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Repellent effect of six medicinal plants against *Tribolium castaneum* (Herbst)

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Abstract
Red flour beetle, *Tribolium castaneum* is a cosmopolitan insect damaging stored food products and cause severe loss. The use of chemical insecticides leads to resistant development in insects and not safe for environment. An alternative method is required to manage this insect without ill-effect to the human being and environment. The botanicals having repellent or insecticidal property will be an alternative method of managing this insect. The acetone and aqueous extracts of six medicinal plants *viz*., *Aloe vera* (Kattarvazha), *Ocimum tenuiflourm* (Tulsi), *Azadirachta indica (*Neem) *Thymus vulgaris (*Thyme) *Piper betel (*Betel leaf) *Acalypha indica (*Kuppaimeni) against *T. castaneum* (Herbst) (Coleoptera: Tenebrionidae) were evaluated for their repellent activities. The various concentrations (2, 4 and 8%) of the plant extracts were used to conduct repellency test using area preference test of adult beetles after fixed period intervals (4, 6 and 8 h). The results revealed that the aqueous and acetone extract of Neem and Kuppaimeni were shown more repellency (90 – 100%) against adult insects. Thus, Gas Chromatography Mass Spectroscopy analysis has been performed for the aqueous extract of these two most repellent plants. The aqueous extract of neem containing Phytol (47.72% peak area) can be considered as repellent. Kuppaimeni leaves containing Myo-Inositol, 4-C-methyl- (24.61% peak area) can also be a promising repellent. These findings confirmed that plant extracts can be a suitable alternative for chemicals in managing stored product insects.

Keywords: medicinal plants, *T. castaneum*, GC-MS analysis, repellent activity

Introduction
Insect infestation is one of the foremost reasons of the losses incurred during processing and storage of grains and their products. Hashemi and Safavi (2012) reported stored food product losses due to insect infestation accumulates to 5-10% and 20–30% in temperate regions and tropical regions, respectively. The main insects which infest the stored product commodities belongs to coleopteran pests with more than 600 species (Yadav, Anand, Sharma and Gupta, 2014), and causes a quantitative loss of 20–30%. Among them, the red flour beetle *T. castaneum* (Coleoptera: Tenebrionidae) is the most dangerous one which is commonly found in food lavatories, such as milling industry, food processing plants, storerooms and retail stocks (Yar, *et al*., 2017)(Scheff *et al.*, 2018). The red flour beetle have been reported to infest and damage a wide variety of commodities (Shafique *et al.*, 2006; Atta *et al*., 2020). One female *T. castaneum* lays 300-400 eggs during its life cycle, which results in heavy infestation if proper control measures were not adopted (Ridley *et al*., 2011). The adults secrete a hazardous chemical (benzoquinone) proving unwanted odor and it also encourages mold growth in flour.

In the past few decades, human population has been growing and it is likely to come near 2.4 billion by 2050 (Modi *et al*., 2017). Simultaneously, there will be increase in food demand. Therefore, measures that safeguard stored food from insect damage can increase the availability of food products for consumption. The toxic chemical insecticides have been used for controlling stored grain insects. But, these chemical insecticides have serious drawbacks on public health and environment. It was also found that the insects developed resistance to insecticides. Therefore, it is necessary to find alternate methods that are readily available, affordable, less toxic to human and safe to the environment (Atta *et al.*, 2020; Rizwan *et al.*, 2019).

Plants including neem, nicotiana, spices like turmeric, clove, cinnamon, ginger, garlic, star anise were used for the management of field insects (Kiruba *et al.*, 2008; Paul *et al.*, 2009; Thacker, 2002). The secondary metabolites are produced by the plants for defence against different insects and hence these plants were found as possible alternative for insect management (Isman and Akhtar, 2007; Mangang *et al.*, 2020). Plant extracts can impact the activities of various storage pests as it possess insecticidal properties to protect storage food products from insects (Belmain and Stevenson, 2001) (Isman, 2000). The essential oils and extracts obtained from plant origin were found to be rich in mixtures of various metabolites and complex volatiles which are natural and safe (Bakkali, *et al.*, 2008). Many phytochemicals have proved their effectiveness against *T. castaneum* (Mangang *et al.*, 2020; Sagheer *et al.*, 2011).

Botanical insecticides are formulations of extract or derivative of plants such as *Azadirachta indica*, *Derris elliptica* (Sae-Yun *et al.*, 2006; Soujanya *et al.*, 2016), *Chrysanthemum cinerariafolium* (Shawkat *et al.*, 2011). Some of the compounds / metabolites of plants are toxic to insects (pyrethrum, nicotine, rotenone), acting as repellents, antifeedants (azadirachtin, rape seed extract) and also act as sterilant (α-asarone) (Ignatowicz and Wesołowska, 1994). In fact, plants contain bioactive metabolites, which act as antifeedants, repellents, and toxicants against a wide range of insects that attack stored products (Manickam *et al.*, 2021; Rajendran and Sriranjini, 2008). In the present study, the repellency of six medicinal plant extracts was screened against red flour beetle, *T. castaneum*.

Materials and Methods

Collection of plants

Fresh leaves of six medicinal plants *Aloe vera* (Kattarvazha), *Ocimum tenuiflourm* (Tulsi), *Azadirachta indica (*Neem), *Thymus vulgaris (*Thyme), *Piper betel (*Betel leaf), *Acalypha indica (*Kuppaimeni) were obtained from the local markets of Thanjavur, Tamil Nadu, India (Table 1). They were used for the repellency study against *T. castaneum.*

Table 1: Description of six medicinal plants used in the study and their parts used for extraction

Sl. No	Local name	Scientific name	Family	Part used
1	Kattarvazha	*Aloe vera*	Liliaceae	Leaves
2	Tulsi	*Ocimum tenuiflourm*	Lamiaceae	Leaves
3	Neem	*Azadirachta indica*	Meliaceae	Leaves
4	Thyme	*Thymus vulgaris*	Lamiaceae	Leaves
5	Betel leaf	*Piper betel*	Piperaceae	Leaves
6	Kuppaimeni	*Acalypha indica*	Euphorbiaceae	Leaves

Extraction of plants

The fresh leaves (50 g) of each plants collected were washed and ground in a mixer with an equal quantity of water (1:1). Similarly, the extract was prepared with acetone in the same amount separately. The ground mixture was stored overnight in a refrigerator maintained at 4°C and filtered using a muslin cloth followed by centrifugation (5000 rpm) and filtration (Whatman filter paper). The filtrate was kept in a 100 ml storage container and kept in refrigerator (0-8°C) for the studies to be conducted. The extract was used as stock solution (Mansoor-ul-Hassan *et al.* 2005) and Sagheer *et al.* 2013).

Insect culture

The mother culture of *T. castaneum* was an original strain that was nurtured for several years at the laboratory in Indian Institute of Food Processing Technology (IIFPT), Thanjavur. These colonies were conserved in the laboratory and they were never exposed and contaminated with any form of insecticides. The adults used in the experiments were reared in 1 L glass container and ventilated by covering with muslin cloth. Each container has a mixture (10:1, $^{w}/_{w}$) of wheat flour and brewer's yeast. Fifty Adults of red flour beetle were released in each container and allowed for egg laying for 24 hrs. Then the adults were removed from the container. The young larvae emerged from eggs and grown in the food material. These larvae were found to have uniform age and size which were used for conducting repellency experiments. The cultures were maintained in ambient condition.

Bioassay repellent activity

The repellency test was conducted by using 2, 4 and 8 per cent concentration of the extracted medicinal plants based on the preliminary studies. Filter paper bioassay using area preference methodology was used to assess the repellent effect of the medicinal plants against adults of *T. castaneum* (Mohan and Fields, 2002). Whatman filter papers (9cm dia.) were cut into halves and one half was applied with the extract while the other was treated with the respective solvent used in extraction. They were dried at room temperature for 30 min, and reattached with their respective half giving 0.5cm spaces in between (Fig.1). *T. castaneum* adults (10 in no.*)* with same age collected from the culture after 3 days of emergence were set on loose in the petri plate and closed. The experiments were performed at ambient conditions and covered with clothes and tray to maintain darkness. The observations were taken on the presence of insects in each half of the filter paper at 4, 6 and 8 hrs. The experiment was replicated three times and the results were given as the percentage of insects present in the untreated part.

$$P = [(Nu - Nt) / (Nu + Nt)] \times 100$$

Where,

P-%repellency; Nu - Number of insects present in untreated area;

Nt - Number of insects present in treated area.

Fig 1: Filter paper bioassay (a) Treated halves of the filter paper before drying (b) Setup of treated half (light yellow) with untreated half (white) inside petri plates

GCMS analysis

The active components of botanicals having more repellency were characterised using Gas Chromatography Mass Spectroscopy (GC-MS) techniques which is a common tool for initial screening which provides spectral output of all the compounds that gets separated from the sample (Hori *et al.,* 2019). The extract was freeze dried (Lark 4kg model) and 1g of the freeze-dried extracts was transferred into a 100 ml volumetric flask and added 25 ml of ethanol. The mix was vortexed for 15 minutes. The contents were centrifuged (Make: Remi; Model: R-8C BL) at rpm and the supernatant liquid was injected in the GC (Make: Bruker; Model: Scion 436-GC). The obtained mass spectra from the GC-MS were compared with the NIST library. The peak area, retention time and molecular formula were used for the confirmation of the presence of phytochemicals (Mangang *et al.,* 2020).

Data analysis

The experiments were laid out in Minitab software. Lately, Turkey's test was conducted to check the significance and determine the plants having the highest repellency. The reported means and standard deviation are of original data given in percentage (Mangang *et al.,* 2020).

Results and Discussion

Repellent effect of six medicinal plant extracts against adults of *T. castaneum* in filter paper test

Effect of aqueous and acetone extract (2%) of plants against T. castaneum

The results showed that the acetone extract (2%) of Kuppaimeni leaves gave the highest repellency (98.33%) against *T. castaneum* after 8 hrs of treatment (p<0.05). Nevertheless, the aqueous extract (2%) of neem (93.33%) and Kuppaimeni leaves (91.67 %) gave significantly different repellent action after 6 hrs of treatment (p<0.05).

However, there is no significant difference between the aqueous plant extracts after 8 hrs of treatment (p<0.05), even with neem leaves obtaining 96.67 per cent repellency (Table 2).

Table 2: Repellency of plant extracts (2%) at various exposure periods (4, 6 and 8 hrs) against adults of *T. castaneum*

Plant extract	Aqueous extract (2%)			Acetone extract (2%)		
	4 hrs	6 hrs	8 hrs	4 hrs	6 hrs	8 hrs
Kattarvazha	73.33±15.28[a]	70.00±10.00[b]	80.00±10.00[a]	76.67±11.55[a]	76.67±11.5[a]	66.67±5.77[c]
Tulsi	80.00±10.0[a]	83.33±2.89[ab]	86.67±5.77[a]	73.33±5.77[a]	90.0±0.00[a]	86.67±2.89[b]
Neem	86.67±5.77[a]	93.33±2.89[a]	96.67±5.77[a]	76.67±5.77[a]	83.33±5.77[a]	95.00±0.00[ab]
Thyme	70.00±0.00[ab]	86.67±5.77[a]	83.33 ±5.77[a]	76.67±5.77[a]	90.0±10.0[a]	90.00±0.00[ab]
Betel leaf	80.00±0.00[a]	85.00±5.00[ab]	90.00±0.00[a]	76.67±5.77[a]	86.67±15.28[a]	90.00±0.00[ab]
Kuppaimeni	80.00±10.00[a]	91.67±2.89[a]	93.33±5.77[a]	86.67±5.77[a]	93.33±5.77[a]	98.33±2.89[a]
Control	46.76±5.77[b]	36.67±5.77c	43.33±5.77[b]	46.67±5.77[b]	43.33±5.77[b]	46.67±5.77[d]

Values with different letters in the same column differ significantly (p<0.05) Repellent effects given as % Mean repellency ± SD in all treatments. Effect of aqueous and acetone extract (4%) of plants against adults of T. castaneum

The results showed that aqueous extract (4%) of Neem and Kuppaimeni were recorded cent per cent repellency after 8 hrs which are significantly highest among other plant extracts (p<0.05) (Table 3). However, there was no significant difference found in repellency among the aqueous extract of Neem and Kuppaimeni during 4 and 6 hrs of treatment. The cent per cent repellency was also shown with acetone extract of Neem and Kuppaimeni after 6 and 8 hrs of treatment with significant difference (p<0.05).

Table 3: Repellency of plant extracts (4%) at various exposure periods (4, 6 and 8 hrs) against adults of *T. castaneum.*

Plant extract	Aqueous extract (4%)			Acetone extract (4%)		
	4 hrs	6 hrs	8 hrs	4 hrs	6 hrs	8 hrs
Kattarvazha	83.33±15.28[a]	93.33±5.77[a]	81.67±2.89[c]	70.00±0.00[c]	73.33±5.77[c]	78.33±2.89[c]
Tulsi	73.33±5.77[a]	86.67±5.77[a]	90.00±0.00[b]	80.00±0.00[abc]	90.0±0.00[ab]	90.00±0.00[b]
Neem	86.67±5.77[a]	96.67±5.77[a]	100.0±0.00[a]	93.33±5.77[a]	100.0±0.00[a]	100.0±0.00[a]
Thyme	73.33±5.77[a]	86.67±5.77[a]	88.33±2.89[bc]	73.33±5.77[c]	80.0±0.00[b]	91.67±2.89[b]
Betel leaf	80.00±0.00[a]	93.33±5.77[a]	90.00±0.00[b]	76.67±11.55[c]	83.33±5.77[b]	90.00±0.00[b]
Kuppaimeni	90.00±0.00[a]	100.0±0.0[a]	100.0±0.00[a]	90.00±0.00[ab]	100.0±0.00[a]	100.0±0.00[a]
Control	46.67±5.77[b]	43.33±5.77[b]	36.67±5.77[d]	46.67±5.77[d]	43.33±5.77[d]	43.33±5.77[d]

Values with different letters in the same column differ significantly (p<0.05) Repellent effects given as % Mean repellency ± SD in all treatments.

Effect of aqueous and acetone extract (8%) of plants against adults of T. castaneum

The aqueous extracts of Neem and Kuppaimeni showed significant difference after 6 and 8 hrs (p<0.05) with cent per cent repellency (Table 4). Similarly, acetone extracts of Neem and Kuppaimeni gave significantly highest repellency of cent per cent at 6 hrs (p<0.05).

Table 4: Repellency of plant extracts (8%) at various exposure periods (4, 6 and 8 hrs) against adults of *T. castaneum.*

Plant extract	Aqueous extract (8%)			Acetone extract (8%)		
	4 hrs	6 hrs	8 hrs	4 hrs	6 hrs	8 hrs
Kattarvazha	76.67±5.77[a]	80.0±10.00[b]	86.67±5.77[b]	70.00±0.00[b]	80.00±0.00[c]	90.00±0.00[a]
Tulsi	73.33±5.77[a]	80.00±0.00[a]	86.67±5.77[ab]	80.00±0.00[ab]	80.00±0.00[c]	90.00±0.00[a]
Neem	80.00±0.00[a]	86.67±5.77[ab]	90.00±0.0[ab]	80.00±0.00[ab]	90.00±0.00[abc]	96.67±5.77[a]
Thyme	83.33±5.77[a]	100.0±0.00 [a]	100.0±0.00[a]	90.00±0.00 [a]	100.0±0.00[a]	100.0±0.00[a]
Betel leaf	76.67±5.77[a]	86.67±5.77[ab]	93.33±5.77[ab]	73.33±5.77[b]	86.67±5.77[bc]	93.33±5.77[a]
Kuppaimeni	73.33±5.77[a]	90.00±0.00[ab]	90.00±0.00[ab]	73.33±5.77[b]	96.67±5.77[ab]	90.00±0.00[a]
Kattarvazha	90.00±0.00[a]	100.0±0.00[a]	100.0±0.00[a]	90.00±0.00[a]	100.0±0.00[a]	100.0±0.00[a]
Control	46.67±5.77[b]	43.33±5.77[c]	36.67±5.77[c]	46.67±5.77[c]	43.33±5.77[d]	53.33±5.77[b]

Values with different letters in the same column differ significantly (p<0.05)

Repellent effects given as % Mean repellency ± SD in all treatments.

The phytochemicals used in the study were found to be effective against *T. castaneum*. Bouda *et al.* (2001) reported that nearly 2000 plants were found to possess antifeedant, repellent or insecticidal compounds. Sadek (2003) reported that the plants are abundant source of secondary metabolites that can be utilized for fabricating environmentally safe insect management systems for insect control as an alternative to synthetic insecticide. The results of the present study depicted that the phytochemicals have potential repellent property against *T. castaneum* adults in stored commodities (Fig. 2). The present outcomes are found to be in agreement with prior studies where garlic and ginger extracts were proven effective against *Sitophilus zeamais* and *T. castaneum,* respectively (Epidi and Odili, 2009). Similarly, *C. aequalis* were reported to emit strong resistant aromatic odours that repells weevils (Ntonifor and Monah, 2001). It was reported *Piper* spp. extracts were found to possess repellent and insecticidal properties against many pests (Ntonifor and Monah, 2001). The aromatic bio-active metabolites present in the aqueous-ethanolic extracts of *Inula racemose* also possess repellent and insecticidal properties against the stored product insect, *S. oryzae* (Liu *et* *al.*, 2006). Similar results were obtained in the present study for neem leaf extracts. It was also reported that the plant extracts in the form of powder, essential oil or solvent may possess fumigating and repelling property. They may induce the insects with the metabolites present in the phytochemical leading to poisoning in the stomach, coagulation of water thereby blocking the respiratory system or may act as a barrier against insect infestation (Law-Ogbomo and Enobakhare, 2007; Mulungu *et al.*,, 2007). The medicinal plants used in the research were safe with low mammalian toxicity, easily found and grown in nature and thus can be used in insect management. More advance research can be performed in formulating mixtures of active components present in the plant and their mode of effect in management of insects.

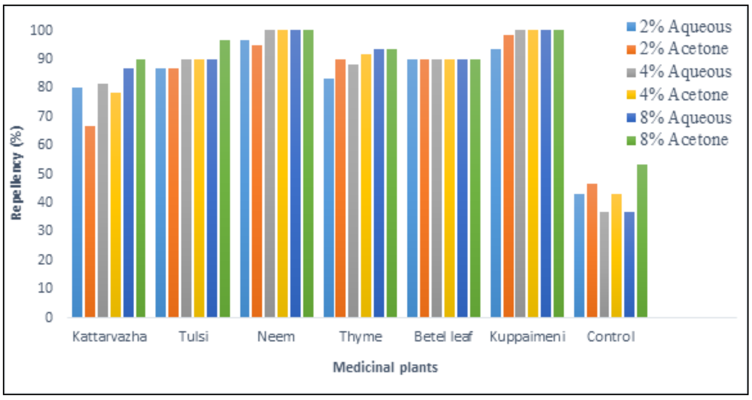

Fig 2: Comparative repellency activity (8hrs) of six medicinal plant extracts at different concentration

Phytochemicals present in Neem and Kuppaimeni

The GC-MS analysis showed that the phytochemicals present in Neem were 1-methoxy-5-hexene, triquinacene and phytol. Among them it was found that phytol has the highest peak area of 47.72 per cent (Table 5). The GC-MS analysis of Kuppaimeni reveals that myo-Inositol, 4-C-methyl-; 1-ethyl-2,6-difluorobenzene; bicyclo [3.1.1] heptane, 2,6,6-trimethyl-, (1.alpha.,2.beta.,5.alpha.)-; n-Hexadecanoic acid; phytol; cis-13-octadecenoic acid; octadecanoic acid were the main active components. It was found that Myo-Inositol, 4-C-methyl- showed the highest peak area of 24.61 per cent followed by 1-Ethyl-2,6-difluorobenzene with 19.87 per cent (Table 6).

Table 5: Major phytochemicals found after GC-MS analysis of neem aqueous extract

Retention time (min)	Phytochemical	Molecular formula	Molecular weight	Peak area (%)
11.47	1-Methoxy-5-hexene	$C_7H_{16}O_2$	116.00	15.85
16.33	Triquinacene	$C_{10}H_{10}$	130.19	4.87
17.40	Phytol	$C_{20}H_{40}O$	296.50	47.72

Table 6: Major phytochemicals found after GC-MS analysis of Kuppaimeni aqueous extract

Retention time (min)	Phytochemical	Molecular formula	Molecular weight	Peak area (%)
13.99	Myo-Inositol, 4-C-methyl-	$C_7H_{14}O_6$	194.18	24.61
14.54	1-Ethyl-2,6-difluorobenzene	C_8H_9F	406.40	19.87
15.13	Bicyclo [3.1.1] heptane, 2,6,6-trimethyl-, (1.alpha.,2.beta.,5.alpha.)-	$C_{10}H_{18}$	138.25	4.24
16.16	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256.42	6.78
17.37	Phytol	$C_{20}H_{40}O$	296.50	5.20
17.58	cis-13-Octadecenoic acid	$C_{18}H_{34}O_2$	282.46	10.42
17.76	Octadecanoic acid	$C_{18}H_{36}O_2$	284.50	4.49

Sagheer *et al.*, (2013) found that the acetone extract of *Nicotiana tabacum* and *Salsola baryosma* showed maximum repellency and larval mortality against *T. castaneum* at higher concentration (10%). In our study, it was found that both acetone and aqueous extract of Neem and Kuppaimeni showed a very good repellent activity. Aqueous being the most polar, easily accessible to the farmers and low cost is highly recommendable than acetone, even though it extracts both polar and non-polar components. Thus, the GC-MS analysis of aqueous extract of Neem and Kuppaimeni was conducted. Although many authors studied the repellency of various botanicals against insects for longer periods (Jayasekara *et al.*, 2005; Mkolo *et al.*, 2011; Wekesa *et al.*, 2011), the present study supported to add new findings and information for developing plant-based formulations for the management of stored product insects.

There has been increased resistance of insects to the existing chemicals used for the stored insect management. The people were aware of the harmful effects and residues formed due to the use of chemicals. The lifestyle and concern about their safety has led to increase preference in chemical free food commodities. The botanical insecticides can be extracted from various plant source and are safe (Freeman and Beattie, 2008). The high volatile nature of compounds in plants possessing insecticidal activity can be effectively utilised for managing stored product insects (Isman, 1997; Regnault-Roger and Hamraoui, 1995). However, further studies are required for developing formulations, evaluations in the real situation and commercialisation.

Conclusion

This present study revealed that aqueous and acetone extracts of six medicinal plant possess the repellent action against *T. castaneum* and could be of potential for utilising in the management of *T. castaneum*. Among the medicinal plants tested, Neem and Kuppaimeni at 2, 4 and 8 per cent showed the good repellent effect against *T. castaneum.* Therefore, leaf extract of Neem and Kuppaimeni can be recommended because they are easy accessibility at farm level, cheap, eco-friendly in nature, safe for humans and also as good alternative to synthetic insecticides.

The effective repellent activities were observed in the plants. In the future, research using these screened botanicals must be carried out and become applicable. For achieving such great heights, on-site studies as well as isolation of the bioactive components must be carried out to endorse the present deductions. After enriching the formulation with the suitable plant species, it is possible to commercialize for the usage of farmers to safe guard the grains free from insect damage in the small and large-scale storages. The botanicals possess potential as a eco-friendly management method to reduce the usage of chemical insecticides in stored grain pest management. Research focusing on simple and effective applicable methods will be needed to utilize the screened botanicals for insect management.

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References

1. Atta B, Rizwan M, Sabir A, Gogi M, Sabar M, Bakhtawar A *et al*. Toxic and repellent characteristics of some plant extracts used against *Tribolium castaneum* (Herbst)(Coleoptera: Tenebrionidae) improve the grain quality of stored wheat. *Journal of Innovative Sciences*,2020:6(1):-11. https://doi.org/10.17582/journal.jis/2020/6.1.1.11
2. Atta B, Rizwan M, Sabir AM, Gogi MD, Ali K. Damage potential of *Tribolium castaneum* (Herbst)(Coleoptera: Tenebrionidae) on wheat grains stored in hermetic and non-hermetic storage bags. *International Journal of Tropical Insect Science*,2020:40(1): 27-37. https://doi.org/10.1007/s42690-019-00047-0
3. Bakkali F, Averbeck S, Averbeck D, Idaomar M. Biological effects of essential oils–a review. *Food and chemical toxicology*,2008:46(2):446-475. https://doi.org/10.1016/j.fct.2007.09.106
4. Belmain S, Stevenson P. Ethnobotanicals in Ghana: reviving and modernising age-old farmer practice. *Pesticide outlook*,2001:12(6):233-238. https://doi.org/10.1039/b110542f
5. Bouda H, Tapondjou L, Fontem D, Gumedzoe M. Effect of essential oils from leaves of *Ageratum conyzoides*, *Lantana camara* and *Chromolaena odorata* on the mortality of *Sitophilus zeamais* (Coleoptera, Curculionidae). *Journal of Stored Products Research*,2001:37(2):103-109. https://doi.org/10.1016/s0022-474x(00)00011-4
6. Epidi T, Odili EO. Biocidal activity of selected plant powders against Tribolium castaneum Herbst in stored groundnut (Arachis hypogaea L.). *African Journal of Environmental Science and Technology*,2009:3(1):001-005. doi: 10.5897/AJEST08.030
7. Freeman BC, Beattie GA. An overview of plant defenses against pathogens and herbivores. *The Plant Health Instructor*, 2008, 94. ttps://doi.org/10.1094/phi-i-2008-0226-01
8. Hashemi SM, Safavi SA. Chemical constituents and toxicity of essential oils of oriental arborvitae, *Platycladus orientalis* (L.) Franco, against three stored-product beetles. *Chilean Journal of Agricultural Research*,2012:72(2):188. https://doi.org/10.4067/s0718-58392012000200004
9. Hori M, Aoki Y, Shinoda K, Chiba M, Sasaki R. Wood volatiles as attractants of the confused flour beetle, *Tribolium confusum* (Coleoptera: Tenebrionidae). *Scientific reports*,2019:9(1):1-8. https://doi.org/10.1038/s41598-019-48073-9
10. Ignatowicz S, Wesołowska B. Potential of common herbs as grain protectans: repellent effect of herb extracts on the granary weevil, *Sitophilus granarius (L).* In *Proceedings of the 6th International Working Conference on Stored-Product Protection*. Canaberra, Australia, 1994, 17-23. http://bru.gmprc.ksu.edu/proj/iwcspp/pdf2/6/790.pdf. Accessed 21 April 2021
11. Isman MB. Neem and other botanical insecticides: barriers to commercialization. *Phytoparasitica*,1997:25(4):339-344. https://doi.org/10.1007/bf02981099
12. Isman MB. Plant essential oils for pest and disease management. *Crop protection*,2000:19(8-10):603-608. https://doi.org/10.1016/s0261-2194(00)00079-x
13. Isman MB, Akhtar Y. Plant natural products as a source for developing environmentally acceptable insecticides. In *Insecticides design using advanced technologies.* Springer, Berlin, Heidelberg, 2007, 235-248. https://doi.org/10.1007/978-3-540-46907-0_10
14. Jayasekara TK, Stevenson PC, Hall DRnd Belmain, SR. Effect of volatile constituents from *Securidaca longepedunculata* on insect pests of stored grain. *Journal of chemical ecolog*y,2005:31(2):303-313. https://doi.org/10.1007/s10886-005-1342-0
15. Kiruba S, Jeeva S, Kanagappan M, Stalin SI, Das SSM. Ethnic storage strategies adopted by farmers of Tirunelveli district of Tamil Nadu, Southern Peninsular India. *Journal of Agricultural Technology*,2008:4(1):1-10. http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.465.6225&rep=rep1&type=pdf. Accesssed 19 April 2021
16. Law-Ogbomo K, Enobakhare D. The use of leaf powders of *Ocimum gratissimum* and *Vernonia amygladina* for the management of *Sitophilus oryzae* (L.) in stored rice. *J. Entomol*,2007:4:253-257. https://doi.org/10.3923/je.2007.253.257
17. Liu C, Mishra A, Tan R. Repellent, insecticidal and phytotoxic activities of isoalantolactone from Inula racemosa. *Crop Protection*,2006:25(5):508-511. https://doi.org/10.1016/j.cropro.2005.05.008
18. Mangang IB, Tiwari A, Rajamani M, Manickam L. Comparative laboratory efficacy of novel botanical extracts against *Tribolium castaneum*. *Journal of the Science of Food and Agriculture*,2020:100(4):1541-1546. https://doi.org/10.1002/jsfa.10162

19. Manickam L, Mangang IB, Rajamani M. Phytochemicals for the Management of Stored Product Insects *Sustainable Bioeconomy*. Springer, 2021, 171-182. https://doi.org/10.1007/978-981-15-7321-7_8
20. Mansoor-ul-Hassan MAS, Sagheer M, Aleem M. Comparative efficacy of ethanol leaf extracts of *Amaranthus viridis* l. and *Salsola baryosma* (schultes) and Cypermethrin against *Trogoderma granarium* (everts). *Pakistan Journal of Agricultural Sciences*,2005:42:3-4.
21. Mkolo N, Sako K, Olowoyo J, Ndlovu S, Magano S. Variation in the repellency effects of the leaves of *Mentha piperita* against adults of *Amblyomma hebraeum*. *African Journal of Biotechnology*,2011:10(55):11426-11432. doi: 10.5897/AJB11.524
22. Modi S, Vargas-Garcia CA, Ghusinga KR, Singh A. Analysis of noise mechanisms in cell-size control. *Biophysical journal*,2017:112(11):2408-2418. https://doi.org/10.1016/j.bpj.2017.04.050
23. Mohan S, Fields PG. A simple technique to assess compounds that are repellent or attractive to stored-product insects. *Journal of Stored Products Research*,2002:38(1):23-31. https://doi.org/10.1016/s0022-474x(00)00039-4
24. Mulungu L, Lupenza G, Reuben S, Misangu R. Evaluation of botanical products as stored grain protectant against maize weevil, *Sitophilus zeamays* (L.) on maize. *Journal of Entomology*,2007:4(3):258-262. https://doi.org/10.3923/je.2007.258.262
25. Ntonifor NN, Monah IM. Use of three spices to protect stored maize against *Sitophilus zeamais*. *Tropical Science*,2001:41(2):74-77.
26. Paul UV, Lossini JS, Edwards PJ, Hilbeck A. Effectiveness of products from four locally grown plants for the management of *Acanthoscelides obtectus* (Say) and *Zabrotes subfasciatus* (Boheman)(both Coleoptera: Bruchidae) in stored beans under laboratory and farm conditions in Northern Tanzania. *Journal of Stored Products Research*,2009:45(2):97-107. https://doi.org/10.1016/j.jspr.2008.09.006
27. Rajendran S, Sriranjini V. Plant products as fumigants for stored-product insect control. *Journal of Stored Products Research*,2008:44(2):126-135. https://doi.org/10.1016/j.jspr.2007.08.003
28. Regnault-Roger C, Hamraoui A. Fumigant toxic activity and reproductive inhibition induced by monoterpenes on *Acanthoscelides obtectus* (Say) (Coleoptera), a bruchid of kidney bean (Phaseolus vulgaris L.). *Journal of Stored Products Research*,1995:31(4):291-299. https://doi.org/10.1016/0022-474x(95)00025-3
29. Ridley A, Hereward J, Daglish G, Raghu S, Collins P, Walter G. The spatiotemporal dynamics of *Tribolium castaneum* (Herbst): adult flight and gene flow. *Molecular Ecology*,2011:20(8):1635-1646. https://doi.org/10.1111/j.1365-294x.2011.05049.x
30. Rizwan M, Atta B, Rizwan M, Sabir AM, Shah ZU. Hussain M. Effect of the entomopathogenic fungus, *Beauveria bassiana*, combined with diatomaceous earth on the red flour beetle, *Tribolium castaneum* (Herbst) (Tenebrionidae: Coleoptera). *Egyptian Journal of Biological Pest Control*,2019:29(1):1-6. https://doi.org/10.1186/s41938-019-0131-y
31. Sadek MM. Antifeedant and toxic activity of *Adhatoda vasica* leaf extract against *Spodoptera littoralis* (Lep., Noctuidae). *Journal of applied entomology*,2003:127(7):396-404. https://doi.org/10.1046/j.1439-0418.2003.00775.x
32. Sae-Yun A, Ovatlarnporn C, Itharat A, Wiwattanapatapee R. Extraction of rotenone from *Derris elliptica* and *Derris malaccensis* by pressurized liquid extraction compared with maceration. *Journal of Chromatography A*,2006:1125(2):172-176. https://doi.org/10.1016/j.chroma.2006.05.075
33. Sagheer M, Ali K, Rashid A, Sagheer U, Alvi A. Repellent and toxicological impact of acetone extracts of *Nicotiana tabacum*, *Pegnum hermala*, *Saussurea costus* and *Salsola baryosma* against red flour beetle, *Tribolium castaneum* (Herbst). *Pakistan Journal of Zoology*,2013:45(6):1735-1739. doi:0030-9923/2013/0006-1735
34. Sagheer M, Hasan M, Ali Z, Yasir M, Ali Q *et al*. Evaluation of essential oils of different citrus species against *Trogoderma granarium* (Everts)(Coleoptera: Dermestidae) collected from Vehari and Faisalabad districts of Punjab. *Pakistan Entomologist*,2013:35:37-41.
35. Sagheer M, Hasan M, Latif M, Iqbal J. Evaluation of some Indigenous medicinal plants as a source of toxicant, repellent and growth inhibitors against *Tribolium castaneum* (Coleoptera: Tenebrionidae). *Pakistan Entomologist*,2011:33(2):87-91.
36. Scheff DS, Sehgal B, Subramanyam B. Evaluating penetration ability of *Plodia interpunctella* (Hübner)(Lepidoptera: Pyralidae) larvae into multilayer polypropylene packages. *Insects*,2018:9(2):42. https://doi.org/10.3390/insects9020042
37. Shafique M, Ahmad M, Chaudry MA. Feeding preference and development of Tribolium castaneum (Herbst.) in wheat products. *Pakistan Journal of Zoology*,2006:38(1):27. doi:0030-9923/2006/0001-0027
38. Shawkat MS, Khazaal AQ, Majeed MR. Extraction of Pyrethrins from *Chrysanthemum cinerariaefolium* petals and study its activity against beetle flour Tribolium castanum. *Iraqi Journal of Science*,2011:52(4):456-463.
39. Soujanya PL, Sekhar J, Kumar P, Sunil N, Prasad CV, Mallavadhani U. Potentiality of botanical agents for the management of post-harvest insects of maize: a review. *Journal of food science and technology*,2016:53(5):2169-2184. https://doi.org/10.1007/s13197-015-2161-0
40. Thacker JR. An introduction to arthropod pest control. Cambridge University press, 2002, 337. https://doi.org/10.1017/s0014479703301334
41. Wekesa I, Onek L, Deng A, Hasanali A, Othira J. Toxicity and repellant potency of *Hyptis spicigera* extracts on *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). *Journal of Stored Products and Postharvest Research*,2011:2(6):113-119.
42. Yadav DN, Anand T, Sharma M, Gupta R. Microwave technology for disinfestation of cereals and pulses: An overview. *Journal of food science and technology*,2014:51(12):3568-3576. https://doi.org/10.1007/s13197-012-0912-8

43. Yar MA, Hassan MW, Ahmad M, Ali F, Jamil M. Effect of packaging materials and time period for damage in packaging and weight loss in packed wheat flour (*Triticum aestivum* L.) by red flour beetles *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae). *Journal of Agricultural Science*,2017:9(4):242-247.
44. Arshad A, Munawar A, Mastoi MI, Sohail S, Liang C. Evaluation of Phosphine and Cypermethrin resistance in field collected strains of Rhyzopertha Dominica (Coleoptera: Bostrichidae) and Tribolium castaneum (Coleoptera: Tenebrionidae) under laboratory conditions. Evaluation. 2019 Sep;4(5).