IOSR Journal of Environmental Science, Toxicology and Food Technology (IOSR-JESTFT)
e-ISSN: 2319-2402,p- ISSN: 2319-2399.Volume 8, Issue 7 Ver. I (July. 2014), PP 37-43
www.iosrjournals.org

Effects of two eco-friendly insecticides (*Dennettia tripetala* & *Piper guineense*) against *Dermestes maculatus* (Degeer) on smoked dried *Clarias gariepinus* (Pisces: Clariidae)

J.O. Olayinka-Olagunju
Department of Environmental Biology & Fisheries, Adekunle Ajasin University,
PMB 001 Akungba-Akoko Ondo State

Abstract**:** *The efficacy of pulverized and extracts of Dennettia tripetala and Piper gunieense indigenous to Nigeria on larvicidal and adult deterrents of fish beetle Dermestes maculatus on stored dried smoked catfish was investigated. The beetles were reared separately in the laboratory at ambient temperature 30±2^{o}C and relative humidity of (65±5%). The pulverized plants parts at 0.5, 1.0, 1.5, and 2.0g and 0.100, 0.125 and 0.150ml of extracts were used against 20g of smoked fish. Adults and larvae stages of D. maculatus were introduced into different aerated jars containing smoked catfish treated with pulverized plant parts and extracts. The jars were left in the laboratory for 168hours. Results showed that the two plants had varying degree of insecticidal activities but were both effective against the beetles. It was observed that the extracts were more effective than pulverized powder when the results were compared. The plants materials can therefore serve as alternative to synthetic chemicals because of they were effective.*
Keyword*: black pepper, dermestid beetle, catfish, Pepper fruit, insecticides*

I. Introduction

The use of synthetic insecticides against stored products is not usually safe on human health and the environment. This is because accumulated percentage of synthetic pesticides results in different types of pollution in the environment. Examples of some synthetic pesticide that have received most attention include Pentachlorophenol (PCPs), Polychlorinated biphenyls (PCBs), Atrasine (S-triazines), Organochlorines (OCs), organophosphates (Ops) and Carbamates, since they are widely used and are highly persistent compounds [1]. [2]Reveal that in order to overcome pollution problems in the environment and to avoid toxic effects of synthetic on non-target organism many natural products of plants origin are being investigated in the field of plant pathology. However, many plants that eco-friendly and readily available in the environment can be used in the control of insects in stored products. Previous studies have shown the use of plant extracts in the control of pest as important natural chemicals and as possible sources of non-phytotoxic insecticides [3] [4], while many of the plants are very effective because of the active ingredient present in them [5].

Fish is a very rich source of protein due to the presence of vitamin D and other minerals such as copper need for melanin [6]. In many African countries, smoked dried fish is being used increasingly to correct deficiency in the normal diet and often serves as alternative to fresh fish because of its nutritive and economic importance. In Nigeria, smoked dried catfish is highly favoured in many traditional dishes and healing of various ailments [7] [8] [9]. Dried fish is one of the highly digestible and respectable sources of proteins and essential minerals in the tropics and also highly susceptible to insect pest infestation [10]. Smoked dried fish is readily attacked by several species of pests of insects including *Dermestes maculatus*, *D. fruchii, D. ater* and *Necrobia rufipes* [11]. These insects-pests are generally infested during storage, transportation and marketing [12].

Dermestid beetles larvae are very destructive pests of stored products especially hides and skins. Unfortunately, dermestid larvae are less susceptible to widely used insecticides than many other beetles that attacked stored products and the use of insecticides may render the fish unattractive and unpalatable to consumers. In a bit to reduce the insecticidal activities of dried fish, many researches have employed the use of alternative, eco-friendly and cheaper insect-pest control using plant parts as powder and extracts [5] [13] [14]. The inhibitory effect of the extracts may also indicate that the plant possesses bioactive compounds which are soluble in ethanol, cold and hot water.

Many Nigerian medicinal plants species have been cited as very important in pests control of stored grains, legumes and dried smoke fish [15] [16] [17] [18] The leaves, barks, fruits and roots of some plants have been highly appraised for their medicinal purposes. Although many natural protectants are regarded as safe because they are also commonly used traditionally as spices and herbal medicines, studies have shown that some of such plants contain noxious compounds which may render them unsafe for man's consumption [3]. For example, the toxicity of *P. guineense* to the nymph and adult of grasshopper, *Zonocerus variegatus* (L.) and rat,

respectively, have been reported [19] [20]. The aim of this study was to investigate the efficacy of graded concentrations of powders and extracts of two tropical plant materials *P. guineense* and *D.tripetala* against the larva and adults stages of *D. maculatus* (hide beetles) on *C. gariepinus* under laboratory conditions.

II. Materials and method

2.1 Insect culture

The insects were cultured at the Environmental Biology and Fisheries Laboratory, Adekunle Ajasin University, Akungba-Akoko, (AAUA) Ondo State, Nigeria. Several males and females of *D. maculatus* were obtained from Oja Oba market, Ikare- Akoko, Ondo State, Nigeria. They were reared separately in jars covered with muslin cloth under laboratory conditions and kept at 30±2^{o}C and relative humidity of 65±5% as described by [21]. 20g smoked catfish (*C. gariepinus*) was disinfected in the laboratory by heat in Gallenkamp oven at 60^{O}C for 60 minutes and was later air dried to prevent mould growth [15] and each weighed into different jars. New generations of *D. maculatus* were prepared by removing adults of each insect from a stock culture. The stock culture was maintained by introducing six unsexed adult species into each culturing medium and this was covered with muslin cloth held tightly by rubber band. Water was supplied with piece of soaked cotton wool for moisture.

2.2 Preparation of Plant Powders

Fresh fruits of two plant materials commonly used as food condiments in diets namely *D. tripetala* (pepper fruit) and P. guineense (black pepper) were purchase from Ode-Irele and Ibaka markets respectively in Ondo State, Nigeria. *D. tripetala* commonly known as pepper fruit belongs to the family annoaceae while *P. gunieense* commonly called black pepper belongs to the family piperaceae. Each of the plant samples was washed, sun-dried and pulverized into powder using an electric 5.0Hp kitchen grinder and was sieved through a 40holes/mm^2 mesh screen. Each of the plant powders was kept in a separate plastic container with a tightly fitted lid until use.

2.3 Preparation of plant extracts

The extracts of grounded plant materials were prepared at the Department of Chemical sciences of AAU, Akungba-Akoko, Nigeria. Cold extraction method was employed using three different solvents namely, hexane, ethyl acetate and ethanol.

2.3.1 Cold extraction method

10ml of hexane was added to 100g of grounded *D. tripetala* and *P. guineense* respectively, the mixture left on the bench for seven days was shaken twice a day and the cover is slightly open to expel oil from the mixture. Thereafter, the mixture was filtered using bulker funnel, conical flask, aspirator flask and suction pump. Lastly, the filtrate was extracted using simple distillation method and the extract was kept in air tight flask until used. The procedure was repeated for ethyl acetate and ethanol.

III. Experimental Procedures

3.1Effect of Plant Powder on adult *D. maculatus*

Six newly emerged adults of *D. maculatus* were introduced into separate 15 jars containing disinfected dried fish that had been thoroughly mixed with each of the plant powders at 0.5, 1.0, 1.5 and 2.0g per 20g of dried fish. Each treatment was in triplicate and was carried out at ambient temperature of 28-32^{o}C and relative humidity of 65±5%. Three similar containers also in triplicate containing untreated fish with six adult beetles were used as control. The jars were covered with muslin cloth to prevent the beetles from escaping or entry of other insects like ants and allowing aeration for the beetles. Adult mortality was recorded at 24, 72 and 168hours after treatment. Percentage mortality was calculated and recorded.

3.2 ` Effect of Plant Powders on *D. maculatus* larvae

Six 3rd instar larvae of *D. maculatus* were introduced into separate plastic jars containing 20g of disinfected dried fish which were thoroughly mixed with each plant powder at 0.5, 1.0, 1.5, and 2.0g concentrations. Tests were in triplicate of each treatment per insect species at 28-32^{o}C and relative humidity of 65±5%. Similar containers, also in triplicates, containing untreated fish and beetles were used as control. The plastic containers were labelled and covered with muslin cloth so as to allow aeration and prevent other insects from entry. Mortality was recorded at days 24, 72 and 168hrs after treatment. Percentage mortality was recorded.

3.3 Effect of plant Extracts on Adult *D. maculatus*

Six newly emerged adults of *D. maculatus* were introduced into separate jars containing 20g of disinfected dried fish which had been thoroughly mixed with plant extracts of hexane, ethyl acetate and ethanol at 0.100, 0.125 and 0.150ml per 20g of dried fish. Each treatment was in triplicate and was carried out at ambient temperature of 28-32^{o}C and relative humidity of 65±5%. Three similar containers also in triplicate containing untreated fish with six adult beetles were used as control. The jars were covered with muslin cloth, adult mortality was recorded at days 24hrs, 72hrs and 168hrs after treatment and percentage mortality was calculated.

3.4 Effect of Plant extract on *D. maculatus* larvae

Six 3rd instar larvae of *D. maculatus* were introduced into separate plastic jars containing 20g of disinfected dried fish which had been thoroughly mixed with plant extracts from hexane, ethyl acetate and ethanol at 0.100, 0.125 and 0.150ml concentrations. Tests were in triplicate of each treatment per insect species at 28-32^{o}C and relative humidity of 65±5%. Similar containers, also in triplicates, containing untreated fish and beetles were used as control. The plastic containers were labelled and covered with muslin cloth to prevent other insects from entry and allow for aeration. Mortality was recorded at days 24hrs, 72hrs and 168hrs after treatment and percentage mortality was calculated.

3.5 Data Analysis

Data obtained were analysed using Duncan's test to get mean values.

IV. Results

The toxicity results of plant powders against the larvae and adult stage of *D. maculatus* are shown in Tables 1 and 2 respectively. Table 1 shows the effect of plant powders on the mortality of *D. maculatus* larva. This reveals that mortality increases with increase in powder concentrations increases form 0.5g to 2.0g and also the mortality increases from 16.67% to 72.22%. It was observed that many of the insects died as at 168hours implies that the mortality increase with the length of hours.

Table 2 reveals the effect of plant powder on adult *D. maculatus*. It was observed that 94.45% of the adult insects died at 2.00g in *D. tripetala* and also 80.89% in *P. guineese*. The result showed statistical difference when compared to the control. In addition, it was observed that at 168hrs post treatment high mortality had been recorded in both larvae and adult stages. The powder showed good activities against both the adult and larvae of the fish pests. The level of damage was less in the control and very high mortality was observed in the jar containing 2.00g of the powders.

Table 1 Effect of Plant powders of percentage mortality on the larvae of *D. maculatus* (Mean ± SEM)

Plant powder	concentration g/ 20g fish	Mortality (%) at hour post- treatment		
		24 hrs	72 hrs	168 hrs
D. tripetala	0.00 control	0.00±0.00^{b}	5.56±5.56^{b}	11.12± 0.56^{b}
	0.50	16.67±0.92^{ab}	33.34±0.92^{ab}	61.12±0.55^{ab}
	1.00	16.67±0.96^{ab}	44.45±0.55^{ab}	72.23±0.55^{ab}
	1.50	22.22±0.55^{ab}	44.44±0.55^{ab}	72.22±0.55^{ab}
	2.00	27.78±0.55 ^{a}	61.11±0.56^{a}	72.22±0.56^{ab}
P. guineense	0.00	0.00±0.00^{b}	0.00±0.00^{b}	0.00±0.00^{b}
	0.50	5.56 ±0.56^{ab}	38.89±0.96^{a}	66.68±1.47^{ab}
	1.00	5.56±0.56ab	32.22±0.67a	66.55±0.96a
	1.50	16.67±0.92ab	61.11±0.56a	72.22±0.56ab
	2.00	22.22±0.56^{a}	55.55 ±0.56^{a}	83.33±0.55^{ab}

Means in the same column with different superscripts differ significantly ($p<0.05$)

Table 2 Effect of plant powders on adult *D. maculatus* (Mean ± SEM)

Plant powder	concentration g/ 20g fish	Mortality (%) at hour post- treatment		
		24 hrs	72 hrs	168 hrs
D. tripetala	0.00 control	0.00±0.00b	0.00±0.00b	05.56±0.56a
	0.50	16.67±0.96ab	27.78±0.56b	55.56±0.55a
	1.00	16.67±0.96a	55.56±0.56a	72.23±0.96a
	1.50	16.67±0.00a	50.00±0.00a	72.22±0.55a
	2.00	38.89±1.47a	66.67±0.55a	94.45±0.55a
P. guineense	0.00	5.56 ± 0.56^{a}	11.12 ± 0.56^{b}	22.23 ± 0.56^{b}
	0.50	5.56 ± 0.56^{a}	33.34 ± 0.55^{a}	66.67 ± 0.96^{a}
	1.00	11.11 ± 0.56^{a}	33.33 ± 0.55^{ab}	72.22 ± 0.56^{a}
	1.50	11.11 ± 0.56^{a}	38.89 ± 0.55^{a}	77.78 ± 0.56^{a}
	2.00	16.67 ± 0.96^{a}	50.00 ± 0.96^{a}	80.89 ± 0.56^{a}

Means in the same column with different superscripts differ significantly ($p<0.05$).

Tables 3 and 4 show the effect of hexane extract on both adult and larvae stages of *D. maculatus*. As the 168hrs 100% mortality rate was observed at 168hrs. This result reveal that these plant extracts protect smoked *Clarias* against dermestid beetles.

Table 3: Effect of hexane extracts on *D. maculatus* larvae (Mean ± SEM)

Plant extract	Concentration ml/20g fish	Mortality (%) at day's post- treatment		
		24hrs	72hrs	168hrs
D. tripetala	0.00 control	5.56±0.56a	5.56±0.56a	5.56±0.56a
	0.100	83.33±0.96a	94.44±0.56a	100.00±0.56a
	0.125	66.67±1.67a	83.34±0.96a	88.90±0.56a
	0.150	77.78±0.55a	88.89±0.56a	88.89±0.00a
P.guineense	0.00	0.00±0.00b	5.56±0.56a	11.11±0.56b
	0.100	33.33±0.96a	55.55±0.55a	83.33±0.55a
	0.125	38.89±0.56a	55.56±0.96a	72.23±0.00ab
	0.150	44.44±1.47a	61.11±0.96a	83.33±0.55a

Means in the same column with different superscripts differ significantly ($p<0.05$)

Table 4 Effect of hexane extracts on adult *D. maculatus* (Mean ± SEM)

Plant extracts	Concentration ml/ 20g fish	Mortality (%) at hour's post- treatment		
	24hrs	72hrs	168hrs	
D. tripetala	0.00 control	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	0.00 ± 0.00^{a}
	0.100	61.11 ± 1.47^{a}	88.33 ± 0.55^{a}	99.44 ± 0.56^{a}
	0.125	55.55 ± 1.47^{a}	72.22 ± 0.96^{a}	77.78 ± 0.56^{a}
	0.150	72.22 ± 0.55^{a}	94.44 ± 0.55^{a}	94.44 ± 0.00^{a}
P.guineense	0.00	0.00 ± 0.00^{c}	16.67 ± 0.96^{ab}	33.34 ± 0.96^{a}
	0.100	50.00 ± 0.96^{b}	61.11 ± 0.56^{b}	72.22 ± 0.56^{a}
	0.125	44.44 ± 0.56^{b}	77.77 ± 0.00^{a}	88.88 ± 0.56^{a}
	0.150	77.78 ± 0.55^{a}	88.89 ± 0.56^{b}	100.00 ± 0.00^{a}

Means in the same column with different superscripts differ significantly ($p<0.05$).

Tables 5 and 6 also show the effect of ethyl acetate extract on against *D. maculatus* larvae and adult stages and it revealed the potency of the plant extracts. The highest mortality was 83.34% and it show that the extracts are more effective than the powdered plants.

Table 5 Effect of ethyl acetate extracts on *D. maculatus* larvae (Mean ± SEM)

Plant extracts	Concentration ml/ 20g fish	Mortality (%) at hour's post- treatment 24hrs	72hr	168hrs
D. tripetala	0.00 control	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	0.00 ± 0.00^{a}
	0.100	16.67 ± 0.96^{ab}	50.00 ± 0.00^{a}	77.22 ± 0.55^{a}
	0.125	44.44 ± 0.56^{a}	83.33 ± 0.56^{a}	94.44 ± 0.56^{ab}
	0.150	22.22 ± 0.55^{a}	72.22 ± 0.55^{a}	88.89 ± 0.96^{a}
P.guineense	0.00	0.00 ± 0.00^{a}	5.56 ± 0.56^{b}	5.56 ± 0.00^{b}
	0.100	11.11 ± 0.56^{a}	38.89 ± 0.55^{a}	72.22 ± 0.00^{a}
	0.125	11.11 ± 0.56^{a}	44.44 ± 0.00^{a}	72.22 ± 0.55^{a}
	0.150	11.11 ± 0.56^{a}	55.55 ± 01.11^{a}	77.77 ± 0.96^{a}

Means in the same column with different superscripts differ significantly ($p<0.05$).

Table 6 Effect of ethyl acetate extracts on *D. maculatus* larvae (Mean ± SEM)

Plant extracts	Concentration ml/ 20g fish 24hrs	Mortality (%) at hour's post- treatment 72hr	168hrs
D. tripetala	0.00 control	0.00 ± 0.00^{b}	0.00 ± 0.00^{c}	0.00 ± 0.00^{a}
	0.100	27.78 ± 0.55^{ab}	61.11 ± 0.96^{a}	77.78 ± 0.96^{a}
	0.125	38.89 ± 0.56^{a}	66.67 ± 1.11^{ab}	83.34 ± 0.96^{a}
	0.150	44.44 ± 2.00^{a}	50.00 ± 0.56^{bc}	66.67 ± 1.66^{a}
P.guineense	0.00	0.00 ± 0.00^{c}	0.00 ± 0.00^{c}	0.00 ± 0.00^{b}
	0.100	22.22 ± 0.56^{a}	50.00 ± 0.55^{b}	77.78 ± 0.55^{a}
	0.125	38.89 ± 0.56^{b}	66.67 ± 0.55^{b}	83.34 ± 0.96^{ab}
	0.150	27.78 ± 0.55^{ab}	33.34 ± 0.56^{a}	44.45 ± 1.11^{ab}

Means in the same column with different superscripts differ significantly ($p<0.05$).

Table 7 shows the effect of ethanol extract from *P. guineese* against larvae and adult dermestid. 83.33% of the insect adults and 77.78% of the larvae were dead at 168hrs. However, there were no much difference between the potency nature of *D. tripetala* and *P. guineense* but greater differences between the powders and the extracts.

Table 7 Effect of ethanol extracts on *D. maculatus* adult and larvae (Mean ± SEM)

Plant extracts	Concentration ml/ 20g fish 24hrs	Mortality (%) at hour's post- treatment 72hr	168hrs
P. guineense	0.00 control	5.56 ± 0.56^{a}	5.56 ± 0.00^{b}	5.56 ± 0.00^{b}
(Larvae)	0.100	11.11 ± 0.56^{a}	33.33 ± 0.55^{a}	72.22 ± 0.55^{a}
	0.125	22.22 ± 0.55^{a}	44.44 ± 0.55^{a}	72.22 ± 0.55^{a}
	0.150	16.67 ± 0.96^{a}	38.89 ± 0.55^{a}	77.78 ± 0.56^{a}
P.guineense	0.000	0.00 ± 0.00^{a}	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}
	0.100	22.22 ± 0.55^{ab}	44.44 ± 0.55^{a}	72.22 ± 0.55^{a}
(Adult)	0.125	11.11 ± 0.56^{a}	38.89 ± 0.55^{a}	61.11 ± 0.55^{a}
	0.150	44.44 ± 1.47^{a}	66.66 ± 0.55^{a}	83.33 ± 0.96^{ab}

Means in the same column with different superscripts differ significantly ($p<0.05$).

V. Discussion

The result obtained from this study showed that powders of the two plant materials were effective in controlling the emergence of the adult and the destruction of the larval stages of the *D. maculatus* on smoked catfish. These plant materials are better than the synthetic insecticides in that plants powders and extracts are eco-friendly to human and the environment but synthetics are chemicals which are toxic to human health. [7] revealed that the ability of P. guineese fruit oil to protect dried fish from insect infestation could be due to the presence of Piperine. The use of this extract and powder may be the practicable means for the reduction of the production rate of D. maculatus.

Previous studies shows that the powders and extracts of *P. guineense* and *D. tripetala* inhibited adult emergence of *Callosobruchus maculatus* and *Sitophillus zeamis* completely also, the bioactive ingredient/ agent in *P.guineense* has been attributed to the presence of chavicine [16] while [24] linked the bioactive component of D. tripetala to 2-nitroethyl-benzene or beta-phenyl nitroethane. [25] Revealed that the treatment with *P. guineense* powder causes the highest percentage mortality as well as reduced the numbers of adult emergence

than any of the plant materials used in the control of *S. zeamis* the result is similar to what was obtained from this study. [17]Also reported that pulverized plant materials from *P. guineense* inhibited egg hatchability of *D. maculatus* and smoked catfish during storage.

The efficacy of *P. guineense* and *D. tripetala* powders and extracts on mortality in this work could be due to blocking of spiracles due to volatile component thus causing respiratory impairment, which probably affect their metabolism and consequently other system of the body of the dermestid [18]. The plant materials could therefore serve as alternative to synthetic insecticides because they are food condiments which are readily available. Apart from the numerous use of *P. guineense* and *D. tripetala*, the extract from these fruits are very protective against infestation of dried fish by *D. maculatus*. It is highly toxic to insects and safe for human consumption. They do not affect the quality and palatability of the treated fish.

The use of *D. tripetala* and *P. gunieense* powder or extract in the reduction of damage and controlling *D. maculatus* infestation on dried smoke *C. gariepinus* during storage should be encouraged since they used as food condiments which are not toxic to the consumer but to the insects. Also the fish sellers should be enlightened on the importance of these plant materials in order to avoid using toxic chemicals. Further investigations are required particularly on their isolation and biological evaluation test to determine the efficacy and the active ingredients present in the two plants. However, further studies are needed to evaluate the active ingredients, toxicity and concentrations of these extracts for effective use in controlling insect pests of fish during storage.

VI. Conclusion

This study has shown the potency of *P. guineese* and *D. trepatala* plant powders and extracts against dermtid beetles in smoke fish and the result has also proved that these plants can serve as alternative to synthetic chemicals used in the protection of stored products. The plants are readily available and also serve as spices in many African dishes, eco-friendly and safe for human consumption and the environment.

Acknowledgments

Thanks to Dr. F.O. Akinwumi, Dr. E. Adebayo, Dr F.A. Gbore and Dr. K.D. Ileke and all technologists in the Departments of Chemical Science and Environmental Biology and Fisheries, Adekunle Ajasin University, Akungba-Akoko, Ondo State, Nigeria.

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