

# Insecticidal and Antifungal Properties of Essential Oil of *Bidens Pilosa* Linn. Var. *Radita* (Asteraceae) Towards Stored Bambara Groundnut Insect and Fungi Pests

Augustin Goudoum<sup>1\*</sup>, Léonard Simon Ngamo Tinkeu<sup>2</sup>, Martin Benoît Ngassoum<sup>3</sup> & Carl Moses Mbofung<sup>4</sup>

<sup>1</sup>: The University of Maroua, The Higher Institute of Sahel, Department of Agriculture, Livestock and Derived Products  
P.O. Box 46 Maroua – Cameroon

<sup>2</sup>: The University of Ngaoundere, Faculty of Sciences, Department of Biological Sciences  
P.O. Box 454 Ngaoundere- Cameroon

<sup>3</sup>: The University of Ngaoundere, National High School of Agro Industrial Sciences, Department of Applied Chemistry  
P. O. Box 455 Ngaoundere- Cameroon

<sup>4</sup>: The University of Bamenda, School of Technology, Department of Food and Bioresource Technology  
P.O. Box 39 Bambili – Cameroon

\* Corresponding author E-mail: [goudoumaugust \[AT\] gmail.com](mailto:goudoumaugust[at]gmail.com)

---

**ABSTRACT**---- *The present research studies in on hand the degradation of seeds due to pests: Callosobruchus maculatus and some fungi during 5 months and in other hand the efficiency of essential oil (EO) of fresh leaves of Bidens pilosa (Asteraceae) to prevent these pest attacks on Bambara groundnut (BG) in the Far-North Region of Cameroon. The major constituents of the leaves EO were methyl  $\alpha$ -pinene (14.71%), E-caryophyllene (13.47%),  $\beta$ -ocimene (12.83%) and cadinene (10.05%). Towards insect pest, the effective concentrations of B. pilosa that kill 50%, 80% and 99% population of C. maculatus were respectively 2.48, 3.46 and 4.07 mg/ml. At the LC<sub>50</sub>, EO protects 62.33 g of 100 g of BG after five month storage. The LC<sub>80</sub> and LC<sub>99</sub> of B. pilosa, reduced respectively 85 and 98% of damages of C. maculatus. Concerning fungi, 6 species belonging to 4 gender are isolated on untreated BG seeds after 5 months of storage: Aspergillus, Penicillium, Fusarium and Mucor. EO of fresh leaves of B. pilosa effectively reduced the fungal development. The sample treated with LC<sub>80</sub> and LC<sub>99</sub> have 4 species namely P. roquefortii (13.33 to 16.33%), A. Niger (20 to 24%), A. fumigatus (10 to 13.33%) and Mucor spp. (3.33 to 6%). This composition made clear the insecticidal and antifungal activities of the EO of B. pilosa and indicated its potential to protect BG from pest attacks..*

**Keywords** ---- Bambara groundnut, *Bidens pilosa*, *Callosobruchus maculatus*, essential oil

---

## 1. INTRODUCTION

Bambara groundnut (*Voandzeia Subterranean* (L.) Thouars) is an indigenous African crop grown all around the continent from Senegal to Kenya and from the Sahara to South Africa [1]. Since food shortage still a major problem in Africa, these legumes are promoted more than before in order to reduce malnutrition [2]. The affordability of plant protein source relative to that of animal origin has led to the intensified development of legume processing as a means of enhancing the availability, palatability and diversity of leguminous source of dietary protein [3].

Despite the importance of this crop for smallholders and their households because the Bambara groundnut (BG) are an important source of food security, being nutritious and high in protein in Africa and precisely in Northern Cameroon, the availability of this seeds for consumption is limited by *Callosobruchus maculatus* (F.) attack. Such attacks occur not only at the ripening stage in the field but also in the storage [4]. The bruchids are the main pest of BG during storage, capable of destroying an entire stock before the expected storage period. Successful storage challenges are also reflected by the irregularity of the presence of the speculation on the market throughout the year on the one hand and on the other by the several tools which producers are used to limit losses.

Beside insects attack, fungal contamination is one of the main causes of deterioration of leguminous. From this contamination; parameters controlling fungal growth and toxin production would abound [5]. Mainly the initial load is quoted in microflora, the presence of broken grains, humidity and temperature of grain storage [6, 5]. It has been reported that during the storage, and even without contamination, the increase of the temperature and / or humidity, can cause quality losses, affecting the hardness and virtuousness of grain and its acidity and the quality of its proteins [6]. This results in variations of technological parameters of the grains and can cause considerable losses. The need for quality controls of BG before, during and after storage is required.

Presently, conventional synthetic insecticides are used to reduce losses due to these insects and fungal contamination. Some pesticides registered to protect stored grains are hazardous to consumers and may pollute the environment [7]. Food protected in this way present a real risk to the consumer, it is therefore urgent to react to the food security problem

by providing ingredients for the development of alternative tools to these dangerous pesticides and achieve sustainable production BG that actually contributes to the promotion of consumer health and recovery of malnutrition [8].

In this respect, essential oils of some aromatic plants of the Northern Cameroon are described to have effectiveness in the control of stored grain insect pests [9, 10, 11]. *Bidens pilosa* (Linn. Var. Radita) is one of the cited insecticidal to protect BG to *C. maculatus* attack in the Far-North of Cameroon. *B. pilosa* is an annual, erect and ruderal herb originating from South America and now found in almost all tropical and subtropical region countries [12, 13]. Nevertheless, this plant is also commonly used in the traditional medicine. The study by Deba et al. [14] was the first report on the essential oils composition, antioxidant, antibacterial and antifungal activities of *B. pilosa* from Japan. More specifically this research aims to evaluate potential of *B. pilosa* as natural insecticide and fungicide to protect BG during storage in Far-North Region of Cameroon.

## 2. MATERIAL AND METHODS

### 2.1. Insects rearing

A strain of *C. maculatus* adults used for study was obtained from naturally infested stocks of BG. The culture was maintained and reared in the laboratory (31.48±2.88 °C and 58.56±6.78 % relative humidity) on the dry BG in the breeding bottle covered with piece of muslin cloth. The muslin cloths were held in place with a rubber band to allow good aeration and to prevent the escape of insects. 25 couples of insects were introduced in one-liter jars containing 1kg of untreated BG brought at the Maroua-Kodek market. This phase lasted for 30 days to allow the emergence of new adults. After this operation, the BG contained in the different jars were screened; adult weevils were removed and breeding continued with BG seeds infested by eggs. This second phase lasted 30 days and 3 days emergence insects were used to infest BG of different treatments of study.

### 2.2. Source of the tested seed variety

The most susceptible variety of BG grain namely CM/EN/DW/03 [15] was obtained from Maroua-Kodek marked in the Far-North region of Cameroon. Before allow different treatment, BG grains were stored for 7 days at 4 °C, at the end to eliminate any form of insects. Three repetitions for each variety are made.

### 2.3. Plant collection and extraction of essential oils

Fresh leaves of *B. pilosa* were collected from Moutourwa in Far-Nord Cameroon in June 2015. The Department of Diamare is located in the Far North Region between 10° and 11° north latitude and 14° and 15° East.

Fresh leaves collected were dried at the shade, out of sunlight during 24 hours and cut in pieces. Once dried, 1 kg of leaves of *B. pilosa* was hydrodistilled in a Clevenger-type apparatus for 4 h as describe by [11]. The distilled oil was preserved in sealed sample tubes and stored within refrigerator till analysis.

### 2.4. Analysis of chemical composition of essential oils

The GC/FID (Chromatograph Agilent HP-6820) was carried out with HP-5MS column (5% phenyl methyl siloxane) with 30 m length and 250 µm in diameter and 1µm of thickness. The carrier gas was hydrogen, the oven temperature was programmed from 40 to 230 °C with a rate of 5 °C. min<sup>-1</sup> with a stay at 230 °C during 5 min. The pressure of the carrier gas was 49.9 KPa and the flux at 74.1 mL.min<sup>-1</sup>. Quantification was carried out by percentage of peak area calculation. The identification of single compounds was performed by comparison of the retention indices with reference data [16, 17].

### 2.5. Determination of efficient concentrations

A Balance was used to remove 10, 20, 30 and 50 mg of essential oil of *B. pilosa* and diluted it in 10 ml acetone to formulate insecticides. For each preparation, 0.5 ml was pumped and flowed regularly on a disk of filter paper (Wathmann n°1) placed in a Petri dish. After this application 20 insects were introduced to the dish 4 min later and it was closed. Mortality of insect was noted 24 h after the treatment. For each trial, 3 replications were made. From this experiment, the concentration killing 50, 80 and 99 % of the experimental population were estimated.

### 2.6. Evaluation BG seeds weight losses due to *Callosobruchus maculatus*

At the end of the conservation time (1, 2, 3, 4, 5 months), physical damages were assessed for the evaluation of seeds weight loss. Seeds weight losses were evaluated on 100 g of BG seeds without damage. The BG treated with different concentration (LC<sub>50</sub>, LC<sub>80</sub> and LC<sub>99</sub>) of essential oil of fresh leaves of *B. pilosa* were filled in glass flask (1200 ml) with of 15 couples of *C. maculatus*. The breeding jar were sealed and kept in a dark place for 30, 60, 90, 120 and 150 days. At the end of any of this period, the remaining seeds weights were recorded for each flask using a Kern made balance (precision 1/1000 g). The non-infested and untreated grains are considered as a control sample. Four repetitions were carried out. The percentage of remaining BG seeds were expressed for each period (month).

## 2.7. Evaluation of the biodiversity of microorganisms associated with stored BG seeds

Untreated and treated BG seeds were used for the detection of microorganisms.

## 2.8. Culture of fungal strains

Batches of 100 g of clean and attack BG seeds, stored in a flask (500 ml) for 5 months were used for fungal assessment. After this storage period, for a bottle, 40 BG seeds were taken out at random and placed in a 100 ml beaker containing 30 ml of 10% hypochlorite. 10 minutes after these seeds were rinsed three times during 10 minutes and introduced into four Petri dishes containing Sabouraud gel at a rate of 10 seeds per box. The sealed Petri dishes were brought in a ventilated oven maintained at 30 °C for 5 days. After this period, the Petri dishes were removed and each isolated fungal strain was reared in a new Petri dish containing Sabouraud gel.

## 2.9. Fungal isolation and identification

Molds were isolated on agar Sabouraud medium, seeding was carried out on the surface with transverse grooves and the dishes incubated at 30 °C for 24 to 72 hours [18]. By microscopic observations, the spores were located and isolated. These individual spores were seeded on Sabouraud medium in order to obtain pure strains from which the molds identification is possible.

The different strains were identified from morphological characteristics (color of the colonies, conidia shape). All isolates were grown on Czapeck medium at 25 °C. Microscopic observations were performed in amount between a slide and coverslip mycelial fragment in distilled water or in lactophenol cotton blue 1%.

The immersion objective Ax100 and a micrometer were invariably used for morphological identification of different structures. [19].

## 2.10. Statistical analysis

The mortality data obtained were expressed as a percentage transformed to probit values and the effective concentrations were obtained using the Proinra.2 Software. The results obtained from the evaluation of reduction of BG seeds weight losses during storage were analyzed with analysis of variance (ANOVA) using the XLstat 2014 software (add-on for Microsoft Excel® of Microsoft Corporation, USA). The average values were classified using Duncan Multiple Test with the same software. For the microbiological analysis, the densities obtained for each treatment were analyzed with MANOVA using XLSTAT 2014 software.

# 3. RESULTS AND DISCUSSION

## 3.1. Essential oil composition

The yields of leaf oil obtained from the hydrodistillation procedures, calculated on a dry weight was 0.19% (v/w). GC analyses enabled the identification of twenty seven compounds, accounting for 97.57% of the total oil contents. Table 1 indicates the percentage composition and the identities of the 13 major components which represent 85.53% of total oil contents. Monoterpenes, sesquiterpenes and phenylpropanoids were the main classes of compounds present in both oils [20].

**Table 1:** Chemical composition of essential oil of fresh leaves of *Bidens pilosa*

N°	KI*	Compounds	%
1	978	$\alpha$ -pinene	14,71
2	1026	$\beta$ -ocimene	12,83
3	1028	limonene	2,31
4	1029	$p$ -cymene-8-ol	3,64
5	1050	$\alpha$ -terpinolene	2,45
6	1100	linalool	4,36
7	1336	Bourbonene	3,85
8	1348	$\Delta$ -elemene	2,45
9	1450	$\epsilon$ -caryophyllene	13,47
10	1560	Megastigmatrienone	7,12
11	1588	Caryophyllene oxide	3,54
12	1648	Cubebene	4,75
13	1658	Cadinene	10,05
<b>Total</b>			<b>85,53</b>

\* The compounds presented in this table are those having a proportion higher than 2 %.

The major constituents of the leaf oil were methyl  $\alpha$ -pinene (14.71%), E-caryophyllene (13.47),  $\beta$ -ocimene (12.83%) and cadinene (10.05%). Other significant compounds include Megastigmatrienone (7.12%), Cubebene (4.75%), linalool (4.36%), Bourbonene (3.85%), Caryophyllene oxide (3.54%), p-cymene-8-ol (3.64%), limonene (2.31%) and  $\alpha$ -terpinolene and  $\Delta$ -elemene (2.45%). Interestingly, the major constituents of the present results,  $\beta$ -caryophyllene, cadinene,  $\alpha$ -pinene, limonene,  $\beta$ -transocimene,  $\beta$ -cis-ocimene,  $\tau$ -muurolene,  $\beta$ -bourbonene,  $\beta$ -elemene,  $\beta$ -cubebene,  $\alpha$ -caryophyllene, caryophyllene oxide, and megastigmatrienone were previously reported to be of significant quantities in the essential oils in the leaves of *B. pilosa* [14, 21].

### 3.2. Toxicity of *Biden pilosa*

It appears from the table 1 that *B. pilosa* was toxic to BG weevils *C. maculatus* with a significant rate ( $p < 0.01$ ) between different concentration (Table 2). The real mortality ranged from 5 to 100% respectively from 1 mg/ml to 5 mg/ml. The effective concentrations which kill 50 % (LC<sub>50</sub>), 80 % (LC<sub>80</sub>) and 99 % (LC<sub>99</sub>) of experimental population of *C. maculatus* were respectively 2.48 g/ml, 3.46 g/ml and 4.07 g/ml.

**Table 2:** Effective concentration of essential oil of *Bidens pilosa* against *Callosobruchus maculatus*

	Concentration values (mg/ml)	Confidence interval (CI)
CL50	2.48	2.08<CI>2.88
CL80	3.46	3.11<CI>3.81
CL99	4.27	3.97<CI>4.57

The difference observed among the mortality due to these oils is due to their active volatiles mostly monoterpenes which are very active on insects [22, 10, 11, 23] reported that insecticidal efficiency of essential oils is related to their chemical composition. Some of their compounds such as 1,8-cineole,  $\alpha$  and  $\beta$ -pinene,  $\alpha$ -phellandrene, and limonene have insecticidal activities [22, 24, 25]; they act alone or have synergistic activities upon others [24, 23].

### 3.3. BG weight loss after treatment of *Bidens pilosa*

The results of the BG weight loss from Table 3 were due to physical damages caused by *C. maculatus* evolve with the storage delay. Untreated and infested BG progressively decreased his weight during storage ( $p < 0.001$ ). This weight reduction range from 88.35 g to 24.33 g respectively after one and five month storage. At the LC<sub>50</sub>, essential oil protects 62.33 g of 100 g of BG after five month storage. For the LC<sub>80</sub> the remaining weight of the initial 100 g BG range from 98.33 g to 85.66 g, respectively after one and five month storage. As regards LC<sub>99</sub>, any significant ( $p > 0.02$ ) difference were observed between storage periods. This last concentration protects 98.66 % of BG from insect attacks.

**Table 3:** Reduction BG weight loss treated with essential oil of *Bidens pilosa* due to *Callosobruchus maculatus* attacks during storage.

	1 month	2 months	3months	4 months	5 months
Untreated	88.35±3.85 <sup>a5**</sup>	81.66±5.52 <sup>a4</sup>	61.66±6.84 <sup>a3</sup>	31.66±6.82 <sup>a2</sup>	24.33±8.61 <sup>a1</sup>
CL50	98.45±4.54 <sup>b5</sup>	87.66±5.51 <sup>b4</sup>	81.33±5.16 <sup>b3</sup>	69.85±5.48 <sup>b2</sup>	62.33±6.47 <sup>b1</sup>
CL80	98.33±3.53 <sup>b4</sup>	94.66±3.08 <sup>c3</sup>	90.66±2.52 <sup>c2</sup>	88.32±4.08 <sup>c2</sup>	85.66±4.65 <sup>c1</sup>
CL99	99.51±3.50 <sup>b1</sup>	99.14±0.28 <sup>d1</sup>	98.78±0.78 <sup>d1</sup>	98.66±1.55 <sup>d1</sup>	98.66±2.57 <sup>d1</sup>

\*: Averages followed by the same letter in the same column are not different significantly with  $P < 0.05$  (Test of Duncan).

\*\*.: Averages followed by the same number in the same line are not different significantly with  $P < 0.05$  (Test of Duncan).

It is obvious and shown by Ngamo et al. [10] that treatment with essential oil was more effective and preserved stored grains. However, the weight loss is the parameter which varies more in these conditions. Goudoum et al. [26] shown that some essential oils reduce the taste of stored grains. This impacted the physiology of the pest by the recognition of the volatiles of oils at the level of its chemoreceptors [27].

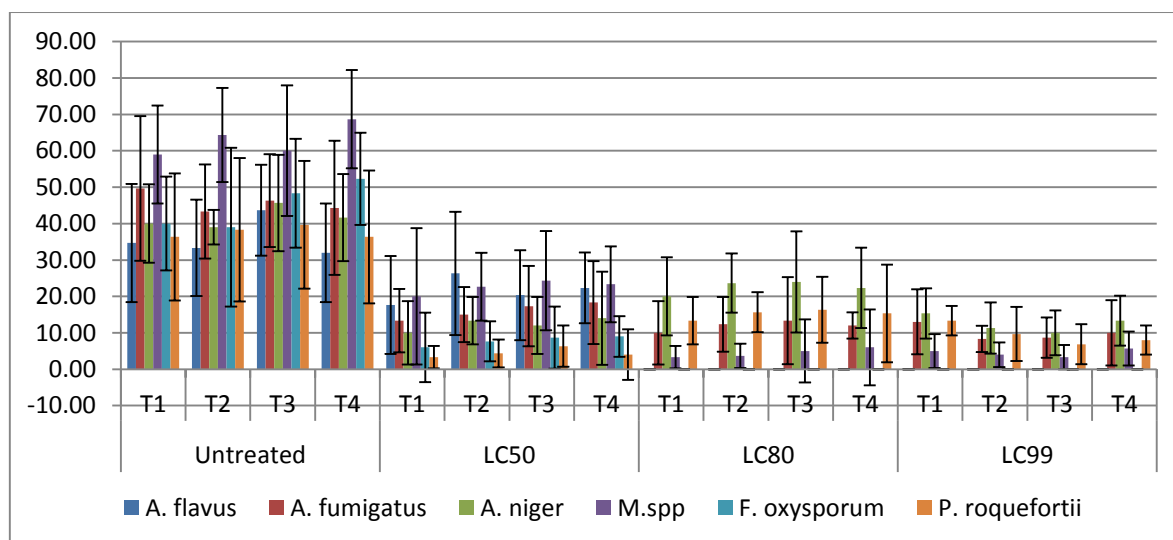
### 3.4. Diversity of fungal populations occurring on stored Bambara groundnuts

Fungal population present on the BG varies in relationship with attack rate. Figure 1 shows the diversity of fungal species isolated on different samples after 5 months of storage. Six species from four genera (*Aspergillus*, *Penicillium*, *Fusarium* and *Mucor*) were isolated on Bambara groundnut, but in different proportions. This is *A. flavus*, *A. fumigatus*, *A. Niger*, *P. roquefortii*, *F. oxysporum* and *Mucor* spp.

The densities of these fungal species as a function of the different batches (T1 to T4) untreated range from 32.00 to 46.67% for *A. flavus*, from 43.33 to 49.67% for *A. fumigatus*, from 39 to 45.67% for *A. Niger*, from 59 to 68.67% for *Mucor* spp., from 39 to 52.33% for *F. oxysporum* and from 36.33 to 39.67% for *P. roquefortii*.

The sample treated with LC<sub>50</sub> present the same species which range from 17.67 to 26, 33% for *A. flavus*, from 13.33 to 18.33% for *A. fumigatus*, from 10.00 to 14.00% for *A. Niger*, from 20.00 to 24.33% for *Mucor* spp., from 6.00 to 9.00% for *F. oxysporum* and from 3.33 to 6.33% for *P. roquefortii*.

The sample treated with LC<sub>80</sub> and LC<sub>99</sub> have 4 species namely *P. roquefortii* (13.33 to 16.33%), *A. Niger* (20 to 24%), *A. fumigatus* (10 to 13.33%) and *Mucor* spp. (3.33 to 6%). A significant difference ( $p < 0.05$ ) was observed between groups (T1-T4) of the same treatment. No interaction between the different concentration and molds was observed ( $p > 0.05$ ).



**Figure 1:** Fungal population present on the untreated and treated BG after 5 months storage

The results in Figure 1 show that *Aspergillus* is the most abundant, followed by *Fusarium* and *Penicillium*. This study provides the level of contamination of grain at the end of conservation in the northern part of Cameroon if no treatment was envisaged. Tabuc [28] Studies showed that the raw materials, including rice, may be contaminated by various fungal species, and that these fungal contamination and mycotoxins range depending on the climate. Indeed, while the *Fusarium* species develop mainly in the field on living plants, *Aspergillus* and *Penicillium* multiply during storage [29, 30]. Mold growth and subsequent mycotoxin production are related directly with the hydrothermal conditions [31, 32]. The northern region of Cameroon is located in the Sudano-Sahelian zone, characterized by a warm climate (temperature between 28 and 40 ° C in the shade, 60-70% relative humidity) [33], presents ideal conditions of these fungal species development. This climate influence fungal species which can grow on stored crops of the region.

From a mycotoxicologic point of view, three of the six isolated gender are potentially toxigenic [34]. Strains owned to these genders may synthesize different mycotoxins: aflatoxins may be produced by *Aspergillus* and *Penicillium*, ochratoxin A. produced mainly by *Penicillium* on stored grain [35, 36]. This study showed that fungal contamination of a particular lot is heavily influenced by the health of the harvested grain. Deba et al. [14] shown that essential oil of *B. pilosa* had inhibitory effects on the growth of fungi and fungitoxic activities at 100 ppm. The same authors demonstrated that  $\beta$ -caryophyllene and caryophyllene oxide detected in *B. pilosa* essential oil may play an important role in antifungal activities. Magiastis et al. [37] demonstrated that  $\alpha$ -pinene has the antifungal activities.

#### 4. REFERENCES

- [1] Atiku AA, Aviara NA, Haque MA. 2004. Performance evaluation of Bambara groundnut Sheller. Agricultural Engineering International: the CIGR, Journal of Science Research and Development 6(4), 1-18
- [2] Okafor PN, Nwabuko CU. 2003. Cyanide, nitrates and nitrite contents of livestock feeds in Umuahia, Nigeria. Biokemistri, 14: 57-63
- [3] Ijarotimi OS, Oyewo MT, Oladji BS. 2009. Chemical, functional and sensory properties of roasted Bambara groundnut (*Vigna subterranean* L. Verdc) and cooking banana (*Musa* spp., ABB genome) weaning diet, African Journal of Food Science, Vol. 3(5). pp. 139-146, May, 2009
- [4] Chukwujindu B, Chinedu FA, Chukwujindu SE. 2013. Evaluation of some accessions of Bambara groundnut (*Vigna subterranean* L. Verdc) for resistance to bruchid infestation, based on grain source and seed coat colour, Journal of Plant Protection Research, Vol. 53(3), 210 – 2014.

- [5] Tahani N, Elamrani A, Serghini-Caid H, Ouzouline M, Khalida. 2008. Isolement et Identification de souches de moisissures réputées toxigènes, Revue de Microbiologie Industrielle Sanitaire et Environnementale, Vol2, N°1, p : 81-91
- [6] Zia-Ur-Rahman. 2006. Storage effect on nutritional quality of commonly consumed cereals, Food Chemistry, 95, 53-57
- [7] Tamgno BR, Ngamo Tinkeu LS. 2013. Utilisation des produits dérivés du neem *Azadirachta indica* A. Juss comme alternatifs aux insecticides synthétiques pour la protection des semences de maïs et de sorgho dans la Vallée du Logone, Sciences, Technologies & Développement, ISSN 1029 – 2225, Volume 15, pp1-9.
- [8] Ngamo Tinkeu LS, Goudoum A, Djakissam W, Ayike Evele N, Madou C, Zilbinkaye M. 2015. Diversité des insectes ravageurs et des méthodes de protection post-récolte du voandzou *Vigna subterranea* (L.) employées par les cultivateurs dans le Nord du Cameroun. 21ème Réunion et Conférence de l'Association Africaine des Entomologistes, Cotonou, Bénin.
- [9] Taponjou LA, Adler C, Bouda H, Fontem DA. 2002. Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles, Journal of Stored Products Research, 38: 395 - 402.
- [10] Ngamo LST, Goudoum A, Ngassoum MB, Mapongmetsem PM, Hance T. 2007. Chronic toxicity of essential oils of 3 local aromatic plants towards *Sitophilus zeamais* Motsch. (Coleoptera : Curculionidae), *African Journal of Agricultural Research*, 2(4): 164-167.
- [11] Goudoum A, Ngamo TLS, Ngassoum MB, Mbofung CM. 2009. Antioxidant activities of essential oils of *Clausena anisata* (Rutaceae) and *Plectranthus glandulosus* (Labiatae) plants used against stored grains insects in North Cameroon, International Journal of Biological and Chemical Sciences, 3: 567-577.
- [12] Geissberger P, Séquin U. 1991. Constituents of *Bidens pilosa* L.: Do the components found so far explain the use of this plant in traditional medicine? *Acta Tropica*, 48, 251-261.
- [13] Grombone-Guaratini MT, Silva-Brandão KL, Solferini VN, Semir J, Trigo JR. 2005. Sesquiterpene and polyacetylene profile of *Bidens pilosa* complex (Asteraceae: Heliantheae) from southeast of Brazil. *Biochem. Syst. Ecol.*, 33, 479-486.
- [14] Deba F, Xuan TD, Yasuda M, Tawata S. 2008. Chemical composition and antioxidant, antibacterial and antifungal activities of the essential oils from *Bidens pilosa* Linn. var. *radiata*. *Food Control*, 19, 346-352.
- [15] Goudoum A, Ngamo Tinkeu LS, Madou C, Djakissam W, Mbofung CM. In press a. Variation of some chemical and functional properties of Bambara groundnut (*Voandzeia Subterranean* L. Thouars) during sort time storage, Food Sciences and Technology (Campinas).
- [16] Davies NW. 1990. Gas Chromatographic Retention Indices of Monoterpenes and Sesquiterpenes on Methyl Silicone and Carbowax 20M Phases. *Journal of Chromatography*, 503, 1-24.
- [17] Kouroussou R, Lanara T, Kokkini S. 1998. Piperitone oxide-rich essential oils from *Mentha longifolia* subsp. *petiolata* and *M. x villosa-nerveta* grown on the Island of Crete (S Greece), *Journal of Essential Oil Research*, 10: 375-379.
- [18] Harrigan WF, Mc Cance ME. 1976. Laboratory methods in food and dairy microbiology, Sciences of Food Agriculture London. *Academic Press*. 32: 1145-1150.
- [19] Pitt JI, Hoking AD. 1977. Fungi and Food Spoilage. *Blackie Academic & Professional*.
- [20] Goudoum A, Abdou Bouba A, Ngamo Tinkeu LS, Ngassoum MB, Mbofung CM. 2016. Antioxidant activities of essential oil of *Bidens pilosa* (Linn. Var. *Radita*) used for the preservation of food qualities in North Cameroon, *Food Science and Nutrition*, doi: 10.1002/fsn3.330.
- [21] Tomczykowa M, Leesczynska K, Tomczyk M, Trynieszewska E, Kalemba D. 2011. Composition of the essential oil of *Bidens tripartita* L. Roots and its antibacterial and antifungal activities, *Journal of Medicinal Food*, Vol 14 (4), 428-433.
- [22] Huang Y, Lam SL, Ho SH. 2000. Bioactivities of essential oil from *Elletaria cardamomum* (L.) Maton. to *Sitophilus zeamais* Motschulsky and *Tribolium castaneum* (Herbst), *Journal of Stored Products Research*, 36: 11-17.
- [23] Goudoum A, Ngamo TLS, Ngassoum MB, Mbofung CM. 2013. Persistence of active compounds of essential oils of *Clausena anisata* (Rutaceae) and *Plectranthus glandulosus* (Labiatae) used as insecticides on maize grains and flour, *African Journal of Food, Agriculture, Nutrition and Development*, 13 (1), 7325-7338.
- [24] Cimanga K, Kambu K, Tona L, Apers A, De Bruyne T, Hermans N, Totté J, Pieters L, Vlietinck AJ. 2002. Correlation between chemical composition and antibacterial activity of essential oils of some aromatic medicinal plants growing in the Democratic republic of Congo, *Journal of Ethnopharmacology*, 79: 213-220.
- [25] Kouninki H, Hance T, Noudjou FA., Lognay G, Malaisse F, Ngassoum MB, Mapongmetsem PM, Ngamo LST, Haubruge E. 2007. Toxicity of some terpenoids of essential oils of *Xylopiya aethiopica* from Cameroon against *Sitophilus zeamais* Motschulsky, *Journal of Applied Entomology*, 131(4): 269–274.
- [26] Goudoum A, Ngamo Tinkeu LS, Ngassoum MB, Mbofung CM. 2012. Variation in technological and nutritional parameters of maize treated with essential oils during storage, *African Journal of Food Science*, 6 (2): 34-40.

- [27] Ducrot PH. 2002. Contribution de la chimie à la compréhension de l'activité biopesticide de produits naturels d'origine végétale. In Les Biopesticides d'Origine Végétales. Edition TEC et DOC 11, Rue Lavoisier F. 75008 : Paris ; 53-65.
- [28] Tabuc C. 2007. *Flore fongique de différents substrats et conditions optimales de production des mycotoxines*. Thèse de Doctorat, Institut National Polytechnique de Toulouse et de l'Université de Bucarest, Spécialité : Pathologie, Mycologie, Génétique et Nutrition. 190 p.
- [29] Miller JD, Marasas WFO. 2002. Ecology of Mycotoxins in Maize and Groundnuts. Supplement to LEISA (Low External Input and Sustainable Agriculture) Magazine, 23–24.
- [30] Wilson CJ, Finch CE, Cohen HJ. 2002. Cytokines and Cognition—The Case for A Head to-Toe Inflammatory Paradigm, *Geriatric Bioscience*, Vol 50(12), 2041-2056.
- [31] Marin S, Magan N, Ramos AJ, Sanchis V. 2004. Fumonisin producing strains of *Fusarium*: a review of their ecophysiology, *Journal of Food Protection*, 67: 1792-1805.
- [32] Ramirez ML, Chulze S, Magan N. 2006. Temperature and water activity effects on growth and temporal deoxynivalenol production by two Argentinean strains of *Fusarium graminearum* on irradiated wheat grains, *International Journal of Food Microbiology*, 106: 291-296.
- [33] Laclavière G. 1979. *Atlas de la République Unie du Cameroun*. Atlas Jeune Afrique, 72p.
- [34] Weindenböner. 2000. Whole wheat and white wheat flour – the mycobiota and potential mycotoxins, *Food Microbiology*, 17, 103-107.
- [35] Jorgensen K, Rasmussen G, Thorup I. 1996. Orchoatoxin A in Danish cereals 1986-1992 and daily intake by the Danish population, *Food Additives and contaminants*, 13S 15-16
- [36] Hajjaji A, Bouya D, Bouseta A, Mathieu F, Collin S, Lebrihi A. 2005. Occurrence de mycotoxines (Ochratoxine A, Déoxinivalénol) et champignons toxigènes dans des céréales marocaines. Impact des facteurs écologiques sur la croissance et la production d'OTA. Symposium Euro-Maghrébin sur les contaminants biologiques, chimiques et la sécurité alimentaire, Fés, Maroc.
- [37] Magistis P, Melliou E, Skaltsounis AL, Chinou IB, Mitaku S. 1999. Chemical composition and antimicrobial activity of essential oils of *Pistacia lentiscus* var. chia, *Planta Medica*, 65, 749-752.