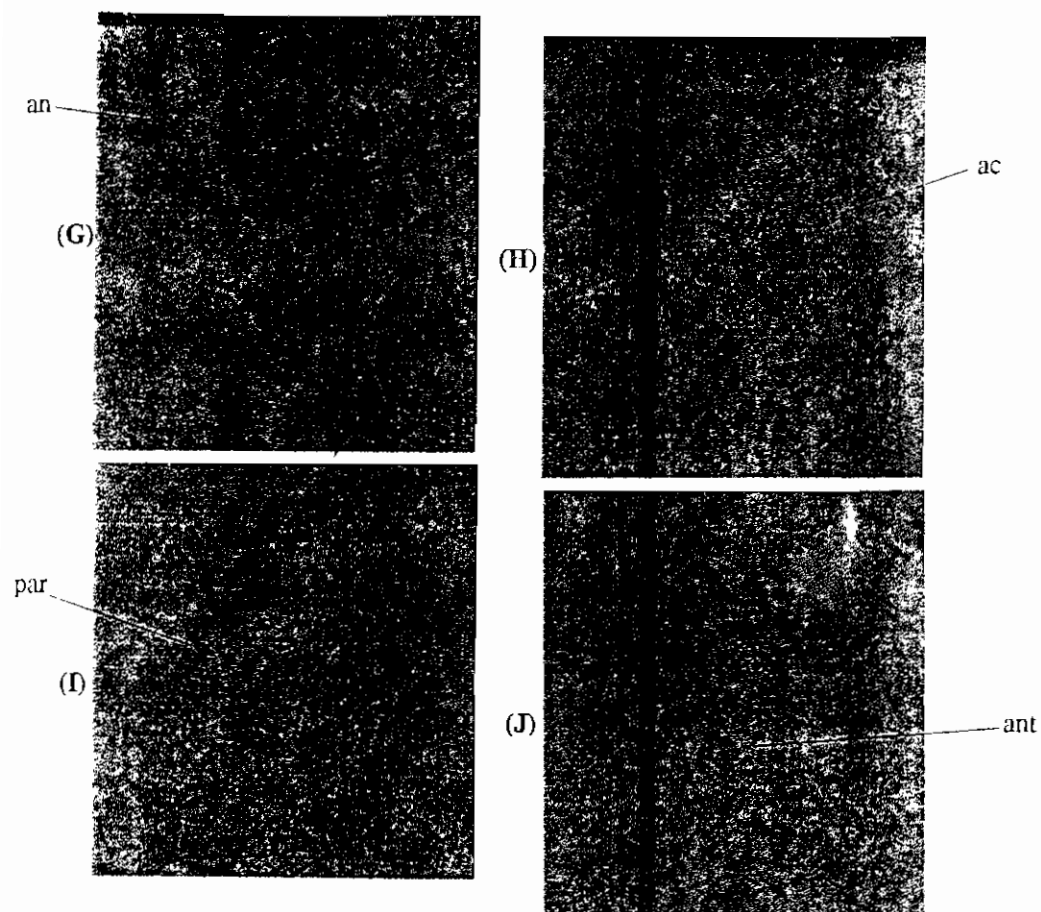


Fig. (2): Cont.

(G) *Prunus*; (H) *Malus*, (I) *Plumeria* and (J) *Citrus* (X400).

Details: ac, actinocytic type; an, anomocytic type; ant, anomotetracytic type.

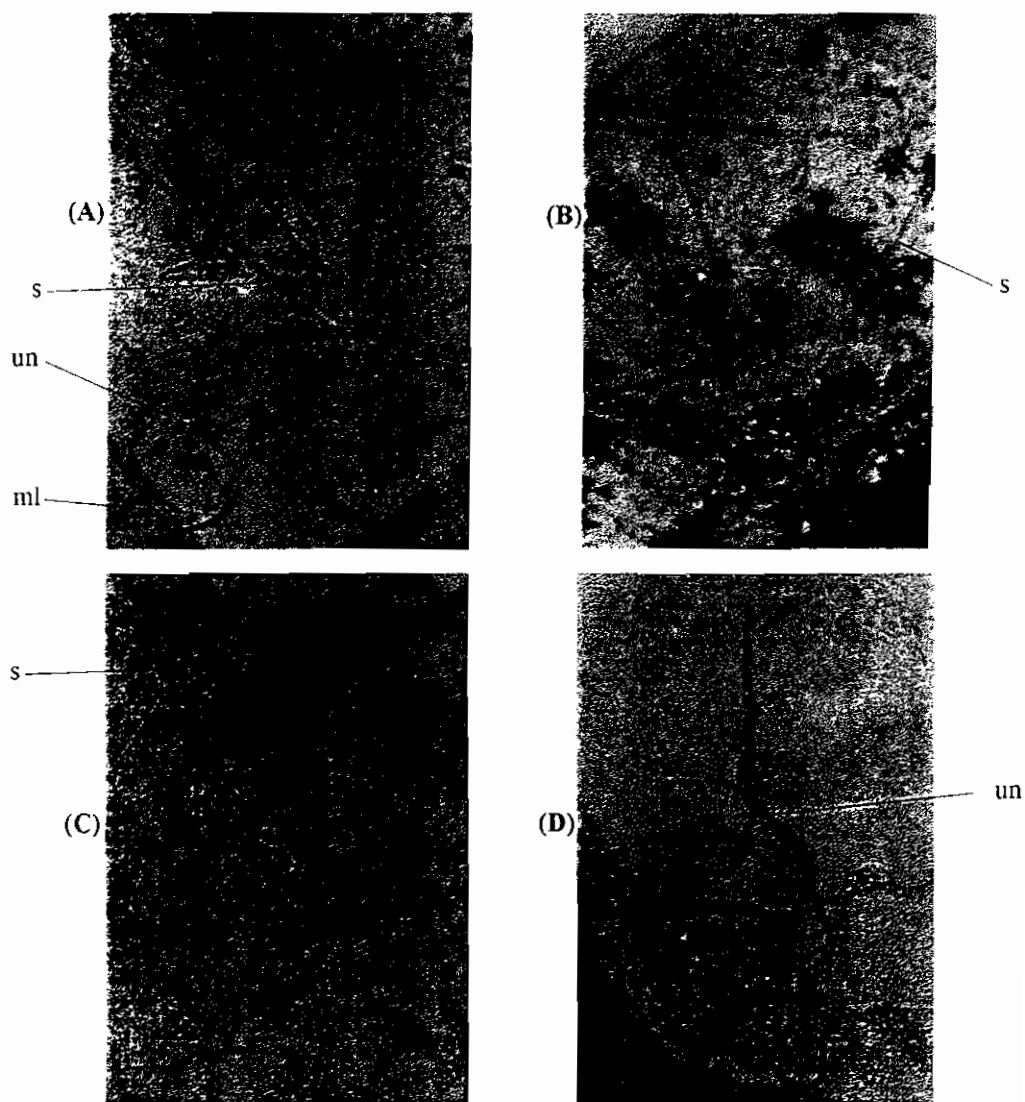


Fig. (3): Different types of epidermal trichomes (X100)
(A) *Phaseolus*, (B) *Gossypium*, (C) *Solanum*, (D) *Malus*
Details ml, multicellular hair; s, stellate hair; un, unicellular hair;

As to the average numbers of stomata (S)/mm², *Citrus* plants recorded the highest numbers (44S/mm²). Since the *Citrus* leaves seem to be the preferable source for feed and reproduction of *Eutetranychus*. Measurement of *E. orientalis* body length and width (60.0 and 57.04 μ) length of legs, (51.16, 42.88, 44.76 and 55.54μ), and stylet 10.16μ, respectively. Generally, for *E. orientalis*, the upper smooth leaf surface is one of the most suitable characters for development and reproduction.

Regarding the correlation between the two studied mites, *T. urticae* and *E. orientalis* and leaf sugar contents, it was found that plant leaves suitable for *T. urticae* development and reproduction contained moderate percentages of total reducing and non-reducing sugars averaging 6.2 – 10.35 %, 2.6 – 5.4 % and 3.30 – 5.45 % in *Ph. Vulgaris*, *G. barbadense* and *S. melongena* respectively (Table 4). This hypothesis, might be applied to *E. orientalis* as its suitable hosts *C. aurantium* and *P. obtuse* also had moderate sugar percentages (Table 4).

Table (4): Concentrations of total, reducing and non-reducing sugars of the leaves of the nine studied genera.

Contents Genera	Total sugars %	Reducing sugars %	Non-reducing sugars %
<i>Phaseolus vulgaris</i>	6.30	2.60	3.70
<i>Gossypium barbadense</i>	6.20	2.90	3.30
<i>Solanum melongena</i>	10.35	5.40	5.45
<i>Ricinus communis</i>	3.50	1.80	1.70
<i>Convolvulus arvensis</i>	31.60	18.80	12.80
<i>Prunus persica</i>	18.04	9.50	8.54
<i>Malus sylvestris</i>	26.60	15.20	11.40
<i>Plumeria obtuse</i>	7.88	3.50	4.38
<i>Citrus aurantium</i>	8.60	3.80	4.80

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تأثير بعض الصفات التركيبية لورقة النبات على دورة حياة وتكاثر نوعين من الحلم التترانيكيدي.

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قسم النبات -كلية الزراعة -جامعة القاهرة -جيزة مصر
قسم الحيوان والنيماتولوجيا الزراعية -كلية الزراعة -جامعة القاهرة -جيزة مصر**

تهدف الدراسة الحالية إلى تأثير بعض الصفات الطبيعية والكيميائية لورقة العائل النباتي مثل سمك الكيوتيكل، الشعيرات المتواجدة على سطح الورقة و التي ربما تؤثر على بيولوجي نوعين من الحلم التترانيكيدي؛ وتترانكس يورتيكا، يوترانكس اورينتاليس . واتضح من الدراسة أن نباتات الفاصوليا والقطن والباذنجان من العوائل المفضلة للاكاروس تترانكس يورتيكا، حيث أسرعت دورة الحياة (9.93، 11.00، 11.37) يوماً (وأطالت فترة حياة الأنثى الكاملة) (15.27، 18.50، 12.24 يوماً) وزادت من تكاثرها (100.34، 70.40، 63.34 بيضة) على التوالي .الحلم تترانكس يورتيكا ويفضل السطح السفلي للنبات وهذا قد يكون راجعاً إلى قلة سمك طبقة الكيوتيكل بالمقارنة بسمكها في النباتات الأخرى غير المفضلة حيث بلغت (1.6) ميكرون .(كما يفضل أيضا النباتات ذات الشعيرات الكثيفة وقد يكون هذا مرتبطاً بالتركيب المورفولوجي للاكاروس مثل ضيق الجسم وطول الفكوك الأبرية. وعلى الناحية الأخرى وجد أن اللارنج، والياسمين الهندي من العوائل المفضلة للاكاروس الأخر يوترانكس اورينتاليس حيث أسرعت دورة الحياة (11.02، 11.48 يوماً) وأطالت فترة حياة الانثى الكاملة (15.40، 12.00 يوماً) وزادت من خصوبتها (25.40، 22.34 بيضة) على التوالي .وتبين أن السطح العلوي للموالح يعتبر من الأسطح المفضلة وهذا ربما يرجع إلى زيادة سمك طبقة الكيوتيكل العليا (3.1 ميكرون)ومن هذا يتضح أن الأسطح الملساء تعتبر العوائل المفضلة لتطور وخصوبة الاكاروس يوترانكس اورينتاليس.

هذا وبتحليل الكربوهيدرات والسكريات المختزلة وجد أن النباتات المفضلة فسي كلا الاكاروسين احتوت على نسبة متوسطة من كل منها.