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# Effect of feeding by two *Neochetina* species or infestation with *Tetranychus urticae* Koch on histological structure of water hyacinth leaves

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## ABSTRACT

Both weevils, *Neochetina bruchi* (Hustache) and *Neochetina eichhorniae* Warner feed on water hyacinth plants as a biocontrol agents. *Tetranychus urticae* Koch infested the plants grown under light and thermostatic controlled aquatic weed green house, and caused repellency of the weevils. Studying some histological structure, of water hyacinth leaves infested with mites, showed increase of the thickness of upper epidermis, number of palisade cell and thickness of leaf petiole, compared with the control and leaves infested with weevils which recorded decrease in the thickness of leaf petiole and increase in lower palisade tissue. The mites infested plants lost chlorophyll, turned brownish and lost size of vacuoles or air chambers used to store the oxygen. The volume of these vacuoles was very small in case of mite infested plants compared with leaves infested with weevils as the same as control. The weevils feeding reflected in increase in both, length and width of fed upon leaves.

**Key words:** Water hyacinth, Histological, *Neochetina bruchi*, *Neochetina eichhorniae* , *Tetranychus urticae*, Egypt.

## INTRODUCTION

Water hyacinth creates serious problems in Egypt. The weed not only affects irrigation, water use, and navigation but it also poses health risks by enabling the breeding of mosquitoes and schistomes (bilharzias) and other human parasites. *Neochetina buruchi* (Hustache) and *Neochetina eichhorniae* (Warner) (Coleoptera: Curculionidae) are two monophagous weevils species, specialized in feeding on the most serious aquatic weed *Eichhorniae crassipes* (Mart) Solms (Fayad, 1982, Fayad *et al.* 2007 and Julien 2001). The adults of these weevils put their eggs in leaves and feed on leaves and petiole causing feeding spots. It was found that *Tetranychus urticae* Koch (Acari:Tetranychidae ) infested water hyacinth plants, grown under light and thermostatic controlled aquatic weed green house caused repellency of these weevils. It was thought that the mite infestation affected the plants to produce some chemicals forcing the weevils not to feed. Family Tetranyidae includes several mite species that infest a wide variety of field crops. They suck the plant sap by inserting their mouth styles through the leaf cuticle and the epidermis. El-Zoghby *et al*. (2008) found that *T. urticae* when infested water hyacinth plants caused changes in leaf contents. Thus it was found to study, some physical and histological structures of the plants leaves. Thus the physical properties of leaves include factors of which the thickness of cuticle layer might affect the mite feeding process. The object of this work was study to the thickness of leaf, thickness of petiole and dimension of leaf.

## MATERIAL AND METHODS

**Samples of leaves infested with mites and insects:**

Three samples of water hyacinth leaves were taken from greenhouse of aquatic weeds of the Department of Biological Control. First sample was infested with *T. urticae*, second sample fed upon with both weevils *N.eichhornia* and *N. bruchi* to be compared with fresh healthy leaves as control as third.

**Anatomical studies:**

Specimens of leaves of water hyacinth, *E. crassipes (*1cm$^2$) were taken from the middle of the lamina including the midrib, while leaf petiole samples were taken from the nearest part of the lamina (2 cm). Specimens were fixed for at least 48 hrs in F.A.A. (10 ml. formalin, 5 ml glacial acetic acid and 85 ml. ethyl alcohol 70%). The selected materials were washed in 50% ethyl alcohol, dehydrated in normal butyl alcohol series, embedded in paraffin wax of 56°C melting point (Sass 1956), sectioned to a thickness of 20 µm, stained with crystal violet erythrosine, then mounted in Canada balsam on glass slides for examination (Nassar and El–Sahhar, 1998). Measurements (µm) of the different tissues were taken and averages of 10 readings were calculated. Examined slides were photomicrographed Microscope with 10 x ocular and 10x, 40 objectives was used for all observations. Anatomical studies were conducted at the Botany Laboratory, Factuality of Agriculture, Cairo University.

Obtained data were statistically analyzed using ANOVA in SAS (Anonymous, 1988). Means separation was conducted using L.S.D. in the same program.

## RESULTS AND DISCUSSIONS

It is obvious from the Table (1) and Fig.(1) that, both plants infested with mites or insects showed an increase in the thickness of upper and lower epidermis of the leaf lamina by 38.9 and 8.9% for mites, and 22.2 and 9.9% for insects compared with control. Fig. (1) clears that the increase of the thickness of upper palisade tissue (9-12) and lower (5-6) layers in case of mites because it increase in the number of the cells. and (The percentage of measurements were recorded for upper and lower palisade tissue, where mites infected plants showed an increase by 35.6 and 70.9%, while insect infested ones recorded 34.4 and 91.0%, compared with control plant (F=192.00 and P = 0.0001) and (F= 686.49 and P= o.ooo1) respectively. This increase of palsied tissue easily penetration of mites and insects. Qaisar *et al*. (2005) mentioned that palisade layer is present on both upper and lower (5-7) layers of cells the lower side beneath the epidermis ones has (2-3) layers. Concerning the lamina thickness, it was found that mites infested one recorded a decrease by 24.8% compared with control plants, while insect infested plants recorded a decrease by 14.2% (F= 56.71 and P= 0.0001). Concerning the measurements, of thickness leaf petiole epidermis (Table1) and Fig.(2), the thickness of epidermis showed an increase by 18.4% for mites infested plants, while that of infest insects recorded a decrease by 12.6%, compared with the control.(F= 161 and P= 0.0001). Concerning the dimensions of the petiole, both mites and insects plants measurements recorded an increase by (9.5 and 1.5%) and (29.6 and 32.1%) for length and width; respectively (F= 132.27 and P= 0.0001), (F= 195.92 and P= 0.0001). Decrease of leaf petiole epidermis thickness giving way of insects could be penetration through it because the lower compared with control, and the insects infested leaf petiole most of leaf. Fig. (2) clears that the leaf petiole of water hyacinth plants, consists of epidermis and

compacted parenchyma and showed very dark stains throughout these cells which are responsible for various metabolic activities in the plants (Rekha and Saroja 2008), there was an increase in case of weevils infestation (C) compare with (B) leaf infested with mites. The leaf of water hyacinth plants infested with mites turned into yellow, bronzing, which affected photosynthesis in the plants as loss in chlorophyll content (Reddy and Baskaran,1991; and Haq and Sumangala 2003) . The result of decreased photosynthesis, result in reduction of carbohydrates and crude protein (El-Zoghby *et al*. 2008). Fig. (2) shows occurrence of vacuoles (air chamber) which are very large in case of weevils (C) compared with mites (B) being pressed and small. The function of these vacuoles is storing oxygen used again in respiration and carbondioxide from respiration, used in photosynthesis (Qaisar *et al*. 2005).

Table (1): Measurements of some histological characters in transverse sections through the leaf lamina and petiole of healthy and infected *Eichhornia crassipes* (Marts) Solms. Plants (with mites and insects) (Averages of 10 readings).

| **Characters** / **Treatments** | | **Control** | **Mites (±% to control)** | **Weevils (±% to control)** | **L.S.D.** | **F Value** |
|---|---|---|---|---|---|---|
| Thickness of leaf: | | | | | | |
| - Upper epidermis | (µm) | 34 c | 47.5 a (+38.9) | 41.8 b (+22.2) | 1.6448 | 204.24 |
| - Lower epidermis | (µm) | 21.3 b | 23.2 a (+8.9) | 23.4 a (+9.9) | 0.9371 | 18.32 |
| - Upper palisade tissue | (µm) | 132.9 b | 180.2 a (+35.6) | 178.6 a (+34.4) | 6.5759 | 192.00 |
| - Lower palisade tissue | (µm) | 45.7 c | 78.1 b (+70.9) | 87.3 a (+91.0) | 2.886 | 686.49 |
| - Lamina | | 534.4 b | 401.6 b (-24.8) | 458.2 a (-14.2) | 17.292 | 56.71 |
| Thickness of leaf petiole epidermis | (µm) | 23.9 b | 28.3 a (+18.4) | 20.9 c (-12.6) | 1.0122 | 161.92 |
| Dimensions of leaf petiole: | | | | | | |
| - Length | (µm) | 3343.8 b | 3662.5 b (+9.5) | 4333.3 a (+29.6) | 151.97 | 132.27 |
| - Width | (µm) | 5125.3 b | 5200.4 b (+1.5) | 6770.8 a (+32.1) | 229.7 | 195.92 |

Means in the same row not followed by the same letter are significantly different (P= 0.0001).

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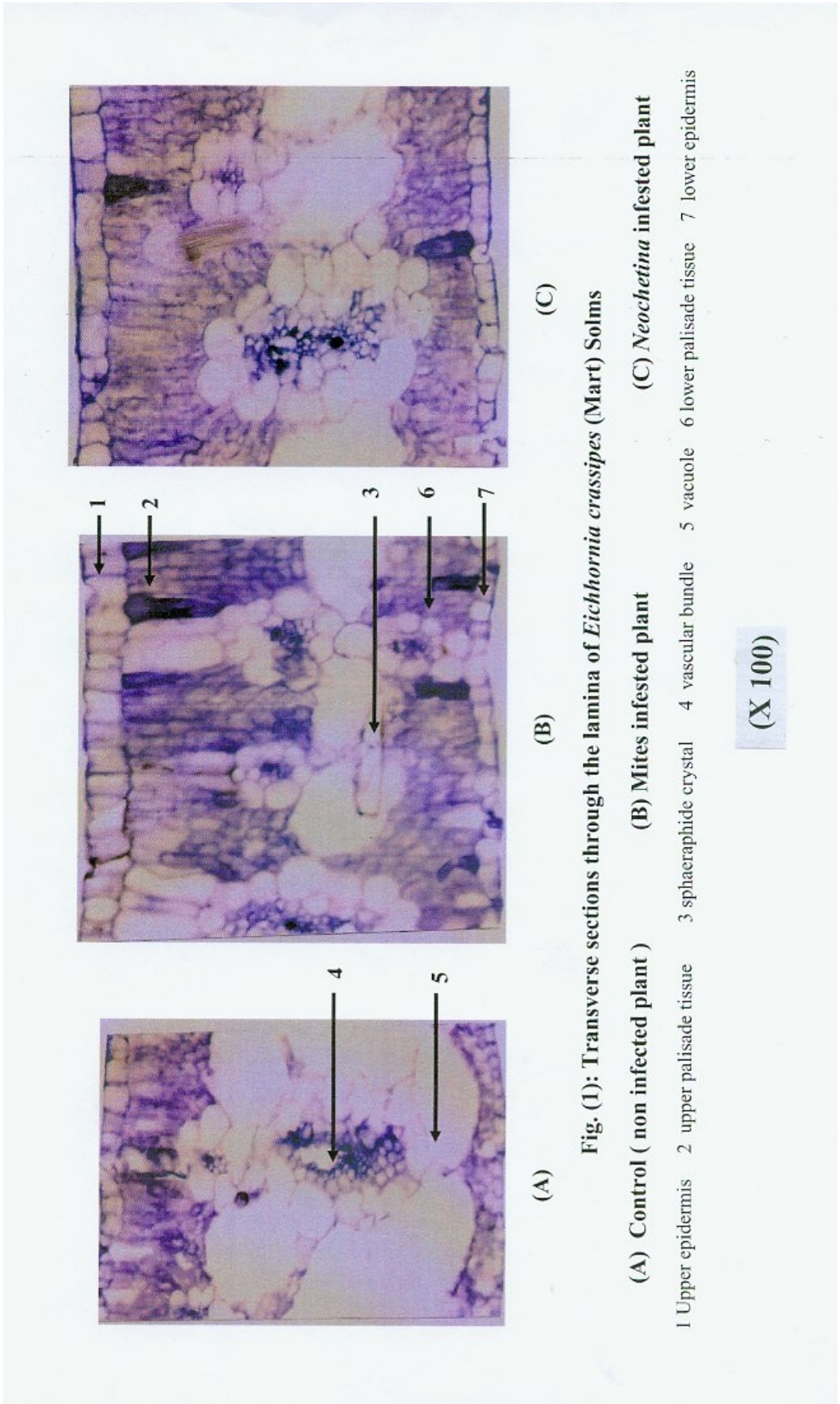


**Fig. (1): Transverse sections through the lamina of *Eichhornia crassipes* (Mart) Solms**

**(A) Control ( non infected plant )** **(B) Mites infested plant** **(C) *Neochetina* infested plant**

1 Upper epidermis 2 upper palisade tissue 3 sphaeraphide crystal 4 vascular bundle 5 vacuole 6 lower palisade tissue 7 lower epidermis

**(X 100)**

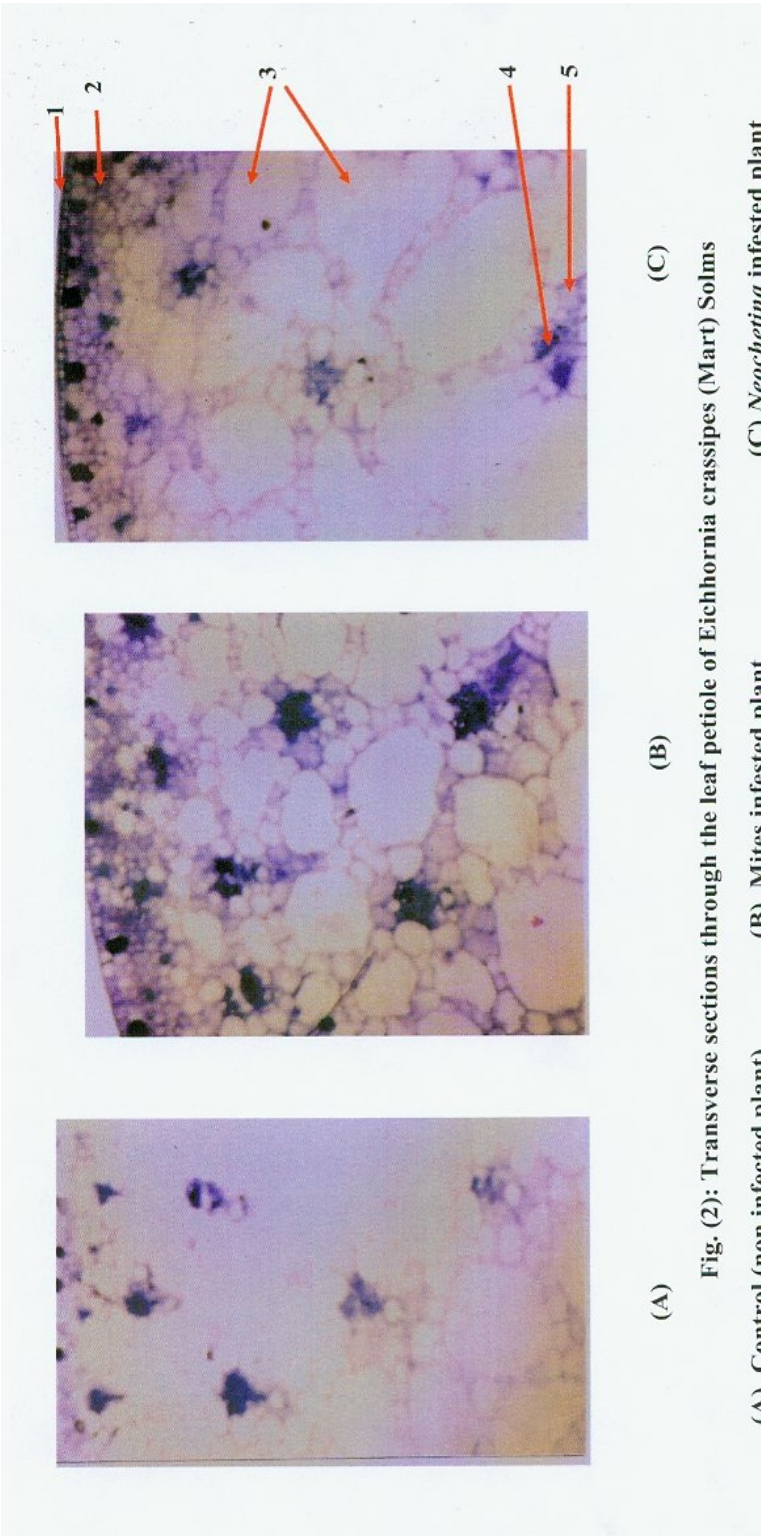


**Fig. (2): Transverse sections through the leaf petiole of Eichhornia crassipes (Mart) Solms**

**(A) Control (non-infested plant) (B) Mites infested plant (C) *Neoacheting* infested plant**

## ARABIC SUMMARY

**تأثير التغذية بنوعين من السوس واصابة العنكبوت الآحمر على بعض التراكيب الهستولوجيه لأوراق نبات ورد النيل**

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تستخدم حشرات النكوتينا بروكى والنكوتينا ايكورنى كعوامل مكافحه بيولوجيه لنباتات ورد النيل حيث انهما متخصصتان فى التغذية علية ويتم تربية هاتين السوستين داخل صوبة الحشائش المائية تحت تحكم فى درجة الحرارة والاضاءة. وعند اصابه العنكبوت الاحمر (تترانكس يوريتيكا) نجد ان الحشرات ترفض البناتات المصابة او السابق اصابتها وتتركها الى نباتات اخرى سليمه.

عند دراسه بعض التراكيب الخاصه بالاوراق المصابه بالحشرات والاخرى المصابه بالعنكبوت الاحمر **مقارنة بالكنترول وجدنا الاتى:** العنكبوت الاحمر يزيد من سمك وعدد خلايا البشره العليا و ايضا يزيد من سمك النسيج العلوى و يزيد من سمك نسيج أعناق الأوراق–ايضا يؤثر على عمليه البناء الضوئى عن طريق فقد الكلوروفيل – فقد فى حجم الغرف الهوائيه التى تحتفظ بالاكسجين ولكن الاصابة الحشرية تؤدى الى ذيادة سمك **الطبقة** السفلى من النسيج و ايضا زيادة فى كلا من عرض و طول الورقة مقارنة بالكونترول.