



Species Diversity and Surveillance on Population Fluctuation of *Bacterocera zonata* and *Bacterocera dorsalis* Fruit Flies (Diptera: Tephritidae) in Citrus orchards of Punjab, Pakistan

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Abstract: Field experiments were conducted to study the species diversity and population fluctuation of *B. dorsalis*, *B. zonata* in three orchards of citrus at Nankana Sahib (Orchard I), Toba Tek Singh (Orchard II), and Sargodha (Orchard III), Punjab, Pakistan through methyl eugenol trap during April 2018 - April 2019. Maximum population of fruit fly (*B. zonata* and *B. dorsalis*) in three orchards was observed in the months August - September 2018 when maximum temperature was 40.52°C. *B. zonata* showed a significant positive correlation coefficient with seasonal average maximum temperature (0.629*,0.695*,0.716**) and significant positive correlation with minimum temperature (0.516*, 0.608*, 0.668*), morning relative humidity showed non-significant positive (0.116,0.165,0.151), afternoon humidity (0.108,0.113,0.124) while *B. dorsalis* also showed a significant positive correlation with maximum temperature (0.591*,0.643*,0.673*), minimum temperature (0.460*,0.523*,0.595*) and non-significant positive correlation with morning relative humidity (0.132,0.215,0.181) and afternoon humidity (0.098,0.134,0.132) in three citrus orchard respectively. It was found that incidence of *B. zonata* was higher with occurrence of 82.71%-90.70% than *B. dorsalis* with 9.30%-17.29% occurrence.

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INTRODUCTION:

Fruit flies (Diptera: Tephritidae) are considered the most destructive insect pests of fruits and vegetables, affecting yield losses, and dropping the value and marketability of horticulture crops. The hosts of these flies belong to wide variety of families of plants, and include many major commercial crops (Vayssières et al., 2008; Saleem and Salam 2013). Without flies control direct damage has been reported from 30 to 80 % depending on fruits, variety, location and fruit season (Mwatawala et al., 2006). True fruit flies comprising over 4500 species distributed globally (White and Elson Harris, 1992). There are 350 to 375 species of genus *Bacterocera* (Weems et al., 2001). Eleven species of fruit flies have been recorded in Pakistan. Among the most frequently present in field are *Bacterocera zonata*, *Bacterocera dorsalis* and *Bacterocera cucurbitae* which are present on mango, guava, citrus, apple, bitter gourd and musk melon (Abdullah et al., 2002; Stone house et al., 2002; Khan et al., 2005). In Pakistan *B. zonata* is distributed in coastal and sub coastal area of Baluchistan and Sindh as well as semi desert areas and northern plains of Punjab, Its presence has also been recorded as a rare pest from

the foothills of Islamabad, Peshawar valley of North Frontier province (Marwat et al., 1992; Sarwar et al., 2006). *B. zonata* has proved serious pest of guava, citrus and mango orchards with estimated 50 to 55 % infestation. The presence of male adult of *B. zonata* trap in orchards of mango, guava and citrus observes as 74.66, 46.62 and 21.03% respectively in Pakistan (Khan et al., 2005). *B. dorsalis*, is destructive pest in throughout tropical and subtropical region (Hasyim et al., 2004). *B. dorsalis* 1st recorded in 1916 Bihar in India and distributed south east asia, including Pakistan, India, Nepal, Burma, Thailand, Sri Lanka and China on different fruit such as citrus, guava, papaya and jaman, due to presence of tropical region it is also known as oriental fruit fly (Drew and Raghu, 2002). There are 50 to 80% fruit infestation recorded in pear, peach and in fig in Pakistan (Dhillon et al., 2005). Various methods have been adopted in controlling male fruit flies, among which, are the use of specific chemical attractant (Ibrahim and Hashim, 1980). Fruit flies can successfully be managed over local area by fruit bagging, field sanitation, protein bait, annihilation technique, growing fly resistant genotypes, augmentation of biological control, and insecticides (Akhtar uz zaman

et al., 1999;; Dhillon et al., 2005; Singh et al., 2013). The present study aimed to assess the species diversity and seasonal fluctuation of male fruit fly in citrus at Sargodha, Toba Tek Singh and Nankana Sahib Pakistan in different seasons and its correlation with a biotic stresses with use of methyl eugenol trap.

MATERIALS AND METHODS

Adult male fruit flies of different species were collected from three different district of Punjab, Pakistan orchards namely orchard-I at Chak Mohammad Pura is a town in east of Nankana sahib. It is almost 25 km from the Nankana sahib city, orchard-II at Chak 336 G.B Saraba is a town in south of Toba Tek Singh. It is almost 7 km from the Toba tek singh city. orchard-III) Chak 86 is a town in west of Sargodha. It is situated almost 3 km from the Sargodha city.

Design of the fruit fly trap and installation

The fruit fly trap was constructed from transparent plastic bottles of approximately 1 liter capacity. Two holes were cut, one in the lid and the other exactly the opposite to facilitate the entry of fruit flies. The holes were fitted with a PVC pipe of about 1/2 inch diameter with a length of 3 inches. The lure was suspended exactly in the center. Ten traps were suspended with the branches of the fruit trees randomly about 2 meters above the soil surface. All the traps were randomly distributed among citrus plants.

Attractant used in the study

Commercial formulation of methyl eugenol, a sex pheromone used specifically for capturing of *B. zonata* and *B. dorsalis* was used. Using a syringe 1 ml of methyl eugenol was applied to cotton and then suspended in the middle of the bottle. The cotton swab was replaced every month with fresh ones.

Data analysis

Data of the ten traps were pooled for a single week and expressed as the number of fruit fly captured/month. The adult fruit flies after narcotization were dried properly to preserve for proper identification. Analysis of Variance (ANOVA) was performed using statix 8.1 and means were compared by Least Square Design (LSD). The differences among means were considered significant at $P \leq 0.05$ level. The meteorological data viz. Maximum temperature, Minimum temperature,

Relative humidity I and II and rainfall were collected from the Pakistan Meteorological Department April 2018 to April 2019 and correlation was done with the incidence of fruit fly through statistical computer programme.

RESULTS

Species diversity

Studies on the species complex under citrus ecosystems using methyl eugenol as attractant revealed the presence of two species viz *B. dorsalis* and *B. zonata*. As an overall seasonal sum up, *B. zonata* had been found to be dominant one. In orchard-I (Nankana Sahib), *B. zonata* was 82.71% and *B. dorsalis* was 17.29%, in orchard- II (Toba Tek Singh) *B. zonata* was 90.70% and *B. dorsalis* was 9.30%, and in orchard-III (Sargodha) *B. zonata* was 90.03% and *B. dorsalis* was 9.97 % respectively (Fig-1). However, some seasonal variations could be noted in the percentage share of the two species.

Incidence pattern of fruit flies with the use of methyl eugenol trap in three different orchards:

The data of incidence pattern of fruit flies in the orchards of three districts Nankana Sahib, Toba Tek Singh and Sargodha is presented in Table-1.

Orchard -I (Nankana Sahib):-

The population of *B. zonata* was found in the orchard throughout the year. The highest number of *B. zonata* was trapped in the August 2018 with trap catches of 395.78 ± 18.03 fruit flies/trap/month. The trap catches slightly declined during month of September (321.44 ± 12.28 fruit flies/trap/ month) and during month of October (203.56 ± 11.02 fruit flies/trap/month). The population declined throughout the year, with lowest trap catches of 4.5 ± 0.84 fruit flies/trap/month during month of February. However the maximum population of *B. dorsalis* was observed during the month of August. The population of *B. dorsalis* was low throughout the study period except august to third week of September. Higher catches was recorded during month of August (106 ± 13.72 fruit fly/trap/month). The trap catches slightly declined during month of September (69.11 ± 8.03 fruit flies/trap/ month). The population declined to 1.78 ± 0.28 fruit flies/trap/month by the month of February. The incidence of *B. dorsalis* was observed again during July 2019 with trap catches of 24.78 ± 3.05 fruit fly/trap/month reached the peak by month of August.

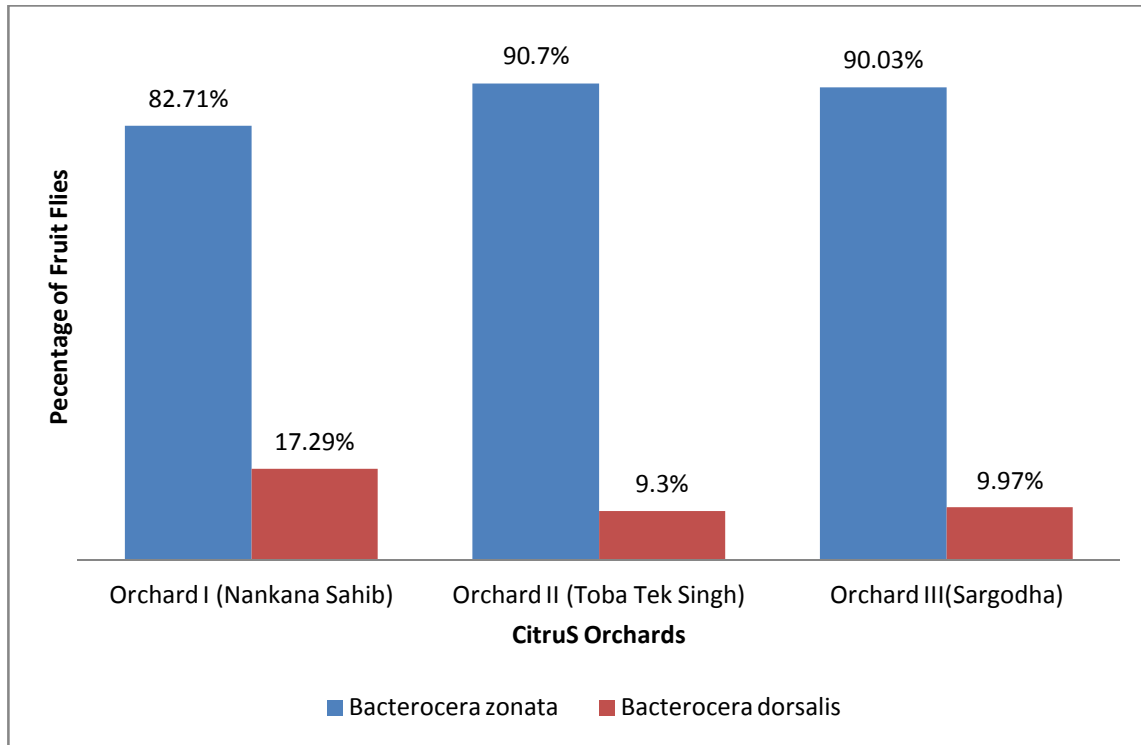


Fig 1: Percentage of different fruit fly species in citrus orchards.

Orchard -2 (Toba Tek Singh):

The population of *B. zonata* was found in the orchard throughout the year. The highest number of *B. zonata* was trapped in the month of August 2018 with trap catches of 445 ± 13.12 fruit flies/trap/month. The trap catches slightly declined during month of September (310.78 ± 6.55 fruit flies/trap/month) and in month July (341.67 ± 14.28 fruit flies/trap/month). The population later declined to reach the lowest of 15.78 ± 1.53 fruit flies/ trap/ month during month of February. The population of *B. dorsalis* was nil throughout the study period except August to September. The higher trap catches was recorded during month of August (57.33 ± 8.34 fruit fly/trap/month). The trap catches slightly declined during month of September (27.89 ± 2.32 fruit flies/trap/ month). The population reached the level of 1.78 ± 0.78 fruit flies/trap/month during of February. The incidence of *B. dorsalis* was again observed during month of July in 2019 with trap catches of 34.78 ± 1.85 fruit fly/trap/month.

Orchard -3 (Sargodha):

The population of *B. zonata* was found in the orchard throughout the year. The highest number of *B. zonata* was trapped in the month of August 2018

with trap catches of 539.44 ± 39.32 fruit flies/trap/month. The trap catches slightly declined during month of September (358.89 ± 20.59 fruit flies/trap/ month) and July (383.11 ± 14.90 fruit flies/trap/month). The population declined to reach the lowest *i.e.* 12.67 ± 2.98 fruit flies/trap/month during month of February. The population of *B. dorsalis* was near to zero throughout the study period except July to September. The higher trap catches was recorded in August (74.56 ± 12.63 fruit fly/trap/month). The population declined to 2.44 ± 0.38 fruit flies /trap /month by the month of February. The incidence of *B. dorsalis* was again observed during month of June in 2018 with trap catches of 11.44 ± 1.40 fruit flies /trap /weeks and reached the peak in July 36.11 ± 3.8 fruit flies/ trap/ month. The more dominant species observed throughout the year from April, 2018 till March, 2019 was *B. zonata*, however the population *B. dorsalis* was most dominant in the month of August to September, 2018.

Table 1. Fluctuation in the fruit fly population (*Bacterocera zonata* and *Bacterocera dorsalis*) at Citrus orchards in relation to Temperature and Humidity.

Months	Number of fruit flies <i>Bacterocera zonata</i> and <i>Bacterocera dorsalis</i> trap /month ± SE ^b in Nankana Sahib, Toba Tek Singh and Sargodha region								
	<i>Bacterocera zonata</i>			<i>Bacterocera dorsalis</i>			Average Temperature (°C)		Humidity(%)
	Orchid I Nankana Sahib	Orchid II Toba Tek Singh	Orchid III Sargodha	Orchid I Nankana Sahib	Orchid II Toba Tek Singh	Orchid III Sargodha	Minimum	Maximum	
January	15.89±1.29 ^b	35±2.73 ^a	15±0.91 ^b	2.89±0.4 ^a	4±0.71 ^a	3±0.65 ^a	7.89	16.48	75.23
February	4.56±0.84 ^b	15.78±1.53 ^a	12.67±2.98 ^a	1±0.37 ^b	1.78±0.28 ^{ab}	2.44±0.38 ^a	12	21.95	66.03
March	37±3.02 ^a	40.89±1.88 ^a	43.89±3.01 ^a	7.78±0.8 ^a	5.89±1.93 ^a	4.89±0.35 ^a	13.64	25.02	64.03
April	37.33±5.05 ^a	49.89±2.4 ^a	46.67±2.85 ^a	5.33±0.9 ^a	5±0.76 ^a	4.44±0.44 ^a	21.02	34	43.86
May	54.44±7.4 ^a	55.89±5.4 ^a	57.67±2.36 ^a	10.89±1.7 ^a	4.89±0.39 ^b	6.22±0.4 ^b	27.45	37.05	27.54
June	81.78±11.92 ^b	88.11±2.1 ^b	117±5.84 ^a	15.78±3.03 ^a	7.67±0.8 ^b	11.44±1.4 ^b	30.02	43.35	38.96
July	130.89±8.54 ^b	341.67±14.28 ^a	383.11±14.9 ^a	24.78±3.05 ^b	34.78±1.85 ^a	36.11±3.8 ^a	35.60	41.25	61.00
August	395.78±18.03 ^b	445±13.12 ^b	539.44±39.32 ^a	106±13.72 ^a	57.33±8.34 ^b	74.56±12.63 ^{ab}	32.60	40.24	65.00
September	321.44±12.28 ^a	310.78±6.55 ^a	358.89±20.59 ^a	69.11±8.03 ^a	27.89±2.32 ^b	37.67±5.09 ^b	27.70	40.52	57.25
October	203.56±11.02 ^a	237.11±7.1 ^a	220.78±11.38 ^a	34±4.61 ^a	19.33±1.48 ^b	19.56±1.57 ^b	23.35	39.63	59.56
November	107±6.56 ^b	141.56±6.41 ^a	124.22±4.4 ^{ab}	15.22±1.89 ^a	12±0.78 ^a	13±0.76 ^a	13.06	28.10	62.50
December	37.44±3.42 ^b	57.56±2.96 ^a	57.67±4.17 ^a	5.33±0.69 ^a	4.44±0.63 ^a	5.56±0.47 ^a	11.21	21.76	62.60

Table 2. Correlation studies between incidence of *Bacterocera zonata* and *Bacterocera dorsalis* fruit flies and weather parameters during April 2018 to April 2019.

Orchard (District)	Fruit fly species	Temperature (°C)		Humidity (%)	
		Maximum	Minimum	Morning	Afternoon
Orchard I (Nankana Sahib)	<i>Bacterocera zonata</i>	0.629*	0.516*	0.116 ^{NS}	0.108 ^{NS}
	<i>Bacterocera dorsalis</i>	0.591*	0.460*	0.132 ^{NS}	0.098 ^{NS}
Orchard II (Toba Tek Singh)	<i>Bacterocera zonata</i>	0.695*	0.608*	0.165 ^{NS}	0.113 ^{NS}
	<i>Bacterocera dorsalis</i>	0.643*	0.523*	0.215 ^{NS}	0.134 ^{NS}
Orchard III (Sargodha)	<i>Bacterocera zonata</i>	0.716**	0.668*	0.151 ^{NS}	0.124 ^{NS}
	<i>Bacterocera dorsalis</i>	0.673*	0.595*	0.181 ^{NS}	0.132 ^{NS}

* = significant at 5%, NS = Non significant

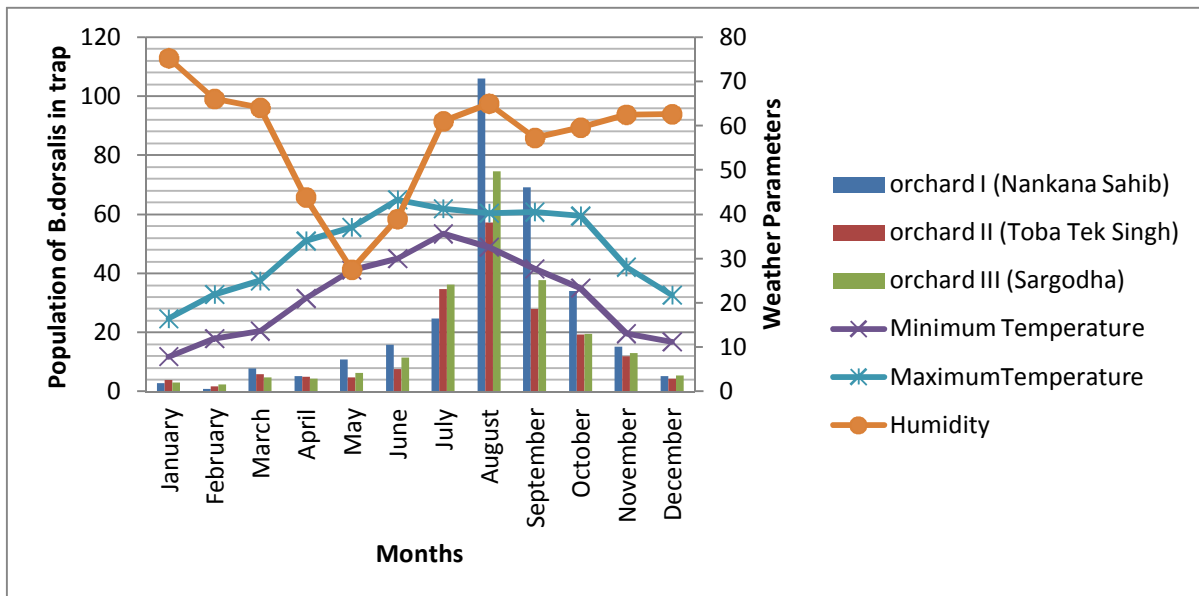


Figure 2. Correlation between *B. dorsalis* population and different weather parameters.

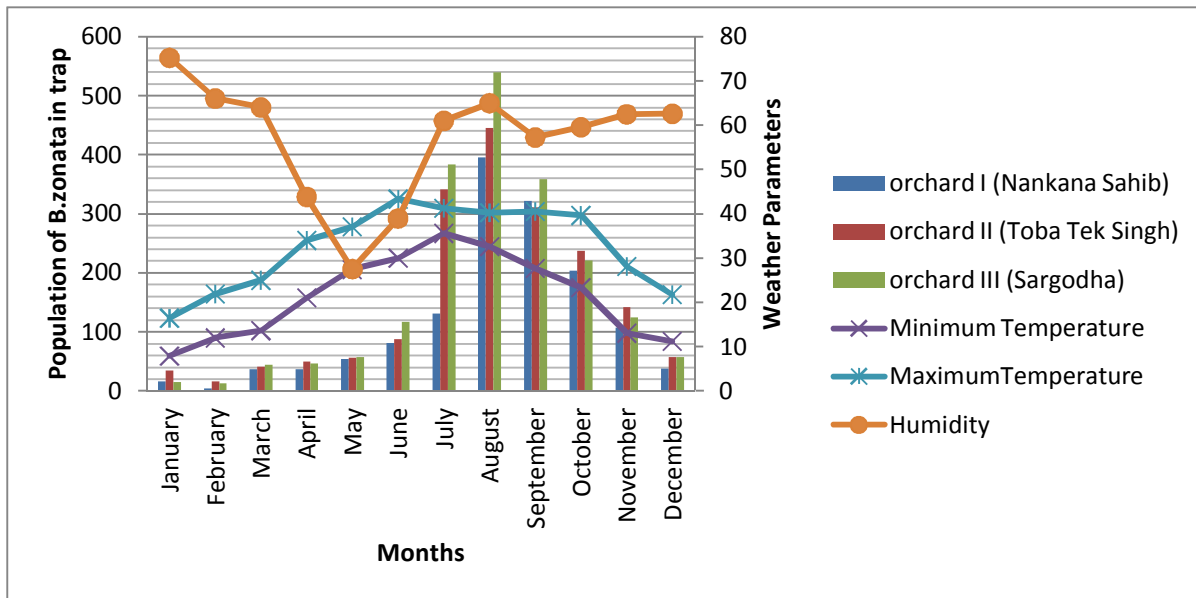


Figure: 3 Correlation between *B. zonata* population and different weather parameters.

Correlation between fruit fly population (*B. zonata* and *B. dorsalis*) and different weather parameters:

The data pertaining to relationship between fruit fly population and weather parameters is presented in the Table-2. *B. zonata* showed a significant positive correlation coefficient with seasonal average maximum temperature (0.629*, 0.695*, 0.716**) and significant positive correlation with minimum temperature (0.516*, 0.608*, 0.668*), morning relative humidity showed non-significant positive (0.116, 0.165, 0.151), afternoon humidity (0.108, 0.113, 0.124) while *B. dorsalis* also showed a significant positive correlation with maximum temperature (0.591*, 0.643*, 0.673*), minimum temperature (0.460*, 0.523*, 0.595*) and non-significant positive correlation with morning relative humidity (0.132, 0.215, 0.181) and afternoon humidity (0.098, 0.134, 0.132) in three citrus orchard respectively.

Discussion

In present study *B. zonata* was found in the citrus orchards with maximum percentage of 82.71%-90.70% was similar with the earlier works of Mahmood et al. (2007) who reported the peak incidence of fruit fly from August-September in chakwal. He also reported that *B. zonata* was the major fruit fly (60%) with presence of *B. dorsalis* (40%) in chakwal. Sushil et al. (1997) was also reported that the population of *B. zonata* peaked during 2nd fortnight of August (453 fruit flies/trap)

and September (483 fruit flies/trap) and thereafter the population declined gradually that results are similar to our finding. Gillani et al. (2002) was also reported the increase in population trend of fruit flies from May to August with peak population in the month of July in the guava orchards of Pakistan. Nandre and Shukla (2014) also studied the population dynamics of fruit fly, *B. dorsalis* on sapodilla *Manilkara achras* and observed that the fruit fly population prevailed throughout the year with maximum activity (172.1 flies per trap) during March to August which are agreement with our finding. The present study are also endorsed by the earlier findings of Seewooruthun et al. (2001); Stonehouse et al. (2002) and Jiji et al. (2009) reported that the peak catch that was observed during the fruit maturity stage (august-september) through methyl eugenol trap. This study also reports that *B. zonata* was found in the citrus orchards with maximum percentage of 89.14%-91.66% while the dominance of *B. dorsalis* in the citrus orchards of Sargodha was only 8-10%

Amice and Sales (1997) observed influence of abiotic factors, such as temperature on the population dynamics of fruit flies. Win et al. (2014) reported a positive correlation ($r=0.33$) of minimum temperature with insect count. Similarly, positive correlation of the fruit fly with temperature was reported by Kannan and Venugopala (2006). Sarada et al. (2001) reported a population of *Bactrocera* spp. showed positive correlation with minimum temperature and weak positive correlation with

maximum temperature. Our results are in agreement with these previous researchers.

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