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Contact activities of *Piper guineense* (Schum and Thonn) and *Eugenia aromaticum* (L). (Merril and Perryl) extracts against larvae of hide beetles, *Dermestes maculatus* (Degger) (Coleoptera: Dermestidae)

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Contact activities of *Piper guineense* (Schum and Thonn) and *Eugenia aromaticum* (L). (Merril and Perryl) extracts in the control of *Dermestes maculatus* larvae infesting stored fish (*Clarias gariepinus*) were investigated under laboratory condition (28 ±3°C and 65±5% RH). The extracts were tested by application of 2.0 µL each to ten third instars larvae using micro pipette at a concentration of 6.00, 10.00 and 20.00% of each of plants extracts (methanol, n-hexane and ethyl acetate). Mortality was recorded at 1, 2, 3 and 7 days of post treatments. The observed mortality was dose and exposure-dependents. All the extracts significantly enhance larval mortality (P>0.05) when compared with control. The n-hexane and ethyl-acetate extracts of *P. guineense* at 20% concentration induced the highest mortality of 86.66%, lowest mortality of 56.66% was observed on methanol fraction treated larvae after 7days of post treatments. The n-haxane of *E. aromaticum* extracts recorded the highest mortality (80.0%), followed by ethyl-acetate (76.66%) and methanolic (7.00%) fractions treated larvae at 20% concentration after 7 days of post-treatments. The results showed strong insecticidal activity in control of larvae of hide beetles infesting dried fish.

Key words: Plant extracts, *Piper guineense*, *Eugenia aromaticum*, *Dermestes maculatus.*

INTRODUCTION

Hide Beetle *Dermestes maculatus* (Degeer) (Coleoptera; Dermestidae) is one of the most destructive insect pests of stored smoked-dried fish in Nigeria (Tejumade, 2019). These pests generally infest dried fish during storage, transportation and marketing, thus responsible for extensive damage to marketed fish leading to enormous weight loss (Don-Pedro, 1989; Amadi and Dimkpa, 2018). Tejumade (2019) reported *D. maculatus* account for about 71.5% of the observed infestation with substantial loss in dry weight of about 43 to 62.7% from both larvae and

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adult.

The control of these pest in Nigeria is primarily dependent upon repeated application of synthetic chemicals such as chloripyripos-methyl, permetrin, cypermetrin, BHC, and “Otapiapia” (locally formulated) onto fish carton for protection against insect pest (Igene et al., 1998; Abolagba et al., 2011). Although many synthetic chemicals are effective, the general use of such chemicals to protect stored fish has been hampered by the report of health hazard, high cost of purchase, lack of availability, illiteracy of fish handler for right application and less susceptibility of Dermestid larvae (Booke et al., 2001; Amusan and Okorie, 2002). With these demerits of synthetic chemicals currently world-wide interest is centered on search for alternative pesticide to stored product by the use of botanical pesticide.

Botanical pesticide tends to have broad spectrum activity which are relatively specific in their mode of action and easy to process and use (Viglianco et al., 2008). To minimize use of synthetic pesticide, several plants extract have been reported as effective against *D. maculatus* on dried fish by several researchers (Fasakin, 2003; Akinwumi et al., 2006, 2007; Akpotu and Adebote, 2013; Olayinka, 2014). These extracts provide a solution to the problem emanating from the use of synthetic chemicals. The present studies have been chosen to investigate the effects of methanolic extracts fractions of *Piper guineense* and *Eugenia aromaticum* against *D. maculatus* larvae as an alternative strategy to synthetic chemicals method of pest control.

MATERIALS AND METHODS

Collection, identification of plant material and preparation of plant powders

The sample of dried fruit of *P. guineense* and *E. aromaticum* were obtained from Sokoto Central Market, Nigeria. The plants were identified and authenticated in the Herbarium of Biological Sciences Department of Usman Danfodiyo University, Sokoto. Voucher specimens (UDUH/ANS/0258 and 0221) were deposited. Samples were milled into fine powders using mortar and pestle, sieved with 0.2 mm mesh following the methods of Adedire and Lajide (2000), Akinwumi et al. (2006) and Jose and Adesina (2014). Each of the plant powders was labeled and kept in a separate plastic container and placed in a cool dry place prior to use.

Preparations of feed

The samples of dried fish, *Clarias gariepinus* was purchased from fish mongers at Sokoto Central Market, identified and authenticated in Hydrobiology Laboratory, Biological Sciences Department of Usmanu Danfodiyo University, Sokoto. The fish samples were disinfected by heat treatment in the laboratory-drying cabinet at 60°C for 1 h and allow cooling at room temperature as adopted by Onu and Baba (2003).

Collection of hide beetle and maintenance of insect culture

Different stages of hide beetle were obtained from naturally infested fish collected from Sokoto Central Market fish stalls. Several adult pairs of *D. maculatus* were obtained and kept in transparent plastic containers (19.0 cm height and 21.2 cm in diameter) fed with dried fish. The containers were covered with Muslin cloth and tight with rubber band. Wet cotton wool was supplied in each jar to provide water requirements for oviposition as suggested by Hill (1990). The adult’s laid eggs which hatched into larvae and changed to pupae, pupae were picked up and transferred into separate container to obtained newly emerged adult, which were used for regular supply of larvae for the experiment in line with Akinwumi et al. (2006).

Preparation of methanol extracts and solvent fractionation

Four hundred grams (400 g) of *E. aromatica* and *P. gueneense* were homogenized with 95% methanol (1 L) in plastic container and kept at room temperature for 24 h and filtered. The methanol crude extract was collected and concentrated almost to dryness in drying cabinet at 40°C for 48 h. The dried extracts were stored in freezing medium until used for fractionation (Akinwumi et al., 2006).

The dried methanol crude extract of *E. aromatica* (19.47 g) and *P. guineense* (26.17 g) were suspended in distilled water and then partitioned with 500 ml of n-Hexane, ethyl acetate and water in increasing order of polarity, following method of Bakele et al. (2016).

Effect of extract fractions on *D. maculatus*

Effects of each plant extract fraction were conducted according to Talukder and Howse (1994). 20% stock solution was prepared for each solvent (methanol, n-haxane and ethyl acetate). Lower concentrations (6 and 10%) were obtained from dilution of the stock solution with distilled water. Ten third instars larvae were chilled for 5 min to immobilize them and then picked up individually by the use of camel hair brush and 2 µl of each of the solution was applied to the dorsal surface of the larvae. Experiments were in three replicates (each replicate contains ten treated larvae). In addition, the same number of larvae (10) was treated with distilled water only as control. After treatment insects were transferred into transparent plastic containers (19.0 cm height and 21.2 cm in diameter) containing dried fish. Observations were made daily and those that did not move or respond to gentle touch was consider as dead. Mortality was recorded at 1st, 2nd, 3rd and 7th days of post treatment.

Data analysis

Data obtained were subjected to one-way analysis of variance (ANOVA) using General Linear Model (SPSS, 2007) and means found to be significant were separated using Duncan multiple range test at 5% level of significant ($p<0.05$).

RESULTS

Effects of methanol fractions on mortality of *D. maculatus* larvae

The effect of *P. guineense* methanolic extracts applied to *D. maculatus* larvae by topical application are presented in Table 1. All the three extract of *P. guineense* exhibited insecticidal activity against *D. maculatus* larvae as dose and time-dependent variables. At day 1 all the three extract of *P. guineense* showed less than 50% mortality

Table 1. Mortality among D*. maculatus* larvae by topical application with *P. guineense* methanolic extracts fractions.

Solvent	No. of Larvea introduced	Mean larval mortality ± SE					
		Period of exposure (in days)					
		Concentration (%)	1st	2nd	3rd	7th	Mortality (%)
Methanol	10	20.00	2.33±0.88^{bcde}	3.33±0.88^{bc}	4.33±0.33^{cd}	5.66±0.88^{cd}	56.60
	10	10.00	2.33±0.33^{bcde}	3.33±0.88^{bc}	4.33±1.20^{cde}	5.66±0.33^{cde}	56.60
	10	6.00	1.33±0.66^{cde}	1.66±0.88^{cd}	2.00±1.73^{ef}	3.33±1.20^{ef}	33.30
N-hexane	10	20.00	4.66±0.33^{b}	6.33±0.33^{ab}	**8.00**±0.57^{ab}	8.66±0.66^{bc}	86.60
	10	10.00	4.33±1.45^{b}	6.00±2.08^{abc}	7.00±2.08^{abc}	8.00±2.00^{abc}	80.00
	10	6.00	1.00±0.57^{cd}	2.00±0.57^{cd}	3.66±0.33^{cde}	5.66±0.33^{de}	56.60
Ethylacetate	10	20.00	6.67±0.88^{a}	**8.00**±0.57^{a}	8.33±0.66^{ab}	8.66±0.33^{bc}	
	10	10.00	4.33±0.33^{b}	5.66±0.33^{ab}	5.66±0.33^{bcd}	7.33±0.33^{abc}	86.60 73.30
	10	6.00	3.66±0.88^{bc}	4.00±1.15^{bc}	5.66±0.33^{bcd}	6.3388^{bc}	63.30
Cypermetrin	10	0.05	3.00±0.57^{bcd}	6.33±0.33^{ab}	9.33±0.33^{a}	9.66±0.33^{a}	96.60
Control	10	-	0.00±0.00^{e}	0.33±0.46^{d}	1.33±0.33^{e}	1.33±0.33^{e}	13.30
p-level	-	-	-	-	-	-	-

Means that have the same super script within a column are not significantly different at 5%level using Duncan's multiple range test.
Source: Author 2020.

of larvae except ethyl-acetate extract at the highest concentration (20%) which caused 66.70% mortality of larvae. At 2nd and 3rd day, mortalities in all the treatment at all concentration increased compared to day 1 of exposure. The ethyl-acetate extract at 20% concentration remain the highest 80% motality of larvae. However, at 7th day of exposure all the three extract at all concentration except 6% concentration of methanol showed a significant ($p<0.05$) mortality of larvae compared to control. The ethyl-acetate extract recorded the highest mortality range of 63.30 to 86.66%, followed by n-Hexane (56.60-
86.6%) and methanol extract (33.30-56.60%).

The contact activity of *E. aromaticum* methanolic extracts fractions applied *D. maculatus* larvae presented in Table 2. All treatments except 6% concentration of methanol and 10 and 6% of n-Hexane were significantly more toxic than control at 1st day of exposure. Efficacy was dosage-dependent with significant higher mortality occurring with increase dosage. No mortality occurred in control (0.00%). Highest mortality was recorded in ethyl-acetate extract at 20% concentration with a percentage mortality of 63.30%. At 2nd day of exposure only 6.0% concentration of methanol extract was statistically similar ($p>0.05$) with control, all other treatment showed significant mortality of larvae compared with control, the highest mortality was recorded in ethyl-acetate (63.33%) followed by n-Hexane (56.60%) and methanol (53.30%). However, at 3rd day of exposure mortality in all the treatment followed a similar trend with 2nd day of exposure with ethyl-acetate extract was the highest with a mortality range of 50 to 70%. At 7th day of exposure, the highest mortality of 80.00% was recorded from n-Hexane fraction at 20% concentration, other concentration of n-Hexane also showed higher mortality of larvae of 53.30 and 46.66%. In addition, the ethyl acetate and methanol extract fraction recorded mortality of larvae ranging from 53.30 to 76.60% and 43.30 to 70.00%, respectively.

DISCUSSION

In the current study, the three (3) extract of *P. guineense* demonstrated contact efficacy to *D. maculatus* larvae. The results indicated that the reported ethanol extract of *D. tripetala* and *P. guineense* resulted in 100% mortality of *D. maculatus* larvae after 3 days of post treatment. Ajayi (2015) stated that acetone extract is more effective in reducing oviposition and adult emergence of *Callosobruchus maculatus* than methanolic and ethanolic extract of the same plant, while methanolic and ethanolic extract were significantly more effective than aqueous extract.

In the current study, the higher activity of ethyl acetate fraction observed might be due to the presence of polar and no polar bioactive component against larval stage of *D. maculatus, a*s ethyl acetate is a semi polar solvent that had ability to extract polar and non-polar compound in the extract of *P. guineense.* Variation in the bioactivities of different solvent fraction observed in the study confirmed the finding of Sun et al. (2001). That crude extract that

Table 2. Mortality among *D. maculatus* larvae by topical application with *E. aromatica* methanolic extracts fractions.

Solvent	No. of Larvea introduced	Mean larval mortality ± SE					
		Period of exposure (days)					
		Concentration (%)	1st	2nd	3rd	7th	Mortality (%)
Methanol	10	20	3.33±o.33^{ed}	5.33±0.33^{abc}	6.00±1.15^{b}	7.00±1.15^{bcd}	70.00
	10	10	4.44±0.66^{bc}	4.00±0.57^{bcd}	5.66±0.33^{b}	6.33±0.66^{bcd}	63.30
	10	6.0	1.33±0.33^{efg}	1.33±0.33^{ef}	2.66±0.66^{cd}	4.33±0.66^{e}	43.30
N-Hexane	10	20	5.33±0.33^{ab}	5.66±0.33^{ab}	6.33±0.33^{b}	8.00±1.00^{ab}	80.00
	10	10	1.00±0.66^{fg}	3.33±0.66^{cd}	4.66±0.66^{bc}	5.33±0.88^{cde}	53.30
	10	6.0	1.66±0.88^{defg}	2.33±0.88 ^{de}	2.33±0.88^{d}	4.66±0.33^{de}	46.60
Ethylacetate	10	20	6.33±0.66^{a}	6.33±0.57^{ab}	7.0±1.15^{b}	7.00±0.57^{bcd}	70.00
	10	10	4.33±1.20^{bc}	6.00±0.57^{ab}	6.33±0.33^{b}	7.66±0.33^{abc}	76.60
	10	6.0	2.00±0.57^{def}	4.33±1.20^{abcd}	5.00±1.15^{b}	5.33±0.88^{cde}	53.30
Cypermetrin	10	0.05	3.00±0.57^{cde}	6.33±0.33^{a}	9.66±0.33^{a}	9.66±0.33^{a}	96.60
Control	10	0.00	0.00±0.00^{g}	0.33±0.33^{f}	0.66±0.33^{d}	1.33±0.33^{f}	13.30

Means that have the same super script within a column are not significantly different at 5%level using Duncan's multiple range test.
Source: Author 2020.

was screened with ethyl acetate, n-butyl alcohol and water fractions of alcoholic extract of leaves and stem of *Vanilla fragrans* against *Culex pipiens* larvae found that n-butyl alcohol and ethylacetate fractions were active in the bio assay, while the water fraction appeared to contain no substance that inhibited the larval growth. Overgaard et al. (2014) showed that mortality rates of mosquitoes declined with increasing polarity of the solvent, the water extract of *Zanthoxylum heitzii* (Rutaceae) produce the lowest adult mortalities whereas its ethyl-acetate and hexane extracts produce high mortalities against *Anopheles gambiae.*

Furthermore, the classes of phytochemical compound contained in the fractions might be responsible for insecticidal actions. Lale and Alaga (2000) reported *P. guineense* extract is known to contain at least three different alkaloids responsible for its insecticidal activity (piperine, chavicine and piperidine).

The result of the study also revealed the efficacy of *E. aromaticum* extracts in which all three extract fractions (was solid, not oily) gave high mortalities of larvae which could be due to its important secondary metabolite such as tarpenes, linoleic acid and oleic identified as the main active compound in *E. aromatica* (Golob et al., 1999). This supports the finding of Akinwumi (2010) who reported 100% mortality of *D. maculatus* adults when 1 ml of oil is mixed with 10.00 g of powder of *E. aromaticum* after seven days of post treatment; the finding also supports the work of Ajayi (2015) who reported that clove and west African black pepper was significantly more toxic to adult of *Tribolium castaneum* than ginger at dosage of 100 mg/50 of seed. Clove and West African black pepper and ginger oil caused 96.3, 100 and 13.2% adult mortalities, respectively and 65.7 and 9.6 larval mortalities, respectively. Akinwumi et al. (2007) also reported 0.5 g of *E. aromaticum* recorded 50.00% larval mortality and 51.67% adult mortality and concentration of 1.0, 2.0 and 2.5 g recorded 100% larval and adult mortality.

Conclusion

The study demonstrated the contact toxicity of *P. guineense* and *E. aromaticum* against *D. maculatu*s larvae. The maximum mortality was recorded on the highest dose of ethyl acetate fraction for both plants, followed by n-Haxane, while methanolic extract recorded the least activity. Hence, ethyl-acetate extract of both plants have potential insecticidal activity against *D. maculatus* larvae.

Recommendations

The findings revealed that both methanolic fractions of *P. guineense* and *E. aromatica* could be used as fish protectant against *D*. *maculatus*. Therefore, the use of this plant extracts for control of *D. maculatus* infestation during processing, storage, transportation and market of smoked dried fish is recommended.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Abolagba OJ, Igene JO, Usifoh CO (2011). Studies of Pesticide Residues in Smoked Cat Fish (*Clarias gariepinus*) in Nigeria: Some Health Implication. Australian Journal of Basic and Applied Science 5(5):496-502.

Adebote DA, Abolude DS, Oniye SJ, Oladodo SS, Hassan MM (2006). Larvicidal and repellant action of *Detarium microcarpum* seed oil against the larvae of *Dermestes lardarius* (Coleopteran: Dermestidae) in dried C*larias gariepinus* fish. Journal of Entomology 38(3):245-253.

Adedire CO, Lajide L (2000). Effect of pulverised plant materials on fish damage and growth performance of fish beetle. *Dermestes maculatus* (Degeer). Journal of Entomology 32:215-218.

Ajayi FA, Peter E (2016). Efficacy of three essential oil in the management of *Cryptolestes ferugineus* (Steph.) Infesting stored Maize grain in Lafiya, Nigeria. Nigerian Journal of Entomology 32:4857.

Ajayi FA (2015). Insect, Plants and Humanity; the Organic Agriculture and Stored Products Protection Axis, 10th in Augural Lecture, Nasarawa State University, Keffi, Onaivi Printing and Pub. Co. Ltd, Keffi Nasarawa State; Nigeria pp. 1-69.

Akinwumi FO (2010). Bioefficacy of Some Oil Mixed Plant Derveratives Against African Mud Catfish *(Clarias gariepinus)* Bettles, Dermestes *maculatus* and *Necrobia rufipes.* World Academy of Science, Engineering and Technology 68:893-900.

Akinwumi FO, Fasakin EA, Adedire CO (2006). Progeny inhibiting effect of four plant product against the leather beetle and copra beetles on smoked African mud cat fish *Clarias gariepinus* Burchell. Journal of Biological Science 6(6):1023-1028.

Akinwumi FO, Fasakin EA, Adedire CO (2007). Toxic and Repellance activities of four extract to *Dermestes maculatus* Degeer on smoked African mud. Catfish, *Clarias gariepinus* Burchell. Journal of Entomology 4(2):149-154.

Akpotu JO, Adebote DA (2013). Repellency Effect of Five plant extracts Against the larvae of *Dermestes maculatus* on smoke Dried *Clarias gariepinus.* Research Journal of Chemical and Environmental Science 1(4):1-4.

Amadi GI, Dimkpa SON (2018). Effect of Indigenous Plants on the Progeny Development of Dermestes maculatus (Degger, 1774) in Dried African Cat Fish (*Clarias gariepinus*). Journal of Biology Agriculture and Health Care 8(6):68-74.

Amusan AAS, Okorie TG (2002). The use *of Piper guineense* Fruit oil (PFO) as protectant of dried fish against *Dermestes maculatus* (Degeer) infestation. Global. Journal of Pure and Applied Sciences 8:197-201.

Bakele D, Habte, T, Zemede A, Bayene P (2016). Bio efficacy of solvent fraction of *Oreosyle africana* and *Piper capense* against the malaria vectors, *Anopheles arabiensis* with High performance liquid chromatographic and ultraviolet visible spectroscopic Analysis. Biochemistry and Analytical Biochemistry 5(3):1-8.

Booke SJ, Vanlon JJA, Vanttuis A, Kossou DK, Dicke M (2001). The Use of Plant Material to Protect Stored Leguminous Seed against Seed Beetle: A Review. The Netherlands: Backhuys Publish P 108.

Don-Pedro KN (1989). Insecticidal activity of some vegetable oil against *Dermestes maculatus* Degeer (Coleoptera; Dermestidae) on dried fish. Journal of Stored Products Research 25(2):81-86.

Fasakin EA (2003). Use of plant oil extract as protectant against storage insect pest, *Dermestes maculatus* Degger in smoked fish: In Eyo, A. A. 18th Annual Conference of Fish. Society Nigeria Proc*.* P 1-6.

Golob P, Moss C, Dales M, Fidegen A, Evans J, Gudrups I (1999). The use of spices and medicinal as bioactive protectants for grains. Food Agriculture organization agricultural services bulletin 137 p.

Hill DS (1990). Pest of Stored Products and their control.Belhaven Press London,74-75.

Igene JO, Abolagba OJ, Ikeme AI (1998). Use of Pesticide on Cured Fish in Nigeria. Report and Proceeding of the Sixth FAO Expert Consultation on Fish Technology in Africa, Kisumu, Kenya pp. 126-130.

Jose AR, Adesina JM (2014). Larval Susceptibility of *Dermestes maculatus* (Degeer) (Coleoptera: Demestidae) to *Secamone Afzelli* K. Schum Leaf on Smoked Dried Fish. International Journal of Aguaculture 4(17):101.

Lale NES, Alaga KA (2000). Exploring the insecticidal, Larvicidal and repellent properties of *Piper guineense* (Schum and Thonn) seed oil for the control of rust-red flour beetle *Tribolium castaneum* (Herbst) in stored pearl millet *Penniseteum glaucum* (L) R. British Journal of plant diseases and protections 108(3):35-313.

Okonkwo EU, Okoye WI (2001). Insecticidal activity of *Dennettia tripetala Baker* (F) and *Piper guineense* Schum. and Thonn against *Dermestes maculatus* Degeer (Coleoptera; Dermestidae) and *Necrobia rufipies* Degeer *(*Coleptera:Cleriadae*) on* dried fish. Nigerian Journal of Entomology 18:109-117.

Olayinka OJ (2014). Effect of Two Eco-Friendly Insecticides (Dennetta Tripetela) and *Piper guineense*) Against Dermestes maculatus (Degeer) on Smoked Dried *Clarias garipinus* (Pisces: Claridae). IOSR Journal of Environmental Science, Toxicology and Food Technology (10SR-J (STFF) 8(7):37-43.

Onu I, Baba GO (2003). Evaluation of neem products *(Azadrachta indica* (A. Juss; Melaceae) for the control of Dermestid beetle. *Dermestes maculatus* (Coleoptera Dermestidae) on dried fish. Nigerian Journal of Entomology 20:105-115.

Overgaard HJ, Sirisopa P, Mikolol B, Malterud WH (2014). Insecticidal activities of bark, leaf and seed extract of *Zanthoxylum heitzii* against the African malaria vector *Anoples gambiae.* Molecules 19:2127621290.

SPSS (2007). Statistical, Packages for the Social Science Version 16 SPSS Inco.

Sun R, Sacallis JN, Chin C, Still CC (2001). Bioactivities aromatics compound from leaves and stem of *Vanilla fragrans.* Journal of Agricultural and Food Chemistry 49:5161-5164.

Talukder FA, Howse PE (1994). Efficacy of pithraj (*Aphanamixis polystachya*) Seed extracts against stored – product pests. Proceeding of the 6th international working conference on stored product Protection 2:848-852.

Tejumade MP (2019). Evaluation of ethanolic extracts of *Piper guineense, Dennettia tripetala and capsicum fruitescense* as protectant of smoked fish, *Clarias gariepinus* (Pisces:Clariidae) against *Dermestes maculatus* (Coleoptera:Dermestidae). The Journal of Basic and Applied Zoology 80(7):1-7.

Viglianco AI, Novo RJ, Crajonolin CI, Nassetta M, Cavallo EA (2008). Atifeedant and Repellent Effect of Extract of Three Plant from Cordobo (Argentina) Against *Stiphilus oryzae* (L). Coleoptera: Curculionidae). Bioassay 3(4):1-6.