

Effect of three different botanical extracts on carbohydrate and phosphorous contents of treated bed bug *Cimex lectularius* L. (Hemiptera:Cimicidae)

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ABSTRACT

The main target of the present study was carried out to evaluate the effect of three different plant extracts (Lantana camara, Melia azedarach and Agave sisalana) at different concentrations on the carbohydrate and phosphorous contents of treated 3rd, 4th nymphal instars and adults of bed bug Cimex Lectuarius.

Statistical analysis of our results clearly indicate that the total carbohydrate of treated 3rd nymphal instar and adult males with M. azedarach seed extracts at concentrations of, 10 and 20% was significantly decreased. However, significant increasing was occurred when treatment carried out on adult females at concentration of 5 %.

Botanical extracts of M. azedarach leaves were more effective on the total phosphorous content than leave extract of L. camara.

The mean value of homogenate body phosphorous content of 3rd, 4th nymphal instars and adult of both sexes treated with A. sisalana stem extracts was decreased with an increase in the concentration of botanical extracts.

INTRODUCTION

The bed bugs can be vector of many parasites, affecting man such as *Rickettsia burenttii*; the cause of Q-fever; and *Leshmania donovani*, *L. tropica*, the cause of cutaneous leshmaniasis (Oriental sore). They are also suspected to be vectors of the hepatitis-B-virus. Bed bugs have also been shown to become infective when allowed to feed on mammals harbouring the virus of yellow fever

In recent years it has been demonstrated that various insect species are affected in their activity, growth and metamorphosis by treatment with many different botanical extracts.

Plants as sources of active chemical compounds used in combating insects and other injurious arthropods are still the subject of interest of many workers all over the world.

The toxicity of the different botanical extracts tested in the present study was established against 1st, 3rd and 4th nymphal instars as well as the adult stage of *Cimex lectularius* by Younes *et al.*, (2000).

The present work deals with the effect of some botanical extracts of different parts of three plant species belonging to different families (Verbenaceae, Meliaceae and Amaryllidaceae) on total carbohydrate and phosphorous contents of *C. lectularius* treated as nymphs and adults.

MATERILS AND METHODS

Rearing procedure of the bed bug *Cimex lectularius* and extraction procedures of different plants under investigation were described by Younes *et al.*, (2000).

Newly moulted nymphs of 3rd, 4th and adults were treated topically just after moulting with concentrations of 5,10 and 20% of extracts of *Lantana camara* leaves, *Melia azedarach* leaves and seeds. and *Agave sisalana* stems.

Samples were taken 48 hours after treatment of moulted 3rd, 4th nymphs instars and adult to determine total carbohydrate and phosphorous contents.

The total carbohydrate content of the whole body was determined according to Singh and Sinha (1977). To determine the total carbohydrate content, 0.1 ml of the aqueous sample diluted with 1 ml distilled water then treated with 5 ml freshly prepared anthron reagent. Blank was prepared by adding 5 ml of anthron reagent to 1.1 ml distilled water. Standard was prepared by adding 0.1 ml of standard solution to 1 ml distilled water and then treated with 5 ml anthron reagent. All test tube of blank, unknown and standard were placed in boiling water bath 10 minutes at room temperature in a dark place. Reading were made at 620 nm. using colourimeter.

Calculation:

$$\frac{\text{absorbance of the sample}}{\text{absorbance of the standard}} \times \text{concentration of the standard}$$

$$\times \frac{1}{\text{Volume} \times \text{Sample used}} \times \frac{100}{\text{wt of sample homogenized}}$$

$$= \text{mg} = \text{carbohydrate} / \text{mg of fresh body.}$$

To determine the total phosphorous content, methods adopted by Zilversmit *et al.*, (1950) and Bogatzki (1938) were used.

Calculation:

$$\text{Concentration of phosphorous} = 5 \times \frac{\text{A sample}}{\text{A standard}}$$

$$= [\text{mg} / 100 \text{ ml}] = \text{phosphorus} / \text{mg} / 100\text{ml}$$

In order to compare the mean of the various groups, the most appropriate test of the significance was found to be the student's "t" test.

RESULTS**Effect of botanical extracts on the total carbohydrate content :-**

The average of homogenate body carbohydrate content of 3rd, 4th nymphal instars and adults of both sexes treated with extracts of *L. camara* leaves was decreased with increasing the concentration of extracts as shown in Table 1. The mean value of total carbohydrate of 3rd, 4th nymphal instars and adult of both sexes treated with concentration of 5 % was significantly increased, it was 2.26, 2.7, 3.29 and 4.31 mg / ml, respectively, as compared with 2.04, 2.38, 2.84 and 3.36 mg / ml in the case of check insects. The corresponding figures of treated nymphal instars and adults with concentration of 10 and 20 % were significantly decreased as compared with insects treated with solvent (check experiment).

It is quite clear from our results that the treatment of 3rd nymphal instar with extracts of *M. azedarach* leaves at concentrations of 10 and 20 % induced significant decrease in the total carbohydrate content, it was 1.79 and 1.4 mg / ml as compared with 2.04 mg / ml in the case of check experiment.

The statistical analysis of the data in Table 2. indicate that the total carbohydrate content of homogenate body of treated 4th nymphal instar with concentrations of 10 and 20 % and treated adults with 5,10 and 20 % was significantly decreased.

Statistical analysis of result presented in Table 3. indicate that the total carbohydrate content of 3rd nymphal instar and adult males treated with *M. azedarach* seed extracts at concentrations of 5,10 and 20% were significantly decreased as compared with 2.04 and 2.84 mg / ml in the case of check insects. On the other hand, significant increase was occurred when treatment carried out on 4th nymphal instars and adult females at concentration of 5 %.

Results in Table 4. shows a gradual decrease in the total carbohydrate content of homogenate body of treated 3rd, 4th nymphal instars and adult of both sexes with an increase in the concentration of *A. sisalana* stems extracts.

Effect of botanical extracts on the total phosphorous content:-

The mean value of homogenate body phosphorous content of treated 3rd, 4th nymphal instar, and adults of *C.lectularius* with different concentrations of varius botanical extracts (leaves of *L. camara* and *M. azedarach*; *M. azedarach* seeds and *A. sisalana* stems) are presented in Tables 5,6,7 and 8.

Data presented in Table 5. clearly indicate that the total phosphorous content of homogenate body of 3rd, 4th nymphal instars and adults was decreased gradually with an increase in the concentration of *L. camara* leave extracts as compared with check insects ($P < 0.001$).

Data presented in Table 6. showed that the botanical extract of *M. azedarach* leaves was more effective on the total phosphorous content than extract of *L. camara* leaves The statistical analysis of results indicate that the total phosphorous content of 3rd, 4th nymphal instars and adults was significantly decreased when treatment carried out with botanical extracts of *M. azedarach* leaves at different concentrations ($P < 0.001$).

Our results clearly indicated that the total homogenate body phosphorous content of 3rd, and 4th nymphal instars treated with *M. azedarach* seed extracts was decreased as compared with check group. The total phosphorous content was 32.99, 22.75 and 17.78

mg / ml for 3rd nymphal instar and 48.31, 36.08 and 30.14 mg / ml for 4th nymphal instar at concentrations of 5, 10 and 20 % respectively, as compared with 137.56 and 154.28 mg / ml in the case of check groups. The corresponding figures for adult males and females were 54.36, 42.28, 42.72 and 31.94, 24.82, 20.67 mg / ml, respectively, as compared with 172.72 and 118.40 mg / ml in the case of check groups (Table. 7).

When 3rd nymphal instar and adult males treated with botanical extracts of *A. sisalana* stems at concentrations of 5 and 10 %, the total phosphorous content was 89.35, 73.81 and 110.97, 70.17 mg / ml, respectively, as compared with 137.56 and 172.72 mg / ml in the case of check groups. The corresponding figures of 4th nymphal instar and adult females were decreased. It was 116.61, 88.70 and 59.27, 47.89 mg / ml respectively, as compared with 154.28 and 118.40 mg / ml in the case of check groups. Statistical analysis indicate that the total phosphorous content of treated stages at different concentrations was significantly decreased as compared with check experiment at $P < 0.001$ (Table.8).

DISCUSSION

The results given in the present work clearly indicate that treatment of 3rd, 4th nymphal instars and adult of both sexes with different concentrations of the botanical extracts induced reduction in the mean of the total carbohydrate, this reduction is directly proportional to the concentration. It is quite clear that the reduction in the total carbohydrate content of 3rd, and 4th nymphal instars was much more pronounced when treatment carried out by extracts of *M. azedarach* leaves and seeds than that treated by *L. camara* leaves and *A. sisalana* stems. This effect interpreted the disturbance in the haemolymph carbohydrates, and rapid decrease of glucose concentration and this was probably induced by high metabolic activity of epidermis, which is known as a utilize trehalose. Our results concur with those obtained by Florkin and Jeuniaux (1964), Mwangi (1982) and Taha *et al.*, (1989) .

It is quite clear from our results that topical treatment of 3rd, 4th and adults of *C. lectularius* at different concentrations of botanical extracts induced significant reduction in the total phosphorous content. These results were clear in the case of treatment with extracts of *M. azedarach* seeds and *A. sisalana* stems. The same trend was also observed when treatment carried out on nymphal

instars and adults with other botanical extracts. Similar response was reported with other insects by many investigators, Salem and El-Maasarawy (1989) and Lenartowicz *et al* (1964).

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Table (1): Total Carbohydrate content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *L. camara* leaf extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet ether	2.04 ± 0.05	-	2.38 ± 0.03	-	2.84 ± 0.05	-	3.36 ± 0.11	-
5.0	2.26 ± 0.04	+10.78	2.70 ± 0.06	+13.44	3.29 ± 0.06	+15.85	4.31 ± 0.25	+28.27
10.0	1.90 ± 0.06	-6.86	2.28 ± 0.04	-13.45	1.51 ± 0.05	-46.83	2.15 ± 0.05	-36.01
20.0	1.66 ± 0.08	-18.63	1.93 ± 0.04	-18.91	1.33 ± 0.05	-53.17	1.78 ± 0.09	-47.02

* Significant
 ** High significant
 *** Very high significant

Table (2): Total Carbohydrate content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *M. azedarach* leaf extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	2.04 ± 0.05	-	2.38 ± 0.03	-	2.84 ± 0.05	-	3.36 ± 0.11	-
5.0	1.93 ± 0.07	-5.39	2.50 ± 0.04	+5.04	1.70 ± 0.05	-40.14	2.84 ± 0.04	-15.48
10.0	1.79 ± 0.05	-12.25	2.08 ± 0.07	-12.61	1.21 ± 0.02	-57.39	2.53 ± 0.07	-25.70
20.0	1.40 ± 0.03	-31.37	1.76 ± 0.03	-26.05	0.85 ± 0.06	-70.07	2.18 ± 0.09	-35.12

* Significant
 ** High significant
 *** Very high significant

Table (3): Total Carbohydrate content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *M. azedarach* seed extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	2.04 ± 0.05	-	2.38 ± 0.03	-	2.84 ± 0.05	-	3.36 ± 0.11	-
5.0	1.96 ± 0.04	-3.92	5.58 ± 0.05	+125.63	1.65 ± 0.05	-41.90	3.47 ± 0.04	+3.27
10.0	1.24 ± 0.04	-39.22	2.42 ± 0.64	+85.71	0.97 ± 0.19	-65.85	3.23 ± 0.05	-3.87
20.0	0.45 ± 0.01	-63.24	3.45 ± 0.20	+44.96	0.41 ± 0.05	-85.56	2.95 ± 0.21	-12.20

- * Significant
- ** High significant
- *** Very high significant

Table (4): Total Carbohydrate content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *A. sisiana* stem extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	2.04 ± 0.05	-	2.38 ± 0.03	-	2.84 ± 0.05	-	3.36 ± 0.11	-
5.0	1.85 ± 0.04	-9.31	2.63 ± 0.05	+ 10.50	2.29 ± 0.05	-19.37	3.10 ± 0.04	-7.74
10.0	1.76 ± 0.04	-13.73	2.42 ± 0.64	+ 1.68	1.94 ± 0.06	-31.69	2.94 ± 0.06	-12.50
20.0	1.48 ± 0.04	-27.45	2.32 ± 0.05	-2.52	1.65 ± 0.05	-41.90	2.68 ± 0.03	-20.24

* Significant
 ** High significant
 *** Very high significant

Table (5): Total phosphorous content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *L. camara* leaf extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	137.56 ± 2.79	-	154.28 ± 0.48	-	172.72 ± 1.80	-	118.40 ± 0.94	-
5.0	111.61 ± 0.66	-18.86	124.58 ± 0.56	-19.25	134.28 ± 1.60	-22.26	111.67 ± 0.53	-5.86
10.0	99.14 ± 2.07	-27.93	106.92 ± 1.76	-30.70	113.36 ± 1.88	-34.37	100.17 ± 1.53	-15.40
20.0	85.39 ± 1.55	-37.93	81.47 ± 0.31	-47.19	88.94 ± 2.75	-48.51	91.17 ± 1.66	-23.00

* Significant
 ** High significant
 *** Very high significant

Table (6): Total phosphorous content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *M. azedarach* leaf extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet ether	137.56 ± 2.79	-	154.28 ± 0.48	-	172.72 ± 1.80	-	118.40 ± 0.94	-
5.0	** 124.55 ± 1.42	-9.46	** 143.28 ± 3.68	-7.13	*** 124.39 ± 0.96	-27.98	*** 90.70 ± 1.70	-23.40
10.0	*** 111.58 ± 1.16	-18.89	*** 136.06 ± 1.22	-11.81	*** 116.47 ± 2.26	-32.57	*** 75.89 ± 1.82	-35.90
20.0	*** 95.56 ± 1.78	-30.53	*** 127.39 ± 1.59	-17.43	*** 98.25 ± 1.34	-43.12	*** 65.08 ± 0.74	-45.03

* Significant
 ** High significant
 *** Very-high significant

Table (7): Total phosphorous content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *M. azedarach* seed extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	137.56 ± 2.79	-	154.28 ± 0.48	-	172.72 ± 1.80	-	118.40 ± 0.94	-
5.0	*** 32.99 ± 1.53	-76.02	** 48.31 ± 1.77	-68.69	*** 54.36 ± 0.94	-68.53	*** 31.94 ± 1.26	-73.02
10.0	*** 22.75 ± 1.19	-83.46	*** 36.08 ± 2.02	-76.61	*** 42.28 ± 1.98	-75.52	*** 24.82 ± 1.68	-79.04
20.0	*** 17.78 ± 1.79	-87.07	*** 30.14 ± 1.41	-80.46	*** 42.72 ± 1.29	-75.27	*** 20.67 ± 1.57	-82.54

* Significant
 ** High significant
 *** Very high significant

Table (8): Total phosphorous content of 3rd, 4th nymphal instars and adults of *C. lectularius* topically treated with *A. sislandia* stem extracts.

Conc. of extract (%)	Nymphal instars				Adults			
	3 rd	Change (%)	4 th	Change (%)	Male	Change (%)	Female	Change (%)
Pet. ether	137.56 ± 2.79	-	154.28 ± 0.48	-	172.72 ± 1.80	-	118.40 ± 0.94	-
5.0	*** 89.35 ± 2.09	-35.05	*** 116.61 ± 1.79	-24.42	*** 110.97 ± 1.69	-35.75	*** 59.27 ± 1.19	-49.94
10.0	*** 73.81 ± 1.82	-46.34	*** 88.70 ± 1.24	-42.51	*** 70.17 ± 1.42	-59.37	*** 47.89 ± 1.67	-59.55
20.0	*** 51.08 ± 1.71	-62.87	*** 80.41 ± 2.31	-47.88	*** 51.22 ± 2.01	-70.35	** 42.17 ± 1.65	-64.38

* Significant
 ** High significant
 *** Very high significant

الملخص العربي

تأثير ثلاثة مستخلصات نباتية على المحتوى الكربوهيدراتي والفسفوري في بق الفراش المعامل سايمكس ليكتيولارس (نصفية الأجنحة - سيميسيدى)

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- الهدف من الدراسة الحالية هو تقدير تأثير ثلاثة مستخلصات نباتية (لانتانا كمارا، الزنزلخت، الصبار) في التركيزات المختلفة على المحتوى الكربوهيدراتي والفسفوري للحوريات في العمر الثالث والرابع، وكذا الطور اليافع لحشرة بق الفراش.
- أوضح التحليل الاحصائي للنتائج ان المحتوى الكربوهيدراتي للحوريات العاملة في العمر الثالث وذكور الطور اليافع في التركيزات المختلفة (5، 10، 20%) لمستخلصات نبات الزنزلخت قد تناقص بصورة معنوية، من جهة اخرى لوحظ زيادة هذا المحتوى عند معاملة الإناث بتركيز 5 % من نفس المستخلص.
- اوضحت النتائج ان المستخلصات النباتية لاوراق نبات الزنزلخت كانت اكثر فعالية وتأثيراً على المحتوى الكلي للفسفور اذا ما قورن بمستخلص اوراق نبات لانتانا كمارا.
- اثبتت النتائج ان المحتوى الكلي للفسفور في حوريات العمر الثالث والرابع والطور اليافع ذكوراً واناثاً قد تناقص معنوياً عند معاملة هذه الاطوار بمستخلصات سيقان نبات الصبار وهذا التناقص مرتبط بزيادة تركيز المستخلص النباتي.
