Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2020; 8(1): 1358-1364

Received: 16-11-2019
Accepted: 18-12-2019

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Bioefficacity of mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on the developing stages of *Callosobruchus maculatus* (Coleoptera: Bruchidae) pest control of *Vigna unguiculata* (Fabaceae) in storage in the Far-North region of Cameroon

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Abstract
The effect of mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on the developing stages of *Callosobruchus maculatus* was investigated. Quantities of 0.25, 0.50, 0.75 and 1.00 g of mixed plant powders were measured: PA= 1/2VS +1/2VL, PB= 1/2VS +1/2SA, PC=1/2VL + 1/2SA and PD=1/3VS+1/3VL+1/3SA (VS= Vepris stem bark, VL= Vepris leaves, SA= *Syzygium aromaticum* and P= Powder). They were applied to 100 g of cowpea seeds, followed by infestation with ten pairs of 48 hours old *Callosobruchus maculatus*. The highest mortality (100%) was obtained with 1.00 g of mixtures PD and PC while the least (82.5%) was recorded with 0.25 g of the mixture PA. The increase in the doses led to the decrease in the number of eggs laid with the highest mean percentage of oviposition deterrence been 98.44% at 1 g/100 g due to powder PB and the least 93.43% with 0.25 g due to powder PA. The mixtures reduced adult emergence of F_1 from 90.83% (powder PB) to 82.5% (powder PA). It also reduced the percentage of attack from 98% (control) to 0% for powder PB which was the best. The rate of germination increased with the quantities of powder applied.

Keywords: Cowpea, oviposition deterrence, storage, germination, *Vepris heterophylla*, *Syzygium aromaticum*

1. Introduction
In Africa, where most of the countries are food-deficient, agriculture occupies a very important status and mobilizes more than 80% of population. Many works have been done by the world community in order to ensure food security which is the basic right of the population [1]. But the difficulties encountered due to consecutively poor rainfall, rising food prices and pest control continue to worsen the situation, leading thus to food insecurity in many African countries [2, 3].

In Cameroon, food insecurity is prevalent in several regions, particularly in rural areas (9.6%). The most affected rural areas are the Far-North region (4.1%) followed by the North region (3.7%) [4]. However, the Far-North region is the agro-ecological zone of optimal development for cereals (millet, sorghum) and legumes (cowpea, soybean, and peanut). The cowpea seed *Vigna unguiculata* (L) is one of the main legumes of importance to farmers. It is a cash crop, whose production is 1.8 t for 1ha or 36 bags of 50 kg [5]. *V. unguiculata* is very rich in proteins (23-43%) and is likely to fill the many protein deficits recorded in the rural areas [6]. Cowpeas therefore have a high potential for contributing to the security and nutritional balance of populations in rural areas. To make it available throughout the year, storage is essential.

Although storage is the main way to make cowpea available, it is subjected to many difficulties such as moisture, shortage of well garner and mostly insect pests. The most redoubtable insect pest of cowpea seed is *Callosobruchus maculatus* (Coleoptera, Bruchidae) which depreciate qualitatively and quantitatively this commodity in few months of storage, hence the huge losses recorded (80 to 100%) [7]. The depreciation begins in the farm and increase in the storage areas. Facing this situation, many methods have been proposed: physical (high temperature, sand, ash), biological (parasitoids), mechanical (PICS bags,

metallic container), resistance variety (use of seed genetically modulated) and chemical (DDforce, poudrox, Force toxin, Unishield). Above all of this, farmers encounter many difficulties and those who have moderate resources are mostly resorting to chemicals synthetic insecticides. However, many problems are associated with these chemicals such as toxic residues in the food, peasant safety and mostly the insect resistance. To reduce these adverse effects, new alternatives are directed towards insecticidal plants [8, 9, 10]. Previous works have been done on the insecticidal plants and the final product obtained was on several forms such as extracts with organic solvent, essentials oils or powders of plants directly [11, 12, 13, 14]. Many of those works implementation was much focused on essential oil of plants such as *Vepris heterophylla*, Azadirachta indica, xylopia aethiopica against *Triboluim castaneum*, *Callosobruchus maculatus* and *Sitophilus zeamais* [15, 11, 16]. But, the damages caused by those insects especially *Callosobruchus maculatus* is still very important. This may be due to the low level of education of farmers or to the accessibility and the volatility of those extractions [16, 13]. In order to optimize the protection of seeds cowpea in the storage, many farmers introduced more than one powder of insecticidal plants directly on stored product in their various area [17, 18, 13, 14]. However, the master of the most efficient insecticidal plants and their combine action towards pest control of cowpea in various region in order to reduce environmental destruction also is still lacking [19, 20, 14]. Thereby, two aromatic plants *Vepris heterophylla* and *Syzygium aromaticum* already used against stored products by farmers were chosen. The aim of the present work was to evaluate the effectiveness of the combined powders of *Vepris heterophylla* and *Syzygium aromaticum* on the developing stages of *Callosobruchus maculatus* in the sudano-sahelian zone of Cameroon.

2. Materials and Methods

2.1 Collection and preparation of plants powders

The study was carried out in the Far-North region of Cameroon. Fresh matured leaves and stem bark of *Vepris heterophylla* were collected from Mokolo (10°44'54"N, 13°47'53"E and 901 m above sea level) in the Mayo Tsanaga Division. The seed of S*yzygium aromaticum* were collected in the main market of Maroua (10°36'23"N, 14°19'53"E and 400 m above sea level) in the Diamare Division.

The collected test plants leave and stem bark were separately air-dried in the laboratory of the Institute of Agricultural Research for Development (IRAD) of Maroua, Cowpea section, at the temperature of 30.9 ± 2.13°C and relative humidity of 35 ± 5.02% for 10 days (leaves) and 20 days (stem bark). The dried plant materials and the flower bud of *Syzygium aromaticum* (clove) were separately milled in an electric miller. The obtained powders were separately sieved through a 0.5 mm mesh net to have fine homogenous powders. Each plant powder was immediately put in separate biodegradable plastic papers, sealed and kept in the refrigerator (4 °C) up to the period of all assessments [15].

2.2 Collection and disinfection of Cowpea seeds

The brown eyed variety of cowpea seeds were purchased from the peasant in the local market of Mouvoudaye, 70 km away from Maroua in the Mayo Danay Division which is the area of predilection. The cowpea was taken to the laboratory where dirty and broken seeds were removed. The un-infested seeds were sterilized by storing in deep freezer at temperature of -5 °C for 48 hours to ensure that all developed stages have been killed. The sterilized seeds were removed, dried on the sun for 2 hours and then acclimated to room temperature during one week before use for all the experiments [29].

2.3 Collection and rearing of *Callosobruchus maculatus*

Brown eyed variety of cowpea seeds infested already by *C. maculatus* were collected from IRAD Maroua and brought to the laboratory. The infested seeds were sieved, *C. maculatus* adults were collected and introduced in three five liters' plastics containers, each already containing 5 kg of sterilized cowpea seeds and covered with mesh net (0.5mm) till the emergence of adults (30.9 ± 2.13 °C and 35 ± 5.02%). Freshly healthy emerged adults were used for further experiments after 48 hours latest.

2.4 Biological assays

Table 1 below shows the stable mixtures of the three powders: PA= 1/2VS +1/2VL, PB=1/2VS +1/2SA, PC=1/2VL + 1/2SA, PD=1/3 VS +1/3VL+1/3SA (VS= Vepris stem bark, VL= Vepris leaves, SA= *Syzygium aromaticum*). From each mixture, 0.25, 0.5, 0.75 and 1 g were measured using a Lutron GM-300p brand electronic scale (300.00x 0.01g). The experiment was shared into six stages, each stage having sixteen glass jars.

Table 1: Equilibrium mixtures of the three powders

Plant powder combination	Quantity used
PA	1/2 VS +1/2VL
PB	1/2 VS +1/2SA
PC	1/2VL + 1/2SA
PD	1/3 VS +1/3VL+1/3SA

(VS= *Vepris heterophylla* stem bark, VL= *Vepris heterophylla* leaves, SA= *Syzygium aromaticum*, P= Powder)

2.4.1 Contact toxicity of the different mixed plant powders towards adults of *Callosobruchus maculatus*

One hundred (100) grams of sterile cowpea were weighed using the same scale and introduced into 500 mL labeled jars. 0.25, 0.5, 0.75 and 1 g of each mixed powders plant were measured and introduced in the jars containing cowpea seeds. Each jar was shake manually in order to ensure uniform coating. After, ten couples of newly emerged cowpea weevils of two days old were introduced into each jar. All the jars were covered with 0.5 mm muslin cloth to enhance aeration and exclude escape of weevils. The control jar was free from any treatment. Each treatment was replicated four times for each powder and each quantity applied.

24 to 72 hours after treatment the dead weevils were recorded from each jar and the bio-insecticidal effect of the different powder against *C. maculatus* was calculated using the formula quoted by Nta and Agbo, Kouninki *et al.* [21, 10]:

%of mortality= (Number of dead weevils/total number of weevils) x 100

2.4.2 Impact of the different mixed plant powders on the oviposition deterrence of *Callosobruchus maculatus*

To investigate whether the different mixed plant powders possesses effect on the oviposition of *C. maculatus* female, another experiment was conducted. Seven days after treatment, ten seeds of cowpea from each jar were randomly taken and the numbers of eggs laid were recorded in each jar. The percentage of oviposition deterrence (POD) was calculated according to Kayombo *et al.* [22]:

POD= [/Ts-Cs/Cs] x100
Ts=Number of eggs laid on treated
Cs=Number of eggs laid on control

2.4.3 Impact of the different mixed plant powders on the emergence of *Callosobruchus maculatus*

Cowpea seeds treated were kept in the laboratory until 24 days in order to assess the first generation (F_1) of *C. maculatus*. From 20th to the 22nd days of treatment, emergence of the new generation was done. The number of F_1 adults from each treatment was noted and the reduction percentage in F_1 adult (RPA) emergence was evaluated as follow [22]:
RPA= [/Ac-At/Ac] x 100
Ac= number of F_1 on the control
At=number of F_1 in the treated

2.4.5 Evaluation of the rate attack on the cowpea seeds after treatment

Three months after treatment, seeds of cowpea having hole of emergence was removed from the 100 seeds in the entire jar and counted. The damage caused by *C. maculatus* on the treated seeds of cowpea was estimated. The percentage of attack (% A) was calculated as follow [23].
%A= (NSH/TNS) x 100
NSH=Number of Seeds Having more than two holes of emergence,
TNS=Total Number of Seeds.

2.4.6 Evaluation of the germinative power of the seed of cowpea before and after treatment

In order to evaluate the influences of the different powders on the rate of germination, two germination tests were performed: the first was carried out with the untreated seed of cowpea and the second three months after being in permanent contact with the different powders in the various quantitie. In a petri dish of 9 cm of diameter, 30 seeds were randomly removed in the different treated jar and introduced. 20 g of pure cotton were measured and introduced in the petri dish aiming to cover all the seeds. 10 ml of water were used every morning to wet the cotton for three days. Four repetitions were done and the seeds in the control jar were free from treatment. On the fourth day, the rootlet was observed and the percentage of germination (% G) was evaluated as follow [24]:

%G= (NGS/TNSS) x 100
NGS=Number of Germinated Seeds of cowpea
TNSS=Total Number of Seed of cowpea Sown

2.5 Data analysis

Abbott's formula was used to correct natural mortality when it was more than 3% in the control jar [43]. The different results obtained were subjected to the analysis of variance (ANOVA) in order to determine the most efficient combination of the plant powders. In the case where there was a significant difference, the complementary test of Newman and Keuls at the level 5% were performed with XLSTAT 2007 version 8.04 in order to determine homogeneous groups.

3. Results and discussion

3.1 Evaluation of the toxicity of the different mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on *Callosobruchus maculatus* adults

The result showed in Table 2 indicates that the mean mortality count of adult *C. maculatus* is higher in cowpea seeds treated with powder PD =1/3VS +1/3VL+1/3SA (100%) and PC = 1/2VL +1/2SA (100%) followed by mixture PB =1/2VS + 1/2SA (90%) while the treatment with PA = 1/2VS +1/2VL (75%) recorded the lowest mortality. The adult mortality increased with the different quantities of plant powder combination used. In the control jar, 1% of mortality was recorded. However, there was significant difference ($P<0.05$) in *C. maculatus* adult mortality 72h after treatment with all the different mixed powders compared to the control.
The mixed powders showed contact toxicity towards *C. maculatus* adult and 100% mortality was achieved with the mixture PD and PC during the exposure period. This result was in agreement with the research of Kouninki *et al*. [25] and Tofel [26] on the essential oils of *Xylopia aethiopica* and some plants powders respectively. Their reports confirmed that aromatic plants were toxic to weevils attacking on storage production and the combination of aromatic plant in different proportions increased the insecticidal potency of the plant thus resulting in their higher mortality. Also, Nta and Agbo [21] showed that the equal mixture of *V. amygdalina* and *O. gratissimum* led to the higher percentage of mortality (86%) during 96 hours of duration. That higher mortality could also be a result of the fumigation effect of the odor of the plant powder which was capable of blocking the spiracles of insects [27]. In addition, the higher mortality obtained could not be attributed only to the action of the major components present in the different powders, but also to the action of the minor compounds. This is in line with the works of Burt, Emeazol *et al*., Gouceem and Kouninki *et al*. [28, 17, 29, 10]. They found that when powders of insecticidal plants are combined no matter their low level in essential oil, they give a heterogeneous group of complex mixtures of organic substances such as polyphenols which acts together and results in a synergistic effect. This finding corroborates with the results of the present study where almost all the different mixtures had insecticidal effect against the mortality of *C. maculatus* comparing to the control jar.

Table 2: Effect of the different mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on *Callosobruchus maculatus* adults

Plant Powder combination	Percentage of mortality of *Callosobruchus maculatus* (%)				
	Quantities used (g)/100 g				
	0	**0.25**	**0.5**	**0.75**	**1**
PA	1	32.5±0.96de	45±0.58c	65±0.58b	75±1.3b
PB	1	35±1.29e	55±0.6cd	55±0.6cd	90±0.96b
PC	1	40±0.82bc	55±0.57ba	65±1.3b	100±0.0a
PD	1	45±1cde	53±0.5cd	60±0.8bc	100±0a

In the same column, means followed by the same letter are not significantly different to the Newman and Keuls test at 5% level.

3.2 Evaluation of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on eggs laid by *Callosobruchus maculatus*.

Table 3 presents the results of the impact of *Vepris heterophylla* and *Syzygium aromaticum* combination on the oviposition deterrence of the female of *C. maculatus*. These results showed that all the four mixtures have reduced the oviposition of *C. maculatus* female comparing to the control jar (0%). The different percentages of oviposition deterrence increased with the different quantities and the maximum was obtained with 1 g/100 g.

The highest number of eggs laid was recorded in the control jar with a mean average of 104 eggs. It was significantly different (P<0.05) from the number of eggs laid in all other treated seeds of cowpea. From 0.25 to 1 g/100 g of the mixed powder used, the activity of the different powder which was to inhibit the females to lay eggs was very fast when looking at the percentage of oviposition deterrence at the dose 0.25 g. All the combinations gave a result varying between 97% and 98% at the dose 1 g/100 g but the highest rate was obtained with the combination PB followed by PD with a mean percentage of oviposition deterrence equaled to 98.44 ± 0.5 and 98.20 ± 0.56% respectively. Significant differences were recorded between the oviposition deterrence observed with the various combinations (PA, PB, PC and PD) of the powders of the two plants when compared with the control jar (0%).

The mixture PB was already efficient in adult mortality. Here it was noticeable that many weevils in the treated jar were dying in other to accomplish their copulation and laying. Knowing that clove has a very persistent odor [30], this result may be due to the changes induced by the different odor in the physiology and behavior of *C. maculatus* female or either on the males also inhibited them to copulate with the females compared with their egg laying capacity in the control jar. It is in agreement with the finding of many authors such as Rajapakse, Musa *et al*., Agbo *et al*., Nta and Agbo, Kouninki *et al*. [31, 32, 33, 21, 10]. Their research reveals that, the odor of the aromatic plant could affect the behavior of *C. maculatus* through the smell sensilla present in their antenna by then creating a vapor barrier which prevents the cowpea weevils from laying on the cowpea seed. The reduction in oviposition on the treated seed may also be due to the presence of small essential oil film on the seeds, which presents an unsuitable surface for oviposition thus the high percentage of ovoposition deterrence obtained.

Table 3: Effect of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on eggs laid by *Callosobruchus maculatus* females

Plant Powder combination	Percentage of oviposition deterrence (%)				
	Quantities used (g)/100 g				
	0	0.25	0.5	0.75	1
PA	0	93.43±0.95bc	96.56±1.5a	96.70±1.5a	97.18±0.95bc
PB	0	96.25±0.81bc	97.8125±0.95c	96.70±1.5a	98.44±0.5c
PC	0	94.68±1.5ab	96.87±0.57bc	97.3±0.78b	97.2±0.68bc
PD	0	96.87±0.57bc	97.5±1.15c	97.80±0.88c	98.20±0.56c

In the same column, means followed by the same letter are not significantly different to the Newman and Keuls test at 5% level.

3.3 Evaluation of the toxicity of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on *Callosobruchus maculatus* emergence (F_1)

The effect of the mixed powders on *C. maculatus* emergence (F_1) was recorded in Table 4. The maximum percentage of reduction in the adult emergence (90.83%) was observed with the combination PB=1/2VS + 1/2SA and PC= 1/2 VL +1/2SA (90.81%). The lowest percentage of reduction in the adult emergence was 82.5% in dose 1g. It is noteworthy that all the different mixed powders shown more than 70% on the reduction of F_1 adult compared to the control (0%). Also, the efficiency of these selected plant powders was much stronger in the oviposition deterrent activity than in the reduction of the F_1 adult.

The highest percentage of reduction of adult emergence obtained could be due to the reaction of the different compounds present in those powders. The mixed powders reacted twice: either by blocking the young weevils inside the seed through their gap of emergency since it has become very lightweight or by knocked down when they had come out. This result was in line with the earlier findings of Jayakumar *et al*., Sathylaseelan *et al*. and Keita *et al*. [34, 35, 36]. They reported that plant products always have obvious effects on postembryonic survival of the weevils and that postembryonic effect led to the reduction of emergence. The adult emergence was greatly reduced in treated seeds than in control. This result was in agreement with the research of Annie Bright *et al*. [37] and Raja *et al*. [38]. They further stated that, when the eggs were laid on treated seeds, the toxic substance present in the product of the plant may enter into the egg through chorion and suppress their embryonic development. The percentage of reduction was higher than those obtained by Emeazor *et al*. [17] on the mixed leaf powders of *Vernonia amygdalina* and *ocimum gratissimum* in Calabar (Nigeria), proving that *Vepris heterophylla* and *Syzygium aromaticum* had high rate of toxicity against *C. maculatus* emergence.

Table 4: Effect of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on *Callosobruchus maculatus* emergence (F_1)

Plant Powder combination	Percentage of reduction in F1 adult (%)				
	Quantities used (g)/100 g				
	0	0.25	0.5	0.75	1
PA	0	69.16±0.95a	77.5±1.7b	80±0.89b	82.5±1.25bcd
PB	0	81.66±1bc	84.16±1.25bcd	86.25±1.25bc	90±0.83cd
PC	0	85±1.2bcd	87.5±0.95cd	89.25±0.98c	90.81±0.95d
PD	0	76.66±1.15b	79.16±1.25b	88.21±0.68c	88.33±0.57cd

In the same column, means followed by the same letter are not significantly different to the Newman-Keuls test at 5% level.

3.4 Impact of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on the cowpea seeds

The number of holes (more than 2) per seeds on the cowpea three months after treatment was recorded (Table 5). It was found that seeds having more holes were much in the control jar than in other jars. The percentage of attack decreased with the increasing quantity of powders applied depending on the plant powder combination. At the dose of 0.25, 0.5, 0.75 and 1 g/100 g, the highest percentage of attack were 10.75±0.95, 7.25±1.5, 5±1.41 and 4.5±1.29 respectively, recorded essentially with the plant powder combination PA. The percentage of attack in the control jar was 98±0.81 which is very high compared to the percentages obtained with powders PB, PC and PD using various doses. The different powders reduce the percentage of attack from 98 to 4.5% for powder PA, from 98 to 3% for powder PD, from 98 to 1.5% for powder PC and from 98 to 0% for powder PB which was the more efficient.

The jar containing the seeds treated with powder PB has the less damaged seeds and hence the best attack rate of 0%. It was followed by powder PC (1.5±1.29%) and PD (3±1.41%). With all the treatment, there is a significant difference between the treated and the untreated cowpea. This may be due to the fact that the combination of powder of different insecticidal plant increased the active component of the new powder obtained which the synergic effect may either kill the adults of *C. maculatus* or preventing them from laying eggs on the seeds. This action would reduce the number of F1 adult emerged and consequently the percentage of attack. This finding agreed with Kosma *et al.* [24] who reported that the powders of leaves and seeds of *Melia azedarach* reduced the percentage of attack of the seed of cowpea from 96.75 to 0% for the seeds and from 96.75 to 1.5% for the leaves. Also, Ngatanko *et al.* [18] works show that the powder of *X. aethiopica* reduces the percentage attack of maize from 95 to 2.57%.

Table 5: Rate attack of cowpea seed treated with mixed powders of *Vepris heterophylla* and *Syzygium aromaticum*

Plant Powder combination	Percentage attack (%)				
	Quantities used (g)/100 g				
	0	0.25	0.5	0.75	1
PA	98±0.81	10.75±0.95a	7.25±1.5bc	5±1.41cde	4.5±1.29de
PB		3.75±1.25de	2.75±0.95f	2.75±1.25ef	0f
PC		5.25±0.5cd	3.5±1.73ef	2.25±0.5f	1.5±1.29e
PD		8.75±0.95b	6.25±1.5cd	4±1.41de	3±1.41ef

In the same column, means followed by the same letter are not significantly different to the Newman –Keuls test at 5% level.

3.5 Impact of the mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on germination

Seed germination rate before treatment and after three months of treatment was conducted (Table 6). Before treatment the germination rate was 100±0%. After three months, the germination rate recorded varied according to the quantity of the powder applied on the seeds and the different type of powder combination. The result shows that the highest rate of germination was obtained by the combinations PB, PC and PD at 1 g/100 g of the administrated dose although the rate of germination in the control jar where *C. maculatus* was in permanent contact with seeds was 1.5±0.57%.

From this result it is noticeable that seeds treated with the different powders did not lose their viability comparing to the germination rate recorded at the beginning of the test. It might be due to the action of the different component present in the mixed powders which overlain the seeds and confer to them a good protection against cowpea weevils. This is in line with Keita *et al.* [36] and Paul *et al.* [39] research. They reported that powders of some insecticidals plants such as *Azadirachta indica*, Ocimum basilicum and *Ocimum gratissimum* could provide complete protection of seeds in storage against weevils and the rate of germination remained near to 100% after three months of storage. Also, the high rate of germination might link to the few level of essential oil in the powders which makes them permeable to oxygen and consequently good respiration of the seeds. This result is in the contrary with the works of Bell, De Groot and Seignobos [40, 41, 42]. They found out that, many essential oil especially essential oil of neem when overlain on the seeds at certain level, constitute an impermeable barrier of oxygen hence reduce the viability of the seed by knock down and the result is the decrease of the germination rate. More current research shows that the conservation of the germinating power of cowpea seed depends also on the type of seed [12]. Their finding reveals that the cowpea known as FP give the highest rate of germination (88.89%) when it was treated with the extract of *Carissa edulis* and *Vinca rosea*.

Table 6: Germination rate before and three months after treatment with the different mixed powders

Plant Powder combination	Percentage of germination (%)					
	Quantities used (g)/100 g					
	beginning of this test	0	0.25	0.5	0.75	1
PA	100±0	1.5±0.57	85±0.81c	88.75±0.5bc	93.75±0.95ab	95±1.15a
PB			93.75±0.95a	95±0.81a	98.75±0.5a	100±0a
PC			96.25±0.95a	96.25±0.95a	98.75±0.5a	100±0a
PD			97.5±0.57a	98.75±0.5a	100±0a	100±0a

In the same column, means followed by the same letter are not significantly different to the Newman –Keuls test at 5% level.

5. Conclusion

This study target was to investigate on the bioefficacity of mixed powders of *Vepris heterophylla* and *Syzygium aromaticum* on the different developing stages of *Callosobruchus maculatus* pest control of *Vigna unguiculata*. The result of this study shows that the application of mixed powder of *Vepris heterophylla* and *Syzygium aromaticum* increased adult mortality and reduced the rate of emergence

of F1 adult of *C. maculatus*. All the different combination performed well in the control of the developing stages of *C. maculatus*, but the best were the mixture PD=1/3VS+1/3VL+1/3SA (100%) and PC=1/2VS+1/2SA (100%) (VS= *Vepris* stem bark, VL= *Vepris* leaves, SA= *Syzygium aromticum*) as in the mortality rate. For the oviposition deterrence and the reduction of the F_1 adult emergence, the best results were recorded in the mixture PB with 98.44 and 90.83% respectively. All the different mixture shows their efficiency in protecting the cowpea seeds towards their low percentage of attack 0% and their high percentage of germination 100%. According to the obtained results, these plants powders could serve as bioinsecticides for the storage of cowpea seeds. Farmers would be sensitized on the way and the proportion to use in order to best implement it in their various rural areas, since the methods for obtaining the various powders are easy.

6. Acknowledgement

The authors are grateful to the Institute of Research for Agriculture and Development of Maroua for the availability of the materials used during the various experiments. Anonymous reviewers and the editor are also gratefully thanked for their comments and suggestions on the manuscript.

7. References

1. FAO. Knowledge and information for food security in Africa. World food summit plan of action. Food and agricultural Organization of the United Nations, Rome, 1996, 5.
2. IFRC. Africa Responding to Food Crisis. International Federation of Red Croix and Red Crescent societies, 2018, 2.
3. FAO. L'état de la sécurité alimentaire et de la nutrition dans le monde: renforcer la résilience face aux changements climatiques, 2018, 10-20.
4. PAM: Programme Alimentaire Mondiale. Analyse globale de la sécurité alimentaire et de la vulnérabilité au Cameroun, 2011, 7-20.
5. ASSAC: Annuaire Statistique du Secteur Agricole du Cameroun. L'agriculture Camerounaise. Edition, 2015, 232-255.
6. Ngamo LST, Hance T. Diversité des ravageurs des denrées et méthodes alternatives de lutte en milieu tropical. Tropicultura. 2007; 25(4):215-220.
7. Ngamo LST, Ngatanko I, Tamgno BR, Watching D, Madou C, Goudoum A *et al*. Extremely Hazardous and Highly Hazardous Pesticides Registered for Pests Control because of lack of Slightly Hazardous Alternatives. International Journal of Scientific Progress and research. 2016; 19(03):140-145.
8. Magaji BT, Dike MC, Amatobi CI, Onu I. Comparative Efficacy of Some Plant Oils for the Control of *Callosobruchus maculatus* (F.), (Coleoptera: Bruchidae) on Stored Cowpea: M.Sc. Thesis, Ahmadu Bello University, Zaria, Nigeria, 2005, 150.
9. Jazia SE, Olfa B, Nidhal S, Kamel M, Jihad A, Majdi H *et al*. Chemical composition and insecticidal activity of essential oil from coriander fruit against *Tribolium castaenum*, *Sitophilus oryzae*, and *Lasioderma serricorne*, International Journal of Food Properties. 2018; 10(20):1532-2386.
10. Kouninki H, Matseu SGN, Sobda G. Specific insecticidal action of three products of *Balanites aegyptiaca* on developmental stages of *Callosobruchus maculatus* (Coleoptera, Bruchidae). Journal of Entomology and Zoology Studies. 2019; 7(3):1256-1262.
11. Kouninki H, Hance T, Djossou J, Noudjou F, Lognay G, Malaisse F. Persistent effect of a preparation of essential oil from *Xylopia aethiopica* against *Callosobruchus maculatus* (Coleoptera, Bruchidae). African Journal of Agricultural Research. 2010; 5(14):1881-1888.
12. Kongne ML, Kouninki H, Fameni TS, Tchuenguem FFN. Management of *Callosobruchus maculatus* F. (Coleoptera: Bruchidae) using methanol extracts of *Carica papaya*, *Carissa edulis*, *Securidaca longepedonculata* and *Vinca rosea* and impact of insect pollinators on cowpea types in the Far-North region of Cameroon. Journal of Entomology and Zoology Studies. 2018; 6(2):1017-1027.
13. Ngatanko I, Ngamo TL, Ayiki EN, Ngassoum MB, Mapongmetsem PM, Goudoum A. Diversity of plants used to store cereals and leguminous and evaluation of the potential use of three aromatic plants against maize weevil *Sitophilus zeamais* (Coleoptera: Curculionidae). Journal of Entomology and Zoology Studies. 2017; 5(3):1295-1301.
14. Tamgno BR, Ngamo TLS. Potentialisation de l'efficacité insecticide des poudres de feuilles ou amandes de neemier *Azadirachta indica* a. juss par formulation avec la cendre de tiges de mil contre *Sitophilus zeamais* Motsch. et *Sitophilus oryzae* L. (Coleoptera: Curculionidae). African Journal of Food, agriculture, Nutrition and developement. 2018; 18(1):13254-13270.
15. Nukenine EN, Tofel HK, Adler C. Comparative efficacy of Neem Azal and local botanicals derived from *Azadirachta indica* and *Plectranthus glandulosus* against *Sitophilus zeamais* on maize. Journal of Pesticides Sciences. 2011; 84(4):479-486
16. Kayombo MA, Mutombo TJM, Somue MA, Muka MP, Wembonyama OM, Tshibangu BKE *et al*. Effet de la poudre de Basilic (*Ocimum basilicum*) dans la conservation des graines de Niébé (*Vigna unguiculata* L. Walp.) en stock contre *Callosobruchus maculatus* F. à Mbuji-Mayi (RD. Congo) Congo sciences Journal. 2014; 2(2):61-66.
17. Emeazor KC, Emosuirue SO, Ogbuji RO. Preliminary evaluation of the efficacy of mixed seeds powders of *Piper guinneese* (Schum and Thorn) and *Thevelia peruviana* (Person) against *Callosobruchus maculatus* (F.) Colereptera: Bruchidae. Nigerian Journal of Entomology. 2007; 24:114-118.
18. Rozman V, Kalinovic I, Korunic Z. Toxicity of Naturally Occurring Compounds of Lamiaceae and Lauraceae to Three Stored-Product Insects. Journal of Stored Products Research. 2007; 43:349-355.
19. Akunne CE, Afonta CN, Mogbo TC, Ononye BU, Ngenegbo UC. Evaluation of the efficacy of mixed powders of *Piper guineense* and *Zingiber officinale* against *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) American Journal of Biology and Life Sciences. 2014; 2(2):63-67.
20. Suleiman M, Suleiman HY. Control of *Callosobruchus maculatus* (Coleoptera: Bruchidae) using leaf powders of euphorbia *Balsamifera* L. and *Lawsonia inermis*. L International Journal of Science, Environment and Technology. 2014; 3(1):100-109.

21. Nta AI, Agbo BE, Udoekong NS. Bio-insecticidal and Grain Protectant Activities of Mixed Powder of *Ocimum gratissimum* and *Vernonia amydalina* against *Callosobruchus maculatus* (Coleoptera: Bruchidae) in Calabar, Nigeria. Asian Journal of Biology. 2017; 2(3):1-7.
22. Vanmathi JS, Padmalatha C, Ranjitsingh AJA, Suthakarisaac S. Efficacy of selected plant extracts on the oviposition deterrent and adult emergence activity of *Callosobruchus maculatus* F. (Coleoptera: Bruchidae). Global Journal of Sciences Frontier Research. 2010; 10(8):1-7.
23. Kayombo MA, Mutombo TJM, Muka MP, Somue MA, Kalambaie BMME. Effet de la poudre de *Tephrosia vogelii* dans la conservation des graines de Niébé (*Vigna unguiculata* L. Walp.) en stock contre *Callosobruchus maculatus* F. à Mbuji-Mayi (RD. Congo). Journal of Animal & Plant Sciences. 2015; 25(1):3827-3835.
24. Kosma P, Bakop R, Djile B, Abdou Bouba A, Goudoum A. Bioefficacity of the powder of *Melia azedarach* seeds and leaves against *Callosobruchus maculatus*, on cowpea seeds (*Vigna unguiculata*) in storage.E3 Journal of Agricultural Research and Development. 2014; 5(4):072-078.
25. Kouninki H, Mfouapon A, Mohamadou S. Biological activities of *Cassia mimosoides*; *Eucalyptus Camaldulensis*; *Vepris heterophylla* plant extract towards old larvae and adults of *Tribolium castaneum* (coleoptera: tenebrionidae). International Journal of Science, Environment and Technology. 2017; 6(5):3196-3213.
26. Tofel HK. Efficacité insecticide de la poudre de *Plectranthus glandulosus* et des produits de neem à l'égard de *Sitophilus zeamaïs* Motsch. (Coleoptera: Curculionidae). Mémoire de Master en Biologie des Organismes Animaux, Faculté Des Sciences Département Des Sciences Biologiques, Université de Ngaoundéré. 2010, 70.
27. Lale NES. Laboratory assessment of the effectiveness and persistence of powders of four spices on cowpea bruchidae and maize weevil in air tight storage facilities. Journal of Agricultural Research. 1994; 11:79-84.
28. Burt S. Essential Oils: Their Antibacterial Properties and Potential Applications in Foods. International Journal of Food Microbiology. 2004; 94:223-253.
29. Goucem née khalfané K. Etude de l'activité insecticides des huiles essentielles et des poudres de quelques plantes à l'égard de la bruche du haricot *Acanthocelides obtectus* (Coleoptera, Chrysomelidae, Bruchidae) et comportement de ce ravageur vis-à-vis des composés volatils de différentes variétés de la plante hôte (*Phaseolus vulgaris* L). Thèse de Doctorat. 2014, 178.
30. Diego FCR, Claudia Regina Fernandes de Souza, Wanderley PO. Clove (*Syzygium aromaticum*): a precious spice. Asian Pacific Journal of Tropical Biomedicine. 2014; 191(14):60215.
31. Rajapakse RHS. The effect of four botanicals on the oviposition and adult emergence of *Callosobruchus maculatus* (F.). (Coleoptera: Bruchidae). Journal of Entomology. 1996; 21:211.
32. Musa AK, Oyerinde AA, Owolabi FO. Evaluation of the efficacy of mixed leaf powder of *Vernonia amygdalina* and *O. gratissimum* against *Callosobruchus maculatus*. Academic Journal of Entomology. 2009; 2(2):85-87.
33. Agbo BE, Nta AI, Ajaba MO. A review on the use of neem (*Azadirachta indica*) as a bio-pesticide. Journal of Bio-pesticide and Environment. 2015; 2(1):58-65.
34. Jayakumar M, Raja N, Ignacimuthu S. Efficacy of crude extracts of *Hyptis suaveolens* and *Melochia corchorifolia* on pulse beetle *Callosobruchus maculatus.* In: Biological control of Insect Pests. Phoenix Publishing House, New Delhi, 2003, 218-221.
35. Sathyaseelan V, Baskaran V, Mohan S. Efficacy of some indigenous pesticidals plants against pulse beetle, *Callosobruchus chinensis* (L.) on green gram. Journal of Entomology. 2008; 5(2):128-132.
36. Keita SM, Vincent C, Schmit JP, Arnason JT, Belanger A. Efficacy of essential oil of *Ocimum basilicum* L. and *O. gratissimum* L. applied as an insecticidal fumigant and powder to control *Callosobruchus maculatus* (Fab.) (Coleoptera: Bruchidae). Journal of Stored Products Research. 2001; 37(4):339-349.
37. Annie Bright A, Babu A, Ignacimuthu S, Dorn S. Efficacy of *Andrographis peniculata Nzes*. On *Callosobruchus chinensis* L. during post harvest storage of cowpea. Indian Journal of Experimental Biology. 2001; 39:715-718.
38. Raja N, Babu A, Dorn S, Ignacimuthu S. Potential of plants for protecting stored pulses from *Callosobruchus maculatus* (Coleoptera: Bruchidae) infestation. Journal of Biological Agriculture and Horticulture. 2001; 19:19-27.
39. Paul UV, Lossini JS, Edwards PJ, Hilbeck A. Effectiveness of products from four locally grown plants for the management of *Acanthoscelides obtectus* and *Zabrotes subfasciatus* (both Coleoptera: Bruchidae) in stored beans under laboratory and farm conditions in Northern Tanzania. Journal of Stored Products Research. 2009; 45:97-107.
40. Bell A. Emploi des substances végétales comme produits de protection des stocks contre le grand capucin du grain (*Prostephanus truncatus*) et autres ravageurs. GTZ, Eschborn, Allemange. 1994; 12:60-98.
41. De Groot I. Protection des céréales légumineuses stockées. CTA. Document Agronomique. 1996; 18:3-45.
42. Seignobos C. Stratégies de conservation du grain. Atlas de la province d'Extrême-Nord Cameroun, 2002, 12.
43. Abbott WS. A method of computing the effectiveness of an insecticide. Journal of Economic Entomology. 1925; 18:265-267.