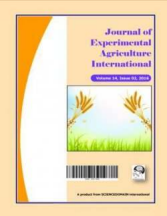

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Toxicity of *Cassia mimosoides* Leaf Extracts Against the Weevil *Callosobruchus maculatus* and Nutritional and Organoleptic Quality Assessment of the Treated *Vigna subterranea* (L.) Verdc

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Authors' contributions

This present investigation was carried with the collaboration of all authors. Authors AM, CS and ENN designed the study. Authors ENN, KHT and CS performed the statistical analysis. Authors AM and CS wrote the protocol. Authors AM and LY wrote the first draft of the manuscript. Authors CS, KHT, LY, KH and ENN managed the analyses of the study. All authors read and approved the final manuscript.

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ABSTRACT

Aims: This present study aimed to assess the toxicity, the inhibition of progeny production and the reduction of weight losses of *Cassia mimosoides* (L.) leaf extracts against *Callosobruchus maculatus* (FAB.) on the bambara groundnut grains as well as the physico-chemical, sensorial and organoleptic characterization of that grains 3 months post-storage.

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Place and Duration of Study: The toxicity tests were done from April to August 2016 in the laboratory of Applied Zoology of the Department of Biological Sciences, University of Ngaoundere. The physico-chemical, sensorial and organoleptic characterization of bambara groundnut were determined in the laboratory of Food Chemical Engineering of the University Institute of Technology, University of Ngaoundéré from October to December 2016.
Methodology: Toxicity of the plant extracts was evaluated by exposing 20 adult weevils to 5, 10, 15 and 20 g/kg concentrations of the plant extracts mixed with bambara groundnut grains and the mortality of *C. maculatus* was monitored at 1, 3 and 6 days post-infestation. The inhibition effect of the plant products was assessed on the production of bambara groundnut pea beetle progeny *F1*. After 3 months of storage, the influence of *V. subterranea* post-storage losses including weight loss, biochemical, sensorial and taste characteristics were also evaluated.
Results: *C. mimosoides* extracts caused a significant mortality of the weevil and also significantly reduced the progeny *F1* production. The hexane extract was revealed the most effective causing 73.65% mortality, reduced completely the progeny *F1* production at the lowest concentration of 5 g/kg. At the same concentration, hexane extract also protected completely bambara groundnut grains up to 3 months from the attack of pea beetle. After 3 months post-storage with the plant extracts, the nutritional value of the bambara groundnut was preserved since no variation in protein, sugar and mineral contents was recorded. Moreover, an improvement in taste, tenderness and crispness of bambara groundnut grains after processing was also noticed.
Conclusion: Thus, *C. mimosoides* products especially hexane extract could be used as a natural safe and biodegradable plant product to protect bambara groundnut grains from the weevil attacks in the storage for at least 3 months without its significant nutritional value degradation and taste change.

Keywords: Toxicity; plant extracts; nutritional value; sensorial characteristics; pea beetle attack.

1. INTRODUCTION

Nowadays, food security is ensured through the protection of agricultural products during storage. In the developing countries especially in sahelian zones, cereals and leguminous plants constitute the basic source of food of the farmers [1,2] and up to 75% the production are stored in the traditional garret [3,4]. Unfortunately, these post-harvest stores are rudimentary [5] and consequently expose the goods to the devastator insects [6]. Among the depreciative insects, *Callosobruchus maculatus* (FAB.) (Coleoptera: Bruchinae) is known as the most cosmopolitan devastator of the leguminous grains and without grain protection measure, can causes around 80 to 90% losses in 12 months [7]. To face the significant problem of the grains losses in storage due to insects, farmers are compelled to search for the prevention measures [8]. For that, synthetic insecticides and fumigants are currently the most widely used to control the postharvest devastator insects [9,10]. Consequently, the misuse of these synthetic insecticides has led to miscellaneous problems including their toxic effect to consumers, their persistence in the environment, and the production of insects resistant to the insecticides [3]. Moreover, these insecticides are relatively costly, scarce and sometimes with doubtful quality (bad preservation condition leading to the loss of the efficacy) in the local markets [5,11,12,13]. The research of the new molecules, taking into account no only the effectiveness of the insecticides becomes a major concern of current time to success an effective grain protection safe for consumers and the environment [14-16]. For these reasons, the uses of plant-based products in the stored products protection program are encouraged [17-19]. Indeed, current studies are aimed to isolate and identify secondary metabolites extracted from the plants, responsible for the anti-insect activity [20]. Previous works were much focused either on essential oils of plants such as *Vepris heterophylla* against *Triboluim castanuim* in the soudano-sahelian zone [4]; of *Eucalyptus saligna, Cupressus sempervirens, Capsicum frutenscens* and *Lantana camara* in the West-Cameroon highland and *Callistemon viminalis* (Myrtaceae) against the adults insects of *Acanthoscelides obtectus* [20] neem oils against *Sitophilus zeamais* [14] as well as vegetable powders [10,22]. However, few works have been carried out on the insecticidal effectiveness of *Cassia mimosoides* secondary metabolites extracted with organic solvents especially against *C. maculatus*. However, the leaves, roots and barks of the plant have already proved their effectiveness as anti-malaria, anti-cancer,

analgesic, antipyretic, antioxidant [23] and *Cassia senna* extracts as anti-insect against *Trogoderma granarium* [24]. Besides, preservation of seeds, the plant metabolite used for preservation could modify the nutritional and organoleptic quality of the treated products. In fact, Rose de Lima et al. [25] reported that the conservation of cowpea with plant essential oils belonging to the family of Myrtaceae improves the organoleptic properties with an improvement in the taste and the flavour of the derived products. It would be thus significant to evaluate the anti-insect activity of this plant extracts against *C. maculatus* and their impacts on the nutritional and organoleptic quality of the preserved products. The objective of this work aims to evaluate the efficacy of *C. mimosoides* leaf extracts against the weevil *C. maculatus* in stored bambara groundnut and also to assess the nutritional and organoleptic qualities of that treated *Vigna subterranea* (bambara groundnut) seeds 3 months post-storage.

2. MATERIALS AND METHODS

2.1 Plant Materials

The fresh leaves of *C. mimosoides* were harvested in September 2014 at Koza (longitude: 13°48'East; latitude: 10°43'North) in the far north region of Cameroon (Fig. 1). The plant was authenticated by the Department of Biological Sciences, Faculty of Science, University of Ngaoundéré, Cameroon. The leaves of *C. mimosoides* were dried under the shade and then crushed in the mortar and passed through 1 mm mesh size sieve. The powder was stored in sealed containers in the refrigerator (-4°C) until uses [14].

The seeds of *V. subterranea* variety used was purchased from the local market of Maroua. The variety could easily be identified based on the grain tegument coloration. After separating from the impurities, bambara groundnut grains were stored at -20°C for 20 days to eliminate living organisms and then acclimated to room temperature during 2 weeks before the use for bioassays.

2.1.1 Plant extraction

The cold maceration method performed of Perry et al. [21] was used for plant extraction. Indeed, 1000 g of the plant leaf powder was macerated in 3000 ml of hexane in 5000 ml glass jar. The maceration was shaking twice a day, during 3 days and then filtrated through Whatman No.1 filter paper to obtain hexane filtrate and residue. Then, the residue was dried and also macerated in 3000 ml of acetone and processed as described previously to obtain acetone filtrate and residue. At last, the dry residue was macerated in the methanol solvent as described previously to obtain methanol filtrate and residue. The filtrates were concentrated using rotary evaporator (BÜCHI R-124) to obtain hexane, acetone and methanol extracts. The dry plant extracts were stored at -4°C until further use. The extraction yield was calculated using the following formula:

Extraction yield (%)=

$$\frac{\text{Weight of the extract obtained (g)}}{\text{Weight of the plant powder used (g)}} \times 100$$

2.1.2 Phytochemical screening of the plant extracts

For the phytochemical screening, of *C. mimosoides* extracts, the presence of alkaloids was tested according to Bidie et al. [26]. N'Guessan et al. [27] method was used to determine total polyphenol compounds. Flavonoids were determined according to Debray et al. [28] method and saponins following the method advocated by Dohou et al. [29]. The presence of steroids, tannins, les coumarins and triterpenoids was assessed according to Fankam et al. [30] method.

2.2 Bioassays

2.2.1 Toxicity of plant products against *C. maculatus* adults

The toxicity of the *C. mimosoides* leaf extracts against *C. maculatus* was evaluated according to Nukenine et al. [14]. Each extract was dissolved in the extraction solvent concentrations of 0.25; 0.5; 0.75 and 1g/mL of hexane, acetone and methanol. The extracts were mixed uniformly with 50 g of bambara groundnut grain in the 900 mL glass bottle. The negative control was made by mixing 50 g of grains with 1 mL of each solvent used separately. The grains in the bottle were left in open air for 2 hours for solvent evaporation and 20 pea beetles (mixed males and females; obtained and reared in the same bambara groundnut bought from Maroua marked), were introduced into each bottle test preparation. Four replicates were maintained for each treatment and the pea beetle mortality was assessed after 1, 3 and 6 days post-treatment.

2.2.2 Effect of plant products in the progeny F1 production

On sixth days after treatment, the dead and live pea beetles were removed from the monitoring bottles and the treated bambara groundnut was kept for observation every week until emergence of pea beetles. The number of pea beetles emerged was recorded every day until 5 last days without emergence of the pea beetles.

2.2.3 Effect of plant products on grain damaged by *C. maculatus* during 3 months storage

For each plant extract, four stock solutions of 0.25; 0.5; 0.75 and 1 g/mL doses were prepared. One mL of each concentration was mixed uniformly with 50 g of pea groundnut in the glass bottles and in the negative control, only solvent used was mixed with bambara groundnut. In each test bottle, 20 male and female pea beetles, 2 days old were added and sealed with muslin cloth. There were four replicates for each dose and after 3 months post-treatment, the number of dead and live pea beetles as well as the number of damaged and undamaged groundnuts was recorded. The weights of damaged and undamaged bambara groundnut grains were also assessed. The weight losses were evaluated according to the method of FAO [31] following the formula of Adam and Schulten [32].

$$P\% = [[(Wu \times Nd) - (Wd \times Nu)] / Wu\ (Nd + Nu)] \times 100$$

Wu= weight of undamaged grains; Wd= weight of damaged grains; Nd= number of damaged grains, Nu= number of undamaged grains.

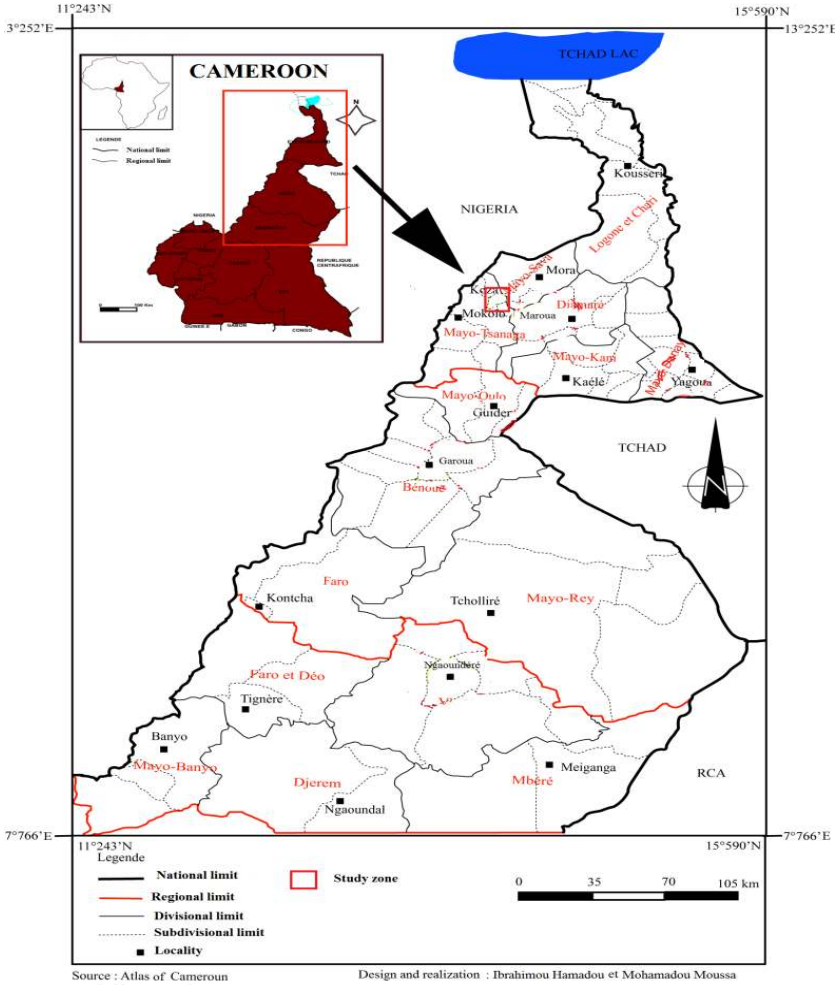

Fig. 1. Map of Cameroon showing Koza (Far north region of Cameroon), the place where the plant material was collected

2.3 Evaluation of Nutritional Value and Organoleptic Characteristic of Damaged and Undamaged Grains Post-storage

The physicochemical characteristics and nutritional values evaluated before and after storage included: the moisture and dry matter [33], ash content [34], proteins [35], lipids [36] and carbohydrates [37]. The method of López-Mejía et al. [38] was used for the determination of the total phenolic contents in the extracts and treated seeds. The total tannin content was determined according to Makkar et al. [39] method.

2.4 Sensorial Analysis of Pea Groundnut

For the sensory analysis, the most appreciated consumed bambara groundnut variety in the various zones of studies which is Kodek variety was used. The undamaged treated seeds with the smallest effective concentration were used. The treated seeds at the end of three months of storages were divided in two samples. Considering the culinary practices of the population for bambara groundnut consumption, one sample was cooked and the other was roasted according to traditional protocols of preparation obtained on the field. The purpose of the sensory analysis study was to analyze and to interpret the organoleptic characteristics of the products as perceived by the sense organs [40] including the color, the odor, the texture and the taste of the products derived from the bambara groundnut. The products thus obtained were submitted to the hedonic test carried out at the sensory analysis laboratory according to the principle of a classification test. That test consists for one characteristic, to arrange by increasing intensity, the samples presented simultaneously to 20 tasters of sexes, ages and different zones as well as their familiarity to consuming these products. At the end of this test, the numerical notations ranging from 1 to 7 in which 1 corresponds to "extremely bad" and 7 "extremely good" were analyzed using test Friedman test.

2.5 Statistical Analyses

Data for mortality, progeny F1 production and damages and weight losses percentages were transformed to arcsin√(x/100) and submitted to ANOVA analysis using statistical analysis software SAS [41,42], Tukey test (P<0,05) was applied for mean separation. Probit analysis was also conducted to determine lethal concentrations that cause 50% (LC_{50}) and 95% mortality (LC_{95}) of *C. maculatus*. Abbott's formula [43] was used to correct for control mortality when mortality in the control is comprised between 3% and 10% before probit analysis and ANOVA. For sensorial analysis, data were analyzed using Friedman test. Fisher coefficient was calculated using Chi-square test (x^2). Different samples comparison by calculating Fc coefficient and compared with "S" value in the Chi-square table at α=5% and degrees of freedom k-1 [44].

3. RESULTS

3.1 Yield of Extraction

The hexane, acetone and methanol extraction yield obtained from cold maceration of 1000 g of plant powder are presented in Table 1. The extraction yield varied from one solvent to another. Acetone and methanol extraction yield values were high compared to the low yield obtained with hexane solvent.

Table 1. Extraction yield of the plant extraction

Extracts	Weight of extract	Extraction yield (%)
Hexane	28	2.8
Acetone	101	10.1
Methanol	81	8.1

3.2 Phytochemical Screening of the Plant Extracts

Results of the phytochemical screening of hexane, acetone and methanol extracts of *C. mimosoides* are presented in Table 2. *Cassia mimosoides* plant extracts possessed a wide variety of phytochemical components distributed in different solvent extraction used. Phenolic compounds and flavonoids were abundant in polar solvent extracts (methanol and acetone) and were absent in the non-polar solvent extract (hexane). Triterpenoids, steroids and alkaloids were abundantly present in both polar solvent (acetone, methanol) and non-polar solvent (hexane) extracts.

3.3 Adult Mortality

The results of the mortality of *C. maculatus* adults exposed to the hexane, acetone and methanol extracts of *C. mimosoides* are presented in Table 3. Compared to the negative control, all extracts of *C. mimosoides* exhibited a significant adult mortality of *C. maculatus*. The

mortality of the pea beetle varied with the different plant extracts and the concentrations applied (F= 2.4 - 3671.4) and the exposure period (*F*= 51.54 - 5032.83 and P < 0.0001). However, 1 d post treatment with the lowest dose of 5 mg/kg, high percentage mortality was recorded with hexane extract compared to the low mortality of pea beetles obtained with methanol and acetone, extracts. At the highest concentration (20 g/kg), hexane and methanol extracts exhibited 100% mortality of *C. maculatus* adults after 3 days post-treatment while, only 50% mortality of pea beetle was noticed with acetone extract at the same period with the dose applied. After 6 days post-exposition, 100% mortality of *C. maculatus* was also observed with hexane and methanol extracts tested at 0.5 g/kg.

Table 2. Phytochemical screening of hexane, acetone and methanol extracts of *Cassia mimosoides*

Phytochemical components	Hexane	Acetone	Methanol
Total phenolic compound	-	++	++
Flavonoids	-	++	++
Tannins	-	+	+
Coumarins	-	-	-
Alkaloids	+	+	+
Triterpenoids	+++	+	+
Steroids	+++	+	-
Saponins	-	+	+

+++ = highly present; ++ = moderately present; + = present; - = absent

Table 3. Corrected mortality percentage and LC_{50} of *Callosobruchus maculatus* exposed to *Cassia mimosoides* extracts on *Vigna subterranea* in soudano- guinean area (Ngaoundéré: Cameroon)

	Hexane extract			
Conc	**DAI**			
	1 days	**3 days**	**6 days**	**F**
0	0.00±0.00eA	0.00±0.00cA	0.00±0.00bA	1.00ns
0.25	73.75±1.25dC	83.36±2.43bB	92.94±1.38aA	29.54***
0.5	81.25±1.25cB	86.36±1.13bB	100.00±0.00aA	111.15***
0.75	80.00±0.00bB	89.87±3.54bB	100.00±0.00aA	8.10**
1	96.25±2.39aA	100.00±0.00aA	100.00±0.00aA	2.45*
F	863.21***	415.16***	562.51***	
LC_{50}	0.11 (0.03-0.1	0.06 -	0.020 -	
Acetone extract				
0	0.00±0.00d	0.00±0.00e	0.00±0.00d	
0.25	2.50±1.44dcB	12.76±2.42dA	17.68±0.43cA	22.23***
0.5	10.00±2.04cB	26.91±1.08cA	33.62±3.84bA	22.14***
0.75	21.25±3.15bC	38.4±1.5bB	79.46±3.67aA	103.5***
1	35.00±2.04aC	50.00±1.07aB	91.34±3.76aA	131.3***
F	51.54***	185.55***	183.20***	
LC_{50}	1.41 (1.1-2.1)	1.03 (0.85-1.4)	0.50 -	
Methanol extract				
0	0.00±0.00eA	0.00±0.00dA	0.00±0.00bA	1.0ns
0.25	18.75±1.25dC	47.30±4.29cB	87.4500±0.00aA	2.55***
0.5	30.00±0.00cC	52.57±1.02cB	100.00±0.00aA	3671.4***
0.75	45.00±2.89bC	80.72±1.45bB	100.00±0.00aA	224.11***
1	56.25±3.15aB	92.30±1.49aA	100.00±0.00aA	135.17***
F	122.57***	271.03***	5032.83***	
LC_{50}	0.87 (0.72-1.16)	0.32-	0.12	

*ns p>0,05; * p<0,05; ** p<0,001; *** p<0,0001. FL: Fiducial Limit. For the same product, mean ± standard error in the row followed by the same capital letter and in the column by the same small letter did not differ significantly according to Tukey test (p=0.05). LC_{50}: Lethal concentration that kill 50% of pea beetle. 50. Each datum represents the mean of 3 replicate values*

The values of LC_{50} varied with plant extracts and decreased with exposition period. From these values of LC_{50}, hexane extract was the most effective followed by methanol and acetone extracts. The LC_{50} values were 0.11, 0.06 and 0.02 g/kg for hexane extract; 1.41, 1.03 and 0.5 g/kg for acetone extract and 0.87, 0.32, and 0.12 g/kg for methanol extract, respectively after 1, 3 and 6 days post-treatment.

3.4 Effect of the Plant Products on the Progeny F1 Production

Table 4 presents the inhibition percentage of the progeny *F1* production of the pea beetle on the bambara groundnut tested with different *C. mimosoides* extracts and their concentrations. From the results, all plant extracts in comparison with the negative control inhibited significantly the progeny *F1* production of *C. maculatus* and this reduction increased with the increasing concentration (Table 4). With the hexane extract at all doses tested, a complete inhibition of Progeny *F1* production was noticed. That complete inhibition of progeny was observed with methanol extract tested at dose of 20 g/kg while at the same dose, acetone extract inhibited 76.09% the progeny production of *C. maculatus*.

3.5 Efficacy of the Plant Extracts on the Reduction of Damages and Grain Losses Caused by *C. maculatus*

Compared to negative control, different plant extracts reduced significantly the percentages of grain damaged and grain weight losses. Treated with hexane extract, pea groundnut was completely protected without damage of grains at all doses applied. With methanol extract, complete protection of pea groundnut grains from damage of *C. maculatus* was also recorded at the highest concentration (20 g/kg) applied. Treated with acetone extract, percentage of grains loses varied from 57.07% at the lowest dose (5 g/kg) to 22.49% at the highest concentration (20 g/kg).

3.6 Biochemical Characteristics of the Treated and Untreated Bambara Groundnut

The biochemical composition of *V. subterranea* variety before and after 3 months storage is presented in Table 6. From the results, no significant (P>0.05) difference between treated and untreated groundnut was observed concerning all biochemical parameters of groundnut assessed. Indeed, the ash content, proteins, sugars total and lipids values showed that the treatment of seeds with the extracts of *C. mimosoides* did not deteriorate the biochemical composition of seeds.

Besides, the increase of the quantity of the antinutritional factors in the treated bambara groundnut was noticed. Total phenolic compounds and tannins quantity increased respectively by 37.78% and 6.11% of dry material of methanol extract treatment (Table 2).

3.7 Organoleptic Properties of the Treated and Untreated Bambara Groundnut

The sensory evaluation of the bambara groundnut cooked and roasted form compared to the control showed that the various extracts of *Cassia mimosoides* modified the sensory characteristics of the derived products (Figs. 2 and 3). The test of Friedman (Table 7) in general confirmed a significant difference (F>9.49 at 5%) between the treated forms and the control product. Indeed, compared to the control, the extracts increased the bambara groundnut grains bitterness, the odor and the color of the two forms of preparation and also decreased the salinity and the crispness of the roasted form

Table 4. Effect of different plant extracts on the reduction of progeny F1 production

Conc	Hexane extract		Acetone extract		Methanol extract	
	Progeny	%Inhibition	Progeny	%Inhibition	Progeny	%Inhibition
0	117.50±6.56a	0.00±0.00b	168.75±0.75a	0.00±0.00d	141.00±6.56a	0.00±0.00d
0.25	0.00±0.00b	100.00±0.00a	153.75±10.38a	8.27±3.79dc	56.25±10.38b	74.04±3.44c
0.5	0.00±0.00b	100.00±0.00a	140.75±1.44ab	39.50±1.46bc	40.50±1.44bc	75.45±0.84bc
0.75	0.00±0.00b	100.00±0.00a	120.00±13.91bc	47.77±4.52ab	19.50±3.19c	96.37±1.07ab
1	0.00±0.00b	100.00±0.00a	102.75±6.01c	76.09±1.67a	0.00±0.00d	100.00±0.00a
F	87.53***	-	12.56***	17.11***	26.56***	93.30***

*** *p<0,0001. FL: Fiducial limit. For the same product, mean ± standard error in the column followed by the same letter and did not differ significantly according to Tukey test (p=0.05). Each datum represents the mean of 3 replicate values*

Table 5. Efficacy of *Cassia mimosoides* extracts on damages reduction caused by *Callosobruchus maculatus*

Conc	% Undamaged grains	% Damaged grains	% losses
		Hexane extract	
0	0.00±0.00b	100.00±0.00a	59.46±1.41a
0.25	100.00±0.00a	0.00±0.00b	0.00±0.00b
0.5	100.00±0.00a	0.00±0.00b	0.00±0.00b
0.75	100.00±0.00a	0.00±0.00b	0.00±0.00b
1	100.00±0.00a	0.00±0.00b	0.00±0.00b
F	-	-	2062***
		Acetone extract	
0	0.00±0.00a	100.00±0.00a	59.46±1.41a
0.25	0.00±0.00a	100.00±0.00a	57.07±1.39ab
0.5	0.00±0.00a	100.00±0.00a	52.08±0.84ab
0.75	0.00±0.00a	100.00±0.00a	36.64±4.52b
1	6.70±1.50a	93.29±3.41a	22.49±3.18c
F	1.00ns	1.00ns	34.85***
		Methanol extract	
0	0.00±0.00b	100.00±0.00a	59.46±1.41a
0.25	0.00±0.00b	100.00±0.00a	46.23±0.79ab
0.5	0.00±0.00b	100.00±0.00a	35.42±3.31bc
0.75	0.00±0.00b	100.00±0.00a	29.14±5.08c
1	100.00±0.00a	0.00±0.00b	0.00±0.00e
F	-	-	48.84***

*ns p>0,05 ; *** p<0,0001. For the same product, mean ± standard error in the column followed by the same letter did not differ significantly according to Tukey test (P=0.05). Each datum represents the mean of 3 replicates values. – F values were not determined because of 100% or no protection at all doses tested*

Table 6. Biochemical characterization of the treated and untreated grains of *Vigna subterranean*

Samples	Parameters					
	Ash (g/100 g)	**Proteins (g/100 g)**	**Lipids (g/100 g)**	**Carbohydrates (g/100 g)**	**TPC (mg/100 g)**	**Tannins (mg/100 g)**
Control	3.54±0.05a	21.53±0.33a	8.64±0.07a	61.24±0.5 0a	565.02	42.03
Hexane extract	3.74±0.040	20.86±0.33ab	7.68±0.69a	61.67±0.50a	-	-
Acetone extract	3.24±0.05a	21.13±0.33a	8.11±0.07a	60.97±0.50a	-	-
Methanol extract	3.84±0.050	21.17±0.9b	8.68±0.15a	61.36±0.36a	778.51	44.60
F	0.00ns	4.58ns	2.97ns	0.928ns		

ns p>0,05. For the same product, mean ± standard error followed by the same letter do not differ significantly according to Tukey test at P=0.05

and the tenderness for the cooked form. It was also noticed that the control and the treated sample products were similar regarding the sweet-salty taste of the roasted form. In general, the control sample product received a better acceptability by the taste volunteers compared to the treated products.

4. DISCUSSION

The hexane, acetone and methanol of *C. mimosoides* demonstrated significantly their efficacy against the post-harvest Bambara divastator *C. maculatus* in the laboratory.

Indeed in this present investigation, extraction yield varied from one solvent to another and acetone and methanol extracts respectively with extraction yield value of 10.1 and 8.1% were high compared to the low yield of 2.8% obtained with hexane solvent. These differences could be attributed to the large number and the high quantity of the phytocomponents in the acetone and methanol extracts compared to the

Table 7. Friedman values calculated from the different sensorial characteristics of cooked and fried *Vigna subterranea* grains treated with *Cassia mimosoides* extracts

Sensorial characteristics	Constances F		Sat 5%	Observations	
	Fried	Cooked		Fried	Cooked
Sweet-salty taste	8.66	153.66	9.49	similar	different
Salty taste	35.66	49.66	9.49	different	different
Bitter taste	82.66	221.66	9.49	different	different
Perfumed odor	129.66	91.66	9.49	different	different
Yellow color	29.66	190.66	9.49	different	different
crisp	13.66	89.66	9.49	different	different
preference	27.66	22.66	9.49	different	different

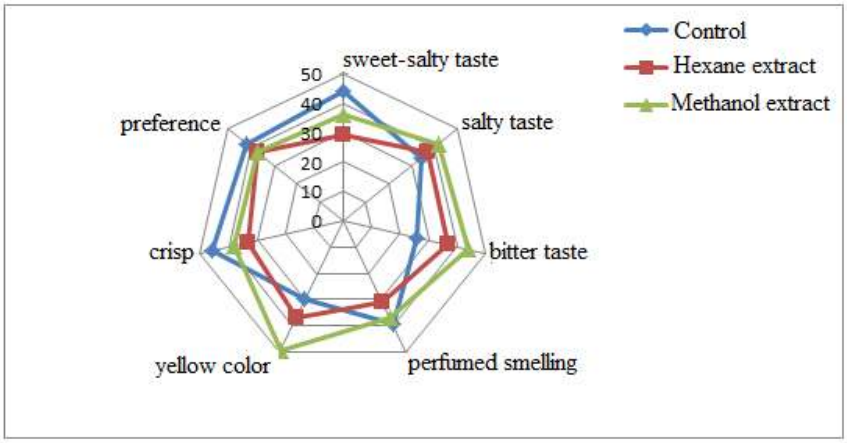

Fig. 2. Sensorial profile of cooked bambara groundnut treated with *Cassia mimosoides* extracts

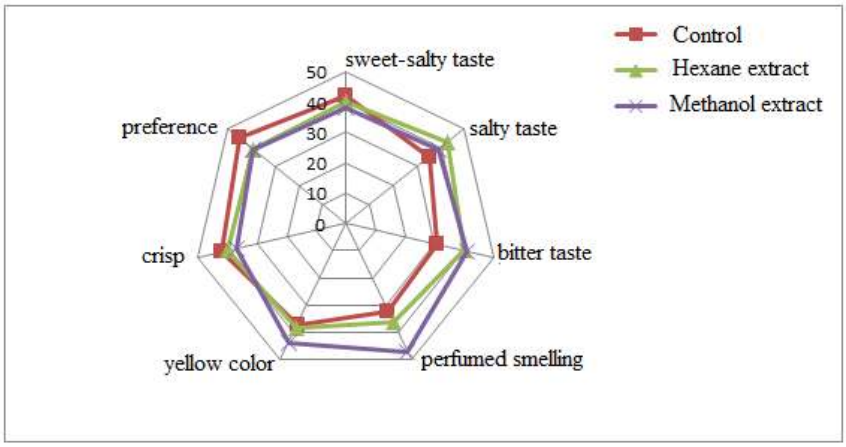

Fig. 3. Sensorial profile of roasted bambara groundnut treated with *Cassia mimosoides* extracts

non-polar solvent hexane extract. However, acetone and methanol extracts contained seven phytocompound groups (total compounds phenolic, flavonoids, tannins, of alkaloids, triterpenes, steroids and saponins) while hexane extract contained only three groups including alkaloids, sterols and triterpenes that could justify moreover their important yield compared to the hexane extract yield. The presence of these compounds in these extracts could be explained by the fact that plants synthesize secondary metabolites during their growth which are implied

in the many physiological processes like the cell multiplication, rhizome genesis, seed germination, fruit maturation and defense against the external aggressions [45,46]. The absence of some compounds in hexane extract could be explained by the fact that hexane is a non-polar solvent incapable to fix some polar compounds. In the same way, the presence of triterpenes and the steroids in acetone and methanol extracts is related to the fact that some polar compounds in their structures are associated to the non-polar compounds like triterpenes**.**

In this present study, all extracts of *C. mimosoides* exhibited a significant adult mortality of *C. maculatus* and varied with the different plant extracts and the concentrations applied as well as the exposition. The mortality recorded with various plant extracts would be due to the presence of the secondary metabolites in these extracts. According to Rubabura et al. [47], the plant secondary metabolites are implied in the various processes including their insecticidal properties. The composition and the nature of the compounds present in each extract depend not only on its solubility in the vegetable powder, but also of the solvent polarity used [48]. The higher mortality noted with hexane extract compared to methanol and acetone extracts could be explained by the chemical composition and the level of sensitivity of the insects towards these various extracts [49]. The alkaloids, saponins, steroids, phenolic compounds and tannins present in the acetone and methanol extracts acted much more like antinutrients hindering thus assimilation of the nutriments [50], whereas the terpenoid compounds acted on the insect nervous system by disorganizing ion exchanges sodium and potassium leading to the insect death in contact [51,52]. The phytochemical screening revealed that hexane extract of the leaves of *C. mimosoides* contained more of the terpenic compounds compared to the two other extracts explaining the strong mortality noted in that extract. Moreover, this difference in mortality could be attributed to the food behavior of *C. maculatus.* Indeed, adults of *C. maculatus* do not feed and the mortality of that insect is attributed to the substances that induced its death by direct contact. The increase in mortality with the dose and the exposure period could be explained by the increase in the quantity of the active ingredients responsible for the insecticidal activity. Our results are comparable to those obtained by Daniel et al*.* [53] who reported a strong mortality of *C. maculatus* with the increasing dose and exposure period with hexane extract of *Gnidia kaussiana* compared to the acetone and methanol extracts. Similarly, Ojimelukwe [54], Tapondjou et al. [55], Obeng-Ofori [56] reported also a strong mortality of *Tribolium confusum, T. castaneum, Sitophilus zeamais, Prostephanus truncatus, Rhyzoperta dominica and Callosobruchus* exposed to the terpenic compounds of essential oils. In contrary, Adeniyi et al*.* [57] obtained a strong mortality of *Acanthscelides obtectus* exposed to ethanol extracts. These results in insect mortalities could be related to the direct food intake mixed with plant products.

In general, all plant extracts inhibited significantly the progeny *F1* production of *C. maculatus* and this reduction increased with the increasing concentration. This reduction would be due not only to the antinutritional substances like tannins, phytates, phenolic compounds contained in the extracts and able to give complex to the availability of the nutriments to the immature stages of *C. maculatus* and also terpenes act directly on the adults in contact by their toxic effect. Rajasekaran and Kumaraswami [58] reported that the seeds treated with the plant extracts reduced significant the offspring of *Sitophilus oryzae*. Similar results were also reported by Thiaw et al. [59] which obtained a significant reduction of the offspring of *Caryedon serratus* on *Vigna subterranea* treated with *Cassia occidentalis* and *Calotropis procera* powders. Daniel et al. [53] obtained also a significant reduction of the offspring of *C. maculatus* on seeds treated with *Ocimum canum* and *Gnidia kaussiana* extracts*.* In contrary Agnes et al. [20] reported that *Callistemon rigidus* acetone extract did not reduce the progeny production of *Acanthoscelides obtecus*. This difference could be explained by the variation in composition and the secondary metabolites content contained of the extract.

Overall, different plant extracts reduced significantly the percentages of grain damaged and grain weight losses. The reduction of seeds damaged and the weight losses could be explained by the presence of the metabolites responsible for the insecticides activity. Similar results were obtained by Nukenine et al. [21] which obtained a significant reduction of the rates of damaged seeds and weight losses of the corn seeds treated with the powders of *Neem Azal, Azadirachta indica* and *Plectranthus glandulosus* against *Sitophilus zeamais*.

Indeed, the ash, proteins, sugars total and lipids contents did not significantly varied showing that the treatment of seeds with the extracts of *C. mimosoides* did not deteriorate the biochemical composition of seeds. The ash contents are similar to those reported by Mazahid et al. [60] who obtained 3.25% with bambara groundnut seeds of bambara groundnut cultivated in Sudan. Similarly, Amartiefio et al. [61] and Abiodum et al. [62] obtained a range from 3.57 g/100 G and 4.85 g/100 g of dry material of seeds collected from Namibia, Swaziland and Nigeria. But these ash contents are different to the low values ranging from 2.55 to 2.98 obtained by Diallo Koffi et al. [63]. According to Amarteifio et al. [64], this difference in the ash contents could be attributed the texture and the composition of the soil which could have an effect on mineral absorption of the plants and the varietal differences. The lipid content obtained in this present study are similar to the values obtained by Amarteifio and Moholo [65], and Boateng et al. [66]. These authors showed that the leguminous plant seeds, except for soya and groundnut, contain in a general way little of lipids. However, the contents obtained are higher than those reported by Abiodun and Adepeju [67]. The total rate of sugars is in agreement with that reported by, Yusuf et al. [68], Boateng et al. [66] and Mazahib et al. [60]. The contents reported by these authors ranged from 54.51 to 65% of dry material. However, our results showed low sugar contents than the average contents of 73.50 % reported by Aremu et al. [69]. These differences observed in the results could be due to the varietal properties and the environmental conditions of their culture [70,66]. The result of the protein content obtained is appreciably equal to the values reported by Amartiefio et al. [71], and Amartiefio et al., [61]. These authors obtained respectively the values ranged from 15.1 to 22.1 and from 17.5 to 21.2 grams of proteins for 100 grams of dry material. This difference might be due to the genotypes and the environmental conditions where these seeds were cultivated [72,70].

Besides, in this present study, the increase of the quantity of the antinutritional factors in the treated bambara groundnut was noticed. This increase could be due to the presence in the extracts of these secondary metabolites. However, the quantity of these antinutritional factors are low compared to the thresholds values of 2700 mg/100g for phenolic compounds and 2000 mg/kg for tannins [73] and could not deteriorate the biodisponibility of the nutriments in the grains. In the same way, Siqueira et al. [74] and Collinaw [75] reported that certain treatments like soaking or cooking could reduce significantly the degree of antinutritional factors. A long cooking process set at 100℃ is also able to eliminate these antinutritional factors [76].

The sensory evaluation of the bambara groundnut cooked and fried treated with *Cassia mimosoides* extracts modified its sensorial characteristics. The increase in the taste, bitterness and low preference of the treated sample products could be explained by the fact that plant extracts contain aromatic compounds which aromatize the derived products compared to the control sample product. Similarly, the presence the bitter plant metabolites compounds like tannins and saponins increase the bitterness of the treated compared to the untreated bambara groundnut. The result of this present investigation is comparable to those obtained by Rose de Lina et al. [25] who reported that the use of essential oils significantly influences the odor and the taste of the cowpea samples stored.

5. CONCLUSION

The extracts of *C. mimosoides* have proven their insecticidal properties against *C. maculatus* during storage. These extracts moreover did not modify the nutritional quality of the derived products and improve the sensory characteristics, in particular the flavour of the treated products. They could thus constitute an effective alternative in the preservation of the bambara groundnut because of their safety for consumers and might then replace synthetic insecticides used for the issue.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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