

Evaluating the Insecticidal and Fungicidal Efficiency of *Acacia nilotica* Pods Extract

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ABSTRACT

The study aimed to identify the potential of phytochemical constituent's occurrence in the extract of *Acacia nilotica* pods and assessment of its bioactivity. Chemical and GC-MS analyses of the ethanolic extract and its fractions of pods identified active compounds such as alkaloids, flavonoids, terpenoids, steroids and tannins. Toxicity of extract proved variable toxicity effects for stages of *Ceratitis capitata* and *Bactrocera zonata*. The extract constituents have demonstrated that the *Ceratitis capitata* egg was more susceptible against hatchability than for *Bactrocera zonata*, and generally, there was ovicidal efficacy noticed for both insects. Also, the essential oil of *A. nilotica* pods was exhibited larvicidal activity for both insects larva at various high concentrations of the tested plant extract. Concerning the adult stage, ethanolic extract of the tested tree pods proved repellent effects and insecticidal toxicity properties causing knockdown against the adults of *Ceratitis capitata* and *Bactrocera zonata*. The ethanol extract of *A. nilotica* shows antifungal activity against plant fungal pathogens; *Aspergillus flavus* and *Sclerotinia sclerotiorum*, where the rate of mycelial growth inhibition was increased by increasing the extract concentration. The extract of *A. nilotica* was found to have promising effects in controlling the investigated pests.

Keywords: *Acacia nilotica*, *Aspergillus flavus*, *Ceratitis capitata*, *Bactrocera zonata*, GC-MS analysis, Plant extract, *Sclerotinia sclerotiorum*.

INTRODUCTION

The deleterious effect of pesticides on human health and environment has necessitated that alternatives be explored which are safe and environmental friendly. The excessive use of pesticides has resulted in accumulation of pesticide residues in the food and fodder besides exercising deleterious effect on the beneficial organisms (Mohan *et al.*, 2011). Some plants contain components that are toxic to pathogens and insects. When extracted from the plant and applied on infested crops, these components are act as alternative for control strategies to reduce dependency on synthetic pesticides. Plants have ability to synthesize aromatic secondary metabolites, like phenols, flavonoids and tannins. The components with phenolic structures were highly active against the pathogen. These groups of compounds show antimicrobial effect and serves as plant defense mechanisms against pathogenic microorganisms (Gurjar *et al.*, 2012).

Acacia nilotica (*A. nilotica*) is one of about 135 thorny Africa *Acacia* species. It is naturally wide spread in arid regions of Africa. It is rich in phenolic compounds and has been reported to possess antimicrobial and phytotoxic activities (Mohana *et al.*, 2011). The bark, leaves and pods of the tree also have many medicinal uses in Africa. Seeds and leaves of *A. nilotica* were reported to contain various active ingredient of multiuse. (Edriss *et al.*, 2012).

Insects affect human and environment beings in a number of ways. Flies (Diptera: *Tephritidae*) are considered the most destructive insect pests of fruit and vegetables in the world. The invasive fruit fly, *Ceratitis capitata* (Wiedemann), [*C. capitata*] and the peach fruit fly, *Bactrocera zonata* (*B. zonata*), are highly polyphagous tephritid that infests 300 species of fruit and vegetables throughout the world uses the various hosts in its environment, moving from one to another as fruit mature, in addition flies may damage plants by laying eggs in fruits tissues (Cohen and Yuval 2000; Sarwar *et al.*, 2013).

For the other hand *Aspergillus* fungus is a large genus composed of more than 180 species. An important group of food borne fungi, *A. flavus* is the main producer of the well-known carcinogenic aflatoxins. The presence of this fungus and aflatoxins is of huge concern in terms of food safety (Perrone *et al.*, 2007). Another important fungal pathogen *Sclerotinia sclerotiorum* (*S. sclerotiorum*), is one of the most devastating and soil-borne plant pathogen. The fungus attacks several plant parts and causes stalk rot/ wilt, rot is a major yield-limiting factor and yield losses can reach up to 100% when the climatic conditions are favorable for the fungus growth (Davar *et al.*, 2013).

Therefore the present study was aimed to evaluate the effect of partially purified fractions of acetone extract of *A. nilotica* on development of *C. capitata*, *B. zonata*, *A. flavus* and *Sclerotinia sclerotiorum*.

MATERIALS AND METHODS

Preparation of plant extract

Egyptian tree; *Acacia nilotica* (*A. nilotica*), Kingdom of Plantae was evaluated in this study. The fruit pods of this tree were bought randomly in dried form from local markets located at Alexandria Governorate. The *A. nilotica* tree was identified by Department of Horticultural Crops, Agricultural Research Center, Alexandria. Pods extracted and fractionated, then, the extract was subjected for phytochemical analysis tests based on the standard methods provided by Mbatchou *et al.*, (2011).

Fractionation of *A. nilotica* pods crude extract by column chromatography

All solvents (ethanol, n-hexane, ethyl acetate and methanol) were analytical grade 98 - 99% - Merck) and the other chemicals; Silica gel (200-400) mesh for chromatography, anhydrous sodium sulfate and phytochemical analysis reagents were purchased from Sigma Chemicals Co. The extract was fractionated using glass chromatographic columns (50 cm L/1 cm i.d) packed in sequence with 1g of anhydrous sodium sulfate, 10 g

normal phase silica gel and covered with 1g anhydrous sodium sulfate. The column was pre-washed with 25 ml of n-hexane and then, 5 ml of the extract was transferred to the column. Elution was employed a step gradient elution solvent system from low to high polarity viz. 1) n-hexane 100%, 2) n-hexane: Ethyl acetate 50:50%, 3) n-hexane: ethyl acetate 25:75%, 4) ethyl acetate 100% , 5) ethyl acetate: methanol 50:50%, 6) ethyl acetate: methanol 25:75%, 7) methanol 100%. (Jebasingh *et al.*, 2011). The fractions obtained were concentrated to 1 ml, and analyzed qualitatively using GC-MS.

Gas Chromatography-Mass Spectrometry (GC-MS) analysis

The fractions obtained were analyzed using GC-MS under the following optimum operating condition: Capillary column TG5-MS (30*0.32 mm*0.25 μ m thickness) was used. Helium used as carrier gas at 1 ml/min, splitless injection mode at 250°C. Oven temperature program was start at 80°C (3 min hold) then elevated at 4°C/min to 280°C (5 min hold). For MS, the mass transfer line temperature was 290°C, and ion source temperature at 220 °C. The MS was operated in the electron ionization mode with electron energy of 70 eV, and quadruple mass analyzer at mass range of 50-500 amu, with solvent delay at 5 min. NIST library-08 was used for compounds identification (in the fractions injected) depending on the mass spectrum of its fragmentation pattern matched with the given library mass spectrum.

Evaluating the Insecticidal and Antifungal activities of *A. nilotica* extract

The bioassay experiments were carried out using the *A. nilotica* extract to study its potential biocidal activity against the tested insects (*C. capitata* and *B. zonata*) and fungi (*A. flavus* and *Sclerotinia sclerotiorum*).

Insecticidal effects of *A. nilotica* pods extract

To conduct toxicity tests, two pests choice of order of Diptera, Tephritidae family that cause a lot of losses for many economic fruit crops, these pests are Mediterranean fruit fly; *Ceratitis capitata* (*C. capitata*) and peach fruit fly; *Bactrocera zonata* (*B. zonata*). Pupae of the two pests were obtained from the Department of Lesions Horticultural Crops - Plant Protection Institute - Agricultural Research Center – Giza, Egypt. Wild flies (pupae of *C. capitata* or *B. zonata*) were collected from infested citrus fruit fields and kept in separate adult cages till eclosion. The old strain of flies was maintained under optimum laboratory conditions before the toxicity assessment experiments as illustrated by Mahmoud (2014). Eggs, larvae, and adult stages of both *C. capitata* and *B. zonata*, were divided into five groups in addition to control group, each consisting of 10 eggs or larvae or adult (flies). Three replicates were performed for each treatment of egg stage of *C. capitata* and *B. zonata* at concentrations of 0, 200, 400, 700, 800 and 1000 ppm of acetone extract. Larvae of both the tested insects extract at concentrations of 0, 200, 300, 500, 750 and 1000 ppm of acetone extract. Six groups of adult flies were treated with 0, 100, 200, 300, 400 and 500 ppm of acetone

extract. Egg, larva and adults mortality percentages were observed with time and corrected for control mortality for estimation of the mortality percentages.

Fungicidal effect of *A. nilotica* extract

The phytopathogenic fungi used in this experiment were *Aspergillus flavus* (*A. flavus*) and *Sclerotinia sclerotiorum* (*S. sclerotiorum*), were obtained from the post-harvest infected guava and orange fruits showing typical decline symptoms. The fungus strains were selected for their implication in the contamination and the deterioration of the foodstuffs by production of mycotoxins. The collected fungi were identified by the Plant Pathology Department, Plant Protection Research Institute, Agricultural Research Center, Alexandria. Both of *A. flavus* and *S. sclerotiorum* colony fungus were prepared as described by the method of Javeria *et al.*, (2014). Mycelium was isolated in the form of small sections (5mm in diameter) from infected host tissue of orange and guava separately, incubated at 25 \pm 1°C for 7 to 10 days under sterilized conditions in petri dishes contain potato-dextrose agar (PDA) media. Mycelium disc of *A. flavus* or *S. sclerotiorum* was treated with different concentrations of extract (100, 250, 500, 750 and 1000 ppm). The treated and control petri plates were inoculated for 7 - 10 days at 25 \pm 1°C. On the other side, Antifungal effect of *A. nilotica* extract against mycelial growth of *A. flavus* or *S. sclerotiorum* was evaluated through the disc diffusion method compared to inhibition growth of control according to the following formula: $IP = [(dc - dt)/dc] \times 100$. Where: (IP) is the inhibitory percentage, (dc) is the mycelium diameter in the control petri dish, and (dt) is the mycelium diameter in the natural extracts treated petri dish.

RESULTS AND DISCUSSION

Phytochemical constituents of extract

The results of chemical analysis for the studied extract of *A. nilotica* pods revealed that the presence of phytochemicals constituent groups such as alkaloids, terpenoids, flavonoids, tannins and steroids, while saponins was absent. This finding is in agreement with many of the previous studies; e.g., Seigler (2003), Jigam *et al.*, (2010), Malviya *et al.*, (2011), Abdul-Wadood *et al.*, (2013), Sarkiyayi and Abdul Rasheed (2013), Auwal *et al.*,(2014) and Bwai *et al.*, (2015).

All, in general, reported that a number of secondary metabolites including alkaloids, terpenes, flavonoids and tannins were identified in the extract of various *Acacia* species. Gas chromatography–mass spectroscopy (GC-MS) analysis

The common identified compounds by GC-MS analysis in the different fractions of the *A. nilotica* pods extract listed in Table (1) were as follows: Piperitone, elemol, ethyl palmitate, stigmaterol, heptacosane, palmitic acid, tetradecanoic acid, ethyl gallate, campesterol, octadecanoic acid, hexadecanoic acid, gallic acid, β -sitosterol, pyrogallol, pyrocatechol, guanosine, desul - phosinigrin, β -eudesmol, ethyl iso-allocholate and estradiol, 3-deoxy.

Table 1. Identified compounds in the fractions of *A. nilotica* pods extract.

Fractions	Eluting solvents system	Identified compounds	Chemical group
F ₁	n-hexane; 100%	Piperitone - Elemol	Terpenoids
F ₂	n-hexane; ethyl acetate; 50:50%	Ethyl palmitate Stigmasterol	Terpenoids Steroids
F ₃	n-hexane: ethyl acetate; 25:75%	Heptacosane - Palmitic acid- Tetradecanoic acid Ethyl gallate Campesterol - Octadecanoic acid Hexadecanoic acid	Terpenoids Tannin Steroids Terpenoids
F ₄	Ethyl acetate; 100%	Gallic acid β-Sitosterol	Tannin Steroids
F ₅	Ethyl acetate: methanol; 50:50%	Pyrogallol - Pyrocatechol Guanosine	Tannin Alkaloids
F ₆	Ethyl acetate: methanol; 25:75%	Desulphosinigrin	Alkaloids
F ₇	Methanol; 100%.	β-Eudesmol Ethyl iso-allocholate - Estradiol, 3-deoxy	Terpenoids Steroids

It was noticed that each fraction contain a major of certain compounds differ than other fractions based on partitioning (or polarity) of the extract components under different solute systems.

As the present study, many of the similar previous researches e.g., Singh *et al.*, (2010) and Malviya *et al.*, (2011) identified variety of phytochemical compounds of the essential oil extracted from different plants including *A. nilotica*. The variability in active compounds and their concentrations in the *A. nilotica* extract is expected to affect in the biological activities against different pests.

Insecticidal effects of treatment

Egg stage of *C. capitata* and *B. zonata*

Table (2) shows some differences in the egg hatchability between *C. Capitata* and *B. zonata*, and illustrates the ovicidal activity of *A. nilotica* extract by explain the percentage of the egg hatch for both insects with different concentrations of extract (0, 200, 400, 700, 800 and 1000 ppm).

The results show that the egg hatchability percentages of *C. Capitata* and *B. zonata* were decreased gradually, and averaged at 26.1 and 33.3%, respectively.

In general, it is clear that the *C. Capitata* eggs more susceptible for *A. nilotica* extract levels than *B. zonata*.

Table 2. Effects of the tested extract of *A. nilotica* pods on egg hatchability for both *C. Capitata* and *B. zonata*.

<i>A. nilotica</i> extract concentration (ppm)	% Egg hatchability (Mean ± SE)	
	<i>C. Capitata</i>	<i>B. zonata</i>
0.0	80.0 ± 1.9	80.0 ± 1.3
200	70.0 ± 1.8	76.7 ± 3.3
400	53.3 ± 2.5	70.0 ± 4.1
700	40.0 ± 1.3	53.3 ± 2.6
800	13.3 ± 1.6	20.0 ± 1.3
1000	0 ± 0	0 ± 0
Average (%)	26.1	33.3

Larvae stage of *C. capitata* and *B. zonata*

The knockdown effect of the extract was tested for larvae of *C. capitata* and *B. zonata* in separate bioassay experiments. The results presented in Tables (3 and 4) shows that the percentage of larval mortality was increased in relation to the concentration and exposure period in all cases as compared to the untreated group, where the percentage of larval mortality was 3.33% at 200 ppm and 100% at 1000 ppm of *A. nilotica* extract after 48 hrs and 72 hrs exposure periods, respectively.

Table 3. Larvicidal effect of different concentrations of *A. nilotica* pods extract on larvae of *C. Capitata*.

<i>A. nilotica</i> extract conc. (ppm)	Mortality percentages (%) at different times									
	12 hrs.	24 hrs.	48 hrs.	72 hrs.	4 days	5 days	6 days	7 days	8 days	9 days
Control (0.0)	0	0	0	0	0	0	3.33	0	6.66	0
200	0	0	3.33	10	26.7	36.7	56.7	80	93.3	100
300	0	0	3.33	13.3	26.6	53.3	73.3	100	0	0
500	0	10	20	40	66.7	86.7	100	0	0	0
750	16.7	40	56.7	83.3	100	0	0	0	0	0
1000	26.6	50	76.6	100	0	0	0	0	0	0

Table 4. LC₅₀ values and their 95% confidence limits for larvae of *C. Capitata* exposed to *A. nilotica* pods extract.

parameters	LC ₅₀ of <i>A. nilotica</i> extract (ppm)	95% Confidence of Lower – Upper Limits (ppm)		Slope ± SE
Exposure period				
48 hrs	714.1	1263.1	1874.6	5.24±0.7
72 hrs	504.4	-	-	1.93±0.2
4 days	389.8	-	-	2.78±0.5
5 days	261.1	600.8	1072.0	3.61±0.5

Moreover, the toxicity of the essential oil against larvae of *C. capitata* showed that the LC₅₀ were; 714.1, 504.4, 389.8 and 261.1 ppm, respectively.

The observations during the experiment appeared the larval period and total development period were found to be prolonged with the tested extract of *A. nilotica*.

Concerning the larvae stage of *B.zonata*, tables 5 and 6 illustrates that the larvicidal activities of the tested *A. nilotica* pods extract on larvae of *B.zonata* through LC₅₀

were; 792.7, 551.8, 372.4 and 278 ppm after exposure times of 48 hrs., 72 hrs., 4days and 5 days, respectively.

Where the lowest concentration; 200 ppm caused

approximately 6.67% mortality in larvae insect population after 72 hrs compared to 100 % mortality at the highest concentration (1000 ppm) after the same period.

Table 5. Effects of the tested extract of *A. nilotica* pods on larvae of *B.zonata*.

<i>A. nilotica</i> extract conc. (ppm)	Mortality percentages (%) at different times									
	12 hrs.	24 hrs.	48 hrs.	72 hrs.	4 days	5 days	6 days	7 days	8 days	9 days
Control (0.0)	0	0	0	0	0	0	0	0	0	0
200	0	0	0	6.67	20	30	46.7	73.3	90	100
300	0	0	0	6.67	23.3	46.7	70	96.7	100	0
500	0	10	20	46.7	73.3	100	0	0	0	0
750	13.3	33.3	50	63.3	90	100	0	0	0	0
1000	13.3	40	63.3	100	0	0	0	0	0	0

Table 6. LC₅₀ values and their 95% confidence limits for larvae of *B. zonata* exposed to *A. nilotica* pods extract.

parameters Exposure period	LC ₅₀ of <i>A. nilotica</i> extract (ppm)	95% Confidence of Lower – Upper Limits (ppm)	Slope ± SE
48 hrs	792.7	1596.8 3354.11	3.95±0.63
72 hrs	551.8	- -	5.52±0.70
4 days	372.4	- -	4.04±0.36
5 days	278	- -	3.61±0.50

Adult stage of *C. capitata* and *B. zonata*

Results listed in tables 7 and 8 shows that the percentage of adult mortality was increased with increasing the concentration and the time of exposure compared with

the untreated control. The tested plant extract have promising repellent activity against tested insects. Results demonstrate the percentage of adult *C. capitata* mortality with time after exposure to different concentration of *A. nilotica* essential oil. Results revealed that this oil at lowest concentration 100 ppm caused approximately 3.33 % mortality in adult insect population after 12 hrs compared to 100% mortality at the highest concentration of 500 ppm after 48 hrs. The insecticidal activity of the essential oil of *A. nilotica* was evaluated on the basis of mortality percentages and the estimated LC₅₀ values of *A. nilotica* pods extract. It was decreased from 1253 to 255 to 131 and to 84 ppm with increasing of *C. capitata* exposure periods; 24, 36, 48 and 60 hrs, respectively, indicating that this extract has a promising insecticidal activity.

Table 7. Effects of the tested extract of *A. nilotica* pods on adult of *C. capitata*.

<i>A. nilotica</i> extract conc. (ppm)	Mortality percentages (%) at different times									
	3 hrs.	6 hrs.	12 hrs.	24 hrs.	36 hrs.	48 hrs.	60 hrs.	72 hrs.	5 days	
Control (0.0)	0	0	0	0	0	3.33	0	0	6.67	
100	0	0	3.33	16.7	26.7	43.3	56.7	79.9	100	
200	3.33	10	16.7	33.3	50	63.3	66.7	86.7	100	
300	0	3.33	6.6	26.6	50	60	83.3	100	0	
400	0	3.33	10	26.6	50	73.3	100	0	0	
500	3.33	16.7	20	43.3	76.6	100	0	0	0	

Table 8. LC₅₀ values and their 95% confidence limits for adults of *C. capitata* exposed to *A. nilotica* pods extract.

parameters Exposure period	LC ₅₀ of <i>A. nilotica</i> extract (ppm)	95% Confidence of Lower – Upper Limits (ppm)	Slope ± SE
4 hrs	1253	- -	0.8133±0.25
36 hrs	255	- -	1.485±0.238
48 hrs	131	1394.77 50215.78	1.148±0.283
60 hrs	84	540.03 5790.87	1.532±0.389

The insecticidal activity of the essential oil of *A. nilotica* was also tested with respected to adults of *B. zonata*. The percentage of observed mortality was illustrated in tables 9 and 10.

The results show toxic efficacy of essential oil to the adults of *B. zonata* and also have repellent activity, so, it has been added buminal as food attractant. The percentage mortality of *B. zonata* adults increased by increasing the time of exposure and the concentration of

the *A. nilotica* pods extract.

Where the lowest mortality was 6.7% after 12 hrs at the concentration of 100 ppm, while the adult's mortality reached to 100% after 60 hrs, with the highest concentration at 500 ppm. Furthermore, the results indicated that the essential oil derived from pods of the experimental tree; *A. nilotica* possesses insecticidal activity against both insect pests, but the adults of *C. Capitata* was noticed as more susceptible than of *B. zonata*.

The results of this study are in agreement with Kamaraj *et al.*, (2011) who evaluated the larvicidal and repellent activities of ethyl acetate extracts of the seeds of *Acacia concinna* against larvae of *Anopheles stephensi* and *Culex quinquefasciatus* at five concentrations; 31.25, 62.50, 125.00, 250.00, and 500.00 ppm under the laboratory conditions. Results revealed that the plant extract showed repellent properties and moderate larvicidal effects after 24 h and 48 h of exposure at 500 ppm of the seed extract.

Table 9. Effects of the tested *A. nilotica* pods extract on adult of *B. zonata*.

<i>A. nilotica</i> extract Conc. (ppm)	Mortality percentages (%) at different times								
	3 hrs.	6 hrs.	12 hrs.	24 hrs.	36 hrs.	48 hrs.	60 hrs.	72 hrs.	5 days
Control (0.0)	0	0	3.33	0	0	0	0	0	6.66
100	0	0	6.7	13.3	20.6	36.7	43.3	53.3	100
200	0	6.7	10	20	23.3	43.3	56.7	80	100
300	0	0	10	26.7	36.7	56.6	83.3	100	0
400	0	3.33	20	33.3	56.6	70	90	100	0
500	6.7	20	26.7	56.7	76.7	90	100	0	0

Table 10. LC₅₀ values and their 95% confidence limits for adults of *B. zonata* exposed to *A. nilotica* extract.

parameters Exposure period	LC ₅₀ of <i>A. nilotica</i> extract(ppm)	95% Confidence of Lower – Upper Limits (ppm)		Slope ± SE
24 hrs	567.62	-	-	1.690±0.270
36 hrs	327.89	-	-	2.19±0.262
48 hrs	191.40	-	-	1.93±0.244
60 hrs	131.28	517.01	919.79	2.37±0.272

The previous similar studies revealed that many of plant extracts has insecticidal, antibacterial, antifungal and antimolluscicidal activities, and antioxidant property. From these, Edriss *et al.*, (2012) studied larvicidal effects of three extracts (water, ethanol and petroleum ether) prepared from *A. nilotica* (leaves and fruits) against *Anopheles arabiensis*. The results showed that the petroleum ether extracts exerted better mortality effects than other extracts, with fruits treatments being superior to leaves in all cases.

There is correlation between the results of this study and the results obtained by Abdul-lahi *et al.*, (2014). They studied the comparative assessment of the efficacy of *A. nilotica* bark and root powder on the mortality of *Sitophilus zeamais*, which is one of the most important pests of maize in storage. Highest mortality of the weevil (100%) was observed after 144 h of treating the maize grain with the highest treatment level (1.5 g) of both bark and root powders when compared with other treatment levels (0.5 and 1.0 g) which recorded 100% of the weevil only after 168 h of treatment.

On the other hand, Gautam and Satwinder, (2015) found that the two partitioned fractions viz. ethyl acetate and water of acetone bark extract of *A. nilotica* were tested for their influence on the development of larvae of *Spodoptera litura*. Both fractions showed a toxic influence on larvae of *S. litura* but the effect was markedly greater with ethyl acetate fraction. The toxic effects were manifested in the form of increased larval mortality, reduced pupal weight, decreased adult emergence and prolongation in larval, pupal and total development period.

Fungicidal activity of the *A. nilotica* extract against *A. flavus* fungus growth.

The antifungal efficacy of *A. nilotica* essential oil against pathogenic fungi of *A. flavus* and *S. sclerotiorum* was carried out under *In vitro* conditions. The obtained results showed in Figure (1) revealed effective inhibitor of *A. nilotica* pods against the tested pathogenic fungi. The antifungal efficacy of ethanolic extract causes weak effective of growth inhibition at percentage ranging from 4.05 to 37.48% when the concentration increased from 100 to 1000 ppm, respectively.

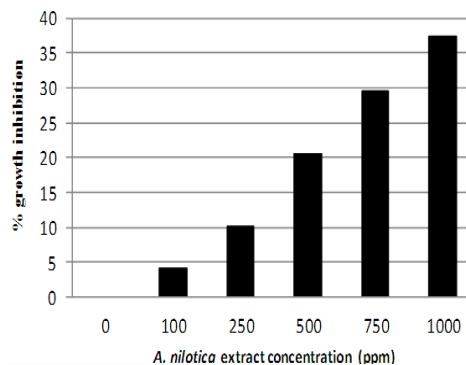


Figure 1. Antagonistic effect of different concentrations of *A. nilotica* extract on *A. flavus* fungus growth

Fungicidal activity of the *A. nilotica* extract Against *S. sclerotiorum* fungus growth

Figure (2) illustrates the inhibition activity of essential oil of *A. nilotica* with different tested concentrations for growth of mycelium *S. sclerotiorum*. The extract from the tested tree pods possess antifungal activity against growth of the tested fungal strain at various concentrations. The minimum inhibitory concentration (100 ppm) of *A. nilotica* extract causes 13.85% of inhibition, while 1000 ppm as maximum of extract inhibit 60% of the mycelium *S. sclerotiorum* growth.

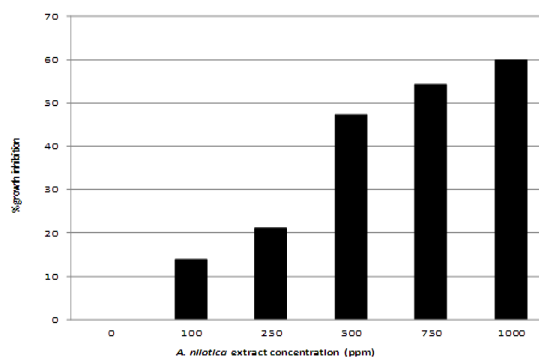


Figure 2. Antagonistic effect of different concentrations of *A. nilotica* extract on *S. sclerotiorum* fungus growth

These results are in agreement with many of the previous researches, e.g., Mahesh and Satish, (2008). They revealed that the bark and leaf methanolic extracts of *A. nilotica* showed significant antifungal activity against *A. flavus* when compared with the root extract. Mohana *et al.*, (2011) showed significant antifungal activity of methanol extract of fresh leaves of *A. nilotica sp.* against pathogenic fungal species; *A. flavus* tested in vitro. Moreover, Prabhakar *et al.*, (2012) resulted that the ethanol extract of

A. nilotica was more fungal inhibitory activity when compared to ethyl acetate extract against *Aspergillus flavus* followed by *Aspergillus fumigatus*. On the other hand, Rathod and Pawar, (2012) explained that the plant extracts of *Azadirachta indica* A. Juss and *Acacia nilotica* (L.) showed inhibitory effect on linear growth of species of fungi belonging *Aspergillus flavus*, *Aspergillus fumigatus*, and *Penicillium chrysogenum*. The results of Abdul Rahman et al., (2014) are confirm the obtained results in the present study. They indicated that the ethanolic and chloroform extracts of *A. nilotica* L. cause 4.91 and 7.0% growth inhibition against *A.niger*, whereas at 4.61% and 10.9% growth inhibition against *A. flavus* fungi. Also, Bwai et al., (2015) showed an increase in the zone of growth inhibition of *Aspergillus niger*, *Aspergillus flavus*, and *Fusarium oxysporum* with increasing the concentration of the *A. nilotica* fruit ethanolic extract. The present study recommend that the phytochemical compounds identified in the tested pods of *Acacia nilotica* are important and have commercial interest to pesticide formulation companies towards the possibilities for replacing the synthetic chemicals by natural products as botanical pesticides for treatment of various crop pests for achieving more safe food for humans and protect our environment.

REFERENCES

- Abdul Rahman, A., Abdul Shakoob, Zaib, G., Mumtaz, A.S., hesham, Y. and Abdul Aziz, N. (2014). Comparative antimicrobial activity of *Acacia nilotica* L. leaves extracts against pathogenic bacteria and fungi. Journal of Medicinal Plant Research, 8(29): 975-982
- Abdul-lahi, N., Umar, I., Tukur, Z. and Babura, S. R., (2014). Comparative efficacy of the bark and root powders of *Acacia nilotica* against maize weevil *Sitophilus zeamais* (Motschulsky) (Coleoptera :Curculionidae) in Kano State of Nigeria. African Journal of Agricultural Research. 9(6) :588 - 592
- Abdul-Wadood, Mehreen, G., Syed, B. J., Muhammad, N., Ajmal, K., Rukhsana, G. and Asnad, (2013). Phytochemical Analysis of Medicinal Plants Occurring in Local Area of Mardan. Biochem Anal Biochem., (2): 2-4
- Auwal, M. S., Saka, S., Mairiga, I. A., Sanda, K.A., Shuaibu, A., Ibrahim, A. (2014). Preliminary phytochemical and elemental analysis of aqueous and fractionated pod extracts of *Acacia nilotica* (Thorn mimosa). Veterinary Research Forum; 5 (2):95-100
- Bwai, M.D., Uzama, D., Abubakar, S., Olajide, O.O. Ikkokoh, P.P. Magu, J., (2015). Proximate, elemental, phytochemical and anti-fungal analysis of *Acacia nilotica* fruit. Pharmaceutical and Biological Evaluations, 2 (3): 52-59.
- Cohen, H and Yuval, B (2000). Perimeter Trapping Strategy to Reduce Mediterranean fruit fly (Diptera: Tephritidae) Damage on Different Host Species in Israel. J.Econ. Entomol. 93(3): 721-725
- Davar, R., Darvishzadeh, R., and Majd, A. (2013). Changes in antioxidant systems in sunflower partial resistant and susceptible lines as affected by *Sclerotinia sclerotiorum*. Biologia., 68(5): 821—829
- Edriss, A. E., Abdalla, A. S., and Zuhair, A. A., (2012). Preliminary studies on phytochemicals and larvicidal effects of *Acacia nilotica* L. extracts against *Anopheles arabiensis* Patton. Scientific Research and Essays, 7(50):4253-4258
- Gautam, S. and Satwinder, K.S (2015). Influence of partially purified fractions of acetone extract of *Acacia nilotica* (L.) on development of *Spodoptera litura* (Fab.). International Journal of Advanced Research, 2 (3):1008-1012
- Gurjar, M. S., Shahid, A., Masood, A. and Kangabam, S. S., (2012). Efficacy of plant extracts in plant disease management. Agricultural Sciences, 3(3):425-433
- Javeria, S., Kumar, H., Gangwar, R. K., Tyagi, S. and Yadav, R. S. (2014). Isolation of Stem rot Disease Causing Organism of Brinjal and their in-vitro Inhibition with Fungicides and Bio-control Agents, 83(9-2):1662-1670
- Jebasingh, E. J. Rajesh, R.P., and Lakshmikandan, M. (2011). Antibacterial activity of seaweed *ulvalactuca* against fish pathogens isolated from marine fish *katsuwonus pelamis*. International Journal of Pharmacy & Technology. 3 (2):2306-2314
- Jigam, A.A., Akanya, H.O., Dauda, B.E. and Okogun, J.O., (2010). Polygalloyl tannin isolated from the roots of *Acacia nilotica* Del. (Leguminosae) is effective against *Plasmodium berghei* in mice. J. Med. Plants Res., 4(12): 1169-1175.
- Kamaraj, C., Abdul Rahuman, Asokan, B., Gandhi, E., Abdul Abduz, Z., and Thirunavukkarasu, S., (2011). Larvicidal and repellent activity of medicinal plant extracts from Eastern Ghats of South India against malaria and filariasis vectors. Asian Pacific Journal of Tropical Medicine, 698-705
- Mahesh, B. and Satish, S., (2008). Medicinal Plant against Plant and Human Pathogens. World Journal of Agricultural Sciences, 4 (5): 839-843.
- Mahmoud, M.F., (2014). New indices for measuring some quality control parameters of the Mediterranean fruit fly, *Ceratitidis capitata* (Wied.). Arthropods, 3(1): 88-95
- Malviya, S., Swati, R., Anil, K. and Meena, V., (2011). Medicinal attributes of *Acacia nilotica* Linn. A comprehensive review on ethnopharmacological claims. International Journal of Pharmacy & Life Sciences. Int. J. of Pharm. & Life Sci., 6(2):830-837
- Mbatchou, V.C., Ayeabila, A.J and Apea, O.B. (2011). Antibacterial activity of phytochemicals from *Acacia nilotica*, *Entada africana* and *Mimosa pigra* L. on *Salmonella typhi*. Journal of Animal & Plant Sciences, 10(1): 1248-1258
- Mohan, M., Haider, S. Z., Andola, H. C. and Purohit, V. K. (2011). Essential Oils as Green Pesticides: For Sustainable Agriculture. Journal of Pharmaceutical, Biological and Chemical Sciences. 2 (4):100-106
- Mohana, D. C., Prasad, P., Vijaykumar, V. and Raveesha, K. A., (2011). Plant extract effect on seed-borne pathogenic fungi from seeds of paddy grown in Southern India. Journal of Plant Protection Research. 2(51): 101- 106

- Perrone, G., Suscal, A., Cozzi, G., Ehrlich, K., Varga, J., Frisvad, J.C., Meijer, M., Noonim, P., Mahakarnchanaku, W. and Samson, R.A. (2007). Biodiversity of *Aspergillus* species in some important agricultural products. *Studies in Mycology* (59): 53–66
- Prabhakar, C., Saleshrani, K., Saranraj, P and Tharmaraj, K., (2012). Studies on the antifungal activity of *Turnera subulata* and *Acacia nilotica* against pathogenic fungal pathogens. *International Journal of Recent Scientific Research*. 3(3):149 -154
- Rathod, L. R. and Pawar, P. V. (2012). Antimicrobial activity of medicinal plant to control seed borne pathogen of soybean. *Current Botany*, 3(2): 10-12
- Sarkiyayi, S. and Abdul Rasheed, K., (2013). Properties of *Acacia Nilotica* leaf extract: A preliminary investigation on anti-typhoid. *International Journal of Current Biochemistry Research*, 1(2): 9- 14
- Sarwar, M., Hamed, M., Rasool, B., Yousaf, M. and Hussain, M (2013). Host Preference and Performance of Fruit Flies *Bactrocera zonata* (Saunders) and *Bactrocera cucurbitae* (Coquillett) (Diptera: Tephritidae) For Various Fruits and Vegetables. *International Journal of Scientific Research in Environmental Sciences (IJSRES)*, 1(8):188-194
- Seigler, D.S. (2003). Phytochemistry of *Acacia sensu lato*. *Biochemical Systematics and Ecology*, (31): 845–873
- Singh, R., Singh, B., Singh, S., Kumar, N., Kumar, S. and Arora, S. (2010). Umbelliferone – An antioxidant isolated from *Acacia nilotica* (L.) Willd. Ex. Del. *Food Chem.*, 120: 825-830

تقييم فعالية مستخلص قرون أشجار القرض كمبيدات حيوية حشرية وفطرية
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شملت هذه الدراسة تقييم مدى كفاءة مستخلص الإيثانول لقرون شجرة القرض ضد آفاتان حشريتان (ذبابة الفاكهة وذبابة الخوخ) وكذلك لتثبيط نشاط فطري إسبرجلس فلافس وإسكلروتينيا إسكلروتينيم. كما تم إجراء الكشف الوصفي لتحديد مختلف مجاميع المركبات الكيميائية الموجودة بمستخلص الإيثانول، وتم التعرف على مجاميع لمركبات الألكالويدات، التيربينويدات، الفلافونويدات، الستيرويدات والتانين، ولم يظهر تواجد لمجموعة الصابونين. كذلك تم إجراء تحاليل تفصيلية بواسطة جهاز كروماتوجرافيا الغاز - مطياف الكتلة، وتم تحديد محتوى مجزئات مستخلص الإيثانول لقرون القرض من المركبات المحتمل فعاليتها حيويًا. أوضحت نتائج التجارب العملية لتقييم مدى سمية مستخلص الإيثانول لقرون القرض على ثلاث مراحل عمرية (الحشرة البالغة – اليرقات- البيض) لآفتي ذبابة الفاكهة وذبابة الخوخ، ولقد دلت النتائج على وجود نسبة من الموت للمراحل العمرية المختلفة للآفتان زادت مع زيادة تركيز ووقت التعرض للمستخلص مقارنة بالمجموعة الضابطة. أما بالنسبة لتقييم مدى فاعلية النبات معملًا على تثبيط فطري إسبيررجلس فلافس وإسكلروتينيا إسكلروتينيم، فقد أظهرت النتائج أن مكونات مستخلص قرون شجرة القرض تمتلك فاعلية ملحوظة مضادة لنمو الفطريات المختبرة.