

Research Article

Application of Two *Ocimum* Species Powders in the Control of Red Flour Beetles, *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae), during Wheat Grain Storage

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Abstract: Pest insects on stored goods are a problem everywhere in the world because they lower grain yields and quality. This has the impact of creating a significant danger to global food security, which could negatively affect people's well-being. In the human endeavor to lessen the threat caused by these insects, the use of synthetic chemical insecticides has unfavorable effects. Recently, there has been advancement in the use of botanicals and agricultural wastes in the control of these pests. This research investigated the effects of *Ocimum gratissimum* (African basil) and *O. basilicum* (common basil) powders against *Tribolium castaneum* infesting wheat grains in storage. Powders were tested singly and mixed at doses of 0.2, 0.4, 0.6, 0.8 and 1.0 g per 20 g of wheat grains. The following parameters were determined: adult mortality, adult emergence rates, seed damage %, weight loss %, and beetle perforation index. The data showed that two *Ocimum* powders were efficacious and their effectiveness increased with increasing the dosage and also the exposure time. The population of adult *T. castaneum* on treated wheat grains was significantly ($p < 0.05$) decreased by the two tested *Ocimum* species powders. After 24 hours of treatment, 1.0 g of *O. basilicum* powder caused 50% mortality of adult *T. castaneum*. However, the combination of *O. gratissimum* and *O. basilicum* powders caused 40% of beetles' mortality. On the other hand, *O. gratissimum* powder was the least toxic on adult *T. castaneum* with 30% only mortality. The lethal doses of both tested powders and their combination required to cause 50% mortality of the *T. castaneum* population after the first day of exposure were 1.09, 1.44, and 1.45 g, respectively. While, the lethal dose required to cause 90% mortality (LD_{90}) was 4.24, 8.44, and 5.38 g after the first day post exposure. This study demonstrated the toxicity of *O. gratissimum* and *O. basilicum* powders on the survival of *T. castaneum* on wheat grains. The least deadly dose was recorded on *O. gratissimum*. Therefore, it is recommended that poor resource farmers and food vendors use *Ocimum* powder as a biocontrol to manage the red-rust flour beetle in stored wheat grains.

Keywords: African basil, common basil, lethal dose, biocontrol, red-rust beetles

1. Introduction

Food loss due to insect infestation during the conservation is a serious problem, especially in developing countries [1-2]. Worldwide, it is estimated that between a quarter and a third of the grain produced per year is wasted during the storage because of the insects' infestation and also diseases. Many grain pest insects typically eat their grain germ; lowering the protein content of feed grain and reducing the percentage of seeds germination [3]. In nations that do not

use the newest storage technologies, such damage can be up to 40% [4].

Wheat grains (*Triticum* spp.) are a significant staple food; contributing 20% of the world's dietary protein and calories besides [5]. Wheat is a great source of energy for farm animals; 42% of wheat production in Europe was utilized for animal feed in 2007 [5-6]. Forage wheat is typically subpar wheat that is used in excess of human needs or is unfit for human consumption (low specific gravity or damaged wheat) [5]. Wheat can be a wise choice when other grains like corn, barley, and sorghum are expensive or when their prices are falling [5]. In order to overcome malnutrition and protein shortages, wheat is rapidly becoming a staple meal for both humans and poultry animals in Africa [5, 7]. If pest insects are not controlled, wheat grains may be eaten by them. *Tribolium castaneum* is one of the harmful insect pests of many stored agricultural products worldwide [8-9]. This insect pest favors goods that are pulverized or powdered and contain flour [5, 8]. Numerous cereals, seeds, and other raw materials such as dried fruits, beans, cocoa beans, almonds, spices, oil seeds, cotton seeds, and processed foods were been reported to be infested by the [10-11]. This beetle is significantly reducing the number and quality of products that are stored all over the world [12].

Farmers are forced to seek out effective control measures to reduce the losses resulting from disease and insect damage to wheat [13-15]. One such method is the use of synthetic pesticides with quick and broad-spectrum insect pest management. Entomologists around the world are concerned about the indiscriminate use of these synthetic pesticides because of their risks to human health, the environment, and the economy due to a lack of funding [16-18]. In addition, many species of insect pests are becoming resistant to the pesticides used over time. In this regard, this study assessed the contact toxicity efficacies and combination effects of *Ocimum gratissimum* and *O. basilicum* powders against *T. castaneum* adults on wheat grains.

2. Materials & methods

2.1 Insects rearing

Tribolium castaneum adults were provided by the Storage Entomology Research Laboratory, Department of Biology, Federal University of Technology, Akure (FUTA). One hundred pairs of the beetle were placed in a 1 L glass Kilner jar containing 1,000 g of wheat flour bought from a supermarket within Akure Metropolis, Akure, Ondo State, Nigeria. The beetle colony was kept at a constant insectarium temperature of 28 ± 2 °C and relative humidity of $75 \pm 5\%$.

2.2 Identification and sexing of *T. castaneum* adult

Red-rust flour beetle was identified and sexed in the Entomology Research Laboratory, Department of Biology, FUTA. Each of the adult male's elytra bears stria punctures and his stiff black seta points backwards. In contrast, the female's seta points forwards [19].

2.3 Collection and preparation of *O. gratissimum* and *O. basilicum* powders

Fresh and insecticide-free leaves of *O. gratissimum* and *O. basilicum* were sourced from a farm in Ifon, Ondo State, Nigeria. The leaves were first identified by a Plant Taxonomist at the Department of Crop, Soil, and Pest Management, FUTA before they were washed with distilled water and air-dried at room temperature for 21 days in the laboratory, crushed with a pestle and mortar, and then ground to a powder in an electric blender (a JTC Omni Blender V) (Model TM-800). A nylon mesh with a 1 mm² opening was used to filter the tiny granules. The powders were then sealed in airtight containers and stored in the fridge at 4 °C to maintain quality until use.

2.4 Collection of wheat grains

Newly stocked and insecticide-free wheat grains were bought from a grain Merchant in Akure, Ondo State, Nigeria. The wheat grains were first cleaned (the debris and seed shaft were removed) before warehoused in a deep freezer at -5 °C for 72 hours to kill any existing insect eggs, larvae, pupae, and adults [2, 20]. The disinfested wheat grains were later air dried naturally in the laboratory for 3 hours to ensure that the grains did not become moldy [20-21].

2.5 Phytochemical screening of the plants

Standard protocols, such as those described by Harborn [22], Trease and Evans [23], and Sofowora [24], Ezeonu and Ejikeme [25] and Ileke et al. [3] were used to conduct chemical assays on crude extracts of the leaves of *O. gratissimum* and *O. basilicum* for the quantitative measurement of phytochemical contents.

2.6 Insect bioassay

2.6.1 Toxicity of single and mixed plants powders on adult mortality and progeny development of *T. castaneum*

Using a precision electronic weighing balance (Model JTC 2101N) in the laboratory, twenty grams (20 g) of disinfected wheat grains were measured and placed in plastic containers (250 mL). Then, wheat grains were mixed with powdered extracts of *O. gratissimum* and *O. basilicum* leaves at concentrations of 0.2, 0.4, 0.6, 0.8, and 1.0 g for each formulation (in three duplicates). The plant powders and wheat grains were properly mixed together by inverting the contain four times. Ten copulating mature adult *T. castaneum* (less than 4 days old) were introduced to each container of the treated wheat grains. Only 20 g of wheat and ten copulating pairs of mature *T. castaneum* were used in the control experiment. The mortality of insects was observed daily for 5 days. Adults that did not move or react to pin probing were considered to be dead. Data on the percentage of adult mortality were corrected using the Abbott (1925) algorithm 5 days after therapy as follows:

$$P_T = \frac{P_o - P_c}{100 - P_o} \times 100$$

Where P_T = corrected mortality (%); P_o = observed mortality (%); P_c = control mortality (%).

Until the first filial generation of adult emergence, daily observations were made using the insect bioassay setup within the insect rearing cage.

The percentage of weight reduction for the wheat grains was calculated as follows:

$$\text{Weight loss \%} = \frac{\text{Change in weight}}{\text{Initial weight}} \times 100$$

Initial weight: weight of the grains before application of treatment and pest.

Change in weight: weight of the grains after termination of the experiment.

The percentage of seed damage in wheat grains was calculated as follows:

$$\text{Seed damaga \%} = \frac{\text{Number of seeds damaged}}{\text{Total number of seeds}} \times 100$$

The Beetle Perforation Index (BPI) was calculated using the method provided by Fatope et al. [26]:

$$\text{BPI} = \frac{\text{treated wheat grains perforated \%}}{\text{control wheat grains perforated \%}} \times 100$$

A BPI greater than 50 was interpreted as indicative of an increase in beetle infestation or a lack of protection by the extract under study.

2.7 Data analysis

Data obtained were analyzed by subjected to one-way analysis of variance (ANOVA) at 5% threshold of significance level and means were separated with Duncan's Multiple Range Test using SPSS version 26. Adulticidal bioassay data were analyzed using the log-Probit model to determine the 50% lethal dose (LD₅₀) and the 90% lethal dose (LD₉₀) using Microsoft Excel Probit analysis work book prepared by [27].

3. Results

3.1 Phytochemical (Quantitative) composition of *Ocimum* species

Table 1 displays the phytochemical components of the two *Ocimum* species (*O. gratissimum* and *O. basilicum*) employed in this investigation. *Ocimum basilicum* had the greatest levels of tannin and flavonoid with 0.24 and 0.51 mg/g, respectively and significantly different from those found in *O. gratissimum* with the rate of 0.17 mg/g and 0.41 mg/g, respectively. The compositions of alkaloid and saponin found in *O. basilicum* (20.47 and 19.52 mg/g, respectively) were higher than those in *O. gratissimum* (18.84 and 17.58 mg/g, respectively).

Table 1. Quantitative analysis of phytochemical present in the tested *Ocimum* species

Phytochemicals	Plant		Sum of Squares	F value	p value
	<i>O. basilicum</i>	<i>O. gratissimum</i>			
Flavonoid	0.51 ± 0.02	0.41 ± 0.03	0.027	9.197	0.016
Alkaloid	20.47 ± 0.19	18.84 ± 0.36	6.642	15.665	0.004
Saponin	19.52 ± 0.21	17.58 ± 0.26	9.449	34.154	0.00
Tannin	0.24 ± 0.02	0.17 ± 0.02	0.012	7.402	0.026

p value: < 0.05 (significant), > 0.05 (non-significant)

3.2 Mortality of *T. castaneum* exposed to two *Ocimum* species powders

Table 2. Mortality of *T. castaneum* exposed to two *Ocimum* species powders

	Dosage (g)	% Mortality (Mean ± Standard error (SE))				
		Day 1	Day 2	Day 3	Day 4	Day 5
<i>O. basilicum</i>	0.2	6.67 ± 3.33 ^{abc}	13.33 ± 3.33 ^{bc}	23.33 ± 3.33 ^d	30.00 ± 0.00 ^c	36.67 ± 3.33 ^c
<i>O. gratissimum</i>		0.00 ± 0.00 ^a	6.67 ± 3.33 ^{ab}	10.00 ± 0.00 ^b	16.67 ± 3.33 ^b	20.00 ± 0.00 ^b
<i>O. basilicum</i> + <i>O. gratissimum</i>		3.33 ± 3.33 ^{ab}	10.00 ± 5.77 ^b	13.33 ± 3.33 ^{bc}	20.00 ± 5.77 ^b	23.33 ± 6.67 ^b
<i>O. basilicum</i>	0.4	13.33 ± 3.33 ^{cd}	23.33 ± 3.33 ^{de}	33.33 ± 3.33 ^e	43.33 ± 3.33 ^d	53.33 ± 3.33 ^{de}
<i>O. gratissimum</i>		3.33 ± 3.33 ^{ab}	10.00 ± 0.00 ^b	16.67 ± 3.33 ^{bcd}	23.33 ± 3.33 ^{bc}	33.33 ± 3.33 ^c
<i>O. basilicum</i> + <i>O. gratissimum</i>		6.67 ± 3.33 ^{abc}	13.33 ± 3.33 ^{bc}	20.00 ± 0.00 ^{cd}	30.00 ± 0.00 ^c	40.00 ± 0.00 ^c
<i>O. basilicum</i>	0.6	26.67 ± 3.33 ^{ef}	36.67 ± 3.33 ^{fg}	53.33 ± 3.33 ^{gh}	63.33 ± 3.33 ^f	73.33 ± 3.33 ^g
<i>O. gratissimum</i>		10.00 ± 0.00 ^{bc}	20.00 ± 0.00 ^{cd}	33.33 ± 3.33 ^e	43.33 ± 3.33 ^d	50.00 ± 0.00 ^d
<i>O. basilicum</i> + <i>O. gratissimum</i>		20.00 ± 0.00 ^{de}	26.67 ± 3.33	43.33 ± 3.33 ^f	53.33 ± 3.33 ^e	60.00 ± 0.00 ^{ef}
<i>O. basilicum</i>	0.8	40.00 ± 0.00 ^g	56.67 ± 3.33	70.00 ± 0.00 ^{hk}	76.67 ± 3.33 ^g	86.67 ± 3.33 ^h
<i>O. gratissimum</i>		20.00 ± 0.00 ^{de}	30.00 ± 0.00 ^{ef}	46.67 ± 3.33 ^{fg}	53.33 ± 3.33 ^e	63.33 ± 3.33 ^f
<i>O. basilicum</i> + <i>O. gratissimum</i>		26.67 ± 3.33 ^{ef}	40.00 ± 0.00 ^g	60.00 ± 0.00 ^{hi}	66.67 ± 3.33 ^f	76.67 ± 3.33 ^g
<i>O. basilicum</i>	1.0	50.00 ± 0.00 ^h	63.33 ± 3.33 ⁱ	76.67 ± 3.33 ^k	90.00 ± 0.00 ^h	100.00 ± 0.00 ⁱ
<i>O. gratissimum</i>		30.00 ± 0.00 ^f	40.00 ± 0.00 ^g	56.67 ± 3.33 ^h	66.67 ± 3.33 ^f	76.67 ± 3.33 ^g
<i>O. basilicum</i> + <i>O. gratissimum</i>		40.00 ± 0.00 ^g	50.00 ± 0.00 ^{hi}	66.67 ± 3.33 ^{ij}	80.00 ± 0.00 ^g	86.67 ± 3.33 ^h
Control	0.0	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a

Each value is the average ± standard error of three separate samples. The Duncan Multiple Range Test (DMRT) indicates that means in a column that are separated by the same letter are not statistically distinct (p > 0.05)

The mortality response of adult *T. castaneum* to *Ocimum* species powders regarding the period of exposure is presented in Table 2. The percentage mortality of *T. castaneum* does not show a significant difference ($p > 0.05$) when comparing the effect of 0.2 and 0.4 g of two *Ocimum* species powder treatments, but there is a significant difference ($p < 0.05$) between the insecticidal effects at different powder concentrations (0.2, 0.4, 0.6, 0.8, and 1.0 g) after the first day of exposure. The insecticidal effects of 0.4 and 0.6 g powder of *O. gratissimum* on adult *T. castaneum* are not significantly different ($p > 0.05$) after the first day of exposure; causing 3.33 and 10.00% mortality respectively. However, the effects of 0.8 g of *O. basilicum* powder and a mixture of both *O. gratissimum* and *O. basilicum* caused 86.67 and 76.67% mortality on *T. castaneum* respectively, and 1.0 g evoked 100 and 86% mortality, respectively which are not substantially different ($p > 0.05$) after the fifth day of exposure.

The lowest mortality was recorded from 0.2 g powder of *O. gratissimum* which evoked 20% mortality on *T. castaneum* after the fifth day of exposure. The highest mortality rate (50%) of *T. castaneum* was registered on 1.0 g of *O. basilicum* powder after the first day of exposure.

3.2.1 Lethal Doses (LD_{50} and LD_{90}) of *Ocimum* species powders against adult *T. castaneum*

One day post-exposure, 1.09 g of *O. basilicum* powder was required to cause 50% (LD_{50}) mortality in *T. castaneum* but 0.38 g was needed on day 4, and 0.31 g for day 5 (Table 3). The LD_{90} for *O. basilicum* powder against *T. castaneum* is 4.24, 1.28, and 1.09 g after the first, fourth and fifth days of exposure, respectively. The linear regression coefficient (R^2) was 0.97, 0.92, and 0.95 on days 1, 4 and 5, respectively.

Table 3. Lethal Doses (LD_{50} and LD_{90}) of *Ocimum* species powders against adult *T. castaneum*

Plant Powder	Exposure Period	Intercept \pm SE	Slope \pm SD	R^2	LD_{50} (LCL-UCL)	LD_{90} (LCL-UCL)	p value
<i>O. basilicum</i>	Day 1	4.92 \pm 0.09	2.19 \pm 0.46	0.97	1.09 (0.72-1.69)	4.24 (2.81-6.39)	0.75
	Day 2	5.27 \pm 0.09	2.14 \pm 0.47	0.95	0.75 (0.50-1.11)	2.99 (2.01-4.44)	0.66
	Day 3	5.65 \pm 0.09	2.17 \pm 0.46	0.94	0.50 (0.34-0.76)	1.97 (1.34-2.91)	0.62
	Day 4	6.04 \pm 0.08	2.48 \pm 0.40	0.92	0.38 (0.27-0.54)	1.28 (0.90-1.82)	0.59
	Day 5	6.20 \pm 0.08	2.36 \pm 0.43	0.95	0.31 (0.21-0.45)	1.09 (0.75-1.59)	0.66
<i>O. gratissimum</i>	Day 1	4.47 \pm 0.08	3.31 \pm 0.30	0.99	1.44 (1.03-2.03)	8.44 (5.03-14.16)	0.89
	Day 2	4.63 \pm 0.12	1.81 \pm 0.55	0.93	1.61 (0.96-2.70)	3.69 (2.46-5.56)	0.60
	Day 3	5.09 \pm 0.09	2.13 \pm 0.47	0.95	0.91 (0.61-1.37)	3.53 (2.51-4.95)	0.63
	Day 4	5.30 \pm 0.09	2.01 \pm 0.50	0.93	0.71 (0.47-1.08)	3.12 (2.05-4.75)	0.55
	Day 5	5.58 \pm 0.09	2.20 \pm 0.46	0.96	0.54 (0.37-0.79)	2.11 (1.44-3.09)	0.78
<i>O. basilicum</i> + <i>O. gratissimum</i>	Day 1	4.64 \pm 0.10	2.28 \pm 0.44	0.95	1.45 (0.95-2.22)	5.38 (3.51-8.25)	0.63
	Day 2	4.88 \pm 0.10	1.89 \pm 0.53	0.91	1.16 (0.73-1.86)	5.69 (3.56-9.11)	0.43
	Day 3	5.38 \pm 0.08	2.36 \pm 0.43	0.93	0.69 (0.48-0.98)	2.45 (1.70-3.52)	0.41
	Day 4	5.68 \pm 0.08	2.41 \pm 0.42	0.94	0.52 (0.37-0.74)	1.80 (1.27-2.56)	0.50
	Day 5	5.96 \pm 0.07	2.62 \pm 0.38	0.96	0.43 (0.31-0.60)	1.34 (0.96-1.86)	0.71

Note: R^2 = Statistical measure of mortality proportion in regression model; SE = Standard error; SD = Standard deviation; LD_{50} = Lethal dosage at which 50% population response; LD_{90} = Lethal dosage at which 90% population response; LCL = Lower confidence limit; UCL = Upper confidence limit; p value = Chi-square (χ^2) Significant

On the other hand, the powder dosage of *O. gratissimum* required to cause 50% (LD_{50}) mortality of *T. castaneum* for the first three days of exposure were 1.44, 1.61 and 0.91 g, respectively, while the last two days were 0.71 and 0.54 g. The lethal dosage required to achieve 90% (LD_{90}) mortality of *T. castaneum* after different days of exposure were 8.44, 3.69, 3.53, 3.12 and 2.11 g. The regression linear (R^2) calculated from the first till the fifth days were 0.99, 0.93, 0.95,

0.93 and 0.96 g, respectively.

The mixture of *O. basilicum* and *O. gratissimum* powders required to kill 50% of *T. castaneum* within five days of exposure were 1.45, 1.16, 0.69, 0.52 and 0.43 g, respectively. The (LD₅₀) mortality of *T. castaneum* were 5.38, 5.69, 2.45, 1.80 and 1.34 g, respectively. The regression linear (R²) calculated for the first, fourth and fifth days were 0.95, 0.94 and 0.96 g, respectively.

3.2.2 Effect of *Ocimum* species powders on adult emergence of *T. castaneum*

There was a statistically significant ($p < 0.05$) difference between the insecticidal effects of the various powder dosage treatments (0.2, 0.4, 0.6, 0.8, and 1.0 g) and the control on *T. castaneum* adult emergence (Table 4). The oviposition of *T. castaneum* was significantly decreased by the plant powders. Wheat beetles lay substantially fewer eggs on treated wheat ($p < 0.05$) compared to untreated ones. At rates of 0.6, 0.8, and 1.0 g of plant powder per 20 g of wheat grains, the emergence of *T. castaneum* was entirely suppressed, and no adults emerged during the growth of the progeny.

Table 4. Effect of *Ocimum* species powders on adult emergence of *T. castaneum*

Plant Powder	Dosage (g)	% adult emergence
<i>O. basilicum</i>	0.2	1.00 ± 0.58 ^a
	0.4	0.33 ± 0.33 ^a
	0.6	0.00 ± 0.00 ^a
	0.8	0.00 ± 0.00 ^a
	1.0	0.00 ± 0.00 ^a
<i>O. gratissimum</i>	0.2	2.00 ± 0.00 ^a
	0.4	1.00 ± 0.00 ^a
	0.6	0.33 ± 0.00 ^a
	0.8	0.33 ± 0.33 ^a
	1.0	0.00 ± 0.00 ^a
<i>O. basilicum</i> + <i>O. gratissimum</i>	0.2	1.33 ± 0.33 ^a
	0.4	0.67 ± 0.33 ^a
	0.6	0.00 ± 0.00 ^a
	0.8	0.00 ± 0.00 ^a
	1.0	0.00 ± 0.00 ^a
Control	0.0	42.67 ± 2.33 ^b

Each value is the average ± standard error of three separate samples. The Duncan Multiple Range Test (DMRT) indicates that means in a column that are separated by the same letter are not statistically distinct ($p > 0.05$)

3.2.3 Percentage of damaged grains, weight loss, and beetle perforation index in wheat grains treated with *Ocimum* species powders by *T. castaneum*

Ocimum basilicum and the combined effects of the two plant powders prevented infestation and damage of the treated wheat grains at rates of 0.6, 0.8 and 1.0/20 g of wheat grains (Table 5). There was neither grain damage nor weight loss recorded in the treated grains, and the beetle perforation index (BPI) was zero. Also, *O. gratissimum* at a rate of 1.0 g prevented the infestation and injury of the treated wheat grains. The BPI of treated grains was significantly different ($p < 0.05$) from that of untreated ones with about 9.31% of the untreated grains being damaged by *T. castaneum* larval feeding.

Table 5. Percentage grains damaged, weight loss and beetle perforation index caused by *T. castaneum* in wheat grains treated with *Ocimum* species powders

Plant Powder	Dosag (g)	Mean no of grains	Mean % of damaged grains	% Weight Loss	Beetle Perforation Index (BPI)
<i>O. basilicum</i>	0.2	473.67 ± 11.68	0.35 ± 0.07 ^{abc}	0.57 ± 0.09 ^{abc}	3.73 ± 0.63 ^{bc}
	0.4	407.67 ± 2.85	0.24 ± 0.00 ^{abc}	0.37 ± 0.03 ^{abc}	2.63 ± 0.14 ^b
	0.6	462.67 ± 10.73	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
	0.8	458.00 ± 16.80	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
	1.0	419.00 ± 6.56	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
<i>O. gratissimum</i>	0.2	428.67 ± 22.85	0.63 ± 0.11 ^c	0.97 ± 0.15 ^c	6.94 ± 1.42 ^d
	0.4	482.00 ± 8.33	0.41 ± 0.01 ^{abc}	0.73 ± 0.03 ^{bc}	4.45 ± 0.14 ^c
	0.6	435.67 ± 28.17	0.16 ± 0.16 ^{ab}	0.27 ± 0.27 ^{ab}	0.00 ± 0.00 ^a
	0.8	458.33 ± 19.03	0.07 ± 0.07 ^{ab}	0.13 ± 0.13 ^{ab}	0.79 ± 0.79 ^a
	1.0	422.33 ± 10.67	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
<i>O. basilicum</i> + <i>O. gratissimum</i>	0.2	427.33 ± 15.45	0.47 ± 0.02 ^{bc}	0.70 ± 0.00 ^{bc}	5.07 ± 0.31 ^c
	0.4	476.00 ± 1.00	0.28 ± 0.07 ^{abc}	0.50 ± 0.15 ^{abc}	2.95 ± 0.58 ^b
	0.6	421.67 ± 8.37	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
	0.8	439.00 ± 8.19	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
	1.0	431.00 ± 9.54	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a	0.00 ± 0.00 ^a
Control	0.0	465.33 ± 6.23	9.31 ± 0.45 ^d	13.00 ± 0.72 ^d	> 50.00 ± 0.00 ^d

Each value is the average ± standard error of three separate samples. The Duncan Multiple Range Test (DMRT) indicates that means in a column that are separated by the same letter are not statistically distinct (P > 0.05)

3.3 Correlation

3.3.1 Correlation between phytochemicals in *O. gratissimum* powders against *T. castaneum* adult emergence, wheat grains damage, and weight loss

The correlation matrices show the interaction among the composition of phytochemical and biological activities of *T. castaneum* in Table 6. The correlation analysis of phytochemicals in *O. gratissimum* powder shows that the adult emergence ($r = -0.208$), grain damage ($r = -0.351$), and weight loss ($r = -0.392$) correlated negatively with flavonoids. The composition of alkaloid's is moderately negatively related to the beetle adult emergence ($r = -0.402$) and grain damage ($r = -0.402$), while the relationship was weak and negatively related to weight loss ($r = -0.356$). The composition of saponin correlated negatively with adult emergence ($r = -0.135$), grain damage ($r = -0.074$), and weight loss ($r = -0.014$), while tannin correlated positively with adult emergence ($r = 0.137$), grain damage ($r = 0.096$), and weight loss ($r = 0.139$).

Table 6. Correlation between phytochemicals in *O. gratissimum* powders on *T. castaneum* adult emergence, wheat grains damage, and weight loss

Variable	Flavonoid	Alkaloid	Saponin	Tannin	% Adult Emergence	% Grain Damage	% Weight Loss
Flavonoid	1	0.592	0.341	0.327	-0.208	-0.351	-0.392
Alkaloid	-	1	0.893*	0.669	-0.402	-0.402	-0.356
Saponin	-	-	1	0.560	-0.135	-0.074	-0.014
Tannin	-	-	-	1	0.137	0.096	0.139
% Adult Emergency	-	-	-	-	1	0.981**	0.962**
% Seed Damage	-	-	-	-	-	1	0.995**
% Weight Loss	-	-	-	-	-	-	1

* Correlation is significant at the 0.05 level (2-tailed)

** Correlation is significant at the 0.01 level (2-tailed)

3.3.2 Correlation between phytochemicals in *O. basilicum* powders on *T. castaneum* adult emergence, wheat grains damage, and weight loss

The correlation coefficient (r) between the composition of phytochemical and biological activities of *O. basilicum* powders against *T. castaneum* is presented in Table 7. There was a weak and positive correlation between the composition of flavonoid and the adult emergence % of *T. castaneum* ($r = 0.090$), while a strong and positive correlation was between flavonoid and grain damage % ($r = 0.685$). Besides, a negatively weak relationship was observed between flavonoid and the percentage of weight loss ($r = -0.209$) caused by the beetles. The correlation between alkaloids composition on adult emergence ($r = -0.341$) showed a weak and negative relation, while it was negative and strong with the weight loss ($r = -0.682$), and positively moderate with the grain damage ($r = 0.418$).

Also, the adult emergence was negatively weak with the composition of saponin in *O. basilicum* ($r = -0.281$), and positively weak with the weight loss ($r = 0.334$), while the relationship was negatively strong with the grain damage ($r = -0.734$). The composition of tannin is negatively weak on the beetle adult emergence ($r = -0.214$), while it is negatively strong on the grain damage ($r = -0.749$), and positively weak with weight loss ($r = 0.060$).

Table 7. Correlation between phytochemicals in *O. basilicum* powders on *T. castaneum* adult emergence, wheat grains damage, and weight loss

Variable	Flavonoid	Alkaloid	Saponin	Tannin	% Adult Emergence	% Grain Damage	% Weight Loss
Flavonoid	1	0.857	-0.649	-0.988**	0.090	0.685	-0.209
Alkaloid	-	1	-0.671	-0.771	-0.341	0.418	-0.682
Saponin	-	-	1	0.623	-0.281	-0.734	0.334
Tannin	-	-	-	1	-0.214	-0.749	0.060
% Adult Emergency	-	-	-	-	1	0.539	0.780
% Seed Damage	-	-	-	-	-	1	0.212
% Weight Loss	-	-	-	-	-	-	1

** Correlation is significant at the 0.01 level (2-tailed)

4. Discussion

In developing countries, such as Nigeria, attention is being drawn to the use of plant spices like the *Ocimum* species as insecticides against the stored product insect pests as a substitute for the use of synthetic chemical insecticides that

pose a risk to the environment's health [28]. Both *O. gratissimum* and *O. basilicum* powders exhibited an insecticidal impact on *T. castaneum*, according to the study's findings. The plant powders of *O. gratissimum* and *O. basilicum*, as well as a combination of the two plants, when used as a contact insecticide were particularly efficient against *T. castaneum* adult.

Both of the oviposition and adult emergence were considerably reduced or inhibited by the powders of *O. gratissimum* and *O. basilicum*, as well as by combining the two plant powders. This was consistent with [29], who found that the essential oil from *Pistacia lentisus* prevents *T. castaneum* from growing. This is also in line with the research of Booker [30], and Adedire and Akinneye [31], who discovered that Eugenia aromatic powder and extract greatly lower or prevent the egg laying and adult *Callosobruchus maculatus* emergence on cowpea seeds. The present results revealed that *O. gratissimum* was less toxic on *T. castaneum* than *O. basilicum* powder. This is in line with the research by Ileke and Adesina [28], who found that *O. gratissimum* and *O. basilicum* extracts were effective in controlling the development of malaria vectors and that *O. basilicum* was more toxic to *Anopheles gambiae*. Adult beetles appeared in all control samples; proving that the plant powders significantly affected the F₁ offspring of the rust flour beetle. Even when low dosages were used for treatment, insects may have survived on treated grains for a short time and produced relatively few F₁ progeny, while the highest levels were completely prevented both insects' young from developing [5]. This study validated the toxicity of the tested plant powders and found that the test insects' ability to reproduce was significantly reduced. Due to their widespread availability, low entry barriers, and a few negative environmental effects, materials like these have the potential to play a substantial role in the integrated pest management (IPM). Several phytochemicals that were once believed to be efficient pesticides have now been discovered to be toxic to animals rather than the ones they were intended to kill. For instance, 14 milliliters of neem oil per kilogram of body weight is the median fatal dosage (LD₅₀) for rats [32].

Insects are most active throughout their larval and adult stages when they gorge themselves on food. This may have resulted in the *O. gratissimum* and *O. basilicum* dosage being toxic enough to cause stomach poisoning and the significant mortality rate shown in the *T. castaneum* employed in this investigation [2, 13-14, 18]. The larvae must have consumed some of the plants' active ingredients during feeding, which resulted in poisoning of the stomach, contacts, and respiratory system [3]. These plant powders may be so deadly to insects because they can block their breathing tubes [20]. A spiracle on the surface of the body serves as the trachea exit for the majority of insects. The adult insects in this experiment may choke and perish as a result of the plant powders clogging the spiracles [18, 20]. The effects of the two *O. gratissimum* and *O. basilicum* powders caused a considerable mortality risk, reduced egg laying and adult emergence, damaged wheat grain, increased beetle perforation indices, and weight loss of the treated wheat grain.

Alkaloids, saponins, tannins, flavonoids, anthraquinone, and phlobotannins are among the phytochemicals found in *O. gratissimum* and *O. basilicum* leaves. According to Fernando et al. [33] and Ileke et al. [3], the majority of plants are known to contain a variety of chemical compounds, including saponins, tannins, flavonoids, and alkaloids, which have been proven to be reasonably efficient against insect pests. Besides, tannins play a role in regulating plant growth and offer protection from predators [3, 34-35].

5. Conclusion

This study concluded that the two *Ocimum* species (*O. gratissimum* and *O. basilicum*) could be used to manage rice red-rust flour beetles, *T. castaneum* and other related insect pests because they had numerous insecticidal effects, particularly *O. basilicum* powder. It was discovered that the plant powders and extracts of these two plants are somewhat safe, degradable, less expensive, and easily accessible in many parts of the world. However, they were proven to be very toxic to *T. castaneum*. Thereby, this study recommends the use of these plants among local farmers for controlling *T. castaneum* and other stored product insect pests as well as no toxic effect on health have been recorded on consumption of treated product with these plants in previous literature at the dosages tested.

Conflict of interest

The authors declare no competing financial interest.

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