Biological Performance of Certain Botanical Fine Dusts, Ash and Sulfur Powders against the Rice Weevil *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae)

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ABSTRACT

Five botanical fine dusts, ash and sulfur powders were tested for their biological performance activity against the rice weevil *Sitophilus oryza*. Their toxicities and delayed effect on the treated insect were determined. The insect was exposed to treated rice grain with each of the evaluated materials and bioassayed for different intervals extended form 3 to 30 days. The insecticidal activity was expressed in terms of the adult insects-mortality due to the contact with the tested materials. The effect of these materials on the inspected damage in rice grains and/or resulted F_I progeny post-treatment had been investigated and expressed in terms of productivity inhibition percentage (PIP).

The complete control of treated insects (100% kill) was recorded at the doses of 0.25 and 0.5 g of black pepper fine dust/50 g rice grains after 21 and 14 days post exposure, consequently. Furthermore, at the higher dose of 0.5 g material/50 g rice grains, camphor and black pepper pronounced complete control of the exposed adult-insects giving 100% kill after an exposure period of 30 days, while Latania fine dust was the least efficient tested material.

The results were also confirmed by the deduced dependent positive strong relationship between the recorded insect mortality and the time needed to cause 50 % kill of the exposed insects (LT $_{50}$) at all the different used doses. Based on LT $_{50}$ values, the superior insecticidal effect was recorded for black pepper, followed by orange peel, then camphor and the other tested materials. Meanwhile, clove and Latania fine dusts were the least efficient materials at all used doses (w/w).

The results proved that camphor fine dust was the most potent material in preventing grains perforation and reducing the damage that can be caused by the adult-weevils of S. oryzae followed by black pepper fine dust. Although, the clove fine dust was less toxic, it showed a potent delayed and completely inhibited the raise of F_I progeny post-parents treatment (100%) at its higher tested dose of 0.5 g powder /50 g rice. There was also a good correlation between the insecticidal activity of black pepper fine dust, either picked up or contacted the exposed insects and its effect on prevention or reduction of grains damage and/or its ability in suppressing the raised individuals of F_I progeny.

INTRODUCTION

Rice is considered to be the main dish among the food consumed by the people in Africa, Asia and other

parts of the world. Rice is subjected to the infestation with different insects. The rice weevil Sitophilus oryzae (L.) (Coleoptera: Curculionidae) is the main insect-pest attacking rice grains and the main source of damage. It attacks most cereal grains (rice, sorghum, wheat, barley and maize) before harvest and in store. It also facilitates the infestation with the other secondary insects and moulds. In badly infested grain, the endosperm may be completely consumed by larvae and adults, leaving only the shell of the grain perforated by feeding and emergence holes (Kranz et al., 1977). Prevention and control of these insect-pests will remain an important objective of agrochemical research. Pesticides are currently the principal method for pest control. An increasing public awareness of the potential contamination and the threat possessed from using pesticides to the environment and to human has motivated the researchers in the different countries to investigate the effects of these chemical insecticides on human, environment, secondary pest outbreaks and development of insects resistance. Chemical pesticides may also contaminate the environment by accumulating as residues in soil, ground water and crops (Levitan et al., 1995; Giannessi et al., 2002; Wauchope et al., 2002); in addition to their adverse effect on the biological agents (parasites and predators). The increasing concern over the level of pesticide residues in food has encouraged researchers to look for alternatives of synthetic pesticides. Plants can be an effective replacement for chemical insecticides to protect stored seeds (Rajapakse, 2006). Botanical powders were evaluated for their effectiveness against certain stored products- insects. Rao et al. (1990) evaluated the efficiency of Sweet flag, ginger, 'jamun' (Syzygium cumini), 'Neem' & mango powders, 'Neem' & sesame cake, sunflower, groundnut & palm oils and ordinary ashes against the Callosobruchus chinensis on pigeon peas. Dales (1996) reviewed the use of plant materials for the protection of stored products, against insect pests. He reported that 11 species of the genus Acorus had a potential usefulness as stored product protectants. The protein-rich pea flour was found to act as an antifeedant and a repellent and was toxic to the rice weevil, Sitophilus oryzae (L.) (Hou and Taylor, 2006).

Nirjara et al. (2010) evaluated the pulverized leaves of Punica granatum (Pomegranate) and Murraya koenigii (Curry trees) for their efficacy against the stored grain pest Tribolium castaneum (Herbst) under controlled laboratory conditions. Five different concentrations of leaf powders (0.05, 0.1, 0.15, 0.5 & 1.0 g/10.0 g wheat grains) were tested against the above mentioned common pest of stored grains for their insecticidal and seed protective properties. Both plant leaf powders have resulted in high mortality, delayed the development and causing a subsequent significant population reduction. Their study suggested that leaves from these trees have insecticidal properties and could be employed as alternatives for chemical pesticides. Ahmed and Din (2009) investigated the effect of leaf powders of Basil (Ocimum basilicum L.), Lantana (Lantana camara L.) and Gardenia (Gardenia jasminoides Ellis) on the biology of Callosobruchus chinensis.

The aim of the present study was focusing on the biological performance of certain botanical fine dusts, ash and sulfur powders against the rice weevil *S. oryzae*. The reduction of caused damage by the tested insect and their effect on the resulted progeny was also investigated.

MATERIALS AND METHODS

Insect

Susceptible strain of *Sitophilus Oryzae* (L.) was obtained from an established laboratory culture reared on disinfested rice grains at ambient conditions of 28 ± 2 °C and $75 \pm 5\%$ R.H in a grain storage research laboratory, Faculty of Agric. (Saba Basha), Alex. Univ., Alexandria, Egypt. The insect was reared for several generations and the age of the adults that have been used for the test was about 7-14 days.

Preparation of the botanical dusts

Black pepper seeds, camphor leaves, ash, dried peels of orange shell, cloves seeds, Latania leaves and sulfur powders were in this investigation. The dried parts of the tested plants (Table 1) were ground and sieved with a 100-mesh sieve to obtain fine dusts before the application to the grains.

Evaluation of tested botanical fine dusts, ash and sulfur against S. oryzae

Rice samples (50 g) were weighed into 1 lb jam jars in an airtight at 25°C for 2 weeks. Samples of the tested botanical dusts were weighed.

Different concentrations of the tested dusts were added to rice the (0.1, 0.25 and 0.5 g/ 50 g rice [w/w]), the jars covered with a piece of Clingfilm® and a lid, and then tilted up and down and rotated by hand for 2 minutes. The jars were kept closed for a few minutes after shaking to allow any free dust to settle. Adult

individuals of S. oryzae (1-2 weeks old) were added (10 insects) to the treated rice and each treatment was replicated four times. The control treatment comprised also 4 replicates containing non of the tested materials. All the jars were then kept in an incubator adjusted at the conditions of 28 \pm 2° C and 70 \pm 5% R.H. Mortality counts were done after 3, 7, 14, 21 and 30 days posttreatment. The insects were categorized as alive or dead (brittle, and showing no movement over a 5 min observation period). The dead insects were removed from the treated rice grain and the survived insects were allowed to oviposite on the grains for another 14 days. The insects were totally removed after that and the resulted progeny was then counted. Progeny reduction percentage in those treated grains compared with the untreated check was calculated according to the formula of Aldryhim (1990) as follows:

$$\begin{array}{c} C-T \\ \text{Progeny reduction\%} = & \begin{array}{c} C-T \\ \end{array} \quad X \; 100 \; , \; \; \text{where} \\ C \end{array}$$

C= No. of emerged adults in control, and T= No. of emerged adults in a treatment

The damage percentage (PD) and weevil perforation index (WPI) of the weevils to the grains were calculated using the methods of Adedire and Ajayi (1996) and Fatope *et al.* (1995), respectively.

PD = <u>Total number of treated grains perforated</u> x 100 Total number of grains

WPI =
$$\frac{\% \text{ of treated grains perforated}}{\% \text{ of control grains perforated}} x 100$$

The means were calculated and the obtained data were statistically analyzed using Analysis Of Variance (ANOVA) to check the significance of differences between treatments.

RESULTS AND DISCUSSION

Response of *S. oryzae* to the tested botanical powders, ash and sulfur

Mortality results of *S. oryzae* adults post 3,7,14 and 21 days after being exposed to rice grains treated with different concentrations of the tested botanical and sulfur powders (0.1, 0.25 and 0.5 g/50 g rice grains) are given in Table 2. The response was varied with the tested material, exposure time, and concentration, adult mortality increased as the concentration and exposure time increased. The treatments of the orange and black pepper fine dusts at the rate of 0.1 g/50 g rice grains were capable to induce cumulative mortality percentages of 52.5 and 40.0%, respectively after 21 days. The clove and Latania fine dusts at 0.1 g/50 g rice gave the lowest mortality after 21 days post-treatment (10.0 and 7.5%, respectively).

abici. The chosen parts of the tested botanical materials						
Common name	Scientific name	Parts Used				
Camphor	Cinnamomum camphora	Leaf				
Black pepper	Piper nigrum	Fruit				
Cloves	Syzygium aromaticum	Seed				
Orange	Citrus sinensis	Fruit Peel				
Latania	Lantana camara L.	Leaf				

Table1. The chosen parts of the tested botanical materials

The highest mortality (complete control) (100%) was recorded at the doses of 0.25 and 0.5 g of black pepper powder /50 g rice after 14 and 21 days post exposure. In this concern, Oparaeke (2007) found that the higher concentrations of an extract from West African black pepper, *Piper guineense* (10% and 20% w/v) and more frequent applications (4 and 6/week) significantly (*P* < 0.05) reduced the numbers of two insect pests (*Vigna unguiculata* and *Maruca vitrata* as two pests of post-flowering period of cowpea) compared to the untreated control in two cropping seasons. Pod damage was significantly reduced and grain yields consequently increased in treated plots compared with the untreated control.

Figure 1 illustrates the cumulative mortality of the rice weevil S. oryzae individuals exposed to different concentrations of the previously mentioned tested materials after 30 days of exposure. Weevil mortality was dosage-dependent and it is noticed that as the concentration of the tested material increases, the mortality also increases. At the low dose of 0.1 g material/50 g rice, the most effective material was orange peel followed by black pepper and sulfur giving mortality percentages \geq 60 while the mortality of the other tested materials was <60%. At the higher concentration (0.5 g material/50 g rice), both of camphor and black pepper pronounced complete control of the tested insect giving 100% kill after an exposure period of 30 days. The least efficient material was Latania which gave the lower mortalities at the different conducted concentrations compared with the other tested materials. Therefore, the insecticidal activity of the tested materials after 30 days of exposure at 0.5g material/50 g rice can be arranged as follows: black pepper = camphor > ash > orange peel > sulfur > cloves> Latania. Rao et al. (1990) evaluated the efficiency of Sweet flag, ginger, 'jamun' (Syzygium cumini), 'Neem' & mango powders, 'Neem' & sesame cake, sunflower, groundnut & palm oils and ordinary ashes against the Callosobruchus chinensis on pigeon peas. Acorus calamus was the most effective, followed by the 'Neem' kernel powder, while the 'Neem' cake and ashes were the least effective tested materials. Ofori et al. (1998) reported that camphor applied either topically, impregnated on filter papers or whole wheat and maize grains was highly toxic to all the four tested species (*S. granarius*, *S. zeamais*, *Tribolium castaneum* and *Prostephanus truncates*).

The results in Table 3 confirm the detected toxic efficiency of tested botanical fine dusts against the rice weevil S. oryzae in comparison with ash and /or sulfur powder. In general, the results showed a dependent-positive strong relationship between the recorded insect mortality and the needed time (days) to kill 50% of the exposed insects (LT₅₀) and the used concentrations of investigated materials. As the concentration of each of the tested materials increases, the LT₅₀ values decrease. These values were ranged between 22.35 – 47.57 days at concentration rate of 0.10 g/50 g rice; and 12.67 & 23.83 at 0.5 g/50 g rice.

The superior toxic effect against the tested insect based on LT_{50s} was obtained for the fine dust of orange peel followed by black pepper indicating LT₅₀ values of 22.35 and 24.82 days, respectively. Increasing the dose of black pepper up to 0.25 and 0.5 g/50 g rice gave the shortest time (LT₅₀) comprised 15.69 and 12.67 days, in respect; followed by orange peel fine dust (21.96 days at the dose of 0.25 g/50 g rice). Meanwhile, the LT_{50s} of camphor, ash, orange peel and sulfur at a dosage rate of 0.5 g/50 g rice were 19.65 -20.69 days. Cloves and Latania fine dusts were proved to be the least efficient tested materials against the treated weevil individuals at all the used concentrations (w/w), despite they showed more increased toxicity at the higher dose of 0.5 g/50 g rice giving a range of LT₅₀ values of 23.19-23.83 days (Table3).

The effect of the tested materials on rice seeds damage

The effect of the tested botanical fine dusts, ash and sulfur powders on the damage caused by these exposed insects to treated rice grains with the tested materials is shown in Table 4. The efficiency of the tested materials can be determined in terms of the amount of the food (rice grains) they consume and as that amount of consumed (perforated) grains decreases, the efficiency of the tested material increases. It is also noticed that as the concentration of the tested material increases the number of perforated grains decreases. The tested materials might have their repellent and lethal effects

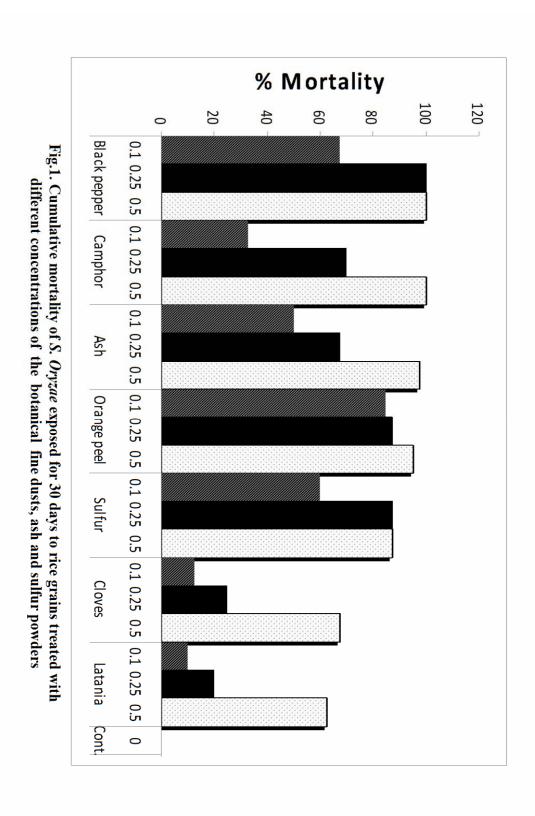
Table 2. Response of the rice weevil Sitophilus oryzae to the different concentrations of tested botanical fine dusts, ash and sulfur at different intervals (days)

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Concentration (g/50 g rice grains)			0.1			0.	0.25			0	0.5	
Days post-treatment												
/	3	7	14	21	သ	7	14	21	သ	7	14	21
Treatment												
Black pepper	0.0	5.0±0.2	25.0±0.7ª @	40.0 ± 0.8^{ab}	2.5±0.2	15.0±0.5ª	52.5±1.1ª	100.0 ± 0.5^{a}	5.0±0.5	55.0 ± 1.7^{a}	100.0 ± 0.8^{a}	100.00
Camphor	0.0	5.0±0.2	17.5±0.4 ^{abc}	25.0±0.2°	2.5±0.2	10.0 ± 0.4^{ab}	27.5 ± 1.1^{bc}	45.0±0.2 ^{bc}	5.0±0.5	20.0 ± 0.2^{b}	42.5±1.0 ^b	70.0 ± 0.2^{a}
Ash	0.0	5.0±0.2	20.0 ± 0.6^{ab}	30.0±0.2 ^{bc}	2.5±0.2	10.0 ± 0.5^{ab}	27.5±0.7 ^{bc}	45.0±0.2 ^{bc}	5.0±0.64	20.0±0.4 ^b	40.0±0.7 ^b	67.5±0.2ª
Orange peel	0.0	7.5±0.4	32.5 ± 1.4^{a}	52.5±0.2ª	2.5±0.2	10.0 ± 0.2^{ab}	35.0±0.5 ^b	55.0±0.2 ^b	5.0±0.5	20.0±0.2 ^b	40.0±1.6 ^b	67.5±0.6ª
Sulfur	0.0	5.0 ± 0.5	22.5 ± 0.8^{ab}	35.0 ± 0.2^{bc}	2.5±0.2	10.0 ± 0.4^{ab}	32.5 ± 0.8^{bc}	55.0±0.4 ^b	5.0±0.2	20.0±0.2 ^b	37.5 ± 1.0^{b}	62.5±0.4 ^{ab}
Cloves	0.0	2.5 ± 0.2	7.5 ± 02^{bcd}	10.0 ± 0.2^{d}	0.0	7.5 ± 0.2^{ab}	17.5±0.2 ^{cd}	22.5±0.4 ^{cd}	2.5±0.2	15.0±0.6 ^{bc}	30.0±0.4 ^b 50.0±0.4 ^b	50.0±0.4 ^b
Latania	0.0	0.0	2.5±0.2 ^{cd}	7.5 ± 0.2^{d}	0.0	5.0±0.5 ^{ab}	10.0±0.2 ^{de}	15.0 ± 0.2^{d}	2.5±0.2	15.0±0.8 ^{bc}	27.5±0.2 ^b	47.5±0.6 ^b
Control	0.0	0.0	0.0 ^d	0.0 ^d	0.0	0.0 ^b	0.0°	0.0^{d}	0.0	0.0°	0.0°	0.0°
Significance		NS	*	*	NS	*	*	*	NS	*	* *	*
@ Means followed with same letter(s) are not significantly different # NS= Not Significant * = Significant and ** Highly Significant	me letter(s)) are not sion	ificantly differen	nt # NS= Not	Sionificant	* = Sionificar	t and ** High	ly Sionificant				

feans followed with same letter(s) are not significantly different. # NS= Not Significant, * = Significant and ** Highly Significant

Table 3. Toxicity of five botanical fine dusts, ash and sulfur against the rice weevil S. oryzae

				Concent	Concentration (g/50 g rice)	g rice)			
Material		0.10			0.25			0.50	
	LT_{50} (Days)	Slop (b)	Correlation	$ ext{LT}_{50} ext{(Days)}$	Slop (b)	Correlation	${ m LT}_{50} \ m (Days)$	Slop (b)	Slop Correlation (b)
Black pepper	24.82 ± 0.45	0.046	0.94	15.69 ± 0.58	0.084	0.95	12.67±0.97	0.063	0.80
Camphor	32.57 ± 0.39	0.033	0.91	23.68 ± 0.35	0.050	0.93	19.65 ± 0.33	0.044	0.92
Ash	27.85 ± 0.38	0.041	0.94	23.83 ± 0.35	0.049	0.93	19.95 ± 0.32	0.043	0.92
Orange peel	22.35 ± 0.41	0.044	0.94	21.96 ± 0.38	0.054	0.93	20.04 ± 0.32	0.043	0.92
Sulfur	26.01 ± 0.41	0.044	0.94	22.07 ± 0.36	0.054	0.94	20.69 ± 0.32	0.042	0.92
Cloves	47.57±0.98	0.029	0.91	40.42 ± 0.28	0.021	0.89	23.19 ± 0.43	0.047	0.89
Latania	47.44 ± 1.30	0.036	0.92	43.49 ± 0.17	0.026	0.97	23.83 ± 0.42	0.045	0.89



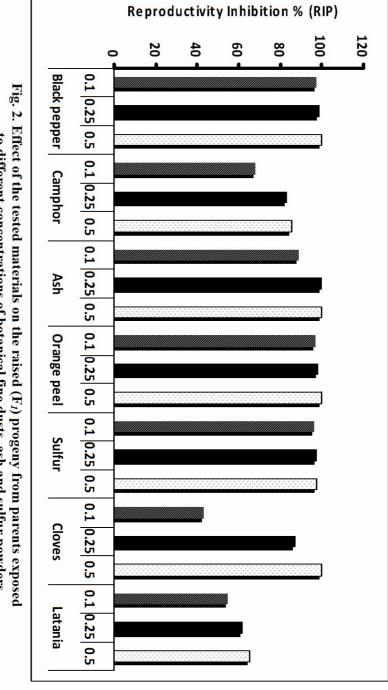


Fig. 2. Effect of the tested materials on the raised (F_I) progeny from parents exposed to different concentrations of botanical fine dusts, ash and sulfur powders

Table 4. Grain damage percentage caused by *S.oryzae* exposed to grains treated with certain botanical fine dusts, ash and sulfur

Treatment	Conc. (g/ 50 g rice grains)	Total No. of grains	No. of perforated grains	No. of Imperforated grains	% grain damage	WPI*
Black pepper	0.10	1801	17	1784	0.94	13.00
	0.25	1776	14	1762	0.78	11.00
	0.50	1755	8	1747	0.45	7.00
Camphor	0.10	1771	16	1695	0.93	13.40
	0.25	1790	7	1783	0.39	6.28
	0.50	1803	4	1799	0.22	3.66
Ash	0.10	1817	23	1794	1.26	17.83
	0.25	1834	12	1822	0.65	10.08
	0.50	1788	10	1778	0.55	8.75
Orange peel	0.10	1711	24	1687	1.40	19.00
	0.25	1807	14	1793	0.77	11.00
	0.50	1817	11	1806	0.60	9.00
Sulfur	0.10	1760	27	1733	1.53	20.00
	0.25	1784	24	1760	1.34	18.00
	0.50	1731	19	1712	1.09	15.00
Cloves	0.10	1793	23	1770	1.28	18.03
	0.25	1820	15	1805	0.82	12.38
	0.50	1809	13	1796	0.71	10.97
Latania	0.10	1815	21	1794	1.15	16.50
	0.25	1819	17	1802	0.93	13.80
	0.50	1811	14	1797	0.77	11.70
Control	0.0	1715	100	1615	5.83	50.00

^{*} WPI = Weevil Perforation Index, where a value above 50 is an indication of negative protectant ability.

which prevent the grains consumption. Camphor was found to be the most effective material to reduce the number of the perforated grains followed by black pepper. On the other hand, sulfur was the least efficient material which allowed the insects to perforate more grains compared with the other tested materials. Nevertheless, the calculation of the Weevil Perforation Index (WPI) showed that all the tested materials had positive protection ability since that index appeared to be less than a value of 50 according to Adedire and Ajayi (1996) & Fatope *et al.* (1995) 's formula. In this respect, it could be said that camphor was the most potent material to prevent grains perforation and to reduce the damage that can be caused by the rice weevil *S. oryzae* followed by black pepper.

The effect of tested materials on the resulted (F_I) progeny from treated parents

The effect of the tested materials on the raised (F_I) progeny resulted from treated parents expressed as progeny inhibition percentage is illustrated in Fig.2. Progeny production was significantly reduced due to the tested materials. It is observed that the number of offspring (adult-insects) of *S. oryzae* emerged from the treated food, on which parent adults were placed, during and after treatment with the all tested materials were

considerably lower than those emerged from the untreated controls. This response was correlated with the increase of the dose. Although, cloves powder did not show a higher insecticidal effect against S. oryzae, it was found to have a high delayed effect and completely inhibited the (F_I) progeny (100%) at the higher tested dose of 0.5 g powder /50 g rice.

On the other hand, the results showed that the least inhibition percentage of F_I adults (43.5), emerged from rice grains treated with cloves at 0.1g /50 g rice, as compared to others. It could be mention that the inhibition percentage depends on the concentration of the tested material. Meanwhile, black pepper and orange peel were found to be effective in reducing and inhibiting the resulted progeny recording an inhibition percentage \geq 95% at the lower tested concentration of 0.1 g powder /50 g rice.

The abovementioned results are in agreement with those of Zewde and Jembere (2010) who evaluated the efficacy of products of orange (*Citrus sinensis*) peels for controlling the stored products beetle *Zabrotes subfasciatus* (L) in stored haricot beans. Powders from ground peels caused significant reduction in progeny emergence of *Z. subfasciatus* (P< 0.05). Also, the results are in accordance with Ashouri and Shayesteh

(2010) who reported that black pepper at 0.5% concentration caused 100% mortality of S. granarius in the first five days. Also R. dominica showed complete mortality at 5% level after 14 days and caused complete reduction in F_I progeny of S. granarius at highest tested doses. Black pepper significantly reduced F_I progeny emergence of R. dominica.

There was a good correlation between the insecticidal activity of black pepper picked up or contacted the exposed individuals and its effect on prevention or reducing grains damage and its ability to suppress the resulted F_I progeny. The F_I progeny might be also affected due to the small number of the alive insects since some of these tested materials caused a complete mortality of the parents. Again, Latania was the least efficient material compared with the other tested ones recording progeny inhibition percentages of 65, 62 and 55 at the concentrations of 0.1, 0.25 and 0.5 g /50 g rice, respectively. Sulfur was found to reduce F_I progeny production by 96.1–97.4%.

It could be said that oviposition, the postembryonic development and adult emergence of *S. oryzae* might be adversely affected when the parents were reared on rice treated with the tested materials.

The results indicate that camphor and black pepper could be used as alternative to chemical insecticides to control S. orvzae. Further investigations are needed to make better use of these materials through the chemical analysis to identify the main components. Therefore, it could be said that the tested botanical powders had a potential usefulness as stored product protectants and can act as safe and effective alternatives to the chemical insecticides since they have insecticidal effect, reducing grains damage and suppressing the new generations. Asawalam et al. (2007) found that the powders of Piper guineense and Capsicum frutescens, tested against S. zeamais had the highest percentage mortality (79.8 and 75.1, respectively). The tested powders significantly (P> 0.05) reduced adult emergence, grain damage and weight loss in the various treatments.

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