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ANTIFEEDING EFFECT AND CHRONIC TOXICITY OF THREE DESERT PLANTS AGAINST THE EGYPTAIN LOCUST, *Anacridium aegyptium* L.

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ABSTRACT

Feeding of fifth instar nymphs of *Anacridium aegyptium* L.on three desert plants; *Calotropis procera* (shrub), *Pulicaria crispa* (bush) and *Zygophyllun simplex* (annual herb) compared with control plant (wheat-seedlings) indicate that the rate of consumption and assimilation of nymphs decreased on the desert plants as compared with wheat seedling. All nymphs fed on the desert plants stop feeding for long time before death The results indicate that the desert plants used had an antifeeding effect and also chronic toxicity. The electrophoresis analysis showed that the protein bands of the nymphs fed on control plant were 16 bands but with those fed on the desert plants it ranged from 14 to 16 bands.

INTRODUCTION

Polyphagous insect species may feed on a great diversity of plant species but certainly do not indiscriminately accept all green plants, *Schistocerca gregaria* (Forsk.), which feed on a wide range of plants belonging to many different families, exhibits pronounced preferences for particular plants, eating some species in small amounts and others in large amounts (Chapman, 1990).

Individuals of the same insect species may show different host plant preferences in different areas of its distribution and even individuals bolonging to the same population may be much more restricted in their choices than the population as a whole (Howard *et al.*, 1994). Some oligophagous or even polyphagous insects should more appropriatoly be considered as monophagous when their host-plant selection is based upon a specific type of plant chemicals (Klausnitzer, 1983).

The ecological triangle of insects host plant and allelochemical constitutes a relationship with profound evolutionary implications. Specialist herbivorous are frequently limited to plants that constitute ``forbidden fruits`` for other phytophagous because these plants contain natural products that are unpalatable and/or toxic for most animall (Fraenkel, 1959). Fourth instar nymphs of *S. gregaria* consumed and assimilated *Schouwia purpurea* just like the wheat seedlings diet. The limited adaptation of the desert locust to plant glucosinolates is compensated by an ability to tolerate high concentrations of allelochemicals for a short period (Mainguet *et al.*, 2000). The quantity of protein available for gorwth must depend not only on upstream processes, such as the rate of consumption of protein and the rate and location of protein break down within the mid-gut, but also on the capacity of the mid-gut to transport digested protein into the body (Arthur and Chamberlin, 1999).Good assimilation of plant protein depends on the type of

allelochemicals in the host plant (Hinks *et al.*, 1993). Significant reduction in haemolymph protein was noted in females of *Euprepocnemis plorans* (Charp.) reared on lupine or horsebean (Elsayed, 1998). Protein bands in the lanes of *E. plorans* nymphs fed on lupine or horse bean is lower as compared with those fed on clover (Elsayed and Elshabrawy, 2002).

This study aimed to evaluate the antifeeding action and/or toxicity of three desert plants to the Egyptain locust.

MATERIALS AND METHODS

Egg masses of *A. aegyptium* were incubated in a closed jar at 30-33 °C until hatching. Nymphs were maintained in rearing cages measuring 33×33×55 cm at 30 °C ,50-70 %RH, and a 12:12 (L:D) cycle. Nymphs and adults were supplied with fresh wheat seedlings, yeast powder and dry wheat bran (Elsayed, 1994).

For quantitative nutritional determination, standard gravimetric techniques of Waldbauer (1968) were followed. Six females were reared on freeze-dried plants of *.C. procera* or *P. crispa* or *Z. simplex* and compared with wheat seedlings as control. On the day of moulting each fifth instar nymph received a third of its body weight of food. The food was replenished every day to maintain the initial quantity all the time . The six nymmphs were kept in a 1 L glass jar covered with wire gauze for ventilation, and containing wet cotton wool to provide water (Ben –Halima *et al.*, 1983) A petri dish for the dry food and a bamboo stick as a moulting support were provided . Each day, faecal pellets were collected from the jar and kept in a deep freezer. At the end of experiment all remains (faecal pellets and uneaten food) were freeze-dried weighed to calculate nutritional indices. To estimate the dry weight of the nymphs, five newly moulted fifth instar nymphs were weighed then dried, and re-weighed to calculate their water content. At the end of experiment newly emerged adults were dried to determine their dry weight. The following seven nutritional indices were calculated according to Elsayed (1998).

Fecundity = Total number of eggs per females.

$$\text{Ovariole yield \%} = \frac{\text{No. of eggs per pod}}{\text{No. of ovarioles per females}} \times 100$$

$$\text{Fertility \%} = \frac{\text{No. of hatched eggs per pod}}{\text{No. of deposited eggs per pod}} \times 100$$

$$\text{Decrease in R.P.\%} = 100-\left(\frac{a \times b}{A \times B} \times 100\right), \text{ where}$$

A= mean egg hatch/control B = mean egg laid / control
a= mean egg hatch / treated b = mean egg laid / treated

$$\text{Phase dependent \%} = \frac{a}{(a+b)} \times 100 \quad \text{or} \quad \frac{b}{(a+b)} \times 100$$

a = main total treatment of solitary b = main total treatment of gregary

Percentage of reduction from control = $\frac{(a-b)}{b} \times 100$

a= treatment b= control

Electrophoresis analysis was carried aiming to identify the general protein pattern in the whole body of the 3-5 old days of fifth nymphs fed on *S. procera*, *P. crispa* or *Z. simplex* and wheat seedlings as control plant. Sampling stock solutions, protein extraction by SDS polyacrylamide gel electrophoresis (SDS PAGE) and detection of the proteins were carried according to the method of Al-Akkad (1997).

RESULTS AND DISCUSSIONS

Consumption and assimilation of fifth instar female nymphs of *A. aegyptium* are presented in Table (1). Locust in this instar consumed and assimilated more wheat seedlings than the three desert plants, *C. procera* or *P. crispa* or *Z. simplex*,' respectively . All nymphs tested on the three desert plants died before moulting to adult females, and their instar periods were longer than those fed on wheat seedlings which moulted naturally to immature adult females .

Table(1): Consumption and assimilation of *Anacridium aegyptium* fifth instar nymphs on wheat seedlings, *Calotropis procera*, *Pulicaria crispa* and *Zygophyllum simplex*.

Food plants	Initial food	Uneaten food	Consu-mption	Assim-ilation	Weight grain	Duration instar (days)	Mortality %
wheat seedlings	2809.5	73.2	2736.3	829.7	128.2	9.0	0.0
C. procera	300.0	120.0	180.0	45.0	0.0	19.6	100.0
P. crispa	600.0	180.0	420.0	118.0	0.0	15.0	100.0
Z. simplex	800.0	200.0	600.0	180.0	0.0	17.6	100.0

Nutritional indices for fifth instar female nymphs of *A. aegyptium* were calculated only for the nymphs fed on wheat seedlings, approximate digestibility (AD) was 30.3%, efficiency of conversion of digested food (ECD) was 4.7% and efficiency of conversion of ingested food (ECI) was 15.5%. But the nutritional indices for the nymphs fed on the three desert plants was 0.0% because all nymphs died before emerging to immature adults (Table 2)

Differences of nymphs body protein fed on wheat seedlings and those fed on the different desert plants, were studied using the gel electrophoresis technique (SDS-PAGE) with a high molecular weight protein marker of 205 and 30 KD.

By examining the protein bands for the fifth instar nymphs shown in Table (3) and Fig (1). It could be concluded that the lane of nymphs fed on wheat seedlings, contained 16 bands ranging from 234 to 23 KD, while the lane of nymphs fed on *C. procera* contained 15 bands ranging from 234 to 23 KD, the lane of nymphs fed on *Z. simplex* contained 14 bands ranging from 145 to 23 KD and the lane of nymphs fed on *P. crispa* contioned 16 bands ranging from 256 to 23 KD.

Table (2): Nutritional indices of of *Anacridium aegyptium* on wheat seedlings, *Calotropis procera, Pulicaria crispa* and *Zygophyllum simplex.*

Food plants	AD%	ECI%	ECD%	GR	RGR	CR	RCR
Wheat seedlings	30.3	4.7	15.5	4.98	0.019	106.2	0.41
C. procera	0.0	0.0	0.0	0.0	0.0	0.0	0.0
P. crispa	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Z. simplex	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table(3). Molecular weight (M.W.) of different protein bands in Egyptian locust fed on different plant foods.

Wheat seedlings		*C. procera*		*P. crispa*		*Z. simplex*	
Band	M.W.	Band	M.W.	Band	M.W.	Band	M.W.
1	234	1	234	1	256	1	-----
2	200	2	207	2	207	2	-----
3	174	3	184	3	-----	3	-----
4	----	4	154	4	-----	4	-----
5	142	5	142	5	145	5	145
6	132	6	-----	6	137	6	136
7	124	7	123	7	128	7	127
8	------	8	-----	8	120	8	118
9	115	9	116	9	113	9	-----
10	109	10	-----	10	-----	10	109
11	-----	11	89	11	-----	11	-----
12	-----	12	82	12	82	12	-----
13	-----	13	-----	13	79	13	78
14	76	14	74	14	75	14	75
15	73	15	-----	15	71	15	-----
16	68	16	69	16	-----	16	-----
17	58	17	59	17	59	17	59
18	51	18	52	18	52	18	52
19	47	19	-----	19	-----	19	49
20	-----	20	-----	20	41	20	42
21	-----	21	-----	21	-----	21	38
22	31	22	32	22	32	22	32
23	23	23	23	23	23	23	23

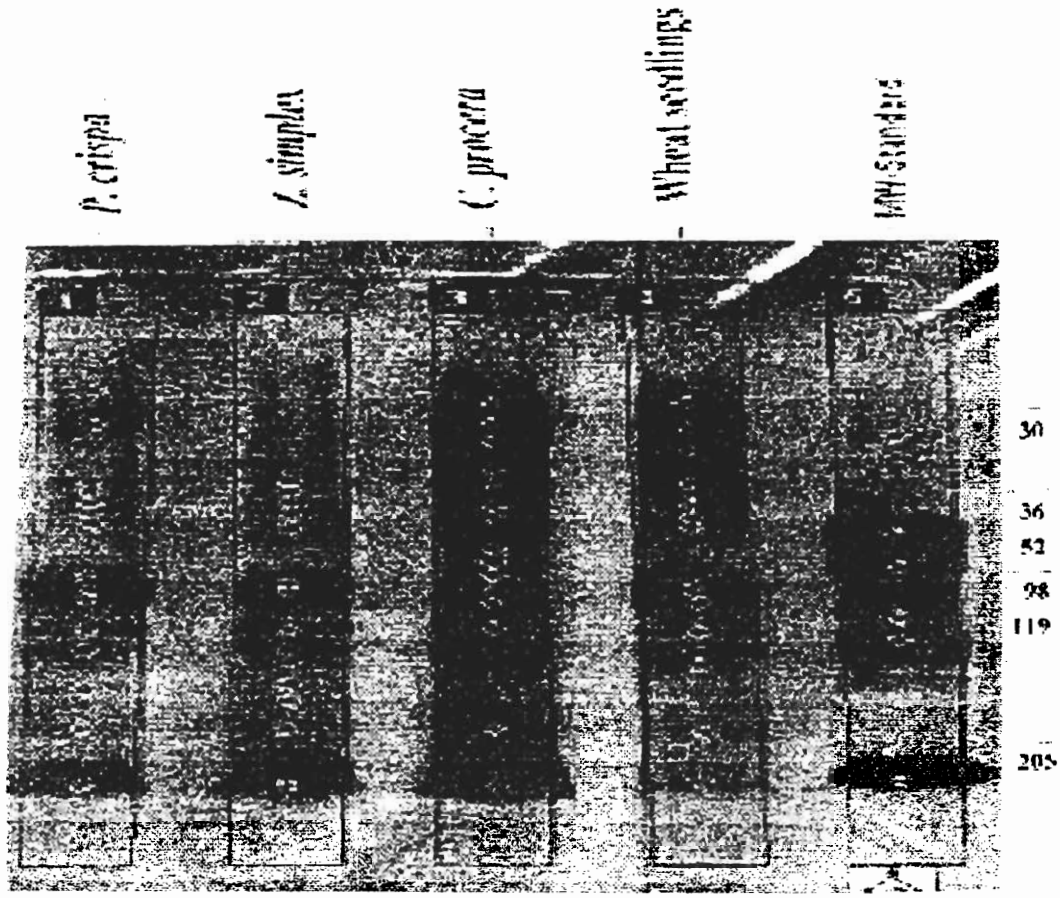

Figure (1). Haemolymph proteins from fifth instar nymphs fed on wheat seedlings, *P. crispa*, *Z. simplex* and *C. procera* were electrophoresis on 5% staking gel and 12% separating gel by used 20 µgm from protein samples and the volume of standard protein was 15 µgm.

It is noticed that one protein band is constant in the four lanes, the molecular weight of this band is 23 KD. But another protein bands in the four lanes are different.

Two protein bands are constant only in the lane of nymphs fed on wheat seedlings and those fed on *C. procera*, the molecular weight of these bands are 234 and 142 KD. One protein band is constant only in the lanes of the nymphs fed on wheat seedlings and those fed on *Z. simplex*, the molecular weight of this band is 109 KD. Three protein bands are constant in the lanes of the nymphs fed on the three desert plants, the molecular weight of these bands are 32,52 and 59 KD. One protein band is constant in the lane of nymphs fed on *Z. simplex* or *P. crispa*, the molecular weight of this band is 145 KD.

Feeding the fifth instar nymphs of Egyptian locust on wheat seedlings resulted in higher consumption and assimilation with shorter duration of development compared with nymphs fed on the three desrt plants, *C. procera* or *P. crispa*, or *Z.. simplex*, before they died. These results may be due to the compounds naturally found in these desert plants, these compounds have had an effect on the digestive enzymes. Approximate digestibility (AD) for second instar grasshopper, *Melanoplus sanguinipes* (F.) fed on oats was significantly lower than the ADs of those fed on wheat or kochia (Hinks and Erlandson, 1995).

Oak tannins inhibit the digestive enzymes in a herbivores gut (Feeny, 1969).

Phenolic acid in Oats can be oxidized to from quinines which react with proteins to produce derivatives with reduced nutritional value (Hinks and Hupka, 1995).

Azadirachtin inhibits the production of trypsin in mid-gut of *Manduca sexta* (Timmins and Reynold, 1992); glucosinolates in *Schouwia purpurea* may inhibit trypsin activity in the gut of *S. gregaria* (Elsayed, 1994), also Elsayed and Elshabrawy (2002) found that, quinolizidin alkaloid or non-protein amino acids in lupine or horse bean have had an effect on the trypsin activity of *E. plorans*. Thomas (1989) reported that the low consumption of red and black spruce by the spruce bud worm, *Choristoneura fumiferana*, may be due to antifeeding and toxic compounds in the diet. Glucosinolates induced gut-irritant effects on the migratory locust, *Locusta migratoria danica* L. (Cotte *et al.*,1988).

Nymphs fed on three different desert plants stay longer time without feeding before death, this result may be attributed to the chronic toxicity on the digestive enzymes caused by allelochemicals in this food. The nymphal development period was prolonged in the grasshopper. *E. plorans* fed on horse bean, which may be considered as another aspect of mal-assimilation of diet (Elsayd,1998).

S. purpurea leaves containing glucosinolates caused chronic toxicity to *S. gregaria* (Elsayed,1994). The percentage of mortality in the nymphs fed on the different desert plants was 100%, this result confirmed that these plants have the allelochemicals which caused the antifeeding and toxic effects After feeding of *E. plorans* on horse bean or lupine, the mortality was increased (Elsayed,1998).

Disappearance of one or two molecular weight of protein bands in the lanes of the nymphs fed on the three desert plants as compared with wheat seedlings, may show that, the plant compounds have had an effect on the protein synthesis. And this result is confirmed by the differences found in the molecular weight of protein bands in the lanes of the nymphs fed on desert plants compared with control plant. The reduction of total proteins and proteins band in the heamolymph of *S. gregaria* fed on *S. purpurea* than those fed on wheat seedlings may be attributed to the glucosionolates finding in plant (Elsayed,1994). Total protein in whole gut homogenates was shgnificantly higher in kochia fed grasshopper in the second instar and significantly lower in those fed on oat in the fifth instar (Hinks and Erlandson, 1995). Six proteins with different molecular weights were separated from

oocytes of *Melanoplus sanguinipes* (F) fed on oats (Hinks *et al.*,1993). Four molecular weights of protein bands disappeared when *E. plorans* nymphs were fed on horse bean or lupine compared with those fed on clover (Elsayed and Elshabrawy, 2002).

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التأثير المانع للتغذية والسمية المزمنة لثلاث نباتات صحراوية علي الجراد المصري

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أوضحت تغذية العمر الحورى الخامس للجراد المصرى علي ثلاث نباتات صحراوية هي نبات العشار *Calotropis procera* وشجيرات النبات *Pulicaria crispa* والعشب الحولي *Zygophyllum simplex* بالمقارنة بتغذيتها علي نبات القمح كشاهد أن معدل الإستهلاك والإستفادة من الغذاء ينخفض علي النباتات الصحراوية بالمقارنة بنبات القمح. توقفت الحوريات المتغذية علي النباتات الصحراوية عن التغذية لفترة طويلة قبل موتها وهذا يدل علي أن هذه النباتات الصحراوية لها تأثير كمانعات للتغذية وتأثير سمى مزمن. تم دراسة معدلات التغذية علي نبات القمح فقط نظرا لوصول حوريات هذه المعاملة فقط إلي حشرات كاملة، وقد تم عمل فرد كهربي للبروتين فى الحوريات المتغذية علي النباتات الصحراوية بالمقارنة بالحوريات المتغذية علي نبات القمح فلوحظ وجود تأثير علي كمية ونوعية البروتين فى الحوريات المتغذية علي النباتات الصحراوية.