# Comparative evaluation of Black seed, *Nigella sativa* powder and Pestox (synthetic pyrethroid) as a suppressant of larval and pupal development of Hide beetle, *Demestes maculatus* in stockfish.

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## Abstract

Black seed, *Nigella sativa* powder and Pestox, a pyrethroid insecticide in powdered formulation were evaluated for the protection of stockfish against hide beetle, *Demestes maculatus* infestation. In the laboratory, both materials were measured at different concentrations ($0.4_g$, $0.8_g$, $1.2_g$, $1.6_g$ and $2.0_g$ per $40_g$). All treatment levels were replicated four times. Male and female of adult *D. maculatus* reared in the laboratory on stockfish were introduced into experimental containers containing the treated stockfish. Mortality and survival levels of treated *D. maculatus* were examined. Results recorded 100% survival of insects treated with black seed in all concentrations, 5days after treatment. Those treated with pestox recorded 50-80% survival (20-50% mortality) within the same period in all concentration levels. Progeny emergence (egg, larvae and pupae) from black seed treated pairs at all levels of treatment was significant ($p< 0.05$). However, larval and pupal numbers compared to normal fecundity of *D. maculatus* was reduced ($p< 0.05$). There was no progeny and no survival in the chemically treated set at all levels from the $5^{th}$ day after treatment. All doses were lethal to the test insect. It is obvious that the chemical, pestox has a better significant protective ability in stockfish against hide beetle infestation compared with the black seed. The dominant compounds found in black seed are known to have significant health benefits to man and no side effects known yet. Considering the results obtained in progeny emergence from the treated adults, it can be inferred that considering higher dosages of *N. sativa* for subsequent trials might not be out of place instead of the use of chemical insecticide (pestox) for the protection of stockfish against *D. maculatus* infestation for safer human consumption of stockfish.

**Keywords**: black seeds, hide beetle, pestox, suppressant, stockfish

## Introduction

Black seed is an annual plant of the ranunculus family, grown for its pungent seeds, which are used as a spice and in herbal medicine (Ali & Blunden, 2003). The black cumin plant is found in southwestern Asia and parts of the Mediterranean and Africa, where it has a long history of use in diverse culinary and medicinal traditions (Hassanien *et al*., 2014). Black cumin plants are hardy annuals that grow from 20 to 60 cm (8 to 24 inches) in height. The branched stems bear fine, deeply divided leaves, and the plant has a developed taproot. The pale blue or white flowers have five petals, numerous stamens, and five or six elongated fused carpels. The seeds have an aroma similar to fennel and have a pungent flavour somewhat similar to nutmeg, though the plant is not related to either. The seeds are commonly roasted and ground as a spice and are widely used in India, the Middle East, and parts of North Africa to season curries, rice, breads, and sweet confections. Black cumin is also important in traditional medicine in many

places and is an esteemed herbal remedy for a wide variety of ailments. The black triangular or pyramidal seeds are borne in a capsule with five or six segments, each of which terminates in an elongated projection. The plants can grow in a variety of soils and readily reseed, becoming weedy in some areas. Sometimes hailed as a panacea, black cumin seeds and their oil are widely used in traditional Islamic medicine and Avurveda to treat a variety of ailments. The seeds are believed to stimulate lactation and have been used for menstrual and postpartum problems. They are commonly used to treat intestinal worms and are said to relieve digestive troubles. The seeds and oil are also used for inflammation and are employed to reduce asthma and bronchitis symptoms and to treat rheumatoid arthritis. A substantial number of studies have been conducted that suggest the seeds may indeed have pharmacological potential. (Aljabre et al, 2009).

The seeds contain a variety of chemicals, but most of the pharmacological properties of black cumin are attributed to the presence of quinine compounds, of which thymoquinone is the most abundant (Kahsai, 2002). There is clinical evidence that the seeds have antimicrobial, antiparasitic, and antifungal properties, and some animal studies have demonstrated tumour suppression. In addition, there is evidence that black cumin may be effective against diabetes and hypertension and may be useful as an anti-inflammatory. Black seed oil is made from the seeds of the black cumin (*Nigella sativa*) plant. Black seed oil is also commonly called black cumin seed oil. Plant species in Nigeria used for insect pest control represent a number of plant families. The most frequently used include *Dennettia tripetala*, *Mondora myristica*, and *Xylopia aethiopicain* the family Annonaceae; *Piper guineense* in the family Piperaceae; and *Citrus aurantifolia*,*Citrus sinensis*, and *Zanthoxylulm zanthoxyloides* in the family Rutaceae. Few attempts, however, have been made to isolate the insecticidal or non-insecticidal constituents of the plants. Species investigated for insect pest control in stored products have generally been those used locally as culinary spices or in traditional medicine. In situations where both the plant and stored grains are consumed daily, the extended consumption of plants and the interactions between the plants and grain could prove toxic, making toxicological studies of materials necessary before any plants could be actively promoted for use as food protectant. The effects of the test materials on seed germination have been examined in some studies, but these tests were seldom done after 6 to 12 months of seed storage, the normal period of seed grain storage. In addition, investigations on use of materials for seed treatment have not studied the human toxicological properties of the plant materials, arguing that such tests are not essential.

Yet, materials registered for seed treatment need to undergo the toxicological evaluations as materials used in other applications because seeds are handled and, in times of shortage, may be eaten. Only a few investigations have done taste trials to ascertain if the stored product treated with an insecticidal plant is acceptable to consumers if prepared in the customary manner. Studies on the essential oils of *Dennettia tripetala*, *Piper guineense*, and *Citrus aurantifolia* indicate these oils are effective against *Callosobruchus maculatus* and *Sitophilus zeamais*for a period of six months if applied to the seed of cowpea and maize at 0.075 ml of essential oil/25 g or 0.1 ml essential oil/100 g. These essential oils also protected millet and sorghum against *Rhyzopertha dominica*, *Sitophilus oryzae*, and *Tribolium castaneum*and dried fish against *Dermestes maculatus*and *Necrobia rufipes* (Okonkwo & Ewete, 2000).

Okonkwo and Ewete (1998) used powdered seed of *Dennettia tripetala* (Pepperfruit) and observed that an admixture of 3 g/25 g maize achieved 100%mortality in *S. zeamais* in 24 h; this result showed that the plant material was more effective than pirimiphos-methyl dust and aluminium phosphide tablet used in storage for 4 months, causing 100% mortality in adult *S. zeamais* within 7 days. The same authors observed that using an admixture of 3 g/25 g cowpea achieved 100% mortality in adult *C. maculatus* in 24h which was more effective in achieving 100% mortality in adult bruchids in 6 months, 7 days post-treatment than pirimiphos-

methyl dust and aluminium phosphide tablet (10% mortality). Stock fish, also known as okporoko, is an unsalted fish. It is typically made from cod, dried by cold air and wind on wooden racks (Thorarinsdottir *et al.*,2004). Nigeria is one of the highest consumers of stock fish in Africa. It is used in many West African dishes and soups. Cod/Stockfish are saltwater whitefish that turns creamy after they are dried. All parts of the fish are essential and edible in many cultures such that no part is wasted but they are rather used for different things (Abdel-Fattah et al, 2000). The heads are sold separately after they are dried, while the roe, liver and tongues and some other parts are sold fresh or canned. In Africa, the boneless creamy fleshes are sold at a very high cost, while some parts of the backbones or cutlets are sold separately at a lower price for various delicacies. Sea foods are best meat for daily consumption from time to time. They are important foods since they provide high quality protein which can be gotten from dietary supplement, dairy products and other super foods. They help greatly in building healthy immune system in man, support blood circulation system, assist in proper weight management, and provide other nutritional functions needed by the body. Such sea foods include, crabs, periwinkles, shrimps, oyster, crayfish, shark, mackerel, haddock, salmon, tuna, sardine tilapia and codfish among other sea foods.

Stockfish is high in omega-3 fatty acids, which is known to be beneficial for proper brain and nervous system function. Codfish has no fibre and carbohydrate content and the fresh codfish has about 72kcal of energy per100g. (Berr *et al.*, 2009). A diet rich in omega 3 fats found in cold water fishlike Cod is known to reduce blood pressure and prevents excess fatty substances in the arteries and in turn lower cholesterol level in the blood. (Berr *et al*., 2009). Feeding by the larvae and adults of *Dermestes spp*. causes considerable quantitative loss of dried cured fish, and also leads to fragmentation. Quality loss may also be caused by the presence of insect bodies and cast skins. The extent and value of quantitative losses caused to dried fish by *Dermestes spp*. have been assessed by various investigators, and estimates range from negligible up to 50% weight loss, depending on length of storage, salt content, moisture content, climatic conditions, and general hygiene during processing and storage. Weight losses due to fragmentation have also been investigated, but the contribution of *Dermestes spp*. to this process has not been separately assessed. These loss estimates have been reviewed by FAO (1981). Additional costs may arise from damage caused to wooden drying racks and store structures by the mature larvae.

The life cycle of *Dermestes maculatus* on either a carcass in dry-decay or in stored animal products requires approximately five to seven weeks completing under optimum conditions. The adults consume the remains of the carcass or the animal product (*Melanie & Mark, 2007*). Pheromones, secreted by males through a gland on the base of the abdomen, are used to attract females. Males and females will mate multiple times and the female will lay eggs within 24 hours of the first mating (Jones *et al*., 2006). Eggs are laid in cracks of the matter on which they are feeding. Females are capable of laying eggs continuously (Jones *et al.*, 2006). *Dermestes maculatus* eggs are typically laid in batches of 3 to 20. The number of eggs a single female can lay over a lifetime varies greatly, ranging from 198 to 845.

The bodies of the larvae are covered in rows of hairs of different lengths, called setae. The underside of the abdomen is typically yellowish-brown while the dorsal surface is typically dark brown, usually with a central yellow line. Two long horn-like protrusions are located on the upper surface of the last segment, partially hidden by surrounding hairs. The protrusions, called urogomphi, curve upward and away from the tip of the abdomen. This distinguishes the larvae from larvae of *Dermestes lardarius*, which has the urogomphi crving downward toward the tip of the abdomen (Brianna & Phillip, 2010). Larvae will pass through five to 11 instars, the number of instars increasing with unfavorable conditions. During the last 10 days of the final instar, the larvae will seek out a place to pupate, typically within the meat or a non-food substance such as wood. Exposed pupae that have failed to find a suitable pupal chamber are

often cannibalized by larvae. Larvae without a suitable place to pupate can delay pupation by over 20 days, but at the cost of lower adult body mass and increased risk to fatal disease (*Melanie & Mark, 2007*). Survivorship for individuals is the highest between 25°C and 30°C (Richardson and Goff, 2001). The last larval skin will usually provide a protective covering for the pupa. The end of the pupal chamber toward the surface can be closed by either debris from the substance which the larvae bore into or from the last larval skin. The pupae are an oval shape, usually smaller than the larvae, and do not have the many long hair-like projections (Kulshrestha and Satpathy, 2001). Currently, the insect pest control practices rely heavily on application of pesticides. This has led to several problems such as health hazards especially to the consumers. *D. maculatus* has also developed some resistance to certain pesticides, which has led to several losses economically. Therefore, there is need to study and ascertain, certain natural protectant of stockfish against *D.maculatus* infestation. This study examines the effect of *N. sativa* on the development of eggs and larvae from adult on treated stockfish.

## Materials and Method

**Stock Culture 1:** Infested head region of stockfishes was bought from Rumuokoro Market in Obiokpor Local Government Area in Port Harcourt, Rivers State. These were placed in plastic containers. The lids were cut open to allow for sufficient aeration. Netting material with fine mesh size was used to cover the contents. For new generations of *D. maculatus* for the experiment, these were kept in the laboratory at ambient temperature and relative humidity.

**Stock culture 2:** From the Laboratory culture of the infested stockfish, larvae of *Demestes maculatus* were picked into test tubes containing pieces of stockfish for further development to pupae and finally to adult stages.

**Black seed powder preparation:** Black cumin (*Nigella sativa*) was purchased from India. The Black seeds were grounded in the Laboratory into a powder. It was sieved with a standard test sieve of 250mic, to get a fine powder particle and was stored in a specimen bottle. This was done prior to the experimental period in order to achieve the desire results.

**Experimental set-up:** The fleshy parts of the stockfish were cut into smaller pieces and the same weight of 40g were weighed for all concentrations and put in plastic containers. It was treated with different concentration of the Black cumin powder, and the chemical (pestox). The treatment levels were ($0.4_g$, $0.8_g$, 1. $2_g$, $1.6_g$, $2.0_g$.), all were replicated 4 times. Male and female adult of *D. maculatus* were introduced into the different concentrations for observations.

## Data Analysis

Data collected were subjected to Mean, Standard Deviation, Percentages, and Chi-Square.

## Results

**Table** 1: Mean comparison of larva emergence and pupa development among the concentration levels

| Concentration Level | Mean±SD | |
|---|---|---|
| | Larva Emergence | Pupa Development |
| Control | 76.67±20.96 | 9.17±11.29 |
| 0.4g/40kg | 50.67±55.14 | 3.83±4.31 |
| 0.8g/40kg | 42.00±46.03 | 6.17±6.82 |
| 1.2g/40kg | 39.67±42.98 | 5.00±5.62 |
| 1.6g/40kg | 51.17±56.12 | 6.33±7.00 |
| 2.0g/40kg | 41.83±46.79 | 6.00±6.60 |

Table 1 shows the mean number of *D. maculatus* larvae and pupal development on stockfish treated with *N. sativa* as a biocontrol agent. The result showed higher number of *D.maculatus* larvae emergence in control followed by smoked fish treated with 1.6 g *N.*

*sativa* though not significantly different from those treated with 0.4 g. the least larval development was recorded on stockfish treated with 1.2 g. similar results were obtained for pupal development (table 1).

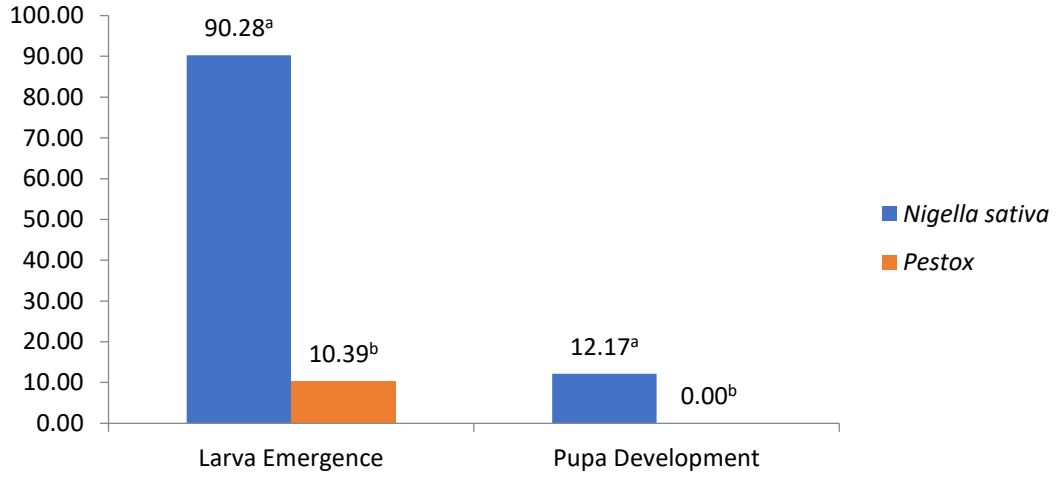



**Fig**.1: Mean comparison of larva emergence and pupa development between *Nigella sativa* and Pestox

From figure 1 when a comparative assessment of black seed (*N. sativa)* powder and pestox (synthetic pyrethroid) was determined on *D. maculatus* raise in stockfish in a laboratory to determine its suppressant ability on the immature stages of *D. maculatus*, the result showed that significantly higher larvae progenies were recorded on *N. sativa* treated stockfish than the pestox treated stockfish and no pupa emerged from the pestox treated stockfish.

**Table**: 2. Mean comparison of frass, weight loss and percentage damage among the concentration levels

| **Concentration Level** | **Mean±SD** | | |
|---|---|---|---|
| | **Frass** | **Weight Loss** | **Percentage Damage** |
| Control | 3.39±0.44 | 35.47±0.46 | 88.27±0.65 |
| 0.4g/40kg | 3.08±0.09 | 33.93±1.01 | 87.04±0.31 |
| 0.8g/40kg | 2.79±0.41 | 33.93±0.96 | 86.84±0.55 |
| 1.2g/40kg | 3.09±0.09 | 35.36±0.56 | 87.87±0.91 |
| 1.6g/40kg | 3.49±0.44 | 34.32±0.83 | 87.90±1.26 |
| 2.0g/40kg | 3.37±0.31 | 34.78±0.58 | 87.44±0.48 |

Table 2 shows the bioactivities of *D. maculatus* bred on stockfish treated with *N. sativa* and the result shows that higher frass was generated by *D. maculatus* bred stockfish treated with 1.6 g *N. sativa* although it was not statistically different from the other treatments while the least frass was generated on stockfish treated with *N. sativa* at 0.8 g. Weight due to the feeding activities of *D. maculatus* on stockfish treated with *N. sativa* was higher in control although not different from those treated with *N. sativa* at 1.6 and 2.0 g and the least was on *D. maculatus* bred on stockfish treated with *N.sativa* at 0.4 and 0.8 g. similar result was recorded on percentage damage attributed to *D. maculatus* on treated stockfish with *N. sativa*.

**Discussion**

The use of plant materials in the management of insects' infestation in store products has been widely reported (Aalonee et al., 2023; Okonkwo, 1998; Dales, 1996; Okonkwo and Okoye, 1996; Don-Pedro, 1996; Okonkwo and Ewete, 1998; Okonkwo and Ewete, 2000). Hence, the evaluation of the application of seed powder of *Nigella sativa* in the management of *Demestes maculatus* infesting stockfish studied in this research. The result of the concentrations of *Nigella sativa* used in this study was not significant in the management of *Demestes maculatus* within 34days of exposure. The result contrasts in effectiveness in other plant materials although not on stockfish. For instance; Okonkwo and Okoye in 1996 worked on powder seed of *Dennettia tripetala* (Pepper fruit) an admixture of $1.5_g/25_g$ dried *Clarias sp*. Fish and 100% mortality in adult *Demestes maculatus* was observed in 7days and *Necrobia rufipes* (Degeer) in 14days. An admixture of $3_g/25_g$ fish completely prevented oviposition and emergence of *D. maculatus* and reduced the member of *N. rufipes* $F_1$ adult; similar result was obtained for P*iper guineense*, however, *D. tripetala* was more toxic than *P. guineense.*

The result indicating a higher level of larvae by *N. sativa* over the progeny emergence may be in agreement with the findings of *Melanie and Mark (2007)* who find out that the number of instars increases with unfavourable conditions and that can make a larva to pass through 5 to 11 instars. Contalamessa (1993) and Grdisa & Grsi (2013) reported that pestox whose active ingredient is cypermethrin is an insecticide that is moderately toxic by dermal absorption or ingestion. Symptoms ranges from loss of bladder control incordination, seizures, burning sensation, adverse effect on the central nervous system and death is in agreement with the position held by this research work because there was no larval survivor nor papae emergence on those treated with pestox. It could also be that the fewer number of papae reported in this research may be as a result of not adhearing to the the findings of Richardson & Goff (2011) who reported that survivorship for individuals is the higher between $25^0$c and $30^0$c.

The frass, weight loss and percentage damage on the controls of *N. sativa* and pestox as well as on different concentration levels were relatively the same indicating that they are both effective in the treatment of *D. maculatus*. As posited by Aalonee et al (2023), the above may be as a result of high phenol and alkaloid chemical compounds isolated from *N. sativa*
Conclusively, plant materials such as *N. sativa* should be used in the management of store product pest since they are both toxic free, economical and easy to use. Application of chemical should be discouraged.

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