Insecticidal Activity of Essential Oils from Three Plant Seeds against Beetle(*Dermestesmaculatus*)in Smoke-dried Catfish (*Clarias gariepinus*)

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Abstract

This study was conducted to determine the insecticidal activity of essential oils extracted from the seeds of *Piper guineense, Monodoramyristica*and*Aframomummeleguet*aagainst adults of the beetle, *Dermestesmaculatus*, on smoke-dried catfish, (*Clarias gariepinus*) during storage.The beetle was least tolerant to *P. guineense*oil which recorded 100% mortality within the first week of the experiment.*M. myristica*oil recorded 100% mortality for beetles on catfish in the second week while *D. maculatus* was most tolerant of *A. melegueta* oil which attained 100% mortality in four weeks. *P. guineense* and *M. myristica* oil extracts were more effective than *A. melegueta* oil extract, though it was observed that *M. myristica* attained 100% mortality in the second week of experiment, the moisture absorption by smoke dried catfish was higher than that of *A. melegueta* which obtained 100% mortality in the fourth week of experiment.

Keywords: Insecticidal activity, essential oils, catfish, *Dermestesmaculatus*.

Introduction

Fish is highly perishable which necessitates its processing and preservation especially in the tropics where high temperature and humidity accelerate the spoilage and bio-deterioration immediately after catch or death. Fish post-harvest losses in Nigeria have been estimated at 30-40%(FAO, 2004). This susceptibility of fish to damage has necessitated the direction of efforts, primarily, towards the preservation of fish for human consumption. However, poor handling, inadequate processing facilities, lack of ice or storage facilities, remoteness of the fishing villages to urban market centres, poor transportation system and poor distribution channels have drastically reduced fish utilization in the tropics (Ames, 1992).

In developing countries, fish is preserved either by salting, smoking or sun-drying and fermentation. Smoke is produced by the process of incomplete combustion of wood in order to impart a characteristic flavour and colour to the fish.Inaddition, smoking increases the shelf life of fish as a result of the combined effects of cooking, dehydration, anti-microbial and antioxidant activity of several of the smoke constituents mainly formaldehyde, carboxylic acid, phenols. Often, smoke-dried fish is eaten without further cooking (Asita, 2000).

Despite the availability of these preservation methods, fish is still highly susceptible to bio-deterioration which does not depend on microbes, enzymes and fat oxidation only but also on insect pest infestation, especially in cured fish (Balogun, 1992). Insect infestation is a serious spoilage agent and fish that is not properly cured can harbour large populations of maggots and blowfly whose developments are enhanced by high moisture content (Dobie et al., 1991).

Infestation by insects could cause up to 50% loss of weight and occurs during different stages of drying of the fish product. As drying continues, the adult *Dermestesmaculatus* and *Necrobraruffipies* lay eggs on fish and the emerging larvae feed on the fish flesh which may break up. Observation suggests that the fish needs to be at least surface-dried before *Dermestesmaculatus* can commence the laying of eggs. After drying, infestation by *Dermestesmaculatus* and *Necrobraruffipies* and mites increases rapidly and this continues during storage and transportation. These pests feed on flesh causing loss in weight and quality, especially with the appearance of insects and maggots. Dried fish can be de-infested by heat treatment such as re-smoking or the use of solar driers. Some beetle larvae are killed by exposure to temperature of 50°C for 15 minutes.

Treatment may include heavy salting which protects the fish against attacks by the larvae of the beetle and decrease the body weight of the adult *Dermestesmaculatus*.

There appears to be some degree of variability in the susceptibility of different fish genera to *Dermestesmaculatus*, for example,Clarias and Tilapia are easily infested while *Heterobranchus*and Synodontis are somewhat more resistant.Fish of fresh water origin are more frequently attacked probably due to the rather poor salt tolerance of this species. Development of the larvae is apparently impossible on fishmeal with a salt level as high as 10%.

Efforts to reduce losses from insect infestation by the use of insecticides have not been fully adopted due to the hazardous nature of these chemicals to health and toxicity at high doses to users (Balogun, 1992). In order to eliminate much of these problems, plant-derived insecticides which are bio-degradable, environment-friendly, cheap, available and affordable to fish farmers and processors have been proposed(Adedire and Lajide, 1999).

In Nigeria, despite the availability of cured fish, high losses still occur due to infestation by beetle (*Dermestesmaculatus*). Peasant farmers are unable to purchase synthetic insecticides due to the high cost but instead tend to substitute these with naturally-occurring insecticides applied mainly as powders or oilsas they are readily available, relatively cheap and safer (Lale, 1995).This study was conducted to evaluate the insecticidal activity of the essential oils extracted from the seeds of common plants: *Piper guineense, Monodoramyristica*and *Aframomummelegueta* against the adults of the beetle (*Dermestesmaculatus*) on smoke-dried catfish (*Clarias gariepinus*) during periods of storage.

Materials and Methods*Insect culture and maintenance*

The founding insect culture of *Dermestesmaculatus* was collected in smoke-dried catfish (*Clarias gariepinus*) that was stored in 3-litre plastic containers at the laboratory of Ekiti State University, Ado Ekiti, Nigeria.

The culturing jars were kept at ambient temperature condition. The jars were disinfected in laboratory drying cabinet at 70°Cfor 1 hour and allowed to cool to room temperature.

Plant materials and extraction of essential oils

Piper guineense (Black Pepper)

West African pepper, also known as Ashanti pepper, Benin pepper, false cubeb, guinea cubeb, uziza pepper or Guinea pepper is a perennial, glabrous woody climber up to 20 m high.The dried fruits of *Piper guineense* are prolate spheroidsand generally bear a reddish tinge. The pepper fruit contains 5.8% piperine which is responsible for the "heat" or pungency; and large amounts of beta-caryophyllene that is being investigated as an anti-inflammatory agent. The fruit also contains significant proportions of myristicin, elemicin, safrole and dillapoil. Research has shown that Ashanti pepper has preservative and anti-oxidant properties and was most effective among three West African pipers for preservation of catfish.

Monodoramyristica (African Nutmeg)

African nutmeg, Jamaican nutmeg or calabash nutmeg (*Monodoramyristica* Dunal) is a tropical tree of the family Annonaceae or custard apple family of flowering plants. The fruit is a berry of 20cm diameter and is smooth, green and spherical and becomes woody. Inside the fruit, the numerous oblongoid, pale brown and 1.5cm long seeds are surrounded by a whitish fragrant pulp. The seeds contain 5-9% of a colourless essential oil.

Aframomummelegueta (Alligator Pepper)

Aframomummelegueta, commonly known as grains of Paradise, melegueta pepper or alligator pepper is a member of the family Zingiberaceae. This spice is an herbaceous perennial plant native to swampy habitats along the coast of West Africa. Its trumpet, shaped, purple flowers develop into 5-7cm long pods containing numerous small, reddish brown seeds. The pungent, peppery taste of the seeds is caused by aromatic ketones (6)-paradol (systematic name: 1-(4-hydroxy-3-methoxypheny D- decan-3-one). The seeds of *Piper guineense*, *Monodoramyristica* and *Aframomummelegueta* were purchased from the market in Ado Ekiti, Ekiti State. The seeds

were dried in a laboratory drying cabinet at 40°Cfor 8 hours and ground in an electric grinder (Retsch Gmbtt, J 657 HAAN). Oil extracts from the milled seeds were obtained in a Soxhlet apparatus using petroleum ether at 40-60°Cand concentrated in the water bath at 60°C.

Preparation of Fish Samples

The smoked fish samples were dried in the laboratory drying cabinet at 60°Cfor 6 hours to ensure adequate drying and the initial weights were recorded.

Experimental Procedure

The whole dried catfish (*Clarias gariepinus*) was put into separate plastic jars and 10 insects were randomly introduced to each jar. The jars were covered with muslin cloth so as to allow aeration for the beetle. The oil extracts were applied to the fish by measuring 1.5ml of each extract and applied to the fish with the aid of a 2ml syringe while the control consisted of introducing the beetles into the fish sample without applying any oil. The treatments were in three replicates and the experiment was conducted at room temperature of 27-30°C, photo period (13L: 11D) and relative humidity (60% ± 5). The experiment was carried out on the laboratory shelf for 30 days.

The fish weight loss and the number of live adult beetles were measured daily for the first 14 days and on the 30th day. The fish weight loss was calculated as the difference between the initial weight and final weight of the fish at the end of the experiment. The data were subjected to analysis of variance (ANOVA) and Duncan's Multiple Range Test was used for the mean separation at 5% probability level.

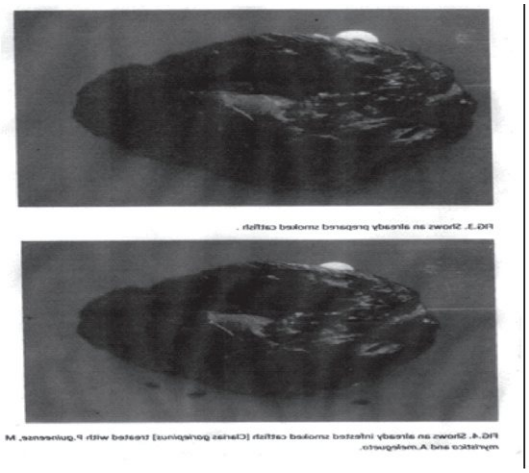
Results

The initial and final weights of smoked catfish (*Clarias gariepinus*) and change in weights (%) caused by *Desmestesmaculatus* infestation under the treatment of different plant oil extracts are shown in Table 1. The initial weight of the fish was 164.73 g in the control and varied between 177.97 to 195.33 g in fish treated with the oil extracts. The final weight of fish in the control was 99.37 g while fish treated with the oil extracts varied between 180.20 and 196.47 g such that the change in weight was negative in the control but positive in the other treatments. The weight change was least in *Piper guineense* and *Aframomummeleguetaa*t 0.385% and 0.578% respectively while *Monodoramyristica* had 1.253% compared to -39.68% in the control treatment (Table 1). The percentage adult mortality of *Dermestesmaculatus* subjected to different plant oil extracts is shown in Table 2 at the end of four weeks. Also, Table 3 presents the effect of test oil extracts on adult mortality of *D. maculatus*

Table 1: Effect of plant oil extracts on weight change of smoked catfish (*Clarias gariepinus*) infested by *Dermestesmaculatus*

Mean	*Control*	*A. melegueta*	*M. myristica*	*P. guineense*
Initial Weight	164.73±6.54	195.33±7.39	177.96±6.962	189.700±11.24
Final Weight	99.367±6.23	196.467±7.89	180.20±10.66	190.43±9.53
Weight change	-65.36	+1.13	+2.23	+0.73
% Weight change	-39.68	0.578	1.253	0.385

Each value is a mean of triplicate samples [±].



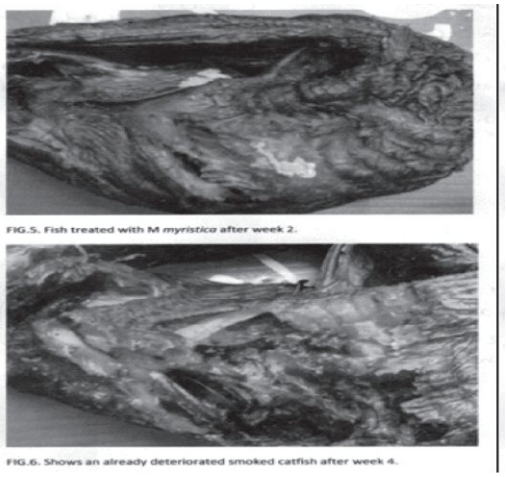


FIG.5. Fish treated with M *myristica* after week 2.

FIG.6. Shows an already deteriorated smoked catfish after week 4.

Table 2: Effect of plant oil extracts on adult mortality of *Dermestesmaculatus*

	Control	*A. melegueta*	*M. myristica*	*P. guineense*
Mean no of adult	10.00	10.00	10.00	10.00
% adult mortality	16.67	100.00	100.00	100.00

Each value is a mean of triplicate sample

In the experiment, it was observed that the mortality rate of the insects introduced in the fish treated with *Piper guineense* was quick because all the insects in the cultured jars died within the first week of experiment, while the insects in the fish treated with *M. myristica* died within two weeks of the experiment and insects in the fish treated with *A. melegueta* died during the fourth week of the experiment. This showed that the extracts from *P. guineense* and *M. myristica* are more effective than that from *A. melegueta*. In the control, it was also observed that 6 insects died naturally

Table 3: Effect of test oil extracts on adult mortality of *Dermestesmaculatus*

Plant Extract	No of Adult Introduced	Mean No. of Adult	No of Dead Adult	% of Mortality
A1	10	10.00	10	100.00
A2	10	10.00	10	100.00
A3	10	10.00	10	100.00
M1	10	10.00	10	100.00
M2	10	10.00	10	100.00
M3	10	10.00	10	100.00
P1	10	10.00	10	100.00
P2	10	10.00	10	100.00
P3	10	10.00	10	100.00
C1	10	10.00	3	30.00
C2	10	10.00	2	20.00
C3	10	10.00	1	10.00

Discussion

Plant-derived pest control agents have been extracted for use for over a century. For example, nicotine, pyrethrin and rotenone have been used as insecticides while several medicinal plants and spices have also been used as pest control agents (Lale, 1995). In the attempts to screen some selected indigenous plants as stored product pest control agents, the use of extracts was generally more potent than the dry powder for the control of *Dermestesmaculatus*. The potential insecticidal effect of vegetable oil extract has been demonstrated (Odeyemi*et al*., 2000). The activity of plant oils might be due to their repellency, chemical toxicity or physical properties rendering change in surface tension within the egg (Bhaduri *et al*., 1990). The thin oil layer restricts oxygen supply to insects causing interferences with normal respiration and so results in suffocation.

From this study, the mortality rate of the beetles in the fish treated with *Piper guineense* was 100% within the first week of the experiment while 100% mortality was observed in the second week with *Monodoramyristica* but the beetles in the fish treated with *Aframomummelegueta* recorded 100% mortality in the fourth week. This performance of *Piper guineenese* is a further proof that plants belonging to the family Piperaceae possess some form of insecticidal activity (Adedire and Lajide, 1999).It has been noted that *Piper guineense* and *Monodoramyristica* possess contact toxicity, fumigant, ovi position inhibition, ovicidal and larvicidal activities (Adedire and Lajide, 1999).Ofuya*et al*.(1992) observed that the crude extract of *Monodoramyristica* has ovicidal, larvicidal and anti-ovipositional effect on the cowpea bruchid (*Callosobruchusmaculatus*). The active ingredient in *Piper guineense* has been attributed to the presence of chavicine, and piperine(Lale, 1995). *Piper guineense* and

Monodoramyristica extracts have hot peppery taste and pungent smell and may act through contact with the insects.

The initial and final weights of fish in the control treatment showed that the loss of weight was substantial. This is because of the high level of insect infestation. The insects had destroyed the fish by feeding on it thereby reducing the weight and subsequently the quality. On the other hand, there was a slight increase in the weights of the fish treated with oil extracts of *Aframomummelegueta*, *Monodoramyristica* and *Piper guineense*. This was due to moisture absorption through the exposed muslin cloth over the jar.

The results show that *Piper guineense* and *Aframomummelegueta* were the most effective against the adult of *Dermestesmaculatus* and prevent fish damages as indicated by the low change in weight while *Monodoramyristica* was least effective. Earlier reports show that the insecticidal activity of essential oils varies in relation to the stage of development of the insect, the species and the plant origin of the essential oil (Tunc*et al*., 2000; Negahban*et al*., 2007). Although *Monodoramyristica* attained 100% mortality in the second week of the experiment, the moisture absorption was higher than with *Aframomummelegueta* which attained 100% insect mortality in the fourth week. Theresults obtained from administering these oil extracts *suggest* an alternative method of environment-friendly pest control of smoked catfish.

Conclusion and Recommendation

The results from this study suggest the possibility of using the oil extracts of *Piper guineense* and *Monodoramyristica* as bio-degradable insecticide against *Dermestesmaculatus* thereby reducing the damage caused by the beetle to smoke-dried fish. The oil extracts of *Piper guineense* and *Monodoramyristica* can provide new materials to be used in bio-rational management of insect pests. It is interesting to note that both *Piper guineense* and *Monodoramyristica* are normally used as spices in food preparations. Hence, the use of these plant materials guarantees safety to fish consumers and simultaneously provides insecticidal activity against *Dermestesmaculatus* due to its biodegradable properties. The use of plant extractives from *Piper guineense*, *Monodoramyristica* and *Aframomummelegueta* could be a plausible means to control insect pest attack of smoked fish and so reduce wastage and damage to fish during storage. The optimum concentration and active ingredients of these plant seeds which are toxic to the various stages of insect development and control in *Dermestesmaculatus* need further investigation.

The use of oil extracts against beetle infestation of smoked-dried catfish may be most acceptable considering the ready availability of the seeds, relatively cheap price and the safety for human consumption compared to the synthetic insecticides. *Piper guineense,* in particular, is recommended for its piperine content whose effect is felt almost immediately after application and so restricts the deterioration of smoked-dried fish and minimizes moisture absorption which is the major problem of smoked preserved fish. In the absence of *Piper guineense, Monodoramyristica* should be used to mitigate deterioration due to the beetle infestation in storage while the weight gain was minimal and would not support bacterial infestation. Thus, smoke-dried fish would be kept for longer period of time as the two oil extracts would guarantee longer shelf-life.

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