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Comparative Bioefficacy of Different Citrus Peel Extracts as Grain Protectant Against *Callosobruchus chinensis*, *Trogoderma granarium* and *Tribolium castaneum*

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Abstract: Efficacy of essential oils extracted from peel of various citrus species; *Citrus reticulata*, *Citrus sinensis*, *Citrus paradisi* and *Citrus grandis* were determined against the adults of *Callosobruchus chinensis* and *Tribolium castaneum* as well as against the larvae of *Trogoderma granarium* in order to find out safe alternatives of high toxics. Four concentrations of each oil; 2%, 4%, 6% and 8% were assayed for their insecticidal activity after exposure durations of 24, 48, 120 and 168 h. The results revealed that the essential oil vapors showed variable toxicity to insects depending on concentration and exposure duration. Efficacy increased as the exposure time and concentration were increased for all insects. All the oils were found to be very successful against adults of *C. chinensis*. Out of which, the oil of *C. sinensis* was the best against *C. chinensis* even at lower concentrations; LD_{50}, LD_{70} and LD_{90} values were 3.49%, 4.70% and 5.91% after 72-h exposure duration followed by the oil of *C. paradisi* that exhibited LD_{50}, LD_{70} and LD_{90} values 2.39%, 4.61% and 6.30%, respectively after 168-h exposure duration. However, the other citrus oils were effective at higher concentrations and required more exposure time. The order of decrease in efficacy was *C. sinensis*>*C. paradise*>*C. reticulata*>*C. grandis.* The oil of *C. sinensis* found to be the most effective at LD_{50}=6% after 168-h among all the oils against *T. castaneum* and the order of efficacy was the same. In case of *T. granarium,* the Citrus oils were found comparatively less effective than other insects.

Key words: Citrus Peel · Essential Oil · Bioefficacy · *Callosobruchus chinensis* · *Tribolium castaneum* · *Trogoderma granarium*

INTRODUCTION

Attack of pulse beetle gram dhora, (*Callosobruchus chinensis)*, khapra beetle *(Trogoderma granarium)* and red flour beetle *(Tribolium castaneum)* cause heavy qualitative and quantitative losses in Pakistan every year [1]. Avesi [2] accounted that about 2-6% food grain production of Pakistan is lost every year during storage by stored grain insect pests. Gujar and Yadav [3] reported pulse grain losses caused by stored grain pests are about 55-60% in seed weight and loss in protein is about 45.50-66.30%. Due to these damages seed become unfit for human consumption as well as for planting.

The use of synthetic compounds to control insect pests has lead to several adverse effects, including water and soil contamination, insect resistance and toxicity to non-target species [4,5] and most of these are mutagenic and carcinogenic [6]. Many neurotoxic insecticides are damaging the environment and/or posing a threat to public via food residues, water, air and soil contamination or accidental exposure [7]. Therefore, additional fumigants and control measures are required and have been studied by some researchers [8-11].

Wink [12] investigated that some plants may provide potential alternatives to currently used insect-control agents because they constitute a rich source of bioactive chemicals. Since these are often active against a limited number of species including specific target insects, are often biodegradable to non-toxic products and are potentially suitable for use in integrated pest management. They can lead to the development of new classes of safer insect-control agents [13-15]. Also the citrus plants have attracted the attention of many workers and thus organic extracts or essential oils of the fruit peels were tested against different species of insect pests [16-21].

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The essential oils extracted from Citrus genus have monocyclic monoterpenoides and its major component is d-limonene (□-mentha-1,8-dene) and they have insecticidal activity against insect pests [22]. Mansour *et al.* [23] already found that Limonene was the dominant compound with an occurrence ranging from 51.97 % to 95.32 % in different citrus species. It was further concluded by many researchers that Limonene alone has a basic biocidal properties [24-29], but its potency may be varied considerably depending on citrus and insect species against which it is being used. According to Choi and Sawamura [28], such variation in limonene content in citrus peel oils may be related to the time of harvesting, the degree of freshness and the size of the fruit. Also, geographical location, fruit variety and method of extraction should be taken into consideration.

Therefore, in this study, it was intended to examine the activity of volatile fractions of essential oils extracted from peel of different citrus species and cultivars, collected from different locations in Faisalabad, Pakistan, on gram dhora, *C. chinensis,* khapra beetle *T. granarium* and red flour beetle *T. castanium* and to evaluate the efficacy of citrus peel essential oils against these insects.

MATERIALS AND METHODS

Heterogeneous samples of adults of Pulse Beetle (*C. chinensis*), Khapra Beetle (*T. granarium*) and Red Flour Beetle (*T. castaneum*) were collected from various locations in Faisalabad district. A culture of these insects was established and maintained in sterilized jars placed in the incubator at 25±2°C and 60-75% R.H for *T. castaneum* and *T. granarium* and at 32±6°C, 45±5% R.H and 12-24 h dark-light cycle for *C. chinensis* to get the homogenous population. The culture medium was the wheat for khapra, wheat flour for red flour beetle and unshelled moong for dhora. Twenty beetles were liberated in each jar. The jars were covered with muslin cloth, tied with rubber bands to avoid the escape of beetle. Beetles were kept in the culture medium for 3 days for egg laying and then removed from the jars by using sieves and fine brush. After separating the adults, the grains along with eggs of the insects were kept again in the same jars. Jars were placed in the incubator at respective temperature and humidity (as given above) for rearing of a homogeneous population of each insect. To prevent the culture from crowding, old seeds were replaced with new ones on monthly basis. Two days old adults of *C. chinensis* and *T. castaneum* while two days larvae of *T. granarium* were used for all bioassays.

Peel from *Citrus reticulata*, *Citrus sinensis*, *Citrus paradisi* and *Citrus grandis* were collected from the local market of Faisalabad. The peel was then cut to splinters. Volatile fraction (essential oil) of the fresh rind tissue (albedo and flavedo) was extracted by steam distillation method as described by Papachristos and Stamopoulos [30]. In this method, steam is produced at greater pressure than the atmosphere and therefore the fruit peel boils at above 100 degrees Celsius which facilitates the removal of the essential oil from the plant material at a faster rate and in so doing prevents damage to the oil. The hot steam helps to release the aromatic molecules from the plant material since the steam forces open the pockets in which the oils are kept in the plant material. The molecules of these volatile oils then escape from the plant material and evaporate into the steam. The steam which then contains the essential oil, is passed through a cooling system to condense the steam, which forms a liquid from which the essential oil and the volatile fraction of the steam distillate is separated from water using a separating funnel. The temperature of the steam is carefully controlled, just enough to force the plant material to let go of the essential oil, yet not too hot as to burn the plant material or the essential oil. With this extraction method, the oil is not damaged. Sukumar *et al.* [31] also reported that extraction of plant chemicals with water or steam exerts a greater influence in the resultant bioactivity than organic extraction. The same method was also used by Negahban *et al.* [32] in order to find out the efficacy of essential oil from *Artemisia scoparia* against *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). After extraction, the essential oil was dried over anhydrous sodium sulphate and stored at 5°C until the onset of bioassays.

The contact effect of the essential oils against adults of dhora *C. chinensis,* red flour beetle *T. castaneum* and larvae of khapra beetle *T. granarium* were evaluated on filter paper discs by treating a whatman No.1 filter paper with the oils diluted in 100% acetone as described by Rehman and Schmidt [33] and Keita *et al.* [34]. The filter papers were placed in polyethylene petri dishes (diameter 9cm; height 1.5cm). 1, 2, 3 and 4 mL volume of each oil were diluted in 50 mL of analytical grade 100% acetone to obtain 2%, 4%, 6% and 8% concentrations respectively. A volume of 5ml solution was applied to each filter paper disc in order to obtain the equal amount of applied essential oil. The 100% acetone was allowed to evaporate for 20 minutes prior to the introduction of 20 adults separately into each polyethylene petri dish. The petri dishes were packed and held with rubber bands. The control petri dishes were treated with 50mL 100% acetone without the oils. Each experiment was replicated three times along with the control. Dead insects were counted 24, 72, 120 and 168 h post application. Insects which did not respond to the gentle touch of a small probe were considered dead [35].

Linear regression was conducted to define dose-response relationships, when correlations were found to be significant. The LT_{50}, LT_{70} and LT_{90} (effective lethal exposure time/durations to oil required to reduce insect adults by 50%, 70% and 90% respectively relative to controls) and LD_{50}, LD_{70} and LD_{90} values (effective doses/concentrations of oil required to reduce insect adults by 50%, 70% and 90% respectively relative to controls) were determined by Probit Analysis [36].

RESULTS AND DISCUSSION

At 2% level of concentration, *C. sinensis* oil showed 100% mortality adults of *C. chinensis* after exposure duration of 120 h while *C. paradisi*, *C. grandis* and *C. reticulata* oils killed only 31.67%, 3.33% and 6.67%, respectively. After 168-h exposure, efficacy of oils of *C. paradisi*, *C. grandis* and *C. reticulata* increased to 45%, 21.67% and 30% of the insects (Fig 1). Oil of *C. sinensis* showed maximum efficacy (88.33%) after exposure duration of 72 h. at 4% concentration as compared to other citrus oils. Mortality percentage of adults of *C. chinensis* was sharply increased to 98.33% and 55% at 4% concentration of oil of *C. sinensis* and *C. paradise*, respectively. On the other hand, oils of *C. grandis* and *C. reticulata* showed very low efficacy even after 120 h. After 168 h, *C. grandis* and *C. reticulata* killed comparatively less adults (46.67% and 33%, respectively) of *C. chinensis* while the oil of *C. sinensis* and *C. paradisi* killed 100% and 55% adults respectively (Fig. 2)

At 6%, Oil of *C. sinensis* showed maximum efficacy (95%) after just 72 h. After 120-h, mortality rate of adults was 100% and 68.33% for *C. sinensis* and *C. paradisi* oil respectively. Whereas, for other oils mortality percentage was lower. Efficacy of oils of *C. paradisi*, *C. grandis* and *C. reticulata* was sharply increased to 98.33%, 46.67% and 60% respectively after 168-h (Fig 3).

At maximum concentration level (8%), after the exposure duration of 72 h, efficacy of *C. sinensis* was maximum (93.33%) while the other citrus oils showed less efficacy. After 120 h, oil of *C. sinensis* killed 100 % insects while the oils of *C. grandis* and *C. reticulata* killed 16% and 46% adults of *C. chinensis* respectively. After exposure period of 168 h, both *C. sinensis* and *C. paradisi* showed 100% efficacy against adults of *C. chinensis* while oils of *C. grandis* and *C. reticulata* killed 75% and 83.33% adults (Fig. 4). It has previously been reported that *C. chinensis* showed lowest resistance for plant derived essential oils followed by *T. castaneum* and *T. granarium.* Udomporn Pangnakorn [37] and Kim *et al.,* [14] found that *C. chinensis* had highest mortality rate among all the insects used in their respective studies. El-Sayed, F.M.A and M. Abdel Razik [38] also found out that peel oil *C. sinensis* had showed complete control of *C. chinensis* among all the insects studied. Same results were concluded by other researchers [39].

In case of *T. castaneum*, oil of *C. sinensis,* at 6% concentration, showed maximum efficacy (45 %) after 168 h among all the oils, whereas, 0, 5% and 3% mortality was recorded for *C. grandis* and *C. reticulata* oil respectively (Fig. 5). By increasing the concentration level to 8%, after the exposure of 72 *C.*, efficacy of *C. sinensis* was slightly increased to 23.33% while the other citrus oils showed no considerable increase in efficacy. After 120 h, *C. sinensis* showed sharp increase in efficacy and killed 68.67% adults. After 168-h, oil of *C. sinensis* killed 98% insects while the oils of *C. paradisi*, *C. grandis* and *C. reticulata* killed 15%, 3.33% and 6.67% adults of *T. castaneum*, respectively (Fig. 6). Same results were concluded when *Anna senegalensis, Hyptis specigera* and *lippie regosa* were tested by Ngamo *et al.* [40] against the four major stored grain insect pests including *T. castaneum* and he found that *T. castaneum* was least affected by the oils among all the insects studied. Mishra and Tripathi [41] also found that *T. castaneum* was the least sensitive among all the insects studied against the three essential oils. Same results were reported by other researchers.

At the concentration level of 6% and after the exposure duration of 72 h, mortality rate of grubs of *T. granarium* was 13.33% for oil of *C. paradisi* while the other oils showed no efficacy. Oil of *C. paradisi* showed maximum efficacy (18.33%) after 120 h while mortality rate of grubs was 3.33% and 1.67% for *C. sinensis* and *C. reticulata* oil respectively. Efficacy of oils of *C. paradisi*, *C. sinensis*, *C. grandis* and *C. reticulata* was slightly increased to 23.33%, 6.67%, 1.67% and 5%, respectively after 168 h (Fig. 7).

At 8%, oil of *C. paradisi* showed 21.67% mortality of grubs of *T. granarium*, while the other oils killed no insects at this concentration even after exposure duration of 72 h. After 120 h, oils of *C. sinensis*, *C. grandis* and *C. reticulata* showed mortality of 5%. 1.67% and 3.33% of grubs. The maximum mortality was recorded due to the oil of *C. paradisi* (26.67%) after the exposure duration of 120 h at 8 % concentration. Oil of *C. paradisi* showed efficacy of 31.67% after the exposure duration of 168 *C.* at 8 % concentration but the oils of *C. sinensis*, *C. grandis* and *C. reticulata* showed 13.33%, 8.33% and 8.33% mortality (Fig. 8).

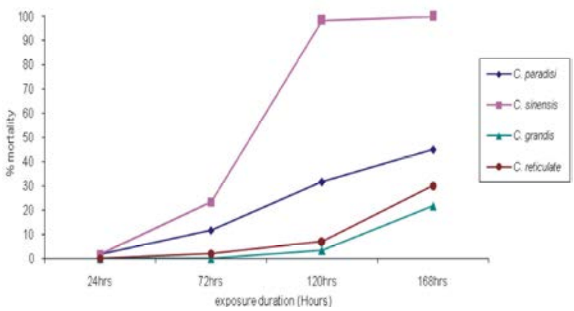

Fig. 1: Performance of 2 % essential oils against adults of *C. chinensis* after exposure duration of 12, 24, 120 and 168 h.

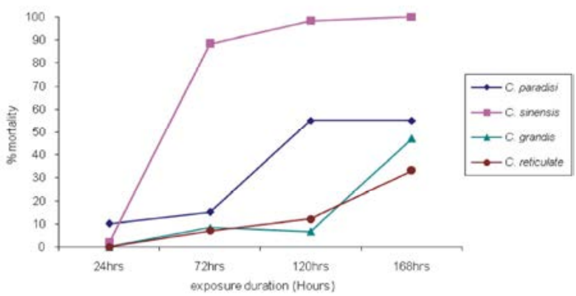

Fig. 2: Performance of 4 % essential oils against adults of *C. chinensis* after exposure duration of 12, 24, 120 and 168 h.

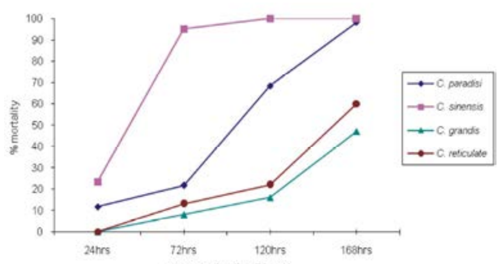

Fig. 3: Performance of 6 % essential oils against adults of *C. chinensis* after exposure duration of 12, 24, 120 and 168 h.

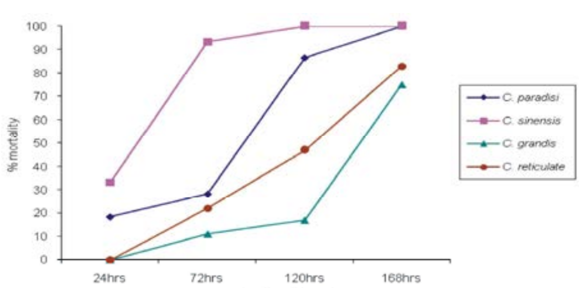

Fig. 4: Performance of 8 % essential oils against adults of *C. chinensis* after exposure duration of 12, 24, 120 and 168 h.

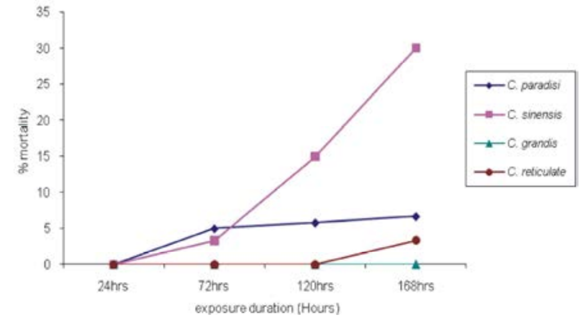

Fig. 5: Performance of 6 % essential oils against adults of *T. castaneum* after exposure duration of 12, 24, 120 and 168 h.

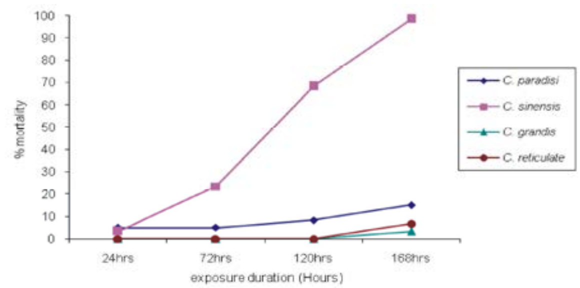

Fig. 6: Performance of 8 % essential oils against adults of *T. castaneum* after exposure duration of 12, 24, 120 and 168 h.

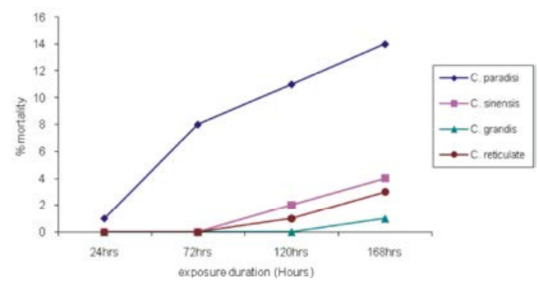

Fig. 7: Performance of 6 % essential oils against grubs of *T. granarium* after exposure duration of 12, 24, 120 and 168 h.

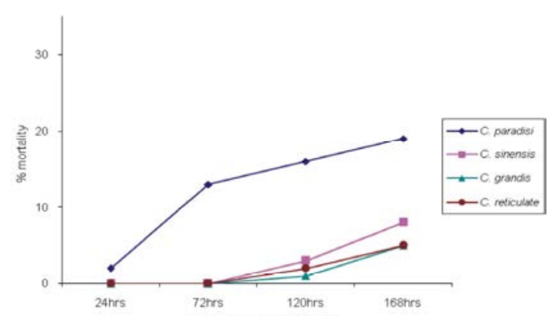

Fig. 8: Performance of 8 % essential oils against grubs of *T. granarium* after exposure duration of 12, 24, 120 and 168 h.

Table 1: Analysis results of linear regression of *C. chinensis* mortality data on exposed periods to four citrus essential oils at different concentrations

Oil	*Concentration (%)*	*Slope*	R^2	$LT_{50}(h)$	$LT_{70}(h)$	$LT_{90}(h)$
C. paradisi	2	0.313	0.893	183	246	309
	4	0.365	0.918	134	180	226
	6	0.639	0.975	96	126	156
	8	0.632	0.956	84	113	142
C. sinensis	2	0.771	0.940	89	112	135
	4	0.635	0.834	72	94	116
	6	0.490	0.807	57	83	110
	8	0.431	0.824	46	78	109
C. grandis	2	0.142	0.848	317	418	520
	4	0.309	0.942	188	245	303
	6	0.309	0.942	188	245	303
	8	0.479	0.886	136	168	201
C. reticulata	2	0.198	0.882	255	334	412
	4	0.219	0.940	246	326	407
	6	0.392	0.944	156	201	246
	8	0.573	0.992	117	151	185

* No mortality was reported

Table 2: Analysis results of linear regression of *T. castaneum* mortality data on exposed periods to four citrus essential oils at different concentrations

Oil	*Concentration (%)*	*Slope*	*R2*	$LT_{50}(h)$	$LT_{70}(h)$	$LT_{90}(h)$
C. paradisi	2	0.045	0.887	887	1235	1583
	4	0.056	0.956	837	1166	1495
	6	0.066	0.981	747	1038	1330
	8	0.069	0.913	596	836	1076
C. sinensis	2			*		
	4			*		
	6	0.306	0.925	192	248	303
	8	0.688	0.990	98	127	155
C. grandis	2			*		
	4			*		
	6	0.010	0.775	2952	4104	5256
	8	0.056	0.868	2952	4104	5256
C. reticulata	2			*		
	4	0.021	0.775	1512	2088	2664
	6	0.021	0.775	1512	2088	2664
	8	0.042	0.775	792	1080	1368

* No mortality was reported

Table 3: Analysis results of linear regression of *T. granarium* mortality data on exposed periods to four citrus essential oils at different concentrations

Oil	*Concentration (%)*	*Slope*	*R2*	$LT_{50}(h)$	$LT_{70}(h)$	$LT_{90}(h)$
C. paradisi	2	0.042	0.894	976	1360	1744
	4	0.056	0.956	837	1166	1495
	6	0.146	0.974	329	459	589
	8	0.188	0.940	233	328	422
C. sinensis	2			*		
	4			*		
	6	0.049	0.944	967	1333	1700
	8	0.094	0.923	509	691	873
C. grandis	2			*		
	4	0.010	0.775	2952	4104	5256
	6	0.010	0.775	2952	4104	5256
	8	0.056	0.868	740	1011	1282
C. reticulata	2			*		
	4	0.035	0.913	1256	1736	2216
	6	0.035	0.913	1256	1736	2216
	8	0.059	0.929	784	1076	1369

* No mortality was reported

Table 4: Analysis results of linear regression and probit analysis of *C. chinensis* mortality data to four citrus essential oils at four exposure duration

Oil	Exposure duration (h)	Slope	R^2	LD_{50}(%)	LD_{70}(%)	LD_{90}(%)
C. paradisi	24	0.373	0.972	19.51	26.84	34.17
	72	0.355	0.996	15.66	22.58	29.49
	120	0.119	0.922	7.36	6.06	8.28
	168	0.087	0.944	2.93	4.61	6.30
C. sinensis	24	0.155	0.952	10.36	13.43	16.50
	72	0.066	0.810	3.49	4.70	5.91
	120			**		
	168			**		
C. grandis	24			*		
	72	0.475	0.910	25.21	34.64	44.06
	120	0.306	0.774	16.00	22.00	28.00
	168	0.113	0.950	5.28	7.53	9.77
C. reticulata	24			*		
	72	0.302	0.992	16.60	22.53	28.46
	120	0.145	0.943	8.88	11.61	14.35
	168	0.103	0.960	4.83	6.82	8.81

** 100% mortality reported at all concentrations after 124 h time interval
* no mortalityreported at all concentration after this exposure time

Table 5: Analysis results of linear regression and probit analysis of *T. castaneum* mortality data to four citrus essential oils at four exposure duration

Oil	Exposure duration (h)	Slope	R^2	LD_{50} (%)	LD_{70} (%)	LD_{90}(%)
C. paradisi	24			*		
	72			*		
	120	0.257	0.774	108	156	204
	168	1.333	0.966	32	46	60
C. sinensis	24			*		
	72	3.676	0.848	13	17	21
	120	11.000	0.883	7	8	10
	168	17.000	0.778	6	7	8
	24			*		
C. grandis	72			*		
	120			*		
	168	0.500	0.952	92	128	164
C. reticulata	24			*		
	72			*		
	120			*		
	168	1.000	0.955	47	65	83

* No mortality was reported

Table 6: Analysis results of linear regression and probit analysis of *T. granarium* mortality data to four citrus essential oils at four exposure durations.

Oil	Exposure duration (h)	Slope	R^2	LD_{50} (%)	LD_{70} (%)	LD_{90}(%)
C. paradisi	24	1.531	0.945	79.45	110.00	140.55
	72	0.315	0.943	16.84	22.95	29.05
	120	3.833	0.974	13.75	18.64	23.52
	168	4.500	0.961	11.71	15.83	19.96
C. sinensis	24			*		
	72			*		
	120	0.926	0.957	51.85	71.41	90.96
	168	2.330	0.941	22.18	29.82	37.45
C. grandis	24			*		
	72			*		
	120	0.254	0.774	124.00	172.00	220.00
	168	1.250	0.871	33.73	45.93	58.14
C. reticulata	24			*		
	72			*		
	120	0.502	0.957	92.00	128.00	164.00
	168	1.255	0.940	37.06	51.18	65.29

*no mortality was reported even after the time interval of 168 h.

The results of Linear regression and Probit analysis showed that there was positive and linear significant relationship between percent mortality and duration of exposure (Lethal Time/LT values) to the essential oil vapors within all concentration levels. Within each essential oil, the slopes of regression (Slope values) of mortality rates on exposure times were smaller in lower concentration than those in higher concentrations (Table 1, 2 and 3). Higher values of R^2 (ranging from 0.775-0.992) indicate that efficacy of the oils increases as the exposure time interval increases, or in other words, increase in mortality rate may be observed at longer exposure time. It can also be verified by decreasing LT values as the concentration level increase. It indicates if higher concentration level of oils is used, the oils would require less exposure time to kill the insects [42,43]. Lower LT_{50}, LT_{70} and LT_{90} values indicate that the oils required less exposure time to kill 50%, 70% and 90% adults respectively and proved to be highly toxic against the insect while the higher LT_{50}, LT_{70} and LT_{90} indicate that the oils required more exposure time to kill 50%, 70% and 90% adults respectively. So the oils that have higher LT values, may also perform better if used in higher concentrations (Tables 1, 2 and 3). Oil of *C. sinensis* proved to be highly toxic against *C. chinensis* in all concentrations as indicated by lower LT values (LT_{90} = 109h and LT_{50} = 46h at 8%) followed by *C. paradisi* (LT_{90} = 84h and LT_{50} = 142h at 8%). The same trend was observed against *T. castaneum*, that the oil of *C. sinensis* (LT_{90} = 155h and LT_{50} = 98h at 8%) was highly toxic among all the oils followed by *C. paradisi* (LT_{50} = 596h at 8%). But in case of *T. granarium*, oil of *C. paradisi* showed the highest toxicity (LT_{50} = 233h at 8%). While other oils showed lower LT values, which indicated their lower toxicity against *T. granarium.* But higher R^2 values emphasize that their efficacy may be higher at longer exposure duration.

Likewise, there was also a linear positive relationship between percent mortality and lethal dose/concentration (LD values) of all essential oils against all insects as indicated by higher values of Regression Coefficient (R^2) and slope. Higher values of R^2 indicate that efficacy of the oils increase as the level of concentration increases. LD values are decreasing sharply within the same oil, as the insects are exposed to the oil for longer period (Tables 4, 5 and 6). It indicates that the oils may kill more insects at the same concentration, if the exposure duration is increased [42,43]. In case of *C. chinensis*, the oil of *C. sinensis* performed best and showed 100% mortality after 120h exposure duration. After 72h exposure, LD_{50}, LD_{70} and LD_{90} values were 3.49%, 4.70% and 5.91% respectively. Following the *C. chinensis*, the oil of *C. paradisi* showed high toxicity (LD_{50}, LD_{70} and LD_{90} values were 2.39%, 4.61% and 6.30% respectively after 168-h), followed by the oil of *C. reticulata* (LD_{50}, LD_{70} and LD_{90} values were 4.83%, 6.82% and 8.81% respectively after 168-h) and the oil of *C. grandis* (LD_{50}, LD_{70} and LD_{90} values were 5.28%, 7.53% and 9.77% respectively after 168-h). Same results were concluded by Ezeonu *et al*. [44] that volatile extracts of *C. sinensis* showed greater insecticidal potency against mosquitoes and housflies, while the cockroach was the most susceptible to the *Citrus aurantifolia* (lime) peels among the three insects studied. Same trend was observed in case of *T. castaneum.* The oil of *C. sinensis* performed best (LD_{50}, LD_{70} and LD_{90} values were 6%, 7% and 8% respectively after 168-h) followed by the oil of *C. paradisi* (LD_{50} = 32% after 168-h), the oil of *C. reticulata* (LD_{50} = 47%, after 168-h) and the oil of *C. grandis* (LD_{50} = 92%, after 168-h). Lower LD_{50}, LD_{70} and LD_{90} values for the *C. sinensis* oil indicate that the oils showed considerable efficacy against the *T. castaneum* and the oils have the potential at lower concentrations to control and kill 50%, 70% and 90% of the insect population respectively. While the higher LD values, as in case of *C. paradisi, C. reticulata* and *C. grandis* oil show that these oils were less toxic against adults of *T. castaneum.* Negahban *et al.* [45] Investigated chemical composition of the essential oil from *Artemisia scoparia* Waldst et Kit and its fumigant and repellent activity against three stored product insects, *Callosobruchus maculatus* (Fab.), *Sitophilus oryzae* (L.) and *T. castaneum* (Herbst). He also found *Callosobruchus maculatus* to be more susceptible than *S. oryzae* and *T. castaneum*. But case was different against adults of *T. granarium.* The oil of *C. paradisi* performed best (LD_{50}, LD_{70} and LD_{90} values were 11.71%, 15.83% and 19.96% respectively after 168-h) followed by the oil of *C. sinensis* (LD_{50}, LD_{70} and LD_{90} values were 22.18%, 29.82% and 37.45% respectively after 168-h), the oil of *C. grandis* (LD_{50} = 33.73%, after 168-h) and the oil of *C. reticulata* (LD_{50}= 37.06%, after 168-h). Higher R^2 values (range: 0.774-0.996) emphasize that their efficacy may be higher at some higher level of concentration. So, it is much worthy to be investigated further by using higher concentration.

CONCLUSION

Comparative efficacy of peel essential oils extracted from the *citrus* species used in this study showed that almost all the four oils may be used as safe alternatives of toxic insecticides being used these days against the stored grains insect pests. Toxicity of all the essential oils was recorded increasing as the exposure duration and level of concentration was increased. All the oils were found to be very successful against adults of *C. chinensis*. Out of which, the oil of *C. sinensis* was the best against *C. chinensis* even at lower concentrations followed by the oil of *C. paradisi*. However, the other citrus oils were effective at higher concentrations and required more exposure time. The order of decrease in efficacy was *C. sinensis*>*C. paradisi*>*C. reticulata*>*C. grandis.* The oil of *C. sinensis* was again found to be most affective among all the oils against *T. castaneum* and the order of efficacy was same. In case of *T. granarium,* the Citrus oils were found comparatively less effective than that against other two insects. The order of decrease in efficacy was *C.paradisi*>*C. sinensis*>*C. reticulata*>*C. grandis* against *T.granarium.*

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