Journal of Biodiversity and Environmental Sciences (JBES)
ISSN: 2220-6663 (Print) 2222-3045 (Online)
Vol. 4, No. 5, p. 222-232, 2014
http://www.innspub.net

RESEARCH PAPER OPEN ACCESS

Seasonal monitoring of fruit fly, *Bactrocera zonata* (Saunders) and its parasitoids *Trybliographa daci* on guava fruit from Sindh Pakistan

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Article published on May 20, 2014

Key words: Seasonal monitoring, *Bactrocera zonata,* infested guava, parasitoid, *Trybliographa daci.*

Abstract

Seasonal monitoring of fruit flies, *Bactrocera zonata* and *Bactrocera dorsalis* was carried out by collecting the infested guava fruits for recording the larval parasitoid under laboratory conditions; and weekly trap catches for fruit fly species were also recorded. The *B. zonata* and *B. dorsalis* pupae were collected from 01.11.2012 to 15.03.2013 at fortnightly interval. The data showed that *B. zonata* and *B. dorsalis* adult emergence was 65.67±1.69 and 5.54±2.35; where the female ratio dominates over the males. On their males (3.00±0.26) adult parasitoids emerged in a fortnight; and weekly fruit flies catches were 616.45±67.19/10 traps. There was significant difference in the adult emergence in larval parasitoid (16.26±1.16%) and female ratio was higher (9.16±0.84) than fruit fly trap catches between weeks (P<0.05). The species-wise insect catches for guava fruit fly indicates that seasonal weekly average fruit flies catches were 605.65 ± 66.32/10 traps. In case of *B. dorsalis,* during 1st week of November 2012, the catches were 21/10 traps which continuously decreased and reached lowest number of catches (6/10 traps) in the 4th week of February. This indicates that *B. zonata* population was markedly (P<0.05) higher than the *B. dorsalis*. There was positive and significant correlation between fruit fly infestation and relative humidity (r= 0.7244**), fruit fly infestation and temperature (r= 0.5299**), between *B. zonata* infestation and relative humidity (r= 0.7368**), between *B. zonata* infestation and temperature (r= 0.5215**), and between *B. dorsalis* infestation and relative humidity (r= 0.4356*), suggests a simultaneous increase in fruit fly infestation with increasing relative humidity and temperature.

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Introduction

Bactrocera is a large genus of tephritid fruit flies, with more than 500 species currently described (Courtice, 2006). Fruit fly, *B. zonata* originates in South and South-East Asia, where it attacks many fruit species (more than 50 host plants), including guavas, mangoes, peach, apricots, figs and citrus. It has spread to other parts of the world, in particular to several countries in the Near East and to Egypt. In recent years, *B. zonata* has become a widespread pest (Cayol, 2008). *B. zonata* is a major insect pest of fruit and vegetables not only in Pakistan but the world over as well. Fruit fly causes 25-50% loss to guava during summer (Syed, 1970 and Hashimi, 2001). The different species of fruit fly cause colossal losses to fruit and vegetables in tropical, sub tropical and temperate regions of the world (Dhaliwal and Heinrich, 1998). They cause direct loss to fruit and vegetables and indirect loss by enhancing expenditure on chemicals which alternately reduce the market value of the product. The infestation may reach up to 50% in summer crop of guava in Pakistan. The peaches, apricot, guava or other fruit attacked by this pest are malformed, mis-shaped, undersized and rotted inside. The damage caused is very heavy and the fruits become unmarketable (Atwal, 1976 and Drew and Raghu, 2002).

Bactrocera zonata (Saunders), is recognized as one of the most important and serious pests attacking fruit crops (James, 2003). It causes serious economic losses, either by direct damage to fruit or by warranting the need for quarantines and insecticide treatments. The control measures adopted rely mainly on contact poisons or baits. Contact poisons may have serious deleterious effect on health as fruits often consumed unwashed in the development countries. Besides, baits and sprays of conventional insecticides also have toxic effects on parasitoids of *B. zonata* (Sheo *et al.* 1990). However, because of the polyandrous and long distance migratory abilities of the flies with high population densities throughout the year, such control measures could not be considered suitable in the continental areas. This dilemma has made it essential to find effective control measures, which are safe to both human beings and non-target biological systems. These methods have a negative impact on the environment, and specifically on the phytoparasitic populations of beneficial organisms. Thus, environment friendly methods of control are much in need (Huang *et al.*,, 2004; Senthil-Nathan *et al.* 2008; Schmutterer and Singh, 1995; Prakash and Srivastava, 2008; James, 2003).

Green *et al.* (1993) monitored behavior of guava fruit flies and reported that flies left fruitless trees soon after release, more flies visited fruit, and flies visited more fruit and spent more time on trees with increasing fruit density, and time between last oviposition and leaving the tree was significantly greater for flies visiting infested fruit than for those visiting uninfested fruit. New parasitoid species are being obtained for tephritids and concluded that the advances in mass rearing of parasitoids has enabled researchers to perform large-scale field testing of these parasitoids to determine the potential of augmentative releases (Purcell, 1998). However, recent successes in rearing other species with different biologies (e.g. egg-pupal parasitoid Biosteres arisanus), the gregarious eulophid, *Tetrastichus giffardianus* and the pupal parasitoid, *Coptera* sp., will enable researchers to broaden the repertoire of parasitoid species for the future (Sangvorn Kitthawee, 2000; Attaullah *et al.* 2002; Rocha *et al.* 2004). Rousse *et al.* (2006) assessed the host specificity of parasitoid, initially reared on *B. zonata*, and then offered for parasitization the eight local tephritid pest species, and concluded that the size of emerging adults is affected by host species and is correlated to pupal weight. *Bactrocera zonata* would be a favorable host to support routine colonization of *F. arisanus* for mass production of this parasitoid. Shanmugam *et al.* (2011) carried out studies on the fruit flies and their parasitoids and concluded that after 2-3 weeks, adult fruit flies and their parasitoids will emerge from infested fruit and can be seen moving around the inside of the clear plastic container and suggested to carefully examine the bottom of the decayed fruits as pupae/flies will often be attached to these areas. Although, several authors have carried significant

work on the fruit fly from abroad but there was nothing was available on this subject from this region that's why an attempt has been made to study this pest along with its parasitoids *trybliographa daci* on guava fruit from Sindh.

Material and methods

Study site

The present research work was carried out during the year 2012-13 on the monitoring of guava fruitflies, *Bactrocera zonata*. During the data recording process, *Bactrocera dorsalis* were also monitored. Pheromone traps were prepared locally, and the method of its preparation is very simple and affordable by average grower. The adult populations of fruitflies were recorded weekly through phramone trap baited with lure toxicant mixture (95% methyl eugenol + 5% thiodon insecticide). The experiment was conducted to observe the fruitfly catches by hanging fruitfly traps at the height on guava trees, ten pheramone traps were installed at different locations of the guava orchard. Other climatic factors like temperature, humidity etc. were also observed.

Weekly interval observations

The study was comprised of a 20 weeks period, started on 01.11.2012, the fruitfly catch of adults were monitored at weekly interval. The natural infestation level of fruitfly, *Bactrocera zonata* and parasitism by larval parasitoid, *Trybliographa daci* were intended by observing the infesting the guava fruits. These attacked fruits were collected at fortnightly interval from guava orchard. Four infested guava fruits were kept in each replication at 26± 2 °C in laboratory. The experiments were replicated three times.

Sifting of pupae

After one week interval, the pupae were sifted and counted. These pupae were kept in Petri dishes and weighed by microbalance and then put under microscopic for further observation.

Statistical analysis

Collected data were subjected to statistical analysis using analysis of variance to know the significance of differences by using and LSD (Least Significance Difference test).

Results

The seasonal monitoring of fruit fly, *Bactrocera zonata* and *B. dorsalis* was carried out during the year 2012-13 by collecting the infested guava fruits for recording the larval parasitoid, *Trybliographa daci* under laboratory conditions. The data in regards to pupal number, pupal weight, total adult emergence (male and female) and percentage of adult emergence were examined in the laboratory for *B. zonata*, *B. dorsalis* and larval parasitoid *Trybliographa daci*. The correlations between fruit fly catches for *B. zonata*, *B. dorsalis* and temperature and relative humidity were also worked out.

Weekly catches of Bactrocera zonata and B. dorsalis

The insect traps were installed at the guava garden of Mahoota Farm for a period of 20 weeks (01.11.2012 to 15.03.2013) to monitor male fruit flies and weekly record of male catches were maintained. There were significant ($P<0.05$) differences in weekly trap catches of both the guava fruit fly species. The data (Table-1) indicated that during 1st week of November 2012, the *B. zonata* catches were 920/10 traps which increased continuously and reached highest number of catches (1211/10 traps) in the 4th week of November and later started decreasing gradually reaching lowest level of catches (265/10 traps) in 3rd week of February. There was increasing trend in the fruit fly catches onwards and at the final observation on 4th week of March, the fruit fly catches were 449/10 traps. The seasonal weekly average fruit flies catches were 605.65 ± 66.32/10 traps. The trap catches record for *B. dorsalis* was also maintained (Table-2) which showed that during 1st week of November 2012, the *B. dorsalis* catches were 21/10 traps which continuously decreased and reached lowest number of catches (6/10 traps) in the 4th week of February and later started increasing reaching lowest 14/10 traps at the 4th week of March. This indicates that *B.*

zonata population was markedly (P<0.05) higher than the *B. dorsalis*.

Adult emergence of Bactrocera zonata

The *B. zonata* pupae were collected from 01.11.2012 to 15.03.2013 at fortnightly interval and the data (Table-2) indicated that out of total 89.33 pupae (avg. of 3 replicates), 54.66 *B.zonata* adults were emerged showing adult emergence of 61.16 percent; while in the 2nd fortnight (15.11.2012), out of 86 pupae, 30 adults emerged indicating 56.14% adult emergence. In the 3rd fortnight (01.12.2012), out of 78 pupae, 51 adults emerged showing 60.38% adult emergence; while in 4th fortnight (15.12.2012), out of 75.33 pupae, 49 adults emerged indicating 67.96% adult emergence; and in the 5th fortnight (01.01.2013), out of 67.33 pupae, 46.66 adults emerged showing 51.60% adult emergence. In the 6th fortnight (15.01.2013), out of 64.33 pupae, 44.66 adults emerged showing 63.88% adult emergence; while in the 7th fortnight (01.02.2013), out of 65.33 pupae, 49 adults emerged showing 75.01% adult emergence. Similarly, in the 8th fortnight (15.02.2013), out of 55.66 pupae, 41 adults emerged showing 73.66% adult emergence; and in the 9th fortnight (01.03.2013), out of 76.66 pupae, 56.33 adults emerged showing 73.50% adult emergence; while in the 10th fortnight (15.03.2013), out of 80.33 pupae, 59 adults emerged showing 73.50% adult emergence. Fortnightly average indicates that out of 72.03±1.95 pupae, 48.93 ± 1.26 adults emerged showing 65.67±1.69 percent adult emergence. The adult emergence was markedly higher in last four fortnights from 1st February 2013 onwards as compared to first six fortnights. Moreover, the female ratio in the adults emerged was markedly higher than the male *B. zonata*.

Adult emergence of Bactrocera dorsalis

The pupae of guava fruit fly, *Bactrocera dorsalis* were collected from 01.11.2012 to 15.03.2013 at fortnightly interval and the data (Table-3) showed 6.33 *Bactrocera dorsalis* adults emerging during 1st fortnight (01.11.12) showing 7.14% adult emergence; and in the 2nd fortnight (15.11.2012), 6.33 adults emerged indicating 9.29% adult emergence; while in the 3rd fortnight (01.12.2012), 4.00 adults emerged showing 5.06% adult emergence. Similarly, in 4th fortnight (15.12.2012), 4.00 adults emerged indicating 5.35% adult emergence; and in the 5th fortnight (01.01.2013), 3.00 adults emerged showing 4.37% adult emergence; while in the 6th fortnight (15.01.2013), 3.00 adults emerged showing 4.57% adult emergence. In the 7th fortnight (01.02.2013), 8th fortnight (15.02.2013), 9th fortnight (01.03.2013) and 10th fortnight (15.03.2013), the emerged *B. dorsalis* adults were 4.00, 3.00, 3.00 and 3.66, indicating adult emergence of 6.03, 5.30, 3.83 and 4.53 percent, respectively. Fortnightly average indicates that a total of 4.03 ± 0.28 adults emerged showing 5.54±2.35 percent adult emergence. The female ratio dominates the adults emerged over the male *B. dorsalis*.

Table 1. Weekly catches of male adult population of *Bactrocera zonata and B. dorsalis* from Guava garden of Mahoota farm during November 2012 to March 2013.

Week observation	No: of fruit flies 10^{-1} traps	No. of fruit flies 10^{-1} traps	Temperature	Relative Humidity
	B. Zonata	***B. dorsalis***		**%**
1st week November	920	21	30.2	52
2nd week	1191	19	29.5	51
3rd week	1076	16	26	50
4th week	1211	18	23	49
Dec: 1st week	713	11	25	51
2nd week	722	10	25	47

3rd week	715	9	23	48
4th week	712	10	22.8	47
Jan: 1st week	492	8	20	49
2nd week	501	7	21.3	46
3rd week	494	8	19.5	46
4th week	491	6	21	49
Feb: 1st week	288	4	21.7	47
2nd week	268	5	23	45
3rd week	265	5	22.4	44
4th week	273	6	22	42
March 1st week	457	12	24.2	44
2nd week	440	15	25	45
3rd week	435	13	25.6	44
4th week	449	14	27	42
Mean ±SD	605.65±66.32	10.85±1.12	23.86±0.64	46.95±0.65

Table 2. Adult emergence of Guava fruit fly, *Bactrocera zonata* collected from infested guava fruits at Mahoota guava orchard from 01.11.2012 to 15.03.2013.

Date	Pupal Nos.	Pupal weight (mg)	Total adults emerged	*Bactrocera zonata*		Adult emergence (%)
				Male	Female	
01.11.12	89.33	0.72	54.66	13.66	41.00	61.16
15.11.12	86.00	0.54	30.00	9.00	29.00	56.14
01.12.12	78.00	0.63	51.00	12.33	38.66	60.38
15.12.12	75.33	0.62	49.00	12.00	37.00	67.96
01.01.13	67.33	0.54	46.66	12.00	34.66	51.60
15.01.13	64.33	0.52	44.66	11.00	33.66	63.88
01.02.13	65.33	0.52	49.00	12.00	37.33	75.01
15.02.13	55.66	0.44	41.00	10.00	31.00	73.66

01.03.13	76.66	0.65	56.33	14.00	43.00	73.50
15.01.13	80.33	0.64	59.00	14.66	44.33	73.50
Mean± SD	72.03 ± 1.95	0.58 ± 0.01	48.93 ± 1.26	12.06 ± 0.35	36.96 ± 0.94	65.67 ± 1.69
LSD 0.05	11.189	0.09	5.50	1.92	3.94	9.88

Table 3. Adult emergence of Guava fruit fly, *Bactrocera dorsalis* collected from infested guava fruits at Mahoota guava orchard from 01.11.2012 to 15.03.2013.

Date	**Pupal Nos.**	**Pupal weight (mg)**	**Total adults emerged**	***Bactrocera dorsalis***		**Adult emergence (%)**
				Male	**Female**	
01.11.12	89.33	0.72	6.33	1.66	4.66	7.14
15.11.12	86.00	0.54	6.33	1.33	5.00	9.29
01.12.12	78.00	0.63	4.00	1.66	2.66	5.06
15.12.12	75.33	0.62	4.00	1.33	3.00	5.35
01.01.13	67.33	0.54	3.00	1.00	2.00	4.37
15.01.13	64.33	0.52	3.00	1.00	2.00	4.57
01.02.13	65.33	0.52	4.00	1.33	2.66	6.03
15.02.13	55.66	0.44	3.00	1.00	2.00	5.30
01.03.13	76.66	0.65	3.00	0.66	2.66	3.83

15.01.13	80.33	0.64	3.66	1.00	2.66	4.53
Mean ± SD	72.03 ± 1.95	0.58 ± 0.01	4.03 ± 0.02	1.20 ± 0.08	2.90 ± 0.23	5.54 ± 2.35
LSD 0.05	11.189	0.09	1.87	0.80	1.48	2.06

Adult emergence of larval Parasitoid, Trybliographa daci

The parasitoid pupae were collected from infested guava fruits attacked by guava fruit flies from 1st November 2012 to 15th March, 2013 and the data (Table-4) indicated emergence of 23 parasitoid adults during 1st fortnight (01.11.12) showing 25.67% adult emergence; and in the 2nd fortnight (15.11.2012), 18.66 adults emerged indicating 26.98% adult emergence; while in the 3rd fortnight (01.12.2012), 16.33 adults emerged showing 20.85% adult emergence. Similarly, in 4th fortnight (15.12.2012), 12.00 adults emerged indicating 15.72% adult emergence; and in the 5th fortnight (01.01.2013), 9.66 adults emerged showing 14.15% adult emergence; while in the 6th fortnight (15.01.2013), 8.33 adults emerged showing 12.81% adult emergence. In the 7th fortnight (01.02.2013), 8th fortnight (15.02.2013), 9th fortnight (01.03.2013) and 10th fortnight (15.03.2013), the emerged parasitoids adults were 6.66, 5.00, 10.33 and 11.33 indicating adult emergence of 10.1, 8.86, 13.36 and 14.0 percent, respectively. Fortnightly average indicates that a total of 12.13 ± 1.09 parasitoid adults emerged showing 16.26±1.16 percent adult emergence in parasitoids. The female ratio was remarkably higher (9.16±0.84) than their males (3.00±0.26) adult parasitoids emerged in a fortnight on average.

Table 4. Adult emergence of larval parasitoid, *Trybliographa daci* collected from infested guava fruits at Mahoota guava orchard from 01.11.2012 to 15.03.2013.

Date	Pupal Nos.	Pupal weight (mg)	Total adults emerged	*Trybliographa daci*		Adult emergence (%)
				Male	Female	
01.11.12	89.33	0.72	23.00	5.33	17.66	25.67
15.11.12	86.00	0.54	18.66	4.33	14.33	26.98
01.12.12	78.00	0.63	16.33	4.00	13.00	20.85
15.12.12	75.33	0.62	12.00	3.00	9.00	15.72
01.01.13	67.33	0.54	9.66	2.66	7.00	14.15

15.01.13	64.33	0.52	8.33	2.00	6.33	12.81
01.02.13	65.33	0.52	6.66	1.66	5.00	10.10
15.02.13	55.66	0.44	5.00	1.66	3.66	8.86
01.03.13	76.66	0.65	10.33	2.66	7.66	13.36
15.03.13	80.33	0.64	11.33	3.00	8.33	14.00
Mean± SD	72.03 ± 1.95	0.58 ± 0.01	12.13 ± 1.09	3.00 ± 0.26	9.16 ± 0.84	16.26 ± 1.16
LSD 0.05	11.189	0.09	5.39	1.70	3.89	4.70

Correlations of fruit fly with climatic factors

Fruit fly v/s Humidity

Correlation between fruit fly infestation and relative humidity was positive and significant (r= 0.7244**) which shows that with increasing relative humidity, the fruit fly infestation increased simultaneously.

Fruit fly v/s Temperature

Correlation between fruit fly infestation and temperature was positive and significant (r= 0.5299**) which indicates that with increase in temperature, the fruit fly infestation increased considerably.

Bactrocera zonata v/s Humidity

The correlation coefficient between *Bactrocera zonata* infestation and relative humidity was positive and significant (r= 0.7368**) which indicates that with increasing relative humidity, the fruit fly infestation increased considerably.

Bactrocera zonata v/s Temperature

The association of *Bactrocera zonata* infestation and temperature was positive and significant (r= 0.5215**) which suggested that with increasing temperature, the *Bactrocera zonata* infestation also increased significantly.

Bactrocera dorsalis v/s Humidity

The relationship between *Bactrocera dorsalis* infestation and relative humidity was positive and significant (r= 0.4356*) which describes that with increasing relative humidity, the *Bactrocera dorsalis* infestation will also increase.

Bactrocera dorsalis v/s Temperature

The interrelationship between *Bactrocera dorsalis* infestation level and temperature was positive and significant (r= 0.8065**) which proposes that with

increasing temperature, the *Bactrocera dorsalis* infestation will also increase considerably.

Table 5. Correlation between relative humidity/temperature and guava fruit fly infestation.

Fruit fly species	Humidity	Temperature
Fruit fly (overall)	0.7244**	0.5299**
Bactrocera zonata	0.7368**	0.5215*
Bactrocera dorsalis	0.4356*	0.8065**

Discussion

The findings of present study showed that *B. zonata* adult emergence was markedly higher in last four fortnights from 1st February 2013 onwards as compared to first six fortnights. The female ratio in the adults emerged was markedly higher than the male in *B. zonata*, *Bactrocera dorsalis* and larval parasitoid, *Trybliographa daci*. Similarly, Seasonal weekly average fruit flies catches were 616.45±67.19/10 traps. There was significant difference in the fruit fly trap catches between weeks ($P<0.05$). *B. zonata* population was markedly ($P<0.05$) higher than the *B. dorsalis*. The interrelationship of fruit fly infestation with relative humidity and temperature was positive and significant, which suggests that with increasing relative humidity and temperature, the fruit fly infestation will increase simultaneously. These results are in accordance with those of Sangvorn Kitthawee (2000) who reported that abundances of fruit and fruit flies per fruit were correlated with seasonal changes in the mean maximum monthly relative humidity. The ratio of female : male parasitoids was 4:3.

Khan (2002) reported that population of fruit flies remained present throughout the fruiting season from flowering to maturity of all the fruits. The maximum infestation of fruit flies in apple orchards was recorded to be 4.61 % on October 1 and was statistically at par with 4.53 and 4.51 % recorded on September 15 and October 15, respectively. The maximum infestation was recorded to be 14.72% on March 1 in 'ber' orchards followed by 11.33 and 10.26% on February 15 and March 15, respectively. Guava fruit showed maximum infestation i.e. 7.56% followed by 5.66, 5.32 and 2.39% on 'ber', mango and apple, respectively. The months of August and September showed maximum infestation (11.38 to 15.36%) in guava orchards. In another study, Alrouechdi and Marouene (2003) concluded that *Bactrocera zonata* causes up to 100% loss when untreated. Rousse *et al.,* (2006) concluded that the size of emerging adults is affected by host species and is correlated to pupal weight. *Bactrocera zonata* would be a favorable host to support routine colonization parasitoid for mass production of this parasitoid. El-Kousy *et al.* (2012) reported that *Bactrocera zonata* exhibited gradually increase in numbers and showed annually peak, with an average of 58.63 and 110.86 individuals/lure trap in October of both 2010 and 2011 seasons, respectively. Peaks of the pest were coincided with the ripening period of guava fruits in both seasons. The pest incidence showed highly significant difference between the examined areas. Infestation patterns of the fruit flies in guava orchards showed that most of the ripening and/or fallen guava fruits were infested by the pest (23.21 pupae/18.54 fruits).

Mean numbers of the emerged adults is equal 75.79% of the collected pupae. The emerged *B. zontata* flies is equal 6.71 fold of *C. capitata*. Ali *et al.* (2012) reported that significant positive correlation (r) of fly incidence was noted with maximum and minimum temperature (r = +0.395 and +0.413 respectively)

with the fruit fly catch per trap for the first year 2008/09 and (r = +0.243 and +0.280 respectively) for the second year 2009/10. During the present study it was also observed that relative humidity have contributed significant role to the fluctuation of fruit flies populations and fruit infestation. Monitoring based on Pheromone traps is necessary to detect the presence of the different species and to evaluate their populations, because both the species and their populations are continuously changing. Angood and Sunaid (2012) found that pest is available almost all the year around, but it reached its peak in September, where the mean pheromone trap catch was 2278 male adults. The number declined when the temperature decreased reaching its minimum (196 male adult) in January and March. 232Darwish *et al.* (2013) reported that a remarkable decrease in the population could be seen from the first week of December until the end of the season (fourth week of December) however, during the present study we have observed that there is significant differences between fruit fly population captured at 1m trap height (Mean = 446.9 ± 97 flies/trap/day) and that captured at 2m trap heights (Mean = 321 ± 72 flies/ trap/day) in different months of the years.

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