Insecticidal Effect of *Jatropha curcas* Oil on the Aphid *Aphis fabae (Hemiptera: Aphididae)* and on the Main Insect Pests Associated with Cowpeas *(Vigna unguiculata)* in Niger

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Summary

The insecticidal activity of Jatropha curcas *has been evaluated on various crop pests. Oil concentrations of 0.5, 1, 2.5, 5, 10 and 15% were first tested on the black bean aphid (*Aphis fabae *Scop.) as part of an efficacy test conducted in the laboratory. During a second stage, the insecticidal efficacy of 5 and 7.5% oil concentrations was evaluated on the main pests infesting cowpea crops (*Vigna unguiculata *L.) as part of a field test conducted at the University of Niamey (Niger). The results obtained in the two tests demonstrate the biocidal effect of the treatments applied, which increases with the concentration. On the black bean aphid, the biocidal effect increases during the hours following the application of oil before reaching a peak after 4 days. On cowpeas,* J. curcas *oil concentrations of 5% and 7.5% make it possible to reduce the level of attack by aphids (*Aphis craccivora*) by 10 and 50% respectively compared to the control. A 50% and 75% fall in the number of thrips (*Megalurothrips sjöstedti*) and bugs (*Anoplocnemis curvipes*), respectively, can be observed under the same conditions. This treatment made it possible to greatly increase yields compared to the untreated control.*

Résumé

Effet insecticide de l'huile de *Jatropha curcas* sur le puceron *Aphis fabae (Hemiptera: Aphididae)* et sur les principaux ravageurs du niébé (*Vigna unguiculata*) au Niger

L'activité insecticide de l'huile de Jatropha curcas *a été évaluée sur divers ravageurs des cultures. Des concentrations de 0,5; 1; 2,5; 5; 10 et 15% d'huile ont été testées sur le puceron noir de la fève (*Aphis fabae *Scop.) dans un essai d'efficacité réalisé en laboratoire. Dans une seconde étape, l'efficacité insecticide des concentrations de 5 et 7,5% d'huile a été évaluée sur les principaux ravageurs du niébé (*Vigna unguiculata *L.) dans un essai en plein champ réalisé à l'université de Niamey (Niger). Les résultats obtenus dans les deux essais mettent en évidence un effet biocide des traitements appliqués qui augmente avec la concentration. Sur le puceron de la fève, l'effet biocide augmente au cours des heures qui suivent l'application de l'huile pour atteindre un niveau maximum après 4 jours. Sur le niébé, les concentrations de 5% et 7,5% d'huile de* J. curcas *permettent de réduire le niveau d'attaque des pucerons (*Aphis craccivora*) de 10 et 50% respectivement par rapport au témoin. Une diminution du nombre des thrips (*Megalurothrips sjöstedti*) et des punaises (*Anoplocnemis curvipes*) de 50 et 75% respectivement est observée dans les mêmes conditions. Ce traitement a permis une augmentation importante du rendement par rapport au témoin non traité.*

Introduction

Jatropha curcas L. is a shrub of the Euphorbiaceae family, which originated in Central America. This succulent plant is highly resistant to drought. The *Jatropha* genus is widespread in the tropical countries (3). In several West and Central African countries, *J. curcas* is used as a means of delimiting fields, in order to protect cereal crops against the wind and grazing by animals (4). *J. curcas* seeds are rich in an oil used as biofuel, which makes this plant an important subject for research into renewable energies. In addition to its use as a biofuel, Jatropha oil can also be used as a bio-pesticide (10). In fact, several authors have tested the use of oil emulsions against insects that attack stored maize grains, *Sitophilus zeamais*, and mung beans, *Callosobruchus chinensis,* at concentrations of 0.5, 1, 2.5 and 10%. After being stored for 2 months, damage to the grains was reduced to 10% when doses of 10% and 5% were applied to *S. zeamais* and *C. chinensis,*

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respectively. In 2000, the same authors also tested the effect of Jatropha oil on various aggressive bio-agents affecting cotton plants (*Amrarsca biguttula*, *Aphid gossypii* and *Helicoverpa armigera*). Doses of 800 ml and 250 ml/ha were compared against commonly used insecticides (Profenofos at 400 g/ha and Deltametrine at 12.5 g/ha). Jatropha oil showed itself to be more effective than Deltamethrine on *A. gossypii,* while the opposite effect could be observed on A. *biguttula*. For *H. armigera*, synthetic insecticides were more effective than Jatropha oil at the start of treatment, as the oil affects only insect growth and its effect is therefore slower (9).

Our study focuses on the toxic effect of *J. curcas* oil on crop predators. It aims to determine the biocidal effect of different oil concentrations on black bean aphids (*Aphis fabae* Scop.) and the main pests affecting cowpea crops (*Vigna unguiculata*).

Materials and method

Jatropha curcas seeds

Jatropha curcas seeds were harvested in Gaya (Niger) by IBS Agro Industrie in October 2010. The grinding was conducted by means of a "Superior" Deklerck hammer mill with a 4 mm sieve. In order to limit the pressing temperature and based on the equipment available locally, a manual hydraulic press of the ADMGA brand from Burkinabé was used.

1. Breeding of aphids

Broad beans (*Vicia faba* L.) were used as a host plant for breeding *A. fabae*. The seeds were sown in 30 cm × 20 cm boxes, which contained a mixture (1:1) of Vermiculite and Perlite. The plants were infested with aphids at two-leaf stage. The aphids were bred under controlled conditions (22 ± 2 °C, 60 - 80% relative humidity and a photoperiod based on 16 hours of light and 8 hours of darkness).

***J. curcas* oil-based formulation**

The Jatropha oil used in the experiments was formulated using 50% oil, 30% pure ethanol as a stabiliser and 20% gum arabic as an adjuvant in order to fix active molecules on the plant. Using this standard solution, oil concentrations of 0.5, 1, 2.5, 5, 10 and 15% were prepared and the insecticidal activity of these formulations on *Aphis fabae* was evaluated and compared in the laboratory to a control (water + alcohol + gum arabic) and a positive control (insecticide: KB Multisect, dosed at 0.05 g/l of Acetamiprid, $C_{10}H_{11}Cl_N4$, systemic).

Sprayer calibration

A trigger-pump sprayer is used to conduct this test. It is suitable for the application of small quantities of specific products. The number of pressings required to wet a young bean plant was pre-determined as being equal to 7. The volume of these seven pressings is measured using a tare flask in order to determine the weight of the sprayed product. This procedure is conducted five times and the average weight of the products collected in the flask is calculated. The average obtained was 13.7 ml.

Infestation of plants and evaluation of mortality

Twenty aphids at the third larval stage were placed in each cage. The aphids were placed on the plants using a moistened brush. The spraying was conducted after the aphids were left to acclimate for one hour. The number of dead aphids was counted after 24, 48, 72, 96 and 120 hours. An aphid is considered dead if it fails to react when touched by the brush. The average mortality of the aphids (M0) is expressed as a corrected mortality (Mc), taking into account the natural mortality observed on the control (Mt) (Table 1) according to Abbott's formula (1):

$$Mc = \frac{M0 - Mt}{100 - Mt} x100$$

In order to calculate lethal concentrations 50 and 90, a binary logistic regression is produced:

(2)

$$\ln\left(\frac{p_i}{1-p_i}\right) = \beta_0 + \beta_1 X_i$$

(pi: probability, β_0 and β_1 predictors, and Xi as doses in our case).

2. Field test conducted on cowpea pests in Niger

J. curcas oil concentrations of 5 and 7.5% were compared to a negative control (water) and a positive control (Decis: Deltamethrine). The cowpea variety used is TN5-78, as selected by the National Institute of Agronomic Research of Niger (INRAN). This is an early and productive variety (cycle duration lasting 40 days and potential yield of 1.5 - 2 t/ha). The test materials consisted of a Fisher block with four repeat tests. Each simple plot is made up of 10 cowpea plants separated by 1 m between the lines and 0.5 m on the lines. The product was applied at three stages of the plants' development: initiation of flowering (35 days after sowing), 50% flowering (45 days after sowing), shoot initiation (60 days after sowing). The product is applied at floral initiation stage, 50% flowering and shoot formation using a ULV sprayer from 6 am or between 4 pm – 5 pm on 10 plants for each repeat test. The quantities of Jatropha oil applied per ha are 800 ml/ha and 1,200 ml/ha, respectively (10). The total volume of spray mixture applied is 22.5 litres/ha. Observations are made twice per day (8 am – 10 am and 4 pm – 6 pm) the day before treatment and the 3rd, 7th and 11th day after treatment. The insects are collected according to the following methods:

- Thrips: three flowers are harvested per plant and placed in a flask containing ethanol at 70° C. The number of thrips is determined using a binocular magnifier
- Bugs: the number of bugs is counted directly on the plants

Table 1
Corrected mortality of aphids

	Observation periods				
Concentrations	24h	48h	72h	96h	120h
0%	0	0	0.6 ± 0.3	0.6 ± 0.3	0.6 ± 0.3
0.50%	13.1 ± 2.4	38.1 ± 2.6	39.4 ± 2.3	43.1 ± 2.8	43.1 ± 2.8
1%	30.0 ± 2.6	38.7 ± 2.5	55.6 ± 1.7	57.0 ± 1.3	57.0 ± 1.3
2.50%	38.1 ± 2.4	50.6 ± 1.5	60.0 ± 2.9	63.1 ± 2.9	66.2 ± 1.2
5%	56.9 ± 2.3	70.0 ± 1.9	76.2 ± 1.7	78.7 ± 1.3	80.0 ± 1.1
10%	81.2 ± 1.2	83.7 ± 1.4	89.3 ± 1.3	89.4 ± 0.8	89.4 ± 0.8
15%	88.1 ± 1.3	95.6 ± 1.2	97.0 ± 1.1	100.0 ± 0	100.0 ± 0
Acetamiprid	100.0 ± 0	100.0 ± 0	100.0 ± 0	100.0 ± 0	100.0 ± 0

Table 2
Binary logistic regression

Logit	24h	48h	72h	96h	120h
Probability	p< 0.001	p< 0.001	p< 0.001	p< 0.001	p< 0.001
Constant (βo)	-1.77	-1.09	-0.84	-0.77	-0.75
Regression coefficient (β1)	0.36	0.32	0.35	0.37	0.38
Odds ratio	1.44	1.39	1.43	1.46	1.47
Wald's test (z)	14.08	12.49	12.04	11.95	11.91
Probability ratio (G)	282.282	223.95	227.51	236.65	239.95

- Aphids: a scale is used to estimate the number of aphids per leaf: 0: 0 - 50 aphids; 1: 51 - 100 aphids; 2: 101 - 200 aphids; 3: 201 - 300 aphids; 4: 301 - 400 aphids; 5: over 400 aphids

Results

1. Insecticidal efficacy of Jatropha oil on *Aphis fabae*

The corrected mortality of *A. fabae* aphids, which were subjected to different treatments of Jatropha oil is shown in table 1. The higher the Jatropha oil concentration, the greater the mortality (p< 0.001).

The mortality of the aphids varies significantly according to the *J. curcas* oil concentrations (p< 0.001).

The mortality also increases with the duration of the treatment. The number of dead aphids rises as time passes after treatment for all concentrations up to 72 hours. It becomes stable between 96 - 120 hours after treatment.

The results of the binary logistic regression are shown in table 2.

At the 5% threshold required for Tukey's test, the tests are significant, regardless of the time of observation. The (positive) regression coefficient sign β_1 shows that aphid mortality increases according to the concentrations and observation times. The odds ratio shows that, if the *J. curcas* oil concentration is increased by one unit, aphid mortality increases by 14.4% for 24 hours, 13.9% for 48 hours, 14.3% for 72 hours, 14.6% for 96 hours and 14.7% for 120 hours after treatment. Observations conducted after 24 and 48 hours provide more detailed information on the resulting regression model. Based on this binary logistic regression, 50 and 90 lethal doses were estimated (Table 3).

Table 3
Lethal concentration estimates

Hours	CL50	CL90
24h	4.85	10.86
48h	3.33	10.01
72h	2.36	8.53
96h	2.06	7.84
120h	1.95	7.61

The 50% and 90% lethal concentrations decrease according to the duration of treatment. The lethal concentration that makes it possible to kill 50% of aphids is approximately 5% after 24 hours and 3.5% after 48 hours. The lethal concentration that makes it possible to eliminate 90% of aphids is approximately 11% of *J. curcas* oil after 24 hours and 10% for 48 hours.

3.2 Insecticidal efficacy of Jatropha oil against cowpea pests in the fields

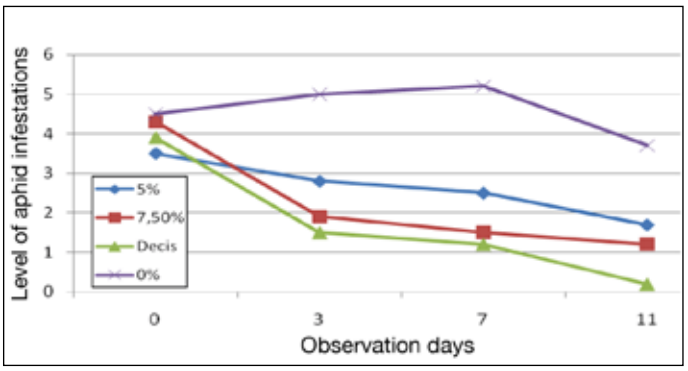

Figure 2: Increased levels of attack by aphids per plant depending on the day of observation after the treatment applied at floral initiation stage

Three categories of insect were evaluated according to development stages of the culture. The aphids were evaluated at floral initiation stage, thrips at 50% flowering stage and bugs at shoot formation stage. Observations were made at each stage: before treatment, and on the 3rd, 7th and 11th days after treatment. Statistical analysis of the data shows that the greater the dose of Jatropha oil, the greater the insecticidal efficacy of this oil ($p < 0.001$).

Increased numbers of aphids were determined depending on the days of observation (Figure 2). Before treatment, the level of aphid infestation was 3.5 – 4.5 for all the plants studied. Three days after the product was applied, reduced aphid infestation was observed with all the treatments. However, for the control sample, the level of infestation increased. Figure 2 shows how the number of thrips increases according to the day of observation.

Flowers were infested by 4 - 5 individual thrips per flower before the products were applied. A fall in the number of thrips is observed until the 11th day after treatment (Figure 3).

Bugs appeared 64 days after sowing, which coincides with the formation of shoots. A fall in the number of bugs per plant can be observed for all the treatments, except for the control sample, for which this number increases (Figure 4).

Table 4 shows average numbers of insects collected during the 3 product applications: aphids at floral initiation stage (35 days after sowing), thrips at 50% flowering stage and bugs at short formation stage. For treatments containing Jatropha oil, a fall in the number of cowpea pests can be observed compared to the control sample, together with an increased yield per plot. This reduction is proportional to the oil content of the formulation applied.

Discussion

Many studies have shown that the toxicity of *J.curcas* oil is due to the presence of phorbol esters (6). These natural organic compounds are tiglianes from the diterpene family. Tiglianes were discovered by Bohm et *al.* in 1934, but their structure was determined in 1967 by Hecker *et al.* [quoted by Mahaela (5)]. According to the model obtained using the binary logistic regression, a 5% *J. curcas* oil concentration would be needed to kill 50% of aphids (*A. fabae*) after 24 hours and 11% to eliminate 90%, whereas an oil concentration of 4% would be required to kill 50% of aphids after 48 hours and 10% to eliminate 90%. These results are comparable to those obtained by Solsoloy *et al.* (10) who tested oil emulsions against insect pests attacking stored maize grain, *Sitophilus zeamais*, and bean weevils, *Callosobruchus chinensis*.

Studies conducted by Ratnadass *et al.* (8) focusing on *Busseola fusca* and *Sesamia calamistis Hampson* (*Lepidoptera: Noctuidae*) on sorghum have shown that the raw oil extracted from *J. curcas* seeds has a larvicidal effect, at concentrations of 0.01% and 1% on *B. fusca* and *S. calamistis*, respectively, and its phorbol ester content at the concentration of 0.025% on *S. calamistis*.

For this reason, the effectiveness of Jatropha oil (1% of the nutrient medium) was compared to that of phorbol esters at 0.025, 0.05 and 0.1% added to the nutrient medium for *S. calamistis* and 0.01, 0.1 and 1% to the

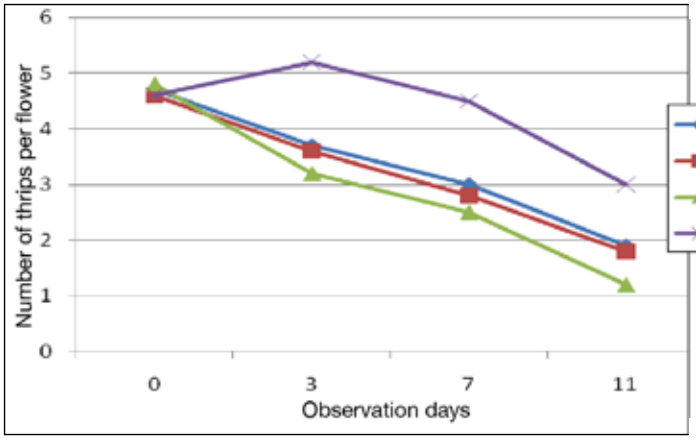

Figure 3: Increased number of thrips per plant depending on the day of observation after application of the treatment at 50% flowering stage

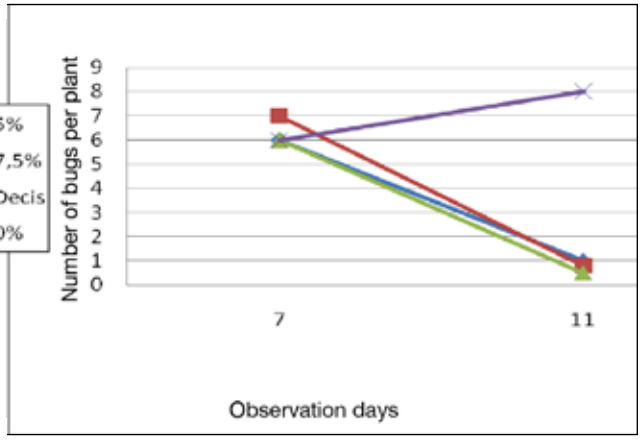

Figure 4: Increased number of bugs per plant depending on the days of observation after the treatment is applied at shoot formation stage on the 56th day after sowing.

Table 4
Average numbers of main pests and plot yields according to *J. curcas* oil concentration

Concentrations	Aphids	Bugs	Thrips	Yield in grains (g/24m²)
0%	4.16 ± 1.30a	6.26 ± 0.20a	6.90 ± 0.29a	30.05 ± 0.96d
5%	3.96 ± 2.73a	3.04 ± 0.15b	3.38 ± 0.18b	160.10 ± 4.16c
7.5%	2.08 ± 1.71b	2.07 ± 0.28c	1.34 ± 0.15c	260.53 ± 8.05b
Decis	1.67 ± 1.67bc	1.23 ± 0.24c	0.85 ± 0.17c	410.10 ± 9.52a

nutrient medium for *B. fusca*. Levels of nymphosis were zero for *S. calamistis* for all doses of the product, for *B. fusca* for treatments using 0.1 and 1.0% oil, while it was 55% for the treatment supplemented by 0.01% and 70% on the control.

Insects represent the major hindrance to cowpea cultivation in semi-arid tropical regions. Effectively combating these pests will make it possible to increase cowpea yields by 10-30% (7). In total, over 100 insect species have been identified as cowpea pests in the world, but only about ten of them have real economic importance in Niger. These pests include aphids (*Aphis craccivora)*; thrips (*Megalurothrips sjöstedti)*; bugs (*Anoplocnemis curvipes)*; stem borers; blister beetles (*Mylabris senegalensis, Coryna argentata* and *Decapotoma affinis)* and weevils (*Bruchidius atrolineatus and Callosobruchus maculatus*) (7). Our study focused on the first three pests. A *J. curcas* oil concentration of 5% makes it possible to reduce aphids by 10% and thrips and bugs by 50%. The 7.5% *J. curcas* oil concentration makes it possible to reduce aphids by 50% and thrips and bugs by 75%. At the same time, increased grain yields of 60-75% were observed when 5 and 7.5% *J. curcas* oil concentrations were used. However, other biological and physical factors may help reduce these insect populations. For example, ladybirds and heavy rain considerably reduce aphids, while blister beetles are also thrips predators (10).

Conclusion

An insecticidal effect by contact has been demonstrated for *J. curcas* oil on aphids (*Aphis fabae*) that attack broad beans (*Vicia fabae*). This toxicity increases as the dose of oil is increased. Insect mortality increases during the hours following the treatment and reaches a peak after 96 hours. *J. curcas* oil has also shown itself to be effective against cowpea (*Vigna unguiculata*) insect pests. On cowpeas, *J. curcas* oil concentrations of 5% and 7.5% make it possible to reduce the level of attack by aphids (*Aphis craccivora)* by 10 and 50%, respectively, compared to the control. A 50 and 75% fall in the number of thrips (*Megalurothrips sjöstedti*) and bugs (*Anoplocnemis curvipes*), respectively, can be observed under the same conditions. This made it possible to significantly increase grain yields compared to the untreated control sample.

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