

RELATIVE EFFICACY OF DIFFERENT CONCENTRATIONS OF MINT (*MENTHA LONGIFOLIA*) AGAINST *TRIBOLIUM CONFUSUM* (COLEOPTERA: TENEBRIONIDAE)

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Abstract

Present study aimed to examine the effect of different concentrations of mint, *Mentha longifolia* (2.00 %, 1.50 %, 1.00 %, and 0.50 %) on *Tribolium confusum* in wheat under laboratory conditions. All the concentrations of mint exhibited the potential to cause mortality of *T. confusum* and their effectiveness against *T. confusum* increased with time. Although, no significant difference was recorded among various mint materials to elicit mortality of *T. confusum* after 24 hours of application. However, a rise in mortality was recorded after 48 hours of application with the highest mortality of *T. confusum* also recorded in 2.00% concentration followed by 1.5 %, 1 %, and 0.5 % concentrations. The mortality of beetles showed further rise afterward and at the end of the week of the mortality experiment, 2.00% mint concentration cause maximum mortality. No mortality of beetles was recorded in the control. Population fluctuation results showed a significant difference in population growth at monthly intervals as a steady growth was observed in the 2%, 1.5% 1%, and 0.5% concentration, but, rapid growth was recorded in the control. At the end of the third month, the highest population of *T. confusum* was recorded in control (88.00±3.79 beetles), whereas, the beetle population observed in 0.50%, 1.00%, 1.50%, and 2% concentrations were (71.33±1.45 beetles), (63.00±3.46 beetles), (53.67±2.91 beetles) and (30.00±3.61beetles) respectively. The overall weight and grain loss were significantly higher in control, whereas, 2% of mint concentration suffered the lowest loss. Therefore, 2% of mint concentration is recommended to be used to manage the population of *T. confusum* in wheat.

Keywords: Botanical, Confused flour beetle, Mint, Population, wheat.

Introduction

Wheat (*Triticum aestivum* L.) is one of the vital cereal crops in the world. Wheat production is an important worldwide target to fill the gap among the growing population of not only in Pakistan but also throughout the country. Therefore, latest and innovative research has been focused during the recent years to improve the yield of wheat to decrease the gap among the world production and food requirements of the global population (Mackowaik *et al.*, 2001; Atiyah *et al.*, 2002; Rehmat *et al.*, 2010). During, 2016-17, wheat was cultivated on an area of 9,052 thousand hectares in Pakistan observing a decline of 1.9 percent related to 9,224 thousand hectares cultivated during the same period last year (GoP, 2018). Food production and its safe storage are some of the

major concerns faced by the world in recent years (Haq *et al.*, 2005). It has been estimated that around 9% of the global production of food grains is lost due to the attack of stored grain insect pests and mites, whereas, only larvae of *Triobolium* sp. can reduce the weight of individual grains up to 14.6% (Tooba *et al.*, 2005). Moreover, favorable climate and poor storage facilities aggravated the situation and increase the losses (Rahman *et al.*, 2009, Degri and zainab 2013). Generally, the major insect pests of stored wheat are lesser borer (*Ryzopertha dominica*), red flour beetle (*Tribolium castaneum* Herbst, 1797), confused red flour beetle (*Tribolium confusum* Herbst, 1797), maize weevil (*Sitophilus zeamais* Motschulsk, 1855), larger grain borer (*Prostephanus truncates* Horn, 1878), granary

weevil (*Sitophilus granarium* Linnaeus, 1758), rice weevil (*Sitophilus oryzae* Linnaeus, 1763) and Angoumois grain moth (*Sitotroga cerealella* Olivier, 1789). These pests cause severe problems in store grain houses, particularly in the under developing countries due to poor sanitation and the use of improper storage facilities all support the attack of pests (Lee *et al.*, 2002). The confused flour beetle, *Tribolium confusum* (Coleoptera: Tenebrionidae), is a common pest insect known for attacking and infesting stored flour and grain. Adults of this beetle are easily confused with other *Tribolium* species. (Papachristos and Stamopoulos, 2002). *Tribolium confusum* is a world-wide pest, especially in cooler regions. *Tribolium confusum* damages a variety of stored products including dried foods, grain products and fruits, chocolate, pet foods, and dried flowers (Athanasidou *et al.*, 2016). Economic losses consist of reduced weight and quality, difficulties in baking, reduced marketability of infested products, and an accompanying unpleasant smell. Besides, *T. confusum* is an allergen that may cause allergies in bakers (Alanko *et al.*, 2000). It has been observed that it mostly attacks on damaged grains that intact ones (Trematerra *et al.*, 2000). Use of synthetic chemicals poses great threats to humans, causes damage to the ozone, increases environmental pollution, and toxic to non-target organisms along with creating resistance among stored pests (Isman, 2006). Thus, such huge concerns to humans and their environment necessitate to look for the eco-friendlier and target specific control measures (Shaaya *et al.*, 1997; Isman, 2000). Accordingly, plants and their derivatives could have gained the huge potential in the management of not only field crops but also many noxious stored pests as many of them are selective with little or no harmful influence on non-target organisms and the environment (Shaaya *et al.*, 1997; Rajendran and Sriranjini, 2008). Among various plants evaluated for their toxicity against stored grain pests, about 25 to 30 species of genus *Mentha* (family Lamiaceae or Labiatae), widely located in diverse ecological zones of the world, hold prominent importance (Lange and Croteau, 1999). Wild mint, *Mentha longifolia* (L.), is a perennial herb that has shown very important and diversified biological and pesticidal activities against many harmful crop

pests (Hajlaoui *et al.*, 2008). Such toxicity of mint may be because of the presence of a lot of monoterpenoids such as piperitone, piperitone oxide, pulegone, piperitenone, d-limonene, menthone, carvone, caryophyllene, 1,8-Cineole and menthol (Oyededeji and Afolayan, 2006). As mint has not been exploited much against the stored grain pests in general and *T. confusum* in particular, therefore, this study was done to determine the effect of various mint concentrations on its mortality and population development.

Materials and Methods

Insect collection and rearing: The culture of *T. confusum* was obtained from the Stored Grain Research Laboratory, Department of Entomology, Sindh Agriculture University, Tandojam. *Tribolium castaneum* population was further reared on the pure wheat flour under controlled temperature (28 ± 2 °C) and 75 ± 5 % R.H in the laboratory.

Selection of plant materials and extract preparation: The leaves of Mint *Mentha Longifolia* was collected from the surroundings of the Sindh Agriculture University, Tandojam. The collected leaves were washed thoroughly and air-dried under shade. The dried plant materials were then ground using an electric blender (GEEPAS China GCG289) and the obtained powder was sieved using a muslin cloth. Four different concentrations of mint powder i.e., 2%, 1.5%, 1%, and 0.5% along with control (without mint powder) were used in the experiment. In the mortality determination bioassay, each concentration was applied in 20 grams of wheat, whereas, the same concentrations were applied in 200 gm of wheat for the population fluctuation experiment. After mixing, the powder was shaken thoroughly in seeds for about 3 minutes to create a homogenous mixture. In each container for the mortality experiment, ten beetles, whereas, twenty freshly emerged beetles of *T. confusum* were transferred for population fluctuation experiment. Each jar was tightened with an elastic rubber band to restrict beetles inside.

Determination of pest damage parameters: The efficacy of the plant materials to cause mortality was recorded after 24 hours, 48 hours, and 7 days. Population fluctuation of *T. castaneum* was

observed at monthly intervals for three consecutive months. At the end of the experiment, weight loss was recorded by using the formula below:

Total weight loss % = (Original weight – Observed weight)/Original Weight*100

Grain weight loss % = (Original grain weight – Observed grain weight)/Original grain weight*100

The collected data were analyzed using ANOVA and means with a significant difference were separated by the Least Square Difference test at the 0.05 probability level. All the analysis was done using STATISTIX 8.1 software.

Results

Effect of different concentrations of mint on mortality of *T. confusum* at different intervals:

The mortality rate of *T. confusum* in wheat due to the application of different concentrations of mint i.e., 2.00 %, 1.50 %, 1.00 %, and 0.50 % at various time intervals (24 hours, 48 hours and 1 week) after their application is given in Table 1. Although, all mint concentrations caused mortality immediately after their application the non-significant difference (F=3.22, P <0.0607) was recorded among various mint materials to elicit mortality of

T. confusum after 24 hours of application. After 24 hours, the highest mortality was recorded in 2.00 % mint concentration (2.00±0.58 beetles) and followed by 1.50 % concentration (1.67±0.67 beetles), whereas, no mortality was recorded in control. A rise in mortality was recorded after 48 hours of application as significantly (F =16.2, P = 0.0002) the highest mortality of *T. confusum* was observed in 2% concentration (74.67±0.6 beetles), followed by 1.5 % (4.00±0.58 beetles), 1 % (1.67 ±0.33 beetles) and 0.5 % (1.00±0.58 beetles) concentrations. A similar trend in the effectiveness of various mint concentrations regarding their effectiveness was recorded after 72 hours and one week of application. After one week of the application, 2.00% mint concentration cause maximum mortality (96.70%), followed by 1.50 (73.30%), 1.00 (46.70%) and 0.50% (40.00%) concentrations. No mortality of *T. confusum* individuals was recorded in the control. Thus, after one week, a significant difference (F = 33.30, P < 0.001) was observed in different mint concentrations to cause mortality among individual beetles of *T. confusum*

Table 1. Effect of different mint concentrations on mortality of *T. confusum* at different intervals under laboratory conditions.

Treatments	Population at different observation Intervals				Mortality %
	Pre-treatment	24 hours	48 hours	One week	
0.50%	10	0.67±0.33ab	1.00±0.58bc	4.00±1.00c	40.00%
1.00%	10	1.33±0.33ab	1.67 ±0.33b	4.67±0.33c	46.70%
1.50%	10	1.67±0.67a	4.00±0.58a	7.33±0.88b	73.30%
2.00%	10	2.00±0.58b	4.67±0.67a	9.67±0.33a	96.70%
	10	0.00b	0.00c	0.00d	0.00%

Effect of different mint concentrations on the population fluctuation of *T. confusum*:

Results for the population fluctuation of *T. confusum* in wheat treated with various mint extracts are given in Table 2. A significant difference was recorded in the population of *T. confusum* in different mint extracts treated wheat after first month (F =18.5, P < 0.0001), second month (F =53.2, p < 0.0000) and third month (F =46.5, P < 0.0000) of observations. After one month, the highest mean population was

recorded in control (21.00±1.73beetles), followed by 0.50% (15.00±1.53 beetles), 1.00% (11.33±0.88 beetles), 1.50% (8.67±1.45 beetles) and 2.00% (6.00±11.00 beetles). In the remaining two months of observations, steady growth was observed in the 2%, 1.5% 1%, and 0.5% concentration; however, rapid growth was recorded in the control. Accordingly, at the end of month three, the highest population of *T. confusum* was recorded in control (88.00±3.79 beetles), whereas, the beetle

population observed in 0.50%, 1.00%, 1.50%, and 2% concentrations were (71.33±1.45 beetles), (63.00±3.46 beetles), (53.67±2.91 beetles) and (30.00±3.61beetles) respectively.

Table 2. Effect of different mint concentrations on the population fluctuation of *T. confusum* in wheat grains under laboratory conditions.

Treatments	Population at different observation Intervals			
	Pre-treatment	1 st Month	2 nd Month	3 rd MONTH
0.05%	20	15.00±1.53b	46.33±2.60b	71.33 ±1.45b
1%	20	11.33±0.88bc	34.00±4.36c	63.00 ± 3.46bc
1.05%	20	8.67±11.45cd	25.33±1.86c	53.67 ± 2.91c
2%	20	6.00±11.00d	13.67±1.76d	30.00 ±3.61d
Control	20	21.00±1.73a	70.67±3.53a	88.00 ±3.79a

The overall weight loss in wheat due to the feeding of *T. confusum* under the effect of different mint concentrations mixed with wheat is given in Table 3. Results indicated that overall weight loss in various concentrations showed a highly significant difference ($F = 44.2$, $P < 0.0000$) due to feeding of *T. confusum*. Accordingly, at the end of the experiment, the highest weight loss was observed in control (47.00 grams or 23.50%), followed by

0.50% (39.33 grams or 19.67%) and 1.50% (24.33 grams or 12.17%) concentrations. The lowest overall weight loss was observed in 2.00% mint concentration (11.33 grams or 5.67%). Similar to overall weight loss, significantly ($F = 69.90$, $P < 0.001$) the lowest and highest grain weight loss was observed in 2.00% mint concentration (17.33 grams or 8.67%) and control (54.00 grams or 27.00%), respectively.

Table 3. Weight loss of wheat grains mixed with different mint concentrations by *T. confusum* under laboratory conditions.

Treatments	Initial weight (grams)	Final weight (grams)	Grain weight (grams)	Over all weight loss (grams / %)	Grain weight loss (grams / %)
0.50%	200	160.67± 1.45d	152.67±0.88d	39.33g (19.67%)	47.33g (23.67%)
1.00%	200	167.67 ±1.45c	160.33±0.88c	32.33g (16.17%)	39.67g (19.83%)
1.50%	200	175.67 ±1.45b	169.33±1.20b	24.33g (12.17%)	30.67g (15.33%)
2.00%	200	188.67 ±0.88a	182.67±0.88a	11.33g (5.67%)	17.33g (8.67%)
Control (0%)	200	153.00±3.79e	146.00±2.52e	47.00g (23.50%)	54.00g (27.00%)

Discussion

Although, not much research has been carried out on the insecticidal properties of mint, but in different regions of the world, various locally available plants were evaluated against noxious insect pests for their detrimental effects. Among these studies, Khani and Rahdari (2012) proved that essential oil extracted from coriander seeds was found effective against adults of *T. confusum* and *C. maculatus* as significant mortality was recorded in 1 to 7-day-old adults of both the species. The mortality of pests increased with the

rise in the concentration of the oils from 43 to 357 µL/L air and exposure timing from 3 to 24 hours. (Sener *et al.*, 2009) also described the insecticidal properties of essential oils obtained from *R. officinalis*, *S. officinalis*, *O. syriacum*, and *T. vulgaris* against *T. confusum* in stored wheat. The γ -terpinene, β -thujone, 1,8-cineole, and p-cymene, respectively were the main constituents of the plant materials used in the study. Different concentrations i.e., 2, 4, 8, 16, and 32 µL/100 mL jar were applied on the filler paper for various plant essential oils. The results obtained indicated that

with an increase in the concentrations of the oils, a rise in the mortality of the targeted beetles was recorded. Among the constituents of the four plants, 1,8-cineole obtained from rosemary and common sage caused the highest mortality, followed by β -thujone from common sage and p-cymene from thyme. Another study also highlighted the potential of the powders and the extracts of hexane and methanol of seeds and leaves of *A. indica*, *B. aegyptiaca*, and *O. basilicum* against *Trogoderma granarium* and *T. castaneum*. All the extracts were applied against 1st to 4th larval instars along with adults both species at 0, 1, 2, 4, and 5-gram doses of the various plant parts extracts and the insecticide primiphos (Actellic 50% EC). After 14 days of exposure, 100% mortality of adult *T. granarium* was recorded with the application of all the concentrations of *B. aegyptiaca* kernels. However, after 14 days of exposure, comparatively lower mortalities were observed in the 1st and 4th larval instars and adults of *T. castaneum* when *B. aegyptiaca* was applied at the above concentrations. Laloub kernels hexane extracts showed 100% mortality of the 1st larval instar and adult stage of *T. granarium* and *T. castaneum* after 14 days at all concentrations, while the 4th larval stage showed few percentage mortalities (Mohamed, 2015). (Hanif et al., 2016) determined the repellency and mortality of *T. castaneum* due to the application of tobacco, bitter apple, and neem using the filter papers. The concentration of three botanicals used in the study as was 5.0%, 10%, 15%, whereas, observations were recorded after 24, 48, 1 week, one, two, and three months of the application. All the tested plants showed good results to cause repellency among the individuals of *T. castaneum* with repellency of 26.56%, 25.67%, and 23.89% recorded for bitter apple, neem, and tobacco, respectively. The maximum mortality results were obtained with the use of tobacco at 10% concentration that caused 86% mortality after 48 hours of application. Neem extracts exhibited more repellency effects than to cause mortality. (Khan et al., 2016) also studied the growth-inhibiting properties of botanicals i.e., *Verbena tenuisecta*, *Parthenium hysterophorus*, *Calotropis procera*, and *A. indica* as growth inhibitors of *T. castaneum*. Various extracts were prepared using either ethanol or acetone. Extracts of *V. tenuisecta*

at a concentration of 500 ppm was proved effective in reducing the egg-laying of beetles as minimum fecundity (61.00) was recorded in it, followed by *A. indica* (77.50 eggs) at 1000 ppm concentration. The maximum mean oviposition (139.67) was recorded in wheat flour treated with *P. hysterophorus* at 500 concentrations, followed by the same plant extracts (144.67 eggs) when applied at 250 ppm concentration. *P. hysterophorus* was also found the most efficient in the fertility of the *T. castaneum* followed by *V. tenuisecta*, both applied at 500 ppm. However, *C. procera* at 1000 ppm concentration caused the maximum larval inhibition, but not significantly different from remaining treatments including control. Maximum pupal inhibition was recorded in flour treated with *V. tenuisecta* extracts. It was also observed that the fecundity of *T. castaneum* was more affected by the acetone extracts of various plants, whereas, other parameters were more influenced by ethanol extracts.

Conclusion

All the mint concentrations exhibited the potential to elicit mortality and influence the population of *T. confusum*. While 2.00% of mint concentration caused the highest mortality and support less population development of *T. confusum*. The lowest mortality and maximum population growth were recorded in 0.50% mint concentration. Overall, the maximum and minimum weight loss was recorded in control and 2.00% mint concentration.

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