
Chemical Compositions and Fumigation Effects of Essential Oils Derived from Cardamom, *Elettaria cardamomum* (L.) Maton, and Galangal, *Alpinia galanga* (L.) Willd Against Red Flour Beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

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Article

Chemical Compositions and Fumigation Effects of Essential Oils Derived from Cardamom, *Elettaria cardamomum* (L.) Maton, and Galangal, *Alpinia galanga* (L.) Willd Against Red Flour Beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

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Abstract: The prevention of destruction caused by red flour beetle, *Tribolium castaneum* (Herbst), using essential oils derived from certain herbal plants presents an interesting alternative to synthetic insecticides, known for their harm to both consumers and the environment. The research aimed to investigate the chemical compositions and fumigating effects of essential oils derived from cardamom (*Elettaria cardamomum* (L.) Maton) and galangal (*Alpinia galanga* (L.) Willd) against *T. castaneum*. Chemical compositions were analyzed using GC-MS, while fumigation bioassay was conducted via a vapor-phase test. Experimental setup followed factorial design in CRD with 5 replications. Factor A encompassed 4 types of essential oils: the manually extracted essential oils from cardamom leaves (MCL) and galangal leaves (MGL), the commercially produced essential oils from cardamom seeds (CCS) and galangal rhizomes (CGR). Factor B consisted of 7 concentrations: 0, 50, 100, 150, 200, 250, and 300 µL/L air. Results indicated that MCL contained eucalyptol, trans-calamenene, and isospathulenol as its main compounds. Similarly, CCS was characterized by camphene and eucalyptol. MGL exhibited caryophyllene, aciphyllene, and α-bisanolene. Lastly, CGR was composed of methyl cis-cinnamate, safrole, and p-vinylphenyl isothiocyanate. Study noted that 250 µL/L air over 168 hours of CGR, demonstrated 94% fumigation efficiency against adult *T. castaneum*. These results suggest the potential of galangal rhizome derived essential oil as an insecticide for managing *T. castaneum* populations in agricultural product storage.

Keywords: Zingiberaceae; essential oil; stored insect pest; plant secondary metabolite; GC-MS

1. Introduction

Stored-product insects threaten global grain stores, causing losses ranging from 10% in temperate regions to nearly 50% in humid tropical areas [1]. Grains and flour, vital sources of carbohydrates worldwide, are particularly prone to infestation, leading to reduced quantity and quality. Post-harvest storage, a critical phase in the agricultural supply chain, directly impacts stored commodities' quality and quantity, but it is often disrupted by pest infestations. These pests exploit favorable conditions within warehouses, evading detection and causing both quality degradation and quantitative losses, commonly known as shrinkage [2]. Various insect species attack grain stores, causing significant losses estimated between 5% to 30% of total global agricultural production [3]. Notably, the red flour beetle poses a serious threat by contaminating grain with shed skins, feces, and

dust, affecting cereal flour and stored foods. Postharvest losses due to insect pests can reach 30% to 40%, posing a significant threat to human and livestock health [4].

The red flour beetle, *Tribolium castaneum* (Herbst, 1797), is a highly damaging storage pest, threatening stored products and the food economy. Despite its classification as a secondary pest, *T. castaneum* infests damaged grains during harvesting, affecting various stored food items, leading to significant economic losses [5]. This infestation causes agglomeration, discoloration, and spoilage, with profound economic implications [6]. It is recognized as one of the most destructive cosmopolitan pests of stored products due to its broad host range and rapid infestation capabilities. Additionally, *T. castaneum* infestations raise temperature and humidity within storage environments, promoting bacterial and fungal growth [7]. Exposure to *T. castaneum* may trigger allergic responses and contribute to product deterioration [8]. The development of resistance to various insecticides, including malathion and phosphine, poses a significant challenge for *T. castaneum* control, increasing the risk of food contamination. With its ability to breed year-round in warm climates, *T. castaneum* poses a continuous threat to global food security, especially in tropical and subtropical regions [9]. Effective monitoring and control measures are essential to mitigate its impact on the cereal value chain and ensure food safety.

The extensive use of chemical insecticides to safeguard stored products from infestation has resulted in insect resistance, leading to environmental pollution and harm to non-target organisms [10]. While phosphine, malathion, and deltamethrin effectively control pests like *T. castaneum*, their excessive application has caused resistance, prompting the need for alternative approaches [11–13]. Insects pose significant threats to global food security, necessitating safer alternatives due to resistance and adverse effects associated with current insecticides [10]. Despite phosphine's widespread use as a fumigant, resistance in *T. castaneum* has emerged, indicating the need for safer options. Dependence on chemical pesticides has caused harm to non-target organisms and environmental pollution [14]. Integrated pest control strategies, which integrate plant essential oils with chemical pesticides, offer a promising approach to reduce environmental impact and ensure sustainable pest management.

Plant secondary metabolites, including essential oils, play a crucial role in plant-insect interactions, enhancing plant resistance to insects. These oils possess various pest control properties such as larvicidal, repellent, and antifeedant effects, making them valuable for integrated pest management [11,15]. They degrade into non-toxic compounds, minimizing risks to non-target organisms and humans, highlighting the importance of natural insecticides for food safety. Essential oils from aromatic plants effectively combat insect pests through fumigation, contact, repellency, and antifeedant actions [16], primarily due to their volatile compounds, particularly monoterpenes and sesquiterpenes, which act as fast-acting neurotoxins in insects, disrupting various biological processes, including growth and reproduction [17]. Concerns about the drawbacks of synthetic insecticides, such as resistance and environmental pollution, have driven interest in botanical insecticides, especially essential oils [10], offering specificity, biodegradability, and low mammalian toxicity, thus promising for integrated pest management. Their natural composition and low persistence mitigate the risk of resistance development [18].

Elettaria cardamomum (L.) Maton, commonly referred to as cardamom, is an aromatic herb from the Zingiberaceae family, globally recognized for its commercial significance, earning it the title "Queen of spices." While native to India and Sri Lanka, cardamom cultivation has expanded to diverse tropical and subtropical regions worldwide [19]. The unique aroma and bioactive compounds of cardamom contribute to its pharmaceutical and nutraceutical properties, rendering it a valuable botanical resource [20,21]. It finds applications across various sectors, including food, perfumery, and traditional medicine, with traditional uses spanning stimulating, stomachic, diuretic, carminative, and anti-infective properties [22]. Renowned for its diverse biological effects, cardamom exhibits therapeutic properties such as antispasmodic, stimulant, anthelmintic, antimicrobial, antiviral, antioxidant anticancer, anti-inflammatory, and insecticidal effects [23–26]. The essential oil of cardamom, primarily comprising 1,8-cineole and α -terpinyl acetate, demonstrates toxic effects on various stored-product pests, including coleopteran and lepidopteran insects, through contact and fumigant actions [27].

Alpinia galanga (L.) Willd., commonly known as galangal, is an aromatic herb belonging to the Zingiberaceae family. It has a rich history of diverse applications, including alleviating stomachache,

displaying antibacterial and antifungal properties, exhibiting antitumor effects, aiding in antiulcer activity, showing antiallergic effects, possessing antioxidant properties, antiplasmid, antimicrobial activities, and demonstrating insecticidal activity [28–31]. Extensively cultivated in Southeast Asian countries such as the Philippines, Indonesia, Thailand, India, and China, it holds significance in various traditional medical systems, including Thai, Ayurveda, Unani, and Chinese folk medicine [32]. It is traditionally considered an important source for medication, culinary products and cosmetics in Asia. For medical purposes, galangal has been widely using as encompass anti-inflammatory, antipyretic, emmenagogue, carminative, abortifacient, and aphrodisiac properties [33]. Moreover, studies have investigated the toxicity of essential oil extracted from *A. galanga* rhizomes against *Bactrocera dorsalis* (Hendel), providing insights into its potential as a natural insecticide [31]. Therefore, the aim of this study was to assess the chemical compositions and fumigating effects of essential oils derived from cardamom (*Elettaria cardamomum* (L.) Maton) and galangal (*Alpinia galanga* (L.) Willd) against adults of the red flour beetle, *Tribolium castaneum* (Herbst). Additionally, gas chromatography-mass spectrometry (GC-MS) analysis was conducted to determine their chemical composition.

2. Results

2.1. Chemical Composition

The manually extracted essential oil from cardamom leaves (MCL) contained a total of 28 chemical components, accounting for 90.77 %. The primary compounds were eucalyptol (25.20%), trans-calamenene (13.49%) and isospathulenol (13.09%). Additionally, it included ylangenal (4.96%), α -guaiene (4.13%), ledol (4.10%), carveol (3.18%), α -cubebene (3.13%), junenol (2.71%), isoaromadendrene epoxide (1.93%), epicubenol (1.63%), trans-verbenol (1.42%) and caryophyllene oxide (1.36%) (Table 1). The commercially produced essential oil from cardamom seeds (CCS) comprised a total of 34 constituents, accounting for 98.09%. The main compounds include camphene (46.06%) and eucalyptol (31.21%), along with linalyl acetate (4.72%), α -pinene (4.69%), linalool (2.49%), terpineol (1.93%) and nerolidol (1.06%) (Table 2).

Besides cardamom, eucalyptol, also known as 1,8-cineole, is also found in other plants such as eucalyptus (*Eucalyptus globulus* Labill.) and rosemary (*Rosmarinus officinalis* L.). Ashokkumar et al. [24] note that all cardamom accessions contain 1,8-cineole as the second most significant monoterpene constituent, ranging from 15.2% to 49.4%. Nonetheless, certain studies have noted lower proportions of α -terpinyl acetate and linalool, accompanied by higher percentages of 1,8-cineole in cardamom essential oil originating from various countries, with 1,8-cineole ranging from 25.6% to 26.71% [34,35]. Similarly, Noshad and Behbahani [36] identified 23 compounds in *E. cardamomum* essential oil, with eucaluptol being the major compound at 31.51%. Alanazi et al. [37] reported that GC/MS revealed 19 chemical constituents, indicating 98.2% of *E. cardamomum* essential oil, with the most compounds were monoterpenes constituents, such as 1,8-cineole (34.3%), α -terpinyl acetate (23.3%), and α -pinene (17.7%). The variation in the chemical composition of *E. cardamomum* essential oil may be attributed to factors such as chemotypes, geographical locations, season at the time of plant collection, stage of plant development, culture climate, extraction techniques, plant varieties, and plant part used, which can affect subsequent biological activities [38–40] observed fluctuations in volatile constituents of cardamom essential oil grown in distinct zones of Idukki hills, with changes in α -terpinyl acetate and 1,8-cineole levels across different zones as the season progressed.

Table 1. The chemical compositions of the manually extracted essential oil from cardamom, *E. cardamomun*, leaves (MCL).

No.	Compound	Retention Time	% Peak Area
1	eucalyptol	8.071	25.20
2	carveol	13.841	3.18
3	trans-verbenol	15.560	1.42
4	α -guaiene	26.043	4.13
5	α -cubebene	26.381	3.13
6	trans-calamenene	27.587	13.44

7	caryophyllene oxide	29.656	1.36
8	isopathulenol	30.307	13.09
9	ledol	30.747	4.10
10	junenol	30.945	2.71
11	ylangenal	31.783	4.96
12	isoaromadendrene epoxide	31.950	1.93
13	epicubenol	32.058	1.63
Total			80.28

Table 2. The chemical compositions of the commercially produced essential oil from cardamom, *E. cardamomun*, seeds (CCS).

No.	Compound	Retention Time	% Peak Area
1	α -pinene	6.012	4.69
2	eucalyptol	8.026	31.21
3	linalool	10.291	2.49
4	terpineol	14.046	1.93
5	linalyl acetate	16.388	4.72
6	camphene	20.907	46.06
7	nerolidol	28.971	1.06
Total			92.16

The manually extracted essential oil from galangal leaves (MGL) consisted of 25 components, accounting for 95.74%. The predominant compounds were caryophyllene (24.17%), aciphyllene (18.31%) and α -bisanolene (10.69%). Additionally, it contained ζ -elemene (7.67%), α -farnesene (6.98%), farnesyl butanoate (6.08%), humulene (4.52%), caryophyllene oxide (4.15%), n-hexadecanoic acid (2.30%), cedrene (1.89%), 11,11-dimethyl-4,8-dimethylenebicyclo[7.2.0]undecan-3-ol (1.57%) and ζ -himachalene (1.37%) (Table 3). The commercially produced essential oil from galangal rhizomes (CGR) included 27 compositions, accounting for 98.73%. The key compounds were methyl cinnamate (47.28%), saffrole (19.82%) and p-vinylphenyl isothiocyanate (11.52%). It also contained eucalyptol (7.65%), α -pinene (5.96%), α -phellandrene (1.46%) and terpeneol (1.12%) (Table 4).

The chemical composition analysis of galangal leaves, as reported by Menon [41], revealed β -caryophyllene (40.5%) and fenchyl acetate (20.7%) as the main compounds. Conversely, cubenol (28.4%), carotol (26.7%), and α -fenchyl acetate (30.5%) were predominant in stem, rhizome, and root. In northern India, *A. galanga* from the Himalayan region showed 1,8-cineole (39.4% and 32.5%) and β -pinene (11.9% and 22.7%) as primary compounds in its rhizome and leaf oils, respectively, with camphor (12.8%) also present in the leaf oil [42,43]. 1,8-cineole serves as the marker compound for *Alpinia* spp., with eucalyptol identified as the major constituent in *A. galanga*, as reported by Singh et al. [44]. Additionally, Abeywickrama et al. [45] confirmed 1,8-cineole as the major constituent in the essential oil of *A. calcarata*. In the oil from *A. galanga* in Tenom, Sabah, Malaysia, Nampoothiri et al. [46] identified major components in *A. galanga*, including 1,8-cineole, α -terpineol, and germacrene-D. Wu et al. [47] identified 51 components in the essential oil extracted from *A. galanga* rhizomes, with eucalyptol and pinenes being prominent. Various studies have investigated the chemistry of galangal rhizome, including analyses conducted by De Pooter et al. [48]. These findings highlight the need for further research on plant cultivation and essential oil standardization to account for genetic, geographic, physiological, and harvest timing factors that contribute to variations in essential oil composition within the same plant species [49].

Table 3. The chemical compositions of the manually extracted essential oil from galangal, *A. galanga*, leaves (MGL).

No.	Compound	Retention Time	% Peak Area
1	γ -elemene	22.766	7.67
2	caryophyllene	24.405	24.17

3	humulene	25.185	4.52
4	aciphyllene	27.159	18.31
5	α -farnesene	27.415	6.98
6	α -bisanolene	27.822	10.69
7	cedrene	28.145	1.89
8	caryophyllene oxide	30.217	4.15
9	11,11-dimethyl-4,8-dimethylenebicyclo[7.2.0]undecan-3-ol	32.096	1.57
10	ζ -himachalene	32.814	1.37
11	farnesyl butanoate	39.247	6.08
12	n-hexadecanoic acid	44.776	2.30
Total			89.70

Table 4. The chemical compositions of the commercially produced essential oil from galangal, *A. galanga*, rhizomes (CGR).

No.	Compound	Retention Time	% Peak Area
1	α -pinene	6.200	5.96
2	α -phellandrene	6.973	1.46
3	eucalyptol	7.924	7.65
4	terpineol	14.044	1.12
5	safrole	22.488	19.82
6	p-vinylphenyl isothiocyanate	22.658	11.52
7	methyl cis-cinnamate	23.274	47.28
Total			94.81

By comparing the chemical composition of the essential oils from cardamom, *E. cardamomun*, and galangal, *A. galanga*, it can be seen that the essential oils from MCL and CCS contained eucalyptol as a component and importantly, they were the same (Table 5). Plant extracts and essential oils commonly contain insecticidal monoterpenoids like limonene, linalool, and terpineol, carvacrol, which are effective against stored product insects [50,51]. These natural products exhibit insecticidal and acaricidal effects by disrupting biological pathways and inducing tissue injuries that may lead to the generation of free radicals [52]. Several monoterpenoids have been identified for their fumigant toxicity against various insects, including α -terpineol against *T. confusum* [53], β -pinene against *Sitophilus oryzae* [54], and 1,8-cineol against both *S. oryzae* and *T. confusum* [55,56]. Additionally, α -pinene was found toxic to *S. oryzae* [55], while p-cymene, α -terpinene, α -terpineol, and terpinene-4-ol showed potential fumigant toxicity to *S. oryzae* as well [50]. Moreover, γ -terpinene and terpinene-4-ol were reported as promising fumigants against *T. confusum* and *Ephesia kuehnielle* [57]. Additionally, α -pinene and β -caryophyllene were found to have repellent action against *S. zeamais* [58,59], while α -pinene also exhibited potential fumigant activity against *S. zeamais* [60].

Table 5. Compare the chemical compositions of cardamom, *E. cardamomun*, and galangal, *A. galanga*, essential oils in both the manually extracted and the commercially produced types.

No.	Compound	MCL	CCS	MGL	CGR
1	eucalyptol	25.20	31.21	-	7.65
2	camphene	-	46.06	-	5.96
3	methyl cis-cinnamate	-	-	-	47.28
4	caryophyllene	-	-	24.17	-
5	safrole	-	-	-	19.82
6	aciphyllene	-	-	18.31	-
7	trans-calamenene	13.44	-	-	-
8	isospathulenol	13.09	-	-	-
9	p-vinylphenyl isothiocyanate	-	-	-	11.52
10	α -bisanolene	-	-	10.69	-

11	caryophyllene oxide	1.36	-	4.15	-
12	humulene	-	-	4.52	-
13	terpineol	-	1.93	-	1.12
14	linalool	-	2.49	-	-
15	α -pinene	-	1.05	-	-

Acetylcholinesterase (AChE) is crucial in insect and tick nervous systems, breaking down acetylcholine to transmit neuronal signals [61]. Plant-derived volatiles, such as those affecting the octopaminergic mode of action, have neurotoxic effects [62]. While some studies suggest monoterpenoids may inhibit AChE activity [63], others argue that terpenoid toxicity doesn't always correlate with AChE inhibition [54]. The mode of action of monoterpenes can vary, potentially acting on the octopaminergic nervous system or inhibiting cytochrome P450-dependent mono-oxygenases [64]. Auti and Kulkarni [65] found significant AChE inhibition with *E. cardamomum* essential oil, suggesting neuroprotective effects [66]. Terpenoid compounds like 1,8-cineole, α -pinene, β -pinene, and carvone are identified as primary agents inhibiting AChE activity in essential oils [65]. Additionally, camphene attracts both sexes of *Rhynchophorus ferrugineus* adults, leading to mortality by blocking their air holes, causing asphyxiation [67]. Essential oils affect insect respiratory, nervous, and hormonal systems, often leading to death [68]. Linalool inhibits AChE and interacts with receptors, showing insecticidal activity [69]. Various compounds like menthol, methonene, limonene, and linalool are toxic to *S. oryzae* by inhibiting acetylcholine esterase enzyme [54]. Moreover, 1,8-cineole inhibits AChE activity in *T. castaneum* larvae and has insecticidal properties against post-harvest pests [56]. Linalool affects insect nervous systems by influencing ion transport and acetylcholine esterase release [70]. Caryophyllene oxide's fumigant toxicity may result from inhibiting the mitochondrial electron transport system, affecting insect respiration [71]. Lastly, menthyl cinnamate's moderate activity stems from its double bond and benzene ring configuration [72]. It's worth noting that the activity and mode of action of essential oil compounds can differ from those of individual components [73].

2.1. Chemical Composition

The types of essential oil had a significantly different ($P < 0.01$) effect on the mortality of adult *T. castaneum* at 24, 48, 72, 96, 120, 144, and 168 hours. CGR resulted in the highest mortality of adult *T. castaneum* in all tested periods, with 7.14 \pm 5.76%, 11.14 \pm 7.90%, 17.14 \pm 11.01%, 22.57 \pm 15.98%, 36.00 \pm 23.30%, 53.71 \pm 31.80%, and 64.57. \pm 37.54 %, respectively. Significant differences were also observed when compared with MCL, CCS and MGL (Table 6). The concentrations of essential oils exhibited a statistically significant difference ($P < 0.05$) in their effect on the mortality of adult *T. castaneum* at 48, 72, 96, 120, 144, and 168 hours. A concentration of 300 μ L/L air showed the highest mortality 10.50 \pm 7.72%, 13.50 \pm 9.00%, 21.50 \pm 11.82%, 32.00 \pm 19.39%, 45.50 \pm 27.29%, and 54.00 \pm 33.92 %, respectively. However, no statistical difference was found when compared with a concentration of 250 μ L/L air. Additionally, a significantly difference ($P < 0.01$) was observed at 24 hours, with the highest mortality at a concentration of 300 μ L/L air being 6.50 \pm 3.00%, but no statistical difference was found when compared with a concentration of 250 μ L/L air (Table 6).

The interaction between types of essential oils and concentrations was found to have no effect on the mortality of adult *T. castaneum* within 24-72 hours after the fumigation bioassay. However, significant differences ($P < 0.05$) were observed in the mortality of adult *T. castaneum* at 96 hours due to the interaction between essential oil types and concentrations. CGR at the concentrations of 150 and 250 μ L/L air resulted in the highest mortality of 38%, but no difference was observed when compared to a concentration of 300 μ L/L air, nor was there a significant difference when compared to CCS at the concentrations of 250 and 300 μ L/L air. Furthermore, the effect of interaction between essential oil types and concentrations on the mortality of adult *T. castaneum* was found to be significantly different ($P < 0.01$) at 120, 144, and 168 hours. CGR at a concentration of 150 μ L/L air exhibited the highest mortality at 120 hours, with 64.00 \pm 33.62%, but no difference was observed compared to the concentrations of 250 and 300 μ L/L air, which resulted in mortalities of 56.00 \pm 13.42% and 54.00 \pm 33.62%, respectively. At 144 hours, CGR at the concentrations of 250 and

300 µL/L air had the highest mortality of adult *T. castaneum*, with 80.00±7.07%, and 80.00±22.36%, respectively, with no significant difference observed compared to a concentration of 150 µL/L air, where the mortality was 78.00±23.87%. Finally, at 168 hours, CGR at a concentration of 300 µL/L air resulted in the highest mortality of adult *T. castaneum*, at 96.00±5.48%. However, no difference was observed compared to the concentrations of 150, 200, and 250 µL/L air, which had the mortality 84.00±15.17%, 84.00±11.40%, and 94.00±5.48%_{ab} respectively (Table 6).

Wang et al. [74] support the notion that the fumigant effect of essential oils was closely linked to dosage and exposure duration. Result indicated that the essential oil of CGR at 250-300 µL/L air gave the highest mortality (94-96%) of adult *T. castaneum*. Our results concurred with Wu et al. [47] demonstrated the potent fumigant activity of *A. galanga* rhizome essential oil against *Lasioderma serricorne*, surpassing other essential oils studied previously like those from *Pistacia lentiscus* L., *Elsholtzia stauntonii* Benth., and *Agastache foeniculum* (Pursh) Kuntze in fumigant toxicity against cigarette beetles [75–77]. This enhanced efficacy may be attributed to key compounds like methyl cis-cinnamate found in *A. galanga* rhizomes, which are widely used in pharmaceuticals, cosmetics, and the food industry and have documented repellent, insecticidal, and larvicidal properties [78–81].

Specifically, methyl cinnamate has shown insecticidal effects against *S. oryzae* and *Musca domestica* adults, while ethyl cinnamate exhibits antifeedant effects against *Spodoptera littoralis* and *Hylobius abietis*, and propyl cinnamate displayed insecticidal effects on *M. domestica* adults [79,82–84]. Fujiwara et al. [85] highlighted the superior larvicidal activity of methyl cinnamate against *Ae. aegypti*, suggesting the potential of cinnamates as alternatives to conventional insecticides. Moreover, *A. galanga* rhizome essential oil contains primary constituents like 1,8-cineol and α-terpineol. Several other studies have identified 1,8-cineole as effective insecticidal and oviposition repellents across various insect species [86–88]. Research on essential oils from diverse plants has revealed the deterrent effects of 1,8-cineole on neonate larvae of the codling moth and its efficacy in repelling or controlling adult stages of urban insect pests [72,89].

Table 6. Mortality of adult *T. castaneum* on cardamom, *E. cardamomun*, and galangal, *A. galanga*, essential oils in both the manually extracted and the commercially produced types.

Treatments	Mortality of adult <i>T. castaneum</i> (%)						
	24 h	48 h	72 h	96 h	120 h	144 h	168 h
Essential oils							
MCL	3.14±3.02b	4.00±3.27b	4.86±3.63b	6.86±5.40c	10.57±9.57c	15.43±12.79c	25.71±16.07c
CCS	1.71±1.80 b	3.71±3.55b	8.00±6.73 b	13.14±9.72b	24.86±15.44b	34.57±20.26b	43.43±24.73b
MGL	1.42 ±1.90b	2.86±2.54b	3.71±3.73 b	4.86±4.14c	6.57±3.98c	9.43±5.00c	12.86±5.76d
CGR	7.14±5.76a	11.14±7.90a	17.14±11.01a	22.57±15.98a	36.00±23.30a	53.71±31.80a	64.57±37.54a
<i>F-test</i>	**	**	**	**	**	**	**
LSD	2.87	3.94	4.93	5.41	6.78	6.66	6.38
Concentration							
ns							
0 µL/L air	0.00±0.00d	0.00±0.00d	0.00±0.00c	0.00±0.00c	0.00±0.00e	0.00±0.00e	0.00±0.00e
50 µL/L air	2.50±1.91bcd	3.00±2.00cd	4.00±2.83bc	4.00±2.83c	9.00±6.83d	12.00±8.64d	18.00±7.12d
100 µL/L air	4.00±1.63abc	7.00±2.58abc	11.00±7.75a	12.50±6.40b	17.50±9.93cd	28.50±21.60c	38.50±23.35c
150 µL/L air	2.50±3.79bcd	5.00±7.57bcd	9.00±9.66ab	14.00±16.73b	25.50±27.87abc	34.00±31.71c	41.50±31.97bc
200 µL/L air	2.00±2.83dc	4.00±2.83bcd	8.00±5.89ab	12.00±5.74b	22.00±13.00bc	35.00±24.49bc	49.00±30.70ab
250 µL/L air	6.00±8.16ab	8.50±7.19ab	13.50±11.36a	19.00±15.87ab	30.50±21.00ab	43.00±30.61ab	55.00±33.32a
300 µL/L air	6.50±3.00a	10.50±7.72a	13.50±9.00a	21.50±11.82a	32.00±19.39a	45.50±27.29a	54.00±33.92a
<i>F-test</i>	*	**	**	**	**	**	**
LSD	3.80	5.21	6.53	7.16	8.97	8.81	8.44

Essential oils x Concentrations							
MCL 0 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	0.00±0.00i	0.00±0.00i	0.00±0.00i
MCL 50 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	0.00±0.00i	0.00±0.00i	10.00±12.25i
MCL 100 µL/L air	6.00±5.48	8.00±4.47	8.00±4.47	8.00±4.47def	12.00±8.37ghi	14.00±11.40gh	32.00±10.95efg
MCL 150 µL/L air	2.00±4.47	4.00±8.94	4.00±8.94	6.00±8.94ef	6.00±8.94hi	14.00±5.48ghi	20.00±12.25gh
MCL 200 µL/L air	2.00±4.47	4.00±5.48	8.00±8.37	12.00±8.37cdef	16.00±5.48fgh	20.00±10.00fg	32.00±16.43efg
MCL 250 µL/L air	4.00±5.48	4.00±5.48	6.00±5.48	8.00±4.47def	20.00±12.25ef	26.00±11.40fg	44.00±16.73e
MCL 300 µL/L air	8.00±4.47	8.00±4.47	8.00±4.47	14.00±5.48cdef	20.00±7.07efg	34.00±15.17ef	42.00±19.24e
CCS 0 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	0.00±0.00i	0.00±0.00i	0.00±0.00i
CCS 50 µL/L air	4.00±8.94	4.00±8.94	6.00±8.94	6.00±8.94ef	12.00±8.37ghi	16.00±11.40gh	24.00±23.02fgh
CCS 100 µL/L air	2.00±4.47	4.00±5.48	4.00±5.48	10.00±0.00def	18.00±10.95fgh	34.00±19.49ef	38.00±21.68ef
CCS 150 µL/L air	0.00±0.00	0.00±0.00	6.00±5.48	12.00±10.95cdef	28.00±19.24d	36.00±18.17de	48.00±13.04de
CCS 200 µL/L air	0.00±0.00	2.00±4.47	6.00±8.94	12.00±8.37cdef	36.00±15.17d	50.00±12.25cd	64.00±11.40cd
CCS 250 µL/L air	2.00±4.47	10.00±10.00	20.00±7.70	26.00±5.48abc	38.00±13.04bc	54.00±11.40c	66.00±5.48c
CCS 300 µL/L air	4.00±8.94	6.00±13.42	14.00±16.73	26.00±15.17abc	42.00±25.88c	52.00±27.75cd	64.00±25.10cd
MGL 0 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	0.00±0.00i	0.00±0.00i	0.00±0.00i
MGL 50 µL/L air	2.00±4.47	4.00±8.94	6.00±13.42	6.00±13.42ef	8.00±13.04hi	12.00±10.95gh	14.00±11.40hi
MGL 100 µL/L air	4.00±5.48	6.00±8.94	10.00±14.14	10.00±14.14def	10.00±14.14hi	10.00±14.14gh	14.00±13.42hi
MGL 150 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	4.00±8.94hi	8.00±8.37hi	14.00±11.40hi
MGL 200 µL/L air	0.00±0.00	2.00±4.47	2.00±4.47	4.00±5.48f	6.00±5.48hi	8.00±8.37hi	16.00±11.40ghi
MGL 250 µL/L air	0.00±0.00	2.00±4.47	2.00±4.47	4.00±8.94f	8.00±8.37hi	12.00±10.95gh	16.00±15.17ghi
MGL 300 µL/L air	4.00±5.48	6.00±5.48	6.00±5.48	10.00±7.07def	12.00±4.47ghi	16.00±8.94ghi	16.00±8.94ghi
CGR 0 µL/L air	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00f	0.00±0.00i	0.00±0.00i	0.00±0.00i
CGR 50 µL/L air	4.00±5.48	4.00±5.48	4.00±5.48	4.00±5.48f	16.00±11.40fgh	20.00±14.14fg	24.00±15.17fgh
CGR 100 µL/L air	4.00±5.48	10.00±10.00	22.00±19.24	22.00±19.24bcd	32.00±19.24d	56.00±28.81c	70.00±15.81bc
CGR 150 µL/L air	8.00±8.37	16.00±15.17	26.00±20.74	38.00±14.83a	64.00±33.62a	78.00±23.87ab	84.00±15.17ab
CGR 200 µL/L air	6.00±8.94	8.00±8.37	16.00±11.40	18.00±13.04cde	30.00±15.81d	62.00±14.83bc	84.00±11.40ab
CGR 250 µL/L air	18.00±14.83	18.00±14.83	26.00±16.73	38.00±16.43a	56.00±13.42a	80.00±7.07a	94.00±5.48a
CGR 300 µL/L air	10.00±14.14	22.00±21.68	26.00±24.08	36.00±35.78ab	54.00±33.62a	80.00±22.36a	96.00±5.48a

<i>F-test</i>	ns	ns	ns	*	**	**	**
LSD	-	-	-	14.33	17.94	17.63	16.88

* represents a significant difference at $P < 0.05$, ** represents a significant difference at $P < 0.01$, Means followed by the same column followed by the same letter are not significantly different (LSD: $P > 0.05$).

3. Materials and Methods

3.1. Insect Raring

The red flour beetles, *Tribolium castaneum* (Herbst), were sourced from rice bran commonly utilized as animal feed, specifically collected for experimental purposes. Adult males and females were grouped into 30 pairs. They were fed a mixture of coarsely ground wheat flour mixed and rice bran in a ratio of 7:10, housed within 4 L plastic container, and securely sealed. Ventilation for the containers was scheduled every 3 days. They were maintained within a growth chamber at a 26 ± 5 °C and a relative humidity of $75 \pm 5\%$. The 14-day-old adult red flour beetle will be utilized in all bioassays.

3.2. Preparation of Essential Oils

The leaves of cardamom (*Elettaria cardamomum* (L.) Maton) and galangal (*Alpinia galanga* (L.) Willd) were collected in November 2022 in Maha Sarakham, Thailand ($16^{\circ}10'38''$ N $103^{\circ}18'03''$ E). Plant materials were washed, roughly chopped, and dried in hot air oven at 65 °C for 2 days, after which they were stored in a plastic bag. Following this, 100 g of dried plant parts were placed in a 2000 mL flask, and 1000 mL of distilled water was added. The mixture underwent water distillation at 100 ± 20 °C for 4 hours using an essential oil extractor. Subsequently, the resulting essential oil underwent purification by centrifuging at 10000 rpm for 10 minutes to separate any remaining water from the essential oil. The pure essential oils were then transferred into sealed amber glass vials and stored in a refrigerator at 4 °C until they were ready for further use in bioassays.

The 100% pure essential oils extracted from the seeds of cardamom, *E. cardamomum*, and the rhizome of galangal, *A. galanga*, were obtained commercially from the Aroma & More Shop, 333/19, Moo 19, Bangbuathong-Suphanburi Road, Laharn Sub-district, Bangbuathong District, Nonthaburi Province, 11110, Thailand.

3.3. Chemical Compositon Analysis

The essential oils of cardamom and galangal, both the manually extracted and commercially produced, were analyzed for their chemical composition using the method outlined by Satongrod et al. [90], employing a gas chromatograph-mass spectrometer model Clarus 680 (PerkinElmer USA). The column utilized was an Rtx-5MS capillary type, with a length of 30 m, diameter of 0.32 mm, and thickness of 1 μ m. An essential oil concentration of 100000 ppm, with a volume of 1 μ L, was injected in split mode (split ratio, 1:100 v/v). Helium gas served as the carrier gas at a flow rate of 1 mL/minutes. The injector temperature was set at 280 °C. Under column conditions, an initial temperature of 45 °C was maintained for 5 minutes before increasing at a rate of 10 °C/minutes to 200 °C, where it was held constant for 5 min. For mass spectrometry conditions the electron impact mode was set at 70 eV. Using a quadrupole mass analyzer, the detector temperature was maintained at 250 °C. Spectra were scanned (m/z) from 40 to 1000 amu.

The essential oils identification was made based on their retention indices (RI) determined with reference to homologues series of C₅–C₃₆ (n-alkanes), by comparison of their mass spectra with the reports in the literature using NIST and Wiley version libraries [91], ensuring a quality match of over 80%. Chemical composition data were analyzed by reading retention time and % peak area.

3.4. Effects of Fumigation Bioassay

Fumigation toxicity was assessed through a vapor-phase test conducted in a completely randomized design (CRD) with 5 replicates. Factor A comprised 4 types of essential oils: the

manually extracted essential oil from cardamom leaves (MCL), the commercially produced essential oil from cardamom seeds (CCS), the manually extracted essential oil from galangal leaves (MGL), and the commercially produced essential oil from galangal rhizomes (CGR). Factor B involved 7 concentrations: 0 (control, consisting of 100% acetone), 50, 100, 150, 200, 250, and 300 $\mu\text{L/L}$ air, prepared by diluting essential oils with acetone solvent. The test was conducted in closed 40 mL glass vials. One hundred μL of the test solution was applied onto a 2 cm diameter Whatman No.1 filter paper disc, which was then allowed to dry at room temperature for 2 minutes. The filter paper was attached to the bottom surface of the glass vial cap. Five pairs of adult *T. castaneum* were introduced into each test vial, which was tightly sealed with a screw cap. These vials were then stored in a growth chamber at $26 \pm 5^\circ\text{C}$ and a relative humidity of $75 \pm 5\%$. The number of *T. castaneum* death was recorded at 24, 48, 72, 96, 120, 144, and 168 hours. Mortality was calculated using the formula $[(\text{NC} / \text{NT})] \times 100$ where NC represented the number of dead *T. castaneum* and NT denoted the total number of *T. castaneum* used in the test. If the mortality of *T. castaneum* in the control fell within the range of 5-20%, the mortality of *T. castaneum* in each treatment needed adjustment using Abbott's formula [92]. Statistical data was conducted using the F-test by analyzing variance (ANOVA) based on the experimental plan factorial in CRD and mean comparisons were made using the least significant difference method ($\text{LSD} < 0.05$).

4. Conclusions

The potential of plant-derived essential oils in controlling stored product pests is significant due to their selectivity and minimal impact on non-target organisms, with our study emphasizing the notable adulticidal activity of *A. galanga* rhizome essential oil against *T. castaneum*, suggesting its promise for pest control in stored products.

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