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DOSE-DEPENDENT EFFECTS OF WOOD ASH, KAOLIN CLAY AND CLOVE (*Syzgium aromaticum*) POWDERS ON POST-HARVEST LOSSES AND QUALITY OF STORED COWPEA.

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Abstract

The cowpea weevil (*Callosobruchus maculatus*) is a major pest of cowpea in storage and is responsible for serious significant post-harvest losses in cowpea globally and this is a major challenge to cowpea storage in tropical regions. This study evaluated the dose-dependent effects of wood ash, kaolin clay, and clove powders on post-harvest losses and quality of stored cowpea. The experiment consisted of the three treatment materials applied at 3 dosage levels (2g, 4g and 6g per 100g of the cowpea seeds) alongside untreated control given a total of 10 treatments. Parameters assessed included: adult mortality, adult emergence, ovipostion, grain damage and weight loss. The experiment was arranged in completely randomized design (CRD) and was replicated 3 times. A total of 10 weevils per treatment was introduced into the containers containing 100g of uninfested seeds. Results from the study revealed that all the treatments reduced the weevil infestation and postharvest losses compared to the untreated control. Clove treatment gave the highest mean mortality level of 2.1, 2.5 and 3.3 at (2, 4, and 6g) respectively. This was followed by clay at 6g with a mean mortality level of 2.1 while ash gave a lower number of mortality of 1.6, 0.8 and 1.3 mortality at (2, 4 and 6g) respectively and these were all significantly different at ($p \leq 0.05$) from the control with the least mean mortality level of 0.1. Generally the seeds treated with clove recorded the least no of eggs, least number of adult emergence, least number of damaged seeds and the least percentage weight loss compared to other treatments and these were significantly different from the control with the highest number of eggs, emergence, damaged seeds and percentage weight loss. Conclusively, the three powders tested showed repellent and toxic effects on the adult beetles, with mortality rates and oviposition increasing in association with higher concentrations. Clove emerged as the most effective treatment across all parameters tested, underscoring its potential for comprehensive pest management strategies and therefore is recommended as an alternative to effective management of the cowpea weevil.. **Keywords: Dose-dependent, Kaolin clay, Clove, Wood ash, Post-harvest, Sustainable, Cowpea**

Introduction: Cowpea (*Vigna unguiculata* (L.) Walp) is a popular leguminous crop in Africa particularly in Nigeria, where it serves as a major source of affordable protein, vitamins and minerals. The crop is widely consumed due to its high nutritional value and its ability to survive under different environmental conditions.. The grain is rich in protein and other micronutrients necessary for healthy living (Udo, 2020). Many societies endowed with cowpea have evolved different ways of utilizing the grain for food. In addition to its importance in human nutrition, cowpea contributes significantly to soil fertility improvement through nitrogen fixation, making it an important component of sustainable farming systems (Boukar et al, 2019). Nigeria produces more than two million tons annually (Adeola *et al* ., 2019) which represents 58% of the total world cowpea production annually and yet a substantial amount of harvested grains is lost during storage due to insect pests attack and poor post-harvest handling practices (FAO, 2022). Cowpea grains like other agricultural produce in the tropics are faced with storage problems. Ajeigbe *et al* ., (2020) noted that, the longer cowpea seeds remain in storage, the more susceptible they are to infestation by rodents, insects etc. Infestation of stored cowpea is of great concern to farmers because it results not only in food shortage for human consumption but also in scarcity of planting materials. Insect pests are by far the greatest threat to stored cowpea seeds. One of the most destructive storage pests of cowpea is the cowpea bruchid (*Callosobruchus maculatus* Fabricius). The insect causes serious damage during storage by laying eggs on stored seeds after which the emerging larvae penetrates and feed inside the grains. Infested seeds become perforated, discoloured and reduced in nutritional and market quality. Severe infestation may lead to drastic weight loss, reduced seed viability and total grain deterioration within a few months of storage (Adebayo and Ibikunle, 2021). The Nigerian Institute of Food Science and Technology (NIFEST) estimated that about 40% of cowpea seeds harvested annually is lost as a result of bruchid attack during storage(Mohammad *et al* ., 2013). In Nigeria and other developing countries of the world, ignorance and lack of adequate storage methods have compounded the situation. Carlos (2019) also stated that unprotected cowpea grains are often completely consumed by bruchids in the first 10 to 12 months of storage. *C. maculatus* multiplies rapidly in storage, giving rise to a new generation every month in grains at temperature of 30ºC to 35ºC and 70 % to 90 % relative humidity (RH) (Dales, 2021). Grains attacked have reductions in weight, nutritional compositions, viability and virtually unfit for human consumption (Sharah, 2019). These losses pose serious threat to food security, especially among small holder farmers who depend on stored cowpea for household consumption and income generation. To minmise these storage losses, synthetic insecticides are widely used because of their rapid action and effectiveness against storage pests. However, over dependence on synthetic insecticides has generated growing concerns due to pesticide residues in food, environmental contamination, increasing insect resistance and health risks to humans and livestock (Nboyine et al, 2020). These challenges have intensified the need for safer, eco-friendly and affordable alternatives for protecting stored grains such as the use of botanical materials. Botanical materials are increasingly being explored as sustainable alternatives for storage pest management because of their insecticidal and protective properties against storage pests. Wood ash has been traditionally used by rural farmers because it causes desiccation and restricts insects movement on grain surface. Kaolin clay acts by forming a protective coating around grains, thereby disrupting insect feeding, egg laying and their survival. On the other hand clove powder contains biologically active compounds such as eugenol which possess strong insecticidal, repellent and fumigant properties against storage pests (Mossa, 2018, Udo *et a*l 2021). Udo *et a*l (2021) also reported that botanical powders significantly reduced bruchid infestation and preserved grain quality during storage. Similarly, Nboyine *et a*l (2020) observed that locally available plant materials provided effective protection against storage insects while minimizing environmental and health risk associated with synthetic pesticides. Despite these findings, information regarding the dose-dependent effects of wood ash, clove and kaolin clay powder on stored cowpea remains limited, particularly under local storage conditions. The effectiveness of some of these protective materials is most times influenced by their rate of application. Low doses may not provide sufficient protection against insect pest while excessive doses may affect grain appearance and consumer acceptability. Determining the appropriate dosage capable of minimizing post-harvest losses while at the same time maintaining grain quality is therefore essential for developing sustainable storage technologies for small holder farmers hence, the aim of this study was to evaluate the dose dependent effects of wood ash clove and kaolin clay on post-harvest losses and quality of cowpea in storage. The study seeks to provide safer, cheaper and environmentally friendly alternatives to synthetic insecticide for sustainable cowpea storage management. Post-harvest losses caused by insect infestation remains a major challenge in stored cowpea production in Nigeria and this has led to reduction in the quality of cowpea grain, lower market value and a very high economic losses for both farmers and consumers. Although synthetic insecticides are commonly used for grain protection, their continuous application has raised concerns regarding pesticide residues, insect resistance, environmental pollution, and health risks to consumers. Botanical and mineral materials such as wood ash, clove and kaolin clay powders have shown potentials as safer alternatives, but limited information exists on their dose-dependent effectiveness in preserving cowpea during storage. More so the optimal application rates required to achieve effective insect control while maintaining grain quality have not been adequately established under local storage conditions. Therefore this study seeks to evaluate the dose-dependent effects of wood ash, clove and kaolin clay on post-harvest losses and quality of stored cowpea. Cowpea is one of the most important food crops consumed in Nigeria because of its rich protein content and affordabilty for many households. It is a key food and income source for many **Materials and Methods: Research location:** The study was carried out at the plant Health Management Laboratory of Michael Okpara University of Agriculture, Umudike in the year 2024. Umudike lies on latitude 05˚ 29¹ N and longitude 07° 33¹ E in the rainforest area of South-East agricultural zone of Nigeria).

Insect Culture: Adult *callosobrochus maculatus* were obtained from infested cowpea grains and cultured under laboratory condition with a temperature of 25 – 30°C and relative humidity of 65-75% on uninfected cowpea purchased from the market. This was stored in a transparent bucket, covered with cheese cloth to allow for aeration and allowed to stay until beetles began to emerge and were allowed to multiply.

Sources of Materials Used: Clay and clove were bought from a local market within the environment while wood ash was collected from a completely combusted hardwood from a garri manufacturing industry in the area Uninfested cowpea variety (Ife brown) was bought from the local market, cleaned and was kept in a deep freezer for 48 hours. This was to sterilize and keep it safe from any unwanted contamination

Preparation of Organic Materials Used: The wood ash was sieved by passing it through a fine mesh sieve to remove unwanted particles. After this it was stored in airtight containers before it was used. The purchased cloves were cleaned and air dried under shade for 3 weeks. It was later ground into fine powder using a laboratory mill and was sieved by passing through a fine sieve to ensure uniform particle size. This was then stored in dark airtight containers to preserve its bioactive constituents, while the clay powder was crushed using a morter and pistle. It was sieved and oven sterilized at 105^0c for 4 hours to eliminate microorganism. It was also stored in airtight container prior to use.

Treatments: The following treatments were used for the experiment: a total of 10 treatments were used for the experiment which included: Ash 2g, 4g and 6g, Clove 2g, 4g and 6g, Clay 2g, 4g and 6g, and control

Treatment Pplication: 100g of clean uninfested cowpea grains were weighed into a containers. The plant extracts were added at the rate of 2g, 4g and 6g accordingly as it was labelled in the container and this was thoroughly mixed with the cowpea grains to ensure uniform coating. 10 adult male and female weevils were introduced into each container and this was covered with muslin cloth for proper aeration.

Data Collection and Experimental Methods

The following data were collected:

Adult mortality (%)
This was calculated by counting number of dead and live insects at 24, 48, and 72 hours using the formular: **Percentage mortality** = $\frac{\text{Total number of dead insect}}{\text{Total number of insects}} \times \frac{100}{1}$

Oviposion: This was done after 7 days by counting the number of eggs laid on 10 randomly selected cowpea grains using a hand lens and total number of eggs was recorded and the average was calculated and multiplied by the number deeds used.

C: Adult emergence: This was done by monitoring the containers daily after 2 weeks post-infestation. Newly emerged adults were counted and removed daily to avoid double counting the emerged adults. This was done using the formular

Percentage adult emergence = $\frac{\text{Total number of emerged adults}}{\text{Total number of eggs laid}} \times \frac{100}{1}$

D: Grain damage assessment: This was calculated by counting the numbers of damaged cowpea seeds from each treatment.
E: Percentage (%) weight loss : This was calculated using the Formular:

Percentage weight loss = Initial weight - Final Weight x <u>100</u>

Data Analysis: All data collected were subjected to analysis of variance in an open source environment using "R" version 4.2.2. HSD (Honestly Significant Difference) was used for the mean separation at 5 percent level of significance.
Table 1 shows the effect of Ash, Clay and Clove powders at different doses on the number of beetle mortality at 24, 48, and 72hrs. The result of the study showed that all treatments had some preservative effect on stored cowpea compared to control. However, clove powder consistently performed better across all application rates (2g, 4g and 6g) indicating a stronger protective effects against the pest. In mortality reduction, the seeds treated with clove gave the highest mean mortality level of 2.1, 2.5 and 3.33 at (2, 4, and 6g) respectively. This was followed by clay at 6g with a mean mortality level of 2.1 while ash gave the least number of mortality of 1.6, 0.80 and 1.33 mortality at (2, 4 and 6g) respectively among all the treatments and these were all significantly different at ($p \leq 0.05$) from the control with the least mean mortality level of 0.11. Moratility was observed to be increasing as the dosage and hours keeps increasing in each treatment.

Table 2 shows the effect of ash, clove and clay powders on oviposition, adult emergence, damaged seed and percentage weight loss of the treated and untreated seed. The result showed a significant reduction in oviposition in all treated cowpea seeds compared to the untreated control. Among the treatments cloves consistently recorded the lowest number of eggs laid at the different application rates (2.00. 0.3 and 0.00) at 2, 4 and 6g respectively indicating strong deterrent effects on female weevils. Oviposition decreases as the dosage increases with highest reduction observed at 6g (0.00) while the control recorded the highest level of oviposition (56.00). Adult emergence followed a similar trend as oviposition. Clove treated seeds had the lowest number of emerged adults across all doses indicating that clove not only reduced egg laying, but also interfered with the development of immature stages. This suggests possible ovicidal and lavicidal properties. Clay treatment moderately suppressed adult emergence while ash showed minimal suppression however, both are significantly different at ($p \leq 0.05$) from the control. The control treatment recorded the highest adult emergence reflecting high survival and successful development of the pest in untreated plots.

The number of damaged seeds was significantly lower in treated samples compared to the control. Seeds treated with clove resulted in the least seed damage (1.30, 0.00 and 0.00) at 2, 4 and 6g respectively, indicating effective protection of cowpea grains against feeding activities of the weevil. Clay also provided a very good protection reducing seed damage (2.70 , 0.70. and 0.30) at 2, 4, and 6g respectively while ash have the least protective effects among the treatments (6.0, 2.3 and 2.0) at 2, 4 and 6g respectively while untreated seed (control) generally recorded the highest number of damaged seeds (19.00). The lowest weight loss was observed at the highest dosage (6g) of clove (0.10g) followed by 4 and 2g (2.10 and 2.50g). Clay treatment moderately reduced weight loss while ash showed the least reduction among the treatments while the control recorded the highest (18.50g). All the treated plots recorded significant lower oviposition, lower adult emergence, lower seed damage and lower weight loss at ($P \leq 0.05$) when compared with the control.

Discussion: From the results obtained in this study, all the treatments applied were able to reduce infestation in stored cowpea when compared to the untreated control. However clove stood out as the most effective treatment across all parameters measured especially when applied at the rate of 6g. This performance can be linked to the presence of eugenol which is the major active compound in clove. Eugenol is well known for its strong insecticidal, repellent and fumigant properties. It affects the nervous system of insects, reduces their ability to lay eggs and can even lead to death at higher concentration as was observed in this experiment. This explains why clove treated seeds recorded the lowest oviposition, reduced adult emergence, minimal seed damage and the least weight loss in this study. This finding is in agreement with Isman (2006) who reported that botanical insecticides can disrupt feeding, reproduction and development of insects. It also agrees with the result of Butu *et al* (2020) who reported on the effectiveness of bio-pesticides in reducing the incidence of insect pests and increasing yield during field application. Ash also showed some level of effectiveness, although not as effective as clove. Wood ash also performed better than the control in reducing the post-harvest losses of the cowpea particularly at high doses. The effectiveness of wood ash may be associated with its abrasive and desiccating properties, which disrupt insect movement and cause loss of body moisture, eventually leading to insect mortality. In addition ash particles may block insect spiracles and interfere with respiration. The result supports the findings of Nboyine *et al* (2020) who reported that inert materials can provide physical protection against storage pests by creating unfavourable conditions for insect survival and protection. This also agrees with the findings of Udo (2011) who reported that wood ash can reduce infestation in stored cowpea. Similarly, Kaolin clay reduced insect infestation and grain damage. The protective effect of kaolin clay may be attributed to its ability to coat grain surfaces and create a physical barrier that discourages feeding and oviposition by storage insects. The fine particles may also interfere with insect mobility and reduce their ability to infest grains successfully.This finding supports the report of Mossa (2018), who observed that essential oils and plant derived powders containing bio-active compounds significantly suppressed storage insect pests through repellence, toxicity and inhibition of insect development. Similar observations have been made in studies with inert materials where they provided some level of protection but are generally less effective than plant derived insecticides. The effectiveness of all treatments increased as the dosage increased from 2g to 6g. The findings of this study also supports the growing global interest in integrating botanical and mineral-based materials into post-harvest pest management systems. As concerns over pesticide residues, insect resistance and environmental pollution continues to increase, the adoption of safer and affordable storage technologies becomes increasingly important for attaining sustainable food security. In summary, this study demonstrated that wood ash, clove and kaolin clay powders possess considerable potential for protecting stored cowpea against post-harvest insect damage. Their effectiveness was strongly influenced by dosage with higher application rates providing better protection and improved grain quality preservation.These materials therefore represents alternatives for sustainable cowpea storage management, Particularly in rural farming communities where access to synthetic chemical may be limited.

Conclusion: This study demonstrated that the application of these locally available materials significantly reduced infestation and damage of stored cowpea when compared to the untreated control. All treatments showed some level of protective effects, however their efficacy varied considerably depending on dosage. Clove powders proved to be best option among the materials tested. Based on these findings, the use of clove powder and kaolin clay is strongly recommended for the protection of stored cowpea against cowpea weevil infestation. Farmers are encouraged to apply clove to achieve better protection during storage. Clove even at the lowest rate of 2g still proved very effective in protecting cowpea from weevil attack which shows its strong repellent potential against the weevils. Farmers and grain handlers are encouraged to apply clove to achieve better protection during storage hence it is affordable and at the same time accessible and also very friendly to humans and the environment. Promoting the use of these organic materials can help reduce dependence on synthetic insecticides, lower production costs and support safer and more sustainable agricultural practices. Further research should be carried out to explore longer storage duration to assess how long the treatment remain effective.

Recommendation: Based on the findings this study, clove powder and clay especially at higher doses are recommended as safe, affordable and eco-friendly alternatives to synthetic insecticides for protecting stored cowpea against post-harvest insect damage. Higher application doses were more effective in reducing infestation and preserving grain quality. Farmers should therefore adopt the appropriate dosages such as the equivalent of 6g to 100g of cowpea seeds for sustainable cowpea storage.

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Results

Table 1: Effect of treatment on mortality rate of cowpea Beetle at 24, 48 and 72hrs

	Mortality Rate			
Treatments	**24HRS**	**48HRS**	**72HRS**	**Mean No of Mortality**
ASH2g	0.00 ^{b}	0.33 ^{bc}	1.33 ^{ab}	1.60 ^{cd}
ASH4g	0.00 ^{b}	0.70 ^{bc}	1.70 ^{ab}	0.80 ^{cd}
ASH6g	0.33 ^{ab}	2.0 ^{0bc}	1.70 ^{ab}	1.33 ^{bcd}
CLAY2g	0.33 ^{ab}	1.33 ^{bc}	1.70 ^{ab}	1.10 ^{bcd}
CLAY4g	0.33 ^{ab}	1.70 ^{bc}	2.33 ^{ab}	1.40 ^{abc}
CLAY6g	1.70 ^{ab}	2.00 ^{bc}	2.33 ^{ab}	2.1 ^{abc}
CLOVE2g	1.70 ^{ab}	2.00 ^{bc}	2.70 ^{ab}	2.10 ^{abc}
CLOVE4g	2.00 ^{ab}	2.33 ^{b}	3.33 ^{a}	2.50 ^{ab}
CLOVE6g	4.00 ^{a}	4.70 ^{a}	1.33 ^{ab}	3.33 ^{a}
CONTROL	0.00 ^{b}	0.00 ^{c}	0.33 ^{b}	0.11 ^{d}
HSD	4.00	2.00	2.60	4.40
CV (%)	110.8	41.60	46.30	31.40

HSD: Honestly Significant Difference; CV: Coefficient of Variation. Note: mean values with the same alphabets are not statistically significant ($p \leq 0.05$)

Table 2: Effect of treatment on the number of eggs, adult emergence, damaged seeds and weight loss

Treatments	**No. of Eggs**	**Percentage Adult Emergence**	**No. of Damaged Seed**	**%Weight Loss**
ASH2g	41.00 ^{b}	6.70 ^{a}	6.70 ^{ab}	15.10 ^{ab}
ASH4g	33.00 ^{b}	1.70 ^{b}	2.30 ^{bc}	13.20 ^{abc}
ASH6g	21.00 ^{c}	0.70 ^{b}	2.0 ^{c}	10.70 ^{bcd}

CLAY2g	7.30 [d]	0.70 [b]	2.70 [bc]	8.00 [cde]
CLAY4g	5.70 [d]	0.00 [b]	0.70 [c]	8.70 [cd]
CLAY6g	2.00 [d]	0.00 [b]	0.30 [c]	2.70 [ef]
CLOVE2g	2.00 [d]	0.00 [b]	1.30 [c]	2.50 [def]
CLOVE4g	0.30 [d]	0.00 [b]	0.00 [c]	2.10 [ef]
CLOVE6g	0.00 [d]	0.00 [b]	0.00 [c]	0.10 [f]
CONTROL	56.30 [a]	18.70 [a]	19.30 [a]	18.40[a]
HSD	9.4	3.1	4.6	5.9
CV(%)	19.3	58.9	62.8	24.4

HSD: Honestly Significant Difference; CV: Coefficient of Variation. Note: mean values with the same alphabets are not statistically significant ($p \leq 0.05$)