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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING AND INSECTICIDAL ACTIVITY OF ZINGIBBER OFFICINALE, ALLIUM SATIVUM AND CURCUMA LONGA POWDERS AGAINST CALLOSOBRUCHUS MACULATUS .FAB. OF STORED COWPEA SEEDS

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Abstract

This study focused on the phytochemcial screening and insecticidal properties/activities of the powdered extracts from three different indigenous spice plants on cowpea weevil Callosobruchusmaculatus. The studied plant were Zingibberofficinale, Allumsativumand Curcumalonga.The plant powders contained the entire nutrient proximate tested such as dry matter, crude protein, crude fibre, ash, ether extract and nitrogen free extracts. The phytochemcial analysis of these extracts indicated the presence of some chemcial like alkaloids, saponins, cardiac glycosides, flavonoids, tannins and terpenoids all of which possess insecticidal properties. Tannin was absent in turmeric while terpenoid was absent in ginger and garlic. In the study,50 weevils were counted in a transparent rectangular cage and introduced to each 200guninfected cowpea in four replicated containers for each plant extract treatment.100g each of the three plant extracts were applied each to four different replicates. Data were subjected for analysis of variance (ANOVA) at 5% probability level and the percentage mortality, number of holes/punctures on the cowpea, and longevity of death of weevils for each treatments. The highest mortality of the weevil were observed in the ginger treatment while turmeric had the least mortality activity. The ginger powder reduces the mean population introduced (50) to 16.5 (mean number alive) of C. maculatus,when compared to garlic powder (24.8)and turmeric powder (32.4) respectively. The longevity in days of C. maculatusfor ginger and garlic effects were 7 days and turmeric was 14days after application. There wasno mortality recorded in the control experiment for the 42 days. Ginger powder gave the lowest mean number of holes on been seeds (16 ± 1.33)followed by garlic powder (41 ± 2.13) and turmeric powder (83 ± 3.04). The average weight loss was recorded high in control treatment (31.5 ± 1.87) and was significantly high than the bean seeds treated with turmeric powder (25.7 ± 1.67), garlic powder (15.0 ± 1.29) and ginger powder (6.0 ± 0.82) respectively. Therefore the powered extracts from these three research plants

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represent an alternative to chemical insecticides in preservation of stored cowpea.

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Introduction:-

The cowpea weevil, CallosobruchusmaculatusFab.Coleoptera: Chrysomelidae, is a major pest of economically important leguminous grains, such as cowpeas, lentils, green gram, and black gram (Diouf, 2011&Machacha, et al., 2012). It is a holometabolic insect with the egg and adult stage found on the grainand the larval and pupal stages living inside the grain. The larvae bore into the pulse grains and eat up the endosperms which become unsuitable for human consumption with reduced viability for replanting or for the production of sprouts (Diouf, 2011&Machacha, et al., 2012). They are important pests of pulse crops in Asia and Africa under storage conditions In India the insect breeds freely from March to November and hibernates in the larval stage during winter. The pest causes maximum damage during February to August when all its developmental stages exist simultaneously.

Cowpea production is affected by insect pests and disease infestations which lead to economic losses. Insect damage is the major constraint to cowpea grain production in most cowpeaproducing nations (Singh et al., 2012). The cowpea weevil, Callosobruchusmaculatus(F.) (Coleoptera: Chrysomelidae), is a cosmopolitan field-to-store pest ranked as the principal post-harvest pest of cowpea in the tropics (Caswel, 1981). It causes substantial quantitative and qualitative losses manifested by seed perforation and reductions in weight, market value and germination ability of seeds (Oluwafemi, 2012).

In order to reduce serious losses experienced during storage, various techniques and control methods have been developed and more are still being developed. Management of cowpea seed storage pests relies heavily on the use of chemical insecticides. However, most of the small scale farmers have not adopted these new techniques due to some financial and technical reasons. Insecticides also have negative impact on the environment, humans and non-target organisms. Therefore, there is a need to develop cheap, safe and easy methods of protecting stored cowpeas against cowpea weevil (Oluwafemi, 2012).

Resource-poor farmers in Africa employ a range of traditional methods such as use of ash, sand, dry pepper and botanical extracts. Naturally occurring plant products have been used to protect agricultural products against pests for many years in some parts of the world; many authors have reported insecticidal effects of plant products against a broad range of pests. Some of the techniques that can be explored include the use of plant products such as garlic, peppermint and chilies. Aromatic plants have both medicinal and aromatic properties and contain a variety of volatile oils which have insecticidal, anti-feedant and repellent effects on insect pests. The chemical repellency hypothesis states that non-host plant odors repel herbivores by disrupting their ability to locate or feed on the host plant (Beizhou, 2012) .The plant products used for this study produce odors that are believed to repel weevils, thereby preventing them from attacking cowpea seeds.

There is limited information on the use of the plant products as an alternative control method for controlling weevils in storage. The use of plant products may offer a sustainable, environmentally friendly and safer alternative to synthetic insecticides. This research was aimed at obtaining with practical prove the phytochemical screening and the insecticidal activities of some plant products on cowpea weevil Callosobruchusmaculatus(Fab.) Coleoptera: Chrysomelidae. This study will also create awareness of the value of plant products as an alternative control method for cowpea weevils in smallholder farmers' storage facilities (Oluwafemi, 2012).

This study introduced the application of active toxic agents from some plant extracts as an alternative control strategy for Cowpea (Vignaunguiculata) other than the use of synthetis insecticides with adverse side effect. This was necessary to cub the hazardous effects of synthetic insecticides on the environment and non-target organisms in the curse of control of pest in stored products. It also created awareness of the value of plant products as an alternative control method for cowpea weevils in smallholder farmers' storage facilities.

Materials And Mehods:-

Study area:

The study wascarried out at the Federal Universityof Technology, Owerri between the month of September and October, 2018.Owerri is the capital city of ImoState found on the longitude 50301 north and 70101E. It lies in the tropical rainforest region of South Eastern Nigeria. There are two distinct season in the area; namely: the dry season (November –March) and the rainy season (April -October). The dry season is ushered in by harmattan period and is characterized by hot weather and low humidity. The rainy season is associated with high humidity of about 80-85%; and very heavy rainfall. Temperature varies according to season between 250c to 320c in sunny day. The forest/vegetation in Owerri is characterized by a variety of plants including some medicinal plants such as Curcuma longa, Allium sativum, Zingiberofficinalespecies. These plants grow well and survive under the described climatic conditions. The people of Owerri are predominantly farmers but also practice herbal medicine to save lives in the area. However, there are few traders, private business owners, artisans, civil servants and professionals like Doctors, Engineers and lawyers.

Procurement of Experimental Materials:

The Insect Pest(Callosobruchusmaculates):

Heterogeneous sample of Adult weevil (Callosobruchusmaculates)used for this study were obtained from already infested cowpea bought from Ihiagwa market located in Ihiagwa, Owerri west local government of Imo state, Nigeria.

After selection of the weevil from theheavily infested cowpea (Vignaunguiculata) seeds, theweevilwere maintained in containers with fine muslin cloth covering the top to prevent entry and exit of the insects, and were left under laboratory condition(25–28 °C and 60%–90% relative humidity)prior to the experiment.

Experimental Cowpea (Beans):

Uninfected and undamaged cowpea seeds that were used were also purchased from fromIhiagwa market located in Ihiagwa, Owerri west local government of Imo state, Nigeria. Each seed was examined using a compact hand lens to make sure there were no damages;eggs laid and exit holes on them. They were then kept in clean plaster containers in the laboratory.

Preparation (treatments):

The three plant samples wereGarlic, Tumeric and Gingerwhich were used as treatments for this studyandwere bought from Ihiagwa market close to Federal Universitty of Technology Owerri (FUTO). Each plant sample was cutinto small pieces using a kitchen knife and a chopping board, and air dried. After drying, was grinded into powder with an electric grinderand was sieved. 100g each of Garlic, Tumeric and Gingerpowderwasweighed using a weighing balancegiven the following treatments:

T0= Control (0g)
T1= Garlic powder extract (100g)
T2= Ginger powder extract (100g)
T3=Turmeric powder extract (100g)

Experimental design:

This project research experiment was carried out under Completely Randomized Design (CRD), design in which the treatments are assigned completely at random so that each experimental unit has the same chance of receiving any one treatment.

Below is the diagrammatic illustration of the experiment design.

Laboratory procedure:

These plants sample were tested for its effect on cowpea weevil in which 100gof each of the three plant samples prepared was measured and added to each of the four replicate groups of the three treatment plants with each of these four replicates having200g of uninfected cowpea seeds and fifty live weevils were introduced and left for six weeks for observations. All observations were recorded.

Data Collection:

The total number of dead adult weevil was counted daily for forty-twodays(6 weeks). The number of infested seeds was counted by looking at the exit holes found the infested seeds of all replications.

Mortality of Pest:
The number of dead weevil in each treatment was regularly checkedper day to ascertain the mortality.The number of dead weevil were counted and recorded.

Number of holes/ punctures on the cowpea seeds:
The Cowpea (beans) was thoroughly examined to check forexit holes or puncture on each of the beans in each treatment replicates using a portable hand lens and holes observed were counted.

Weight loss:
The cowpea was examined to determine if there was any weight lost due to infestation by theweevils. This was done by weighing the wholecowpea in each replicate using a weighing balance. The final weight ofcowpeaseeds was subtracted from their initialweight obtained before experiment.

Longevity of storedpest(Life expectancy):
The longevity ofthe pest was a parameter toascertainthelife expectancy of the weevils on the powdered extracts. This was determined by counting the number of days the death of weevils occurred after applicationofthepowdered extracts.

Phytochemical of the study sample:
Phytochemical screening was carried out on the three different plant samplesto detect the chemical constituents which could be associated with the sample using the methods of Edeoga et al (2006).

Statistical Analysis:
Data collectedwereanalysedby one-way ANOVA using the SPSS software (version 20.0). Multiple comparisons were done on least square means. All the comparisons wereconsidered significant when P = 0.05.

Results:-

Death Rate (Mortality) of Callosobruchusmaculatus(Beans weevil):
The control treatment showsno significance effect in mortality in all weeks of observations (1 week to 6 weeks). The ginger plant powder showed significant mortality effect in the reduction of Callosobruchusmaculatuspopulation in all week (Table 1). The ginger population in all weeks (table 1). The ginger powder reduces the mean population introduced (50) to 16.5 (mean number alive) of C.maculatus,when compared to garlic powder (24.8) and turmeric powder (24.8) and turmeric powder (32.4) respectively. The longevity in days of C. maculatesfor ginger and garlic effects were 7 days and turmeric was 14days after application. There was no mortality observed in 42 days of observation in control treatment.

Table 1:- Effect of powder extracts of ginger, garlic and turmeric on C. maculates.

	Plant powder	Grams	Mean number of weevil introduced			Mean number of dead weevil in weeks					Mean number alive	Longevity in days
					1	2	3	4	5	6		
	Control	0	50		0	0	0	0	0	0	50	Over 42
	Ginger	100	50		7.8	8.0	4.5	4.2	4.8	4.2	16.5	7
	Garlic	100	50		4.2	6.5	2.6	4.6	4.2	3.1	24.8	7
	Turmeric	100	50		0	5.2	5.2	3.2	2.0	2.0	32.4	14

Number holes and weight loss of bean seeds:
Ginger powder (100g) gave the lowest mean number of holes on been seeds (16 ± 1.33) with percentage mortality of 67%, followed by garlic powder (100g) with mean number of holes (41 ± 2.13) and percentage mortality of 50.4%, turmeric powder (83 ± 3.04) mean number of holes with 35% mortality (table 2). The average weight loss was recorded high in control treatment (31.5 ± 1.87) and was significantly high than the bean seeds treated with turmeric powder (25.7 ± 1.67), garlic powder (15.0 ± 1.29) and ginger powder (6.0 ± 0.82) respectively

Table 2:- Effect of powder extracts of ginger, garlic and turmeric on number of holes and weight loss of bean seeds.

Plant powders	Grams	Average weight of seed (g)	initial bean	Average weight of seed (g)	final bean	Mean number of holes	Average weight loss	Mean number of dead weevil	Mortality %
Control	0	$200^{a} \pm 1.00$		$168.5^{d} \pm 4.33$		$105^{a} \pm 3.42$	$31.5^{a} \pm 1.87$	$0.0^{d} \pm 0.0$	0.0
Ginger	100	$200^{a} \pm 1.00$		$194.0^{a} \pm 4.64$		$16^{d} \pm 1.33$	$6.0^{d} \pm 0.82$	$33.5^{a} \pm 1.93$	67.0
Garlic	100	$200^{a} \pm 1.00$		$185.0^{b} \pm 4.53$		$41^{c} \pm 2.13$	$15.0^{c} \pm 1.29$	$25.2^{b} \pm 1.67$	50.4
Turmeric	100	$200^{a} \pm 1.00$		$174.3^{c} \pm 4.40$		$83^{b} \pm 3.04$	$25.7^{b} \pm 1.69$	$17.6^{c} \pm 1.40$	35.0

Mean along the column having different letters of superscript differ significantly at P = 0.05 level

Nutrition Proximate Composition and Phytochemcial Constituents of Ginger, Garlic and Turmeric

Tables 3 and 4 show results on proximate composition and phytochemcial constituents of ginger, garlic and turmeric. The plant powders contained all the nutrient proximate tested such as dry matter, crude protein, crude fibre, ash, ether extract and nitrogen free extracts. The phytochemcials found were alkaloids, flavonoid, saponin and cardiac glycoside. Tannin was absent in turmeric while terpenoid was absent in ginger and garlic.

Table 3:- Proximate composition of turmeric, ginger and garlic.

	Constituents	Turmeric	Ginger	Garlic
	Dry matter (%)	51.62	81.87	80.62
	Crude protein	12.51	9.62	8.11
	Crude fibre	12.22	10.57	11.65
	Ash	6.44	8.14	8.13
	Ether extract	8.15	7.33	5.61
	Nitrogen free extracts (%)	49.10	42.40	40.98

Phytochemcial Analysis of Turmeric, Ginger and Garlic:
The phytochemcial analysis of these extracts indicated the presence of some chemcial like alkaloids, saponins, cardiac glycosides, flavonoids, tannins and terpenoids all of which possess insecticidal properties. Tannin was absent in turmeric while terpenoid was absent in ginger and garlic.

Table 4:- Phytochemcial analysis of turmeric, ginger and garlic.

	Parameters	Turmeric	Ginger	Garlic
	Alkaloid	+	++	+
	Cardiac glycoside	+	++	+
	Tannins	+	+	-
	Flavonoids	+	++	++
	Saponins	+	++	+
	Terpenoids	+	-	-

Discussion:-

The results of this study showed that treatments differed in the percent mortality of cowpea weevils, number of exit holes on seeds and weight loss of cowpeas. It has been shown in this experiment that garlic and ginger had similar

detrimental effect on cowpea weevils for the parameters measured. This, therefore, implied that these plant products negatively impacted the weevils more than turmeric and control. It can, therefore, be mentioned that ginger and garlic can better protect cowpea seeds from cowpea weevil damage than if seeds are left or stored unprotected. This even agrees with the work of Nwachukwu et al., (2003) on the effect of Citrus sinensis (Orange) powdered peel extract on Cryptolestsfurringineusof pearl millet Pennisetumglauncum in Imo state. Although turmeric had less negative effect on the weevils than ginger and garlic, it also showed significant reduction in number of cowpea weevils found on seeds, number of exit holes on seeds, weight loss of cowpeas and increased percent mortality of cowpea weevils compared to untreated seeds.

The reduction in the presence of cowpea weevil might be due to toxicity of the plant materials to the weevils rather than deterrence activity of the extract. This is because, as it can be seen from the study, these plant materials caused highest mortality which can be linked to toxicity of the plant materials. Also the phytochemical analysis of the phyto-constituents of these plants sample showed the presence of some active substances with the capability of lysing the cell membrane of the pest by contact action. It should be noted that even though ginger and garlic showed a stronger effect on cowpea weevils when applied on the seeds, they did not eliminate the cowpea weevil's presence or occurrence of their effect on the seeds but rather reduced weevils numbers and their effect on the seeds to significantly low levels as compared to unprotected seeds. The implication of these effects is that garlic and ginger as well as turmeric material used as treatment in this experiment do not offer total exclusion of cowpea weevil but rather they lowered the weevil numbers and their effects on the cowpea seeds.

The mortality of the cowpea weevil followed a similar trend, with higher mortality observed on all treatments compared with the control. The highest mortality was observed on ginger treatment followed by that of garlic treatment and the least was the tumeric. A significant reduction in the exit holes was exhibited on the ginger and garlic treatments compared with the turmeric treatment and control. The highest number of exit holes was observed on the control.

Conclusion and Recommendations:-

Based on my results, it can be concluded that ginger, garlic and, to a lesser extent turmeric, might serve as alternatives to insecticides on rural farms in tropical and subtropical regions. Insecticides are costly and not sustainable in the long run due to environmental contamination (Silver et al., 1994), the use of these plant products would be cost effective and sustainable, especially considering that these plants are easy to grow. The plant products tested contain a range of bioactive chemicals, many of which are selective and have little or no harmful effect on non-target organisms and the environment (Vinayaka et al., 2010). In addition, these products are safe to users as evidenced by the fact that they are used as culinary spices and herbs.

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