

THE CONTROL OF *Sitophilus oryzae* (L.) IN STORED WHEAT WITH *Prosopis juliflora* (SW) D.C. SEED EXTRACTS

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ABSTRACT

The effectiveness of *Prosopis juliflora* (S.W) D.C. seed extracts against *Sitophilus oryzae* (L.) insect reared on wheat grains was investigated in laboratory bioassays. All tested plant extracts (petroleum ether, chloroform, and acetone) effectively controlled adult insects and their toxicity LC_{95} and LC_{50} values averaged (12.0, 5.8ml/kg) for acetone < petroleum ether (8.0, 4.1ml/kg) < chloroform (6.3, 2.2ml/kg), respectively. A high significant effect on oviposition ($P < 0.05$) was found for all tested extracts at LC_{50} levels, while at LC_{95} levels, oviposition was completely inhibited. Thus, a progeny emergence was completely suppressed at LC_{95} levels as well as at LC_{50} of acetone extract. Chloroform extract indicated a slow rate of degradation after 1 month of storage (90% mortality). All tested plant extracts caused loss in grain wheat after 45 days of storage, but chloroform extract was the most effective compound. Most treatments didn't affect water absorption but viability was significantly reduced. Petroleum ether and chloroform extracts caused a significant inhibition effect of AchE in adult insects while acetone extract caused a significant activation effect. All extracts caused significant activation effects on AcP and AlkP except chloroform and acetone extracts treatments that caused a significant inhibition effect on AcP in adults. Moreover, all extracts showed significant decrease in protein and carbohydrate contents of adult insects, except carbohydrate content of adults treated with acetone extract, meanwhile, there was a significant increase of lipid content in adults treated with all extracts and carbohydrate content in adults treated with acetone extract.

Keywords: Grain Protectant, Plant Extracts, Mesquite, *Prosopis juliflora*, *Sitophilus oryzae*, Botanical Insecticide,

INTRODUCTION

Currently the measures to control pest infestation in stored grain products rely heavily upon the use of insecticides which can lead to problems of build up the toxic residues and environmental contamination (Zettler and Cuperus, 1990; White, 1995).

The indigenous materials of botanical origin are an important source of grain protectant because they have been found to possess toxic effects against insects (Arroyo, 1995). Research on the evaluation of available local plants is very necessary to help farmers to use these plants to limit post-harvest losses of their products caused by different insects. Mesquite plant, *Prosopis juliflora* is a common widely spread in most parts of Saudi Arabia (Collenette, 1999; Chaudhary and Al-Jowaid, 1999). The aqueous extracts of the leaves was previously considered to have antibacterial (Satish et al., 1999), antifungal (Ahmed et al., 1997; Gomathi and Kannabiran, 2000; Kamalakannan et al., 2001) activities. Leaf extracts of *P. juliflora* also showed an insecticidal efficiency against *Callosobruchus analis* (Tabassum et al., 1994) and *Plutella xylostella* (Torres et al., 2001).

The present investigation was studied to evaluate the effectiveness of *P. juliflora* seed extracts as a protectant against the rice weevil *Sitophilus oryzae* (L.) by testing the effect of the seed extracts on each of the following aspects: adult mortality, egg laying, adult emergence, grain weight loss, residual effect, grain viability, grain water absorption, enzymes assay and main metabolites.

MATERIALS AND METHODS

Rearing technique:

The insect pest *S. oryzae* were cultured in glass jars on wheat grains under controlled temperature and humidity (27°C and 70% relative humidity). The new cultures were prepared by adding each 200-300 adults (unsexed) from a stock culture to about 500g of wheat grains in a glass jar. After 3 weeks of oviposition period, the parent adults were removed, and one week old insects subsequently emerging were used for the experiments. All experiments were conducted using wheat grains at the same conditions.

Extraction technique:

Mesquite seeds were obtained from the local markets then washed, dried and ground in electric grinding machine. A sufficient quantity of powder was extracted with organic solvents (of increasing polarity), petroleum ether, chloroform and acetone as described by Su (1985). The solvents were sequentially used to extract the plant material for a period of 48h each at room temperature then filtered through anhydrous sodium sulfate. The rotary evaporator was used to remove the solvents. The obtained oils in each case, were stored in labeled plastic bottles at 5°C until required for use (Islam, 1983).

Mixing technique:

For all experiments extract were added to wheat grains in glass jars using solvents as a carrier. Each concentration of every extract was dissolved in his solvent and shaken thoroughly, subsequently allowing solvent evaporation by a stream of air. All treatments were replicated at least 3 times. In all cases, the experiments were performed in constant temperature incubators at 27°C and 70% RH. Three jars of grains were made to each treatment where they were treated with 0.2ml of each solvent and used as control. All treated jars were covered with cloth fastened with rubber bands to prevent the contamination and insects from escaping.

Adult mortality:

Four different concentrations were prepared from each extract after preliminary test. Twenty adults of *S. oryzae* of mixed sex were introduced into each jar containing 10g of wheat grains. The effects of *P. juliflora* seed extracts on the survival of adults rice weevils was assessed by recording mortality at 1, 3, 5, 7, 14 days. Adults were considered as dead when no response was obtained after probing the abdomen with forceps. Percentage insect mortality was calculated for each concentration using the formula

proposed by Abbott (1925). Moreover, LC_{50} and LC_{95} values during the first three days, were calculated according to probit analysis (SPSS, 1999). The data were subjected to analysis of variance. Significant differences between treatment means were separated at $P = 0.05$ by Duncan's multiple range test (1951).

Oviposition and adult emergence:

At this experiment the following parameter were assessed:

- i) Total number of eggs laid.
- ii) Total number of adult emergence.

Concentrations of LC_{50} , LC_{95} and controls of either petroleum ether, chloroform and acetone extracts were applied to each of three replicates consisting of 5g wheat grains in a glass jar. Ten pairs (1-7 day old) of *S. oryzae* adults were added to each jar. They were sexed following the method described by Halstead (1963). Egg laying capacity of weevils was recorded after 10 days. Acid Fuschin stain was employed for the detection of the eggs (Frankenfeld, 1950). The eggs were counted from the stained grain samples. A similar treatment was made to determine total emergence. After 10 days, the insects were removed and from the first day of emergence, the newly hatched weevils were removed and counted daily. The following formula was used to determine the percentage reduction in the number of eggs and offspring:

$$\% = \frac{X}{Y} \times 100$$

Where, X = the number of eggs or adults in the treatment.

Y = the number of eggs or adults in the control.

For comparison between treatment and control, t- test was used.

Grain weight loss:

Percent loss in grain weight was calculated by following the method of Khare and Johari (1984). To determine moisture content (%), 10g samples of wheat grains were grounded finely and heated in oven at 105°C for 18 hours and reweighed. Samples of 10g grains were weighted, transferred into glass jars and treated with seed extracts of *P. juliflora* at LC_{50} and LC_{95} . Each treatment was replicated three times. Solvents were similarly pipette in another jars used as controls. After thorough mixing and evaporation of the solvents, 20 adults of *S. oryzae* were added, and the grains were stored for 45 days. Weight loss in grains was estimated by sieving the contents of each jar by weighing the resultants. Percent weight loss was then calculated after correcting for moisture by the formula:

$$\frac{\text{Initial grain wt} - \text{Final grain wt}}{\text{Initial grain wt}} \times 100$$

All data were statistically analyzed by calculating t-test at 5% level.

Residual effect:

To assess the persistence of the treatments, each tested extract - LC₉₅ concentration including control, was mixed with 1kg samples of grain and stirred continuously for 30 min to ensure even spread of the material over the surface of the grains. Treated grains and controls were allowed to dry for 2hrs before storage. The grains of each treatment were infested every three days with adult weevils of mixed sex. For each 10gm sample, 20 of *S. oryzae* adults were introduced in each replicate where all treatments and control were replicated three times. Mortality counts were made after 3 days, and the data were analyzed by calculating LT₅₀ and LT₉₅ values.

Grain viability and water absorption:

Viability and water absorption tests of the treated seeds were conducted 1 and 30 days after treatment. From each treated grains jars with LC₉₅ concentrations of all extracts, initial and after storage periods, and controls, 30 grains from each treatment and control, were released between 3 petri dishes (3 replicates) on two layers of moistened Whatman filter paper. Treated grains were moistened daily. Germination was recorded after 10 days (Anonymous, 1966).

Water absorption tests were carried out in small jars - three quarters full of distilled water. Two gram of treated and untreated grains were submerged in water for 1, 5, 24 hr (Sighamony *et al.*, 1986). At each period, the seeds were dried with filter paper and re-weighed to estimate the absorbed water.

Reduction in germination and water absorption were determined. Results were analyzed by ANOVA, and means were compared with Duncan's multiple range test.

Enzymes assay:

1- Acetylcholine esterase (AchE)

Acetylcholine esterase (AchE) was measured according to the method described by Simpson *et al.*, (1964), using acetylcholine bromide (AchBr) as substrate.

2- Phosphatases (AcP & AlkP)

Acid phosphatase (AcP) and alkaline phosphatase (AlkP) were determined according to the method described by Powell and Smith (1954).

Main metabolites:

The main metabolites (total proteins, total lipids and total carbohydrates) were determined in the total body homogenates:

1- Total proteins

Total proteins were determined by the method of Bradford (1976).

2- Total carbohydrates:

Total carbohydrates were determined by the method described by Singh and Sinha (1977).

3- Total lipids:

Total lipids were estimated according to Knight *et al.*, (1972) using phosphovanillin reagent.

All results were analyzed by calculating t-test at 5% level.

RESULTS AND DISCUSSION

Effect on adult mortality:

The data in Table 1 showed that *P. juliflora* seed extracts were remarkably effective producing mortality after 3 days with almost all of their concentrations on *S. oryzae*. However, within 1 day of exposure to wheat grains treated with petroleum ether and chloroform extracts, 20% and 25% of the weevil adults killed at 5 ml/kg, respectively. After 5 days of exposure, total mortality was achieved by petroleum ether, chloroform and acetone extracts at 5, 3 and 10 concentrations, respectively. A similar trend in mortality was observed at most of other extract concentrations 7 days after exposure.

Table 1: Average mortality of *Sitophilus oryzae* when exposed to wheat grains treated with different extracts of *Prosopis juliflora* seeds.

Extract	Concentration (ml / kg)	% Cumulative mortality *				
		1 day	3 days	5 days	7 days	14 days
Petroleum ether	3	0 ^{ade}	30 ^{abde}	71 ^{ab}	73 ^{ac}	79 ^a
	4	2 ^{ade}	40 ^{abi}	82 ^{abc}	85	100 ^b
	5	20 ^{bcefg}	60 ^{cglj}	100 ^{ce}	-	-
	6	29 ^{bclg}	85 ^{dij}	100 ^{dij}	-	-
LC50		4.1				
LC90		8.0				
Slope		5.7				
Chloroform	1	0 ^{ade}	20 ^{ah}	54 ^d	70 ^{acd}	74 ^a
	2	0 ^{ade}	30 ^{abeh}	81 ^{abe}	100 ^{be}	-
	3	8 ^{abdef}	57 ^{cglj}	100 ^{ce}	-	-
	5	25 ^{bclg}	95 ^d	100 ^{ce}	-	-
LC50		2.2				
LC90		6.3				
Slope		3.52				
Acetone	4	0 ^{ae}	20 ^{adgh}	52 ^d	62 ^{cd}	73 ^a
	6	4 ^{ade}	46 ^{bcdi}	90 ^{bce}	100 ^{bde}	-
	8	16 ^{bcd}	63 ^{cg}	94 ^{bce}	100 ^{bde}	-
	10	32 ^{bog}	95 ^d	100 ^{ce}	-	-
LC50		5.8				
LC90		12.0				
Slope		5.2				
F-ratio		7.82	30.422	17.127	18.044	16.934
F-tabulated		2.216				

The effect of higher and lower concentrations of tested extracts on adult mortality was significantly different from each other, as well as mortality was directly proportional to the level concentration and time. When each treatment was compared with the control, a high significant difference was obtained ($P < 0.05$).

The slopes of the probit lines were steeper as concentration increased (Table 1). On the basis of relative toxicity at both levels (LC_{50} and LC_{95}), the treatment could be summarized as: chloroform extract (2.2, 6.3 ml/kg) > petroleum ether extract (4.1, 8.0 ml/kg) > acetone extract (5.8, 12.0) at LC_{50} and LC_{95} values, respectively.

The data on adult mortality showed a strong relationship between mortality percentage and extract concentration, where more than 50% of insect mortality occurred at all tested concentrations of all extracts after an exposure period of 5 days. Consequently, the tested concentrations of the plant extracts are sufficient to cause significant insects mortality. The present results agree with those previously reported by Tabassum *et al.*, (1994) and Al-Moajel, (2003).

Previously, Al-Moajel and Al-Dosary (2003) stated that petroleum ether extract was considered the most toxic plant extract, whereas in this study, chloroform extract proved to be the most toxic one against rice weevil.

Effect of oviposition and adult emergence behavior:

Table (2) compares the ovipositional response of *S. oryzae* females exposed to the extracts of *P. juliflora* seeds. The mean number of deposited eggs in grains treated with both lower and higher concentrations of Mesquite seed extracts (LC_{50} and LC_{95}) was observed to be minimum. Maximum determinacy in oviposition was obtained from all extracts at LC_{95} concentrations (98-99%), while 84-92% ovipositional reduction was observed at LC_{50} concentrations. When adult insects were fed with wheat grains treated with solvents only (petroleum ether and chloroform and acetone), the number of eggs laid averaged 76.4, 94.7, 106.7 eggs per 10 pairs, respectively. It is apparently showed in Table 2, a high significant effect on oviposition ($P < 0.05$) was found for all extracts at LC_{50} concentrations, while at LC_{95} concentrations, oviposition was almost inhibited.

Reduction in hatchability of 97% and 85% was observed at LC_{50} concentrations of petroleum ether and chloroform extracts, respectively. T-test analysis showed that there were high significant differences, while progeny emergence was completely suppressed in grains combining LC_{95} concentration of all tested extracts as well as LC_{50} of acetone extract.

Complete reduction in progeny of *S. oryzae* treated with LC_{95} concentrations could be possible because of the observed high adult mortality. At LC_{50} concentration of acetone extract egg laying was reduced by 86%, but progeny emerging was reduced by 100%, thus acetone extract was ovicidal to the eggs of *S. oryzae*.

Some workers have observed a reduction in oviposition of *S. oryzae* weevils on grains treated with plant extracts (Risha *et al.*, 1990; Mahgoub *et al.*, 1998; Ahmed, 2002). These results were confirmed by those of Al-Moajel and Al-Dosary (2003) who reported that *P. juliflora* seed extracts were effective in reducing the oviposition and progeny emergence of *C. maculatus*.

Table 2: Efficacy of different seed extracts of *Prosopis juliflora* against *Sitophilus oryzae* for ovipositional and progeny deterrent properties in wheat grains.

Extract	Concentration (ml/kg)	Av. no. of egg laid / 10 pairs + SE	T-value	Oviposition reduction %	Av. no. of emergence +SE	T-value	Emergence reduction %
Petroleum ether	Control	76.4±0.38			29.7±1.2		
	LC ₅₀ (4.1)	5.2±0.61	-98.68	92*	0.7±0.12	-24.19	97*
	LC ₉₅ (8.0)	0.7±0.06	-194.82 S	99*	0.0±0.00	-	100
Chloroform	Control	94.7±2.4			25.7±0.0		
	LC ₅₀ (2.2)	15±1.20	-29.79	84*	3.7±0.00	-65.99	85*
	LC ₉₅ (6.3)	1.7±0.00	-38.54 S	98*	0.0±0.00	-	100
Acetone	Control	106.7±3.50			34.7±0.93		
	LC ₅₀ (5.8)	17±1.70	-22.79	86*	0.0±0.00	-	100
	LC ₉₅ (12.0)	0.7±0.15	-29.97 S	99*	0.0±0.00	-	100

* High significant differences ($\alpha = 0.05$)**Residual effect:**

All seed extracts at LC₉₅ concentrations gave almost complete protection until 15 days of storage (Table 3).

Table 3: Susceptibility of *Sitophilus oryzae* adults to wheat grains treated with *Prosopis juliflora* seed extracts after different interval of storage.

Intervals of storage (days)	% Adult mortality		
	Pet-ether	Chloroform	Acetone
Initial	100	98	98
3	96	96	95
6	96	95	96
9	95	96	96
12	94	94	94
15	95	96	94
18	93	95	90
21	88	95	85
24	85	95	60
27	82	90	52
30	75	90	40
Slope	-3.7299	-1.8559	-2.2149
LT ₉₅	12	14	8
LT ₅₀	33	104	43

Gradually, the residual toxicity of petroleum ether and acetone extracts decreased with length of storage, so after 30 days of storage the insects mortality found in the treated grains being only 75% and 40%, respectively. While in the treated grains with chloroform extracts, 90% of *S. oryzae* individuals were killed after 30 days of exposure. Consequently, adult

mortality in chloroform extract at various storage duration seems to have retained the initial activity, so the chloroform extract of *P. juliflora* seeds obviously gave the highest degree of protection for stored gains against *S. oryzae*. On the other hand, no mortality was recorded in untreated check. It should be also noted that LT_{95} value of chloroform extract is longer than those in petroleum ether and acetone extracts. Therefore, it can be concluded that chloroform extract showed a long residual activity after one month of storage.

Similar result about the effectiveness of plant extracts for nearly one month of storage were also obtained by Al-Moajel and Abd El-Baki, (2000) with *Brassica rapa* extracts against *R. dominica*, and Ahmed *et al*, (2002) with *Capparis spinosa* extracts against *C. maculatus*. On the other hand, some plant extracts well protected stored products as adulticide over 2-8 months of storage and that was previously confirmed by Mahgoub (1992) with neem extracts against *C. maculatus*; and Al-Moajel (2000) with *B. napus* against *S. granarius*. In respect of *P. juliflora* extracts, these results agree with that of Al-Moajel and Al-Dosary (2002) on cowpea seed extracts against *C. maculatus*.

Effect on grain viability

Data in Table (4) showed that the germination of grains treated with petroleum ether, chloroform and acetone extracts at the rates of 8.0, 6.3, and 12.0 ml/kg (LC_{95}) averaged 92.0%, 88.0% and 84.0%; and 90%, 84% and 81% after the initial and 30 days from application, respectively. All treatments gave a range of germination reduction from 6.2% to 15.6% (Table 4). The lowest germination value was obtained in grains treated with acetone extract, which caused 84-81% reduction at the two experimented intervals (initial and after 30 days of storage), respectively.

Table 4: Effect of *Prosopis juliflora* seed extracts on germination of wheat grains initial and 30 days after application.

Extract and concentration (ml/kg)	Initial		Thirty days after application	
	% Germination +SEM	% Reduction	% Germination +SEM	% Reduction
Control	98±1.15 ^{bcd}		96±1.00 ^{cde}	
Petroleum ether (8.0)	92±0.00 ^{ab}	6.2	90±0.00 ^{abc}	6.3
Chloroform (6.3)	88±1.15 ^c	10.2	84±2.00 ^{ad}	12.5
Acetone (12.0)	84±2.31 ^{ad}	14.2	81±0.58 ^{de}	15.6
F-ratio	17.82		33.19	
F-tabulated	4.0661			

Means within column followed by same letter are significantly different at $P < 0.05$.

These results agree with those of Khaire *et al*, (1992); and Abdel-Latif (2003) who reported that there was adverse effect of plant extract and oil treatment on germination of seeds and grains. On the contrary, others found very negligible effect of other plant extracts on seed germination (Signal & Singh, 1990).

Effect on water absorption

Table (5) showed that one hour after application, chloroform and acetone treatments absorbed significantly more water than control. Also, after five hours from application all three seed extracts significantly absorbed more water than in untreated control. Other treatments at initial time and all treatments after storage period recorded very negligible effect on the amount of water absorption.

This effect has been reported in previous studies (Mahgoub *et al.*, 1998 and Shemais and Al-Moajel, 2000), who found no effect on water absorption. Tembo and Murfitt (1995) reported that there was a significantly effect of some plant oils on water absorption.

Table 5: Effect of *Prosopis juliflora* seed extracts on water absorption of wheat grains initial and 30 days after application.

Extract and concentration (ml/kg)	% Water absorption					
	Initial			Thirty days after application		
	1 hr	5hrs	24hrs	1 hr	5hrs	24hrs
Control	18 ^b	32 ^b	51 ^a	14 ^a	28 ^a	50 ^a
Petroleum ether (8.0)	21 ^{ab}	35 ^a	52 ^a	17 ^a	31 ^a	50 ^a
Chloroform (6.3)	23 ^a	38 ^a	55 ^a	14 ^a	29 ^a	50 ^a
Acetone (12.0)	23 ^a	38 ^a	56 ^a	14 ^a	27 ^a	47 ^a
F-ratio	4.47	37.50	2.96	2.25	13.2	1.59
F-tabulated	4.0661					

Effect on weight loss

Table (6) indicated that the loss percentages in grain weight in different treatments after 45 days of application ranged from 1% to 5%.

Table 6: Grain weight losses caused by *Sitophilus oryzae* weevil on stored wheat grain treated with *Prosopis juliflora* seed extracts.

Extract	Concentration (ml/kg)	% Of loss in weight + SE	T- value	% Protection
Petroleum ether	Control	11.33±0.88	7.91 10.58	75 83
	LC ₅₀ (4.1)	3±0.58		
	LC ₉₅ (8.0)	2±0.00 S		
Chloroform	Control	11.33±0.88	8.32 11.72	66 91
	LC ₅₀ (2.2)	4±0.00		
	LC ₉₅ (6.3)	1±0.00 S		
Acetone	Control	11.33±0.88	6.01 6.29	58 66
	LC ₅₀ (5.8)	5±0.58		
	LC ₉₅ (12.0)	4±0.76 S		

S = significant

While the weight loss percent was maximum in control (11.33%). On the other hand, chloroform extract at LC₉₅ concentration was found to be the best preventing material, which caused only 1% loss. There were significant differences ($P < 0.05$) between the weight loss between treated wheat grains and control after 2 months. The percentage losses in grain weight were significantly lower in grain treated with acetone extract than in the grain treated with other extracts, and the percentage weight loss was significantly higher in grain, treated with chloroform extract at LC₅₀ and LC₉₅ concentrations. Consequently, chloroform extract at LC₉₅ value was the most effective extract in reducing the grain weight loss, which gave 91% reduction. All plant extracts protect the grains against feeding by the insect pest *S. oryzae*.

Effect on esterase and phosphatase enzymes

Table (7) showed that in petroleum ether and chloroform extracts treatments, acetylcholinesterase (AChE) enzyme significantly decreased (1101.48 and 1057.67 $\mu\text{m/g/min}$) for the two extracts, respectively and that compared with 1222.07 $\mu\text{m/g/min}$ in the control. But in case of acetone extract, AChE enzyme slightly increased (1230.41 $\mu\text{m/g/min}$). Acid phosphatase enzyme (AcP) activity was significantly inhibited by using chloroform and acetone extracts (233.65 and 253.85 $\mu\text{m/g/min}$) compared with 281.39 in control, respectively. Acetone extract increased AcP enzyme to an average of 311.63 $\mu\text{m/g/min}$.

On the other hand, alkaline phosphatase enzyme (AlkP) activity increased in all tested extracts, which reached 6.33, 5.41 and 10.95 $\mu\text{m/g/min}$ with petroleum ether, chloroform and acetone extract treatments, respectively. In general, acetone extract caused the highest effect.

Petroleum ether and chloroform extracts caused a significant inhibition effect of AChE. Similar effect of chloroform and acetone treatments was also obtained on AcP. On the contrary, the petroleum ether treatment caused a significant activation effect of both AlkP, and AcP enzymes. Ahmed, (2000) found that after 72 hrs of exposure, *Ricinus communis* seed extracts caused an inhibition effect in *S. oryzae* adults.

Effect on main metabolites

Data presented in Table (8) showed that after 3 days of *P. juliflora* seed extracts application, the total protein content significantly decreased in all treatments. Protein content was at its lowest level (13.78 mg/g) in adults treated with chloroform extract. Slight increase was obtained with petroleum ether and acetone extracts (16.59 and 16.48, respectively) compared with 20.08 mg/g in control. On the contrary, total lipids were significantly increased in all extracts. Total lipids showed the highest level (36.95 mg/g) in chloroform extract, while using petroleum ether and acetone extracts, the total lipids had a normal increase (22.25 and 21.58 mg/g, respectively) compared with 20.02 mg/g in control. Meanwhile, the carbohydrate content was significantly higher in adult insects treated with acetone extract (15.08 mg/g), and had a significant lower levels (6.20 and 11.24 mg/g) in petroleum ether and chloroform treatments, respectively compared with 14.17 mg/g in control.

Table 7: Effect of *Prosopis juliflora* seed extracts (LC₉₅) on the rate of acetylcholinesterase (AChE) and Phosphatases (AcP & AlkP) in *Sitophilus oryzae* treated after 72 hrs.

Extract and concentration (ml/kg)	AChE ($\mu\text{g/g/min}$)				AcP ($\mu\text{g/g/min}$)				AlkP ($\mu\text{g/g/min}$)			
	Mean \pm SE	95% Confidence Interval			Mean \pm SE	95% Confidence Interval			Mean \pm SE	95% Confidence Interval		
		Lower	Upper	T-value		Lower	Upper	T-value		Lower	Upper	T-value
Control	1222.07 \pm 16.15	1190.89	1244.92		281.39 \pm 4.97	179.11	292.74		4.63 \pm 0.16	4.31	4.85	
Pet-ether (4.1)	1101.48 \pm 42.82	1027.39	1175.73	23.39	311.63 \pm 5.55	303.48	322.23	38.14	6.33 \pm 0.23	6.08	6.79	-404.06
	S				S				S			
Chloroform (2)	1057.67 \pm 68.39	950.34	1184.77	14.00	233.65 \pm 8.29	221.34	249.43	16.12	5.41 \pm 0.31	4.92	5.97	-310.01
	S				S				S			
Acetone (5.8)	1230.41 \pm 26.89	1199.89	1284.01	42.05	253.85 \pm 4.19	247.17	261.56	36.76	10.95 \pm 0.39	10.19	11.47	-228.59
	S				S				S			

S = significant ($\alpha = 0.05$)

Table 8: Effect of *Prosopis juliflora* seed extracts (LC₅₀) on the rate of main metabolites in *Sitophilus oryzae* treated after 72 hrs.

Extract and concentration (ml/kg)	Total proteins (mg/g)				Total lipids (mg/g)				Total carbohydrates (mg/g)			
	Mean \pm SE	95% Confidence interval			Mean \pm SE	95% Confidence interval			Mean \pm SE	95% Confidence interval		
		Lower	Upper	T-value		Lower	Upper	T-value		Lower	Upper	T-value
Control	20.08 \pm 0.67	18.85	21.17		20.02 \pm 0.97	18.30	21.65		14.17 \pm 0.43	13.41	14.91	
Pet-ether (4.1)	16.59 \pm 0.60	15.89	17.79	-138.07	22.25 \pm 0.47	21.47	23.09	-165.91	6.20 \pm 0.21	5.8	6.50	-404.06
	S				S				S			
Chloroform (2.2)	13.78 \pm 0.24	13.32	14.12	-362.60	36.95 \pm 1.39	34.32	39.03	-45.43	11.24 \pm 0.50	10.34	12.08	-176.47
	S				S				S			
Acetone (5.8)	16.48 \pm 0.14	16.21	16.69	-589.11	21.58 \pm 1.34	19.39	23.16	-69.15	15.08 \pm 0.44	14.37	15.87	-195.41
	S				S				S			

S = significant ($\alpha = 0.05$)

From the previous results, it can be concluded that all extracts led to a significant decrease in protein and carbohydrate contents of adult insects, except carbohydrate content in those treated with acetone extract. There was also a significant increase of lipid content in adults treated with all extracts and carbohydrate content in those treated with acetone extract.

Generally, the results showed that the lowest amount of protein and the highest amount of lipid were obtained in treatments using chloroform extracts. The lowest amount of carbohydrate was observed in insects treated with petroleum ether extract. These results of reduction in protein and carbohydrate contents and increase of lipid contents by using *P. juliflora* seed extracts may be due to prevention action.

These results of increasing of lipid are in agreement with Mostafa and Sherif (1993) by using different plant powders.

From the previous results, it can be concluded that according to updated information, the activity of seeds of *P. juliflora* as protectant was carefully studied here for the first time (Al-Moajel and Al-Doary, 2002, 2003). It can be stated that seed extracts of *P. juliflora* plant are useful grain protectants because of their following advantages:

It can be used as natural toxicants agent. They may have no toxicity to mammals, because are normally used as human foods and animal fodders (Fleger, 1990). They have also medicinal and pharmacological properties (Rogers, 2000; Kamalakannan *et al*, 2001).

The present results indicate that seed extracts have obvious toxic effect on adult insects and they also reduced insect fecundity and weight loss of exposed grains. Chloroform extract was the most effective extract with low concentration on adult mortality with more active of residual effects and less of weight loss than the other tested extracts. Further research dealing with the constituents and bioactivity of *P. juliflora* seed extracts against other stored product insects is needed, and there is much need to do research on the components of its extracts.

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مكافحة حشرة سوسة الأرز في القمح المخزن باستخدام مستخلصات بذور نبات الغاف

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تم تقييم فعالية ثلاث مستخلصات لبذور نبات الغاف على سوسة الأرز المرباة على حبوب القمح.

كان للمستخلصات الثلاثة : الايثر البترولي والكلوروفورم والأسيتون سمية عالية على الحشرات البالغة ، حيث تدرجت فعالية هذه المستخلصات بالتركيزات القاتلة لـ 95% و 50% من الحشرات على التوالي كالآتي:

الأسيتون (12 و 5.8 مل /كجم) > الايثر البترولي (8 و 4.1 مل / كجم) > الكلوروفورم (6.3 و 2.2 مل / كجم). كما انخفض معدل وضع البيض انخفاضاً معنوياً عالياً في جميع المستخلصات عند المعاملة بالتركيزات القاتلة لـ 50% من الحشرات، بينما لم يكن هناك أي بيض تقريباً عند المعاملة بالتركيزات القاتلة لـ 95% من الحشرات ، وعلى هذا لم يظهر أي نسل عند هذا التركيز ، وكذلك عند المعاملة بمستخلص الأسيتون عند التركيز القاتل لـ 50% من الحشرات . أما عند تخزين الحبوب المعاملة بالمستخلصات الثلاثة فقد كان مستخلص الكلوروفورم أكثر بقاءً، حيث بلغت نسبة الموت 90% بعد شهر من التخزين.

كما كانت جميع المستخلصات ذات فعالية عالية في خفض الفقد في الوزن بعد 45 يوماً من المعاملة ، إلا أن مستخلص الكلوروفورم أكثرها فعالية.

كالمال يمكن لغالبية المعاملات تأثير معنوي على معدل امتصاص حبوب القمح للماء إلا أنها أثرت معنوياً على معدل الإنبات.

وكان لمستخلص الايثر البترولي والكلوروفورم تأثير مثبط لإنزيم الكولين استيراز بينما كان لمستخلص الأسيتون تأثير منشط لهذا الإنزيم ، أما إنزيم الفوسفاتاز الحمضي فقد كان لمستخلصي الكلوروفورم والأسيتون تأثير مثبط له، إلا أن مستخلص الايثر البترولي كان له تأثير منشط كما كان للمستخلصات الثلاثة تأثير منشط للفوسفاتاز القلوي.

كما قلت كمية البروتين والكربوهيدرات في أجسام الحشرات المعاملة معنوياً ما عدا مستخلص الأسيتون فقط زاد كمية الكربوهيدرات في أجسام الحشرات المعاملة معنوياً، بينما زادت معنوياً كمية الدهون في أجسام الحشرات المعاملة بجميع المستخلصات.

ولذا ينصح باستخدام مستخلص بذور نبات الغاف في حماية حبوب القمح من الإصابة بسوسة الأرز إلا أنه لا ينصح باستخدامها في حماية تقاوي الحبوب.