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Effect of Different Plant Extracts on Olfactory Response of *Bactrocera zonata* Saunder and Postharvest Quality of Banana (*Musa acuminata* Colla)

Alia Tajdar^{1,3}, Shaghef Ejaz², Muhammad Shah Zaib¹, Anum Ishfaq¹, Syed Muhammad Zaka^{1*}, Hafiz Muhammad Safeer¹ and Khalid Abbas¹

¹Department of Entomology, Faculty of Agricultural Sciences and Technology, Bahauddin Zakariya University, Multan, Pakistan ²Department of Horticulture, Faculty of Agricultural Sciences and Technology, Bahauddin Zakariya University, Multan, Pakistan

³College of Plant Protection, China Agricultural University, Beijing, China

ABSTRACT

Bactrocera zonata is an important pest of different fruits in tropical and subtropical regions. It damages the fruits at early ripening stage and reduces the quality and the final yield of fruit crop. Moreover, to maintain the quality parameters of fruits after harvesting, is an important aspect of post-harvest management. For the management of fruit fly and to keep the natural quality of fruit after harvesting different methods are used (cultural practices, mechanical practices, biological practices), but chemical control practices are dominant among all. Besides quick and effective outcomes of chemical control. It also has side effects (residues in fruits, human health issues, resistance, damage to non-target population, environment pollution). In the present study the effect of different botanical extracts on the olfactory response of B. zonata and on the post-harvest quality parameters of banana was evaluated. Extracts of Moringa olifera, Mentha pipertia, Zingiber officinale, and Cinnamomum spp. were prepared in methanol, ethanol, and distilled water. The olfactory response of B. zonata was studied in Y-tube olfactometer and effect on the quality parameters (colour, taste, weight loss, pH, and titratable acidity) of banana was evaluated in laboratory conditions. The olfactory results of the present study showed that all botanical extracts exhibited the repellent effect against B. zonata, but the methanolic extract of cinnamon showed the highest repellence activity against B. zonata. Moreover, no side effects were observed on the post-harvest quality of banana treated to different methanolic extracts as compared to control. This shows that botanical extracts could be used for the control of B. zonata and for the better post-harvest management of fruits.

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Authors' Contribution

AT performed the experiment and wrote the manuscript. SE helped in postharvest parameters. MSZ did the statistical analysis. AI performed the experiment. SMZ designed and supervised. HMS helped in experiment and manuscript editing. KA helped in graphical presentation of this study.

Key words

Behaviour response, Botanicals, Postharvest quality, Repellent, Bactrocera zonata

INTRODUCTION

Tephritidae fruit flies of the genus *Bactrocera* are destructive pest that have negative effect on fruit and vegetable production worldwide (De Meyer *et al.*, 2007). *Bactrocera zonata* (Saunder) (Tephritidae, Diptera) commonly known as peach fruit fly is a severe polyphagous pest having more than 50 wild and commercial host plant species (Allwood *et al.*, 1999; White and Elson-Harris, 1992).

^{*} Corresponding author: zaka_ento@bzu.edu.pk 0030-9923/2023/0002-735 \$ 9.00/0



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B. zonata is considered as one of the most damaging and serious pest of guava, mango, and peach in tropical and subtropical climatic conditions (Choudhary *et al.*, 2012; Sarwar *et al.*, 2013; El-Gendy, 2017).

Different control methods have been used for the control of fruit fly such as cultural practices, but these strategies are not very significant as compared to chemical control (Bashir *et al.*, 2005). Several studies have been suggested that different insecticides are used to control the population of fruit flies e.g., malathion (Organophosphates), methomyl (Carbamate), and spinosad (Microbia) (Nadeem *et al.*, 2014). Moreover, the repeated application of insecticides develop resistance in many species of fruit flies (Daane and Johnson, 2010; Jin *et al.*, 2011; Nadeem *et al.*, 2014).

Extract of different plants such as Curcuma longa L., Acorus calamus L., Azadirachta indica A. Saussurea lappa, Valeriana jatamansi, C. B. Clarke and Peganum

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harmala L. has been reported having the repellent, growth inhibition, and oviposition deterrent effect against *B. zonata* (Naheed *et al.*, 2004; Rehman *et al.*, 2009). Acetone extract of yellow kaner, *Thevetia peruviana* (Pers.) K. Schum, datura, *Datura alba* (Nees) and chloroformed extract of sufaida, *Eucalyptus camaldulensis* (Dehnh) have the impact of repulsion and oviposition deterrence against *B. zonata* (Ilyas *et al.*, 2017). Moreover, Siddiqi *et al.* (2006) have reported strong growth inhibition and repellence effect against *B. zonata* of turmeric plant extract extracted in acetone, ethanol, and petroleum ether (Siddiqi *et al.*, 2006).

However, there is a need to develop safer and reliable methods for the control of fruit flies globally. The use of botanicals as alternative control measure is considering to be more reliable, eco-friendly, and less chance of resistance development in fruit flies (Campos *et al.*, 2019; de Oliveira *et al.*, 2014; Isman, 2006; Khan *et al.*, 2017). The aim of present study was to determine the effect of different botanical extracts (horse radish tree, *Moringa olifera* (Lam), peppermint, *Mentha pipertia* (Linnaeus), ginger, *Zingiber officinale* (Linnaeus), and cinnamon, *Cinnamomum* spp. on the olfactory response of *B. zonata* and on the quality parameters of banana fruits.

MATERIALS AND METHODS

Insect collection and rearing

Guava infested with *B. zonata* were collected from the fruit and vegetable market (Sabzi Mandi) Multan. Fruits collected from market were kept in plastic bowls $(14\times14\times7cm)$ for pupation and employed a period of two weeks for pupation. Pupae were sieved and placed in cage (46×46×52 cm). The adults of *B. zonata* were reared on artificial diet as described by Khan (2013). A cotton swab soaked in diet solution (sugar, water and yeast) was offered to adult flies and a cotton swab soaked in water given to adult flies as a moisture source. For oviposition fresh bananas were provided to adult flies. *Bactrocera zonata* was reared under laboratory conditions at $26\pm2^{\circ}C$ and $65\pm5\%$ RH.

Plant material source and extract preparation

Mentha piperita (peppermint) leaves and Zingiber officinale (ginger) root were purchased from the fruit and vegetable market There were no significant (P=0.981) differences observed among the control and the treated fruits until the 8^{th} day of storage duration and Cinnamomum spp. (cinnamon) bark was purchased from the grain market Multan (Pakistan). Moringa olifera (moringa) leaves and bark were collected from the Bio-park of Bahauddin Zakariya University, Multan (Pakistan). The extracts were prepared in different solvents (methanol, ethanol, and distilled water) by following the method as described by Ohia *et al.* (2013).

Olfactometer bioassay

The response of flies to plant extract vs control bananas was evaluated in a Y-tube olfactometer. The olfactometer was made of a glass with 2.5 cm diameter, 22 cm long base, and 21 cm long arms (Abbas *et al.*, 2021). Mix (male and female) population of *B. zonata* was used in the choice test. Arms of the olfactometer were connected to different odour sources via Teflon tube. Purified air was passed through an air pump (8 kPa s⁻¹) into each of the olfactometer arm and exited through a suction pump (Abbas *et al.*, 2021). A 12-watt LED bulb was used as a light source between the two arms of the olfactometer at 24cm distance from the olfactometer. Light source was facilitating the attraction of Fruit fly.

Four flies were released at the base of olfactometer and their response was observed for 2 min. This was repeated ten times. Fruit flies entered in each arm of the olfactometer were recorded by counting. Apparatus was rinsed with the tap water and odour source was changed after each paired olfactory test.

Post-harvest quality test

Banana fruits were purchased from the fruit and vegetable market Multan 'Sabzi Mandi' (N30°1139.2676, E71°3058.0788). Fruits were selected carefully to make sure the uniformity in appearance (colour, shape, and size). Banana were separated from the bunch, washed with distilled water, and dried at room temperature. Banana fruit was dipped in the chlorinated water (5% NaOCl) to disinfect from any contamination for 5 min and then rinsed with distilled water and dried at room temperature (Gomez-Lopez *et al.*, 2009). For experiment banana fruit was selected randomly.

Treatments

Methanolic extract exhibited the good repellence effect against fruit flies at 30% concentration as described in earlier studies (Tajdar *et al.*, 2020), therefore, the effect of methanolic extracts was checked on the postharvest quality of banana. Banana fruits were treated with methanolic extract of *M. olifera*, *Z. officinale*, *M. pipertia*, and *C. verm*. Each treatment was replicated 3 times and each replication had 12 bananas. Treated bananas were kept in growth chamber (Panasonic) at 20°C and 70% RH. Data were collected on alternative days by selecting the three bananas from each replication of each treatment.

Colour

Changes in colour during storage and ripening stage

was determined by using the numerical rating scale ranges 1-7 where (1 = 0 to < 10% yellow, 2 = 10 to < 30% yellow, 3 = 30 to < 50% yellow, 4 = 50 to < 70% yellow, 5 = 70 to < 90% yellow, 6 = 90 to 100% yellow and 7 = blackened / rotten) as described by (Hossain and Iqbal, 2016).

Weight loss (%)

Wight loss of fruits was determined by weighing the fruits on each day and compared the weight of each day with initial weight (day 1 weight) (Shrestha *et al.*, 2018).

pH and titratable acidity (TTA)

The weighed amount of banana pulp was taken and diluted in the distilled water (water quantity was equal to the banana weight). Mixture was blended for 1 min until homogenized and become juicy (Liew and Lau, 2012). pH was determined by using the pH meter (Milwaukee, Mi. 180 Bench meter) (Zahoorullah *et al.*, 2017). Titratable acidity was determined by following the method as described by (Ranganna, 2011).

Taste

Taste of fruits was determined by using the scale of 1-4 where (1= very sweet, 2= sweet, 3= less sweet and 4= taste less) (Bico *et al.*, 2010).

Data recording and analysis

The olfactory data were analysed and compared by using a paired t-test at a 95% confidence level. The data for repellence, oviposition deterrence, and postharvest quality of banana were analysed by using one way analysis of variance (ANOVA), followed by all pair comparison tests of treatment with LSD test (α =0.05). The analysis was performed through SAS v.8 statistical package (SAS institute Inc., 1999).

RESULTS

Flies were given choices between volatile compounds released from the control arm and the arm having volatiles of the treated banana. In all three tests flies preferred the volatiles of control arm over the treated arm significantly. In case of banana treated with EMB, significantly (P=0.047, df=1) higher (26) numbers of flies preferred the control arm while ten numbers of flies opted the treated arm. Similar results were observed when banana treated with MMB and D.W MB was given to flies. In both tests, significantly (P=0.016, df=1; P=0.045, df=1, respectively) higher (24) numbers of flies preferred the control arm while eight and ten numbers of flies selected the treated arm, respectively (Fig. 1A).

In this olfactory test, flies respond to the moringa leaves extract similarly to the moringa bark extract volatiles. When treated banana with EML was given to flies, significantly (P=0.049, df=1) 24 flies preferred the volatiles of the control arm while ten flies were attracted towards the treated arm. Flies exhibited similar response to the banana treated with the MML and D. W ML extracts. In both, significantly (P=0.047, df=1; P=0.034, df=1, respectively) higher (18) number of flies attracted towards the control arm while six and eight number of flies respond to the treated arm, respectively (Fig. 1B).



Fig. 1. Olfactory response Mean±SEM, n=10 of adult *Bactrocera zonata* flies towards an olfactometer arm with odour sources of A, distilled water extract of moringa bark (D.W MB), methanolic extract of moringa bark (EMB). B, distilled water extract of moringa leaves (D.W ML), methanolic extract of moringa leaves (MML), and ethanolic extract of moringa leaves (EML).

Flies respond to the control arm volatiles more significantly than the arm having the volatiles of treated banana. The banana treated with EC and MC was offered to flies, significantly (P=0.011, df=1; P=0.001, df=1, respectively) 26 flies preferred the control arm while six and eight flies opted the treated arm volatiles. Likewise, significantly (P=0.003, df=1) higher number (22) of flies preferred the control arm as compared to the treated arm (Fig. 2A).

When olfaction was done with ginger, flies exhibited strong attraction to the control arm than the treated arm. When the banana treated with EG and MG was given to flies, significantly (P=0.047, df=1; P=0.016, df=1, respectively) higher (22) numbers of flies opted the control arm as compared to the treated arm. Similar response was exhibited by flies when the banana treated with D. W G was offered. Significantly (P=0.009, df=1) more number of flies (24) exhibited strong preference to the control arm over the treated arm (Fig. 2B).

Olfactory results showed that comparatively more numbers of flies preferred the control arm over the treated arm. When flies were given choices between the control and the treated banana with EM and MM, significantly ((P=0.145, df=1; P=0.034, df=1) higher numbers (22 and 18, respectively) of flies preferred the control arm as compared to the treated arm. Likewise, significantly (P=0.242, df=1) higher number of flies (20) preferred the control arm volatiles as compared to the treated arm volatiles (Fig. 2C).



Fig. 2. Olfactory response of Mean±SEM, n=10 adult Bactrocera zonata flies towards an olfactometer arm with odour sources of A, distilled water extract of cinnamon (D.W C), methanolic extract of cinnamon (MC), and ethanolic extract of cinnamon (EC); B, distilled water extract of ginger (D.W G), methanolic extract of ginger (MG), and ethanolic extract of ginger (EG); C, distilled water extract of peppermint (D.W M), methanolic extract of peppermint (MM), and ethanolic extract of peppermint (EM).

Postharvest quality of banana

Colour

The changes in colour were recorded on numerical scale ranges from 1-7, which represents the fruit's yellowness level. The value of colour scale for all untreated and treated banana gradually increased throughout the storage life, but there were no significant differences in the colour values of all the treatments on the 8th day of storage was observed (Table I).

Weight loss (%)

In all treatments increase in weight loss was recorded in storage. There were no significant (P=0.981) differences observed among the control and the treated fruits until the 8th day of storage duration (Table I).

pH and titratable acidity (TTA)

The pH and Titratable acidity (TTA) of banana fruit changes during the storage duration. In all treatments, pH of banana was increased in storage life. There were no significant differences observed in the pH of banana until the 8th day of storage life. The TTA of fruit is an important aspect related with the maturation and ripening of banana. The values of TTA in banana were reduced gradually with storage duration. There were significant differences observed on the 8th day of storage life. The lowest values of TTA were observed in moringa leaves and ginger treatments, respectively, on 8th day of storage as compared to other treatments (Table I).

Taste

The taste scale is an important aspect for fruit taste evaluation related to the sweetness and sourness of the fruit. In storage life the taste scale value was increased significantly. The value of taste scale was reached to maximum in the control, moringa leaves, and peppermint, respectively, on the 6th day of storage life, but there were no significant differences observed in all treatments. Moreover, skin of fruit was ruptured on 8th day of storage life, so, no data were recorded on 8th day of storage (Fig. 3).

DISCUSSION

Fruits are important part of human diet and are good source of vitamins and nutrients (Melse-Boonstra, 2020). Moreover, fruits are good source of foreign exchange and countries can earn a lot of foreign exchange by exporting different fruits (Akgüngör *et al.*, 2002). Orient to the fruit's importance, different control measures were designed and used for the management of pests as well as to increase the shelf life of fruits. In all control practices, chemical control was dominant in both pest management and shelflife enhancement of fruits. Besides quick control, chemical control has side effects (pest resistance, chemical residues in fruits, human health issues, environmental pollution, etc.) (Wojciechowska *et al.*, 2016). Therefore, in the present study, the role of plant extract in pest management and enhancement of fruit shelf life was evaluated. Moreover, the effect of different plant extracts on the olfactory response of *B. zonata* adults was assessed. Plants have potential to control insect pests and can effectively be used as eco-friendly insecticides for the control of pests.



Fig. 3. Effect of different treatments on the taste development of banana in storage life. Bars with similar letters are non-significant (P=0.05) and bars with same colour are representing same analysis.

The olfactory results of the present study revealed that methanolic extracts of *M. olifera* leaves and bark, *Z. officinale, M. pipertia* and *C. verm* have repellent effect on *B. zonata* adults. The cinnamon extract exhibited the highest repellency against *B. zonata* adults in all type of solvents. Extracts prepared in methanol were more repellent as compared to other solvents this could be due to that methanol has capacity to extract those compounds from plants which are repellent to insects. In accordance with Nathan *et al.* (2006) and Prabhu *et al.* (2011), who recorded that methanolic extract of *Melia azedarach* and moringa showed the highest repellency to *Anopheles stephensi* (Liston). Muryati *et al.* (2012) studied that

methanolic extract of citronella grass had no repellent effect against Carambola fruit fly this may be due to the difference in fruit fly species and plant used for extract preparation. However, Palanikumar et al. (2017) recorded that methanolic extract of Callistemon citrinus (Myrtaceae) leaf possessed the larvicidal potential against mosquito Aedes aegypti. Likewise, Hossain and Khalequzzaman (2018) studied that methanolic extract of Azadirachta indica A. have repellent effect while methanolic extract of sweet guard exhibited the oviposition deterrence effect against Bactrocera cucurbitae (Coquillett). Maduagwuna et al. (2020) recorded that the methanolic extract of Caryota no (CN) seeds caused non-significant decrease in total protein levels, AChE activity, negative geotactic behaviour and fecundity of Drosophila melanogaster. The methanolic extract may have compound which repel the dipteran pests such as flies and mosquitoes. Moreover, Ayalew (2020) studied that ethanolic extract of Lantana camara exhibited the highest repellency against Sitophilus zeamais than methanolic extract. The difference in repellency effect to pests could be due to the difference in solvents and their efficiency to extract compounds from different plants. Moreover, this could be due to the difference in pest species and their response to different extracts.

In the current study the effect of methanolic extracts on different physical and chemical properties of banana was evaluated. There were no significant differences observed in the physical and chemical properties of treated and untreated banana (Table I). The colour of fruit is an important parameter which indicates the fruit ripening stage and shows that fruit is fit for consumption. The values of colour scale were increased gradually as days increased in storage, but in all treatments including control there were no obvious difference observed in colour change of banana. The results are in accordance with Mohammadi *et al.* (2021) who studied that there were no significant differences observed in the

Table I. Effect of different treatments on the physical and chemical parameters of banana till the 8th day of storage life.

Treatment	Colour	Weight loss (%)	рН	Titratable acidity
Control	6.63A±0.20	$10.78 \; A \pm 0.79$	$5.57A\pm0.15$	$0.15 \ AB \pm 0.02$
Moringa leaves	6.63A±0.20	$9.89A\pm0.63$	$5.57A\pm0.07$	$0.11 \ B \pm 0.02$
Moringa bark	6.73A±0.13	$10.10 \; A \pm 2.11$	$5.60\ A\pm0.03$	$0.15 \ AB \pm 0.02$
Ginger	6.40A±0.10	$9.82\ A\pm0.50$	$5.40\ A\pm0.06$	$0.11 \ B \pm 0.02$
Peppermint	$6.40A{\pm}0.20$	$9.66\ A\pm1.19$	$5.53 \ A\pm 0.15$	$0.13 \ AB \pm 0.00$
Cinnamon	6.30A±0.20	$9.80A\pm0.62$	$5.40\ A\pm0.03$	$0.17~AB\pm0.02$

Means (\pm S.E) along the columns having same letters are statistically similar (P=0.05).

treated and untreated strawberry colour change in storage conditions. Similar results were recorded by Parsa et al. (2020) that in storage the colour become darker and there was no significant difference observed in the treated and untreated cherries colour. Weight loss was recorded in all treatments and increase in weight loss in storage conditions was observed. No significant difference was observed in the weight loss between the treated and untreated banana until 8th day of storage. The results agree with Contigiani et al. (2020) who studied that non-significant difference in weight loss was observed in plant extracts treated and untreated strawberries. In contrast, chillies coated with ALV gel showed lower weight loss as compared to the control (Ul Hasan et al., 2021). Loss in weight could be due to the fact that fresh fruits respire and as a result loss in moisture occurs. Moisture loss causes the weight loss as well as senescence of fruit. Moisture loss can be reduced through coatings with coating material (Ratra et al., 2016). pH of banana increased as days of storage increased. Moreover, there were no significant differences observed in the pH of treated and untreated bananas. The results coincide with Mohammadi et al. (2021) who recorded that pH of strawberry fruits increased during storage time. Moreover, the treatments did not alter the pH of strawberry significantly. Total titratable acidity (TTA) of all treatments decreased along storage duration except moringa leaves and ginger treatments. The results agree with Nair et al. (2018) who studied that TTA of guava treated with PPE extract decreased gradually. The decrease in TTA of fruits could be due to that the changes in the metabolic process of fruits during the respiratory process (Rasouli et al., 2019).

CONCLUSION

The outcomes of the present research revealed that botanicals can be used as an alternative of synthetic pesticides for the management of *Bactrocera zonata* Saund. (Tephritidae: Diptera) to protect the environment from their hazards. Among all extracts, methanolic extract of peppermint is most effective and it also exhibited no effect on the quality of fruit. However, further research is required for isolation and identification of phytochemicals, responsible for such activity so that this may be exploited for the management of *B. zonata* population in the field.

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Statement of conflict of interest

The authors have declared no conflict of interest.

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