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BIOLOGY AND MORPHOMETRIC OF DIFFERENT LIFE STAGES OF THE ORIENTAL FRUIT FLY (*BACTROCERA DORSALIS* HENDLE*)* (DIPTERA: TEPHRITIDAE) ON THREE VARIETIES OF MANGO OF SINDH, PAKISTAN

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ABSTRACT

A study on the biology and morphometric of different life stages of *Bactrocera dorsalis* (Hendle) was carried out on three varieties of mango, namely Beganpali, Sindhri and Chunsa in laboratory under controlled conditions 26±1°c and 65±5%, at the department of Zoology University of Sindh Jamshoro, from June to August 2013. It was observed that the eggs of *Bactrocera dorsalis* were white, shiny, rice shaped, slightly curved, elongated and tapered at the anterior end; whereas the posterior end is broadly rounded. The average length and width were 0.54±0.11mm and 0.19±0.08mm, respectively. The first and second instars measured 2.6±0.75mm and 0.55± 5.88mm in length respectively, and 0.27±0.82mm and 2.34±0.7mm in width respectively. The 3rd instar was very active, jumping from mango fruits to sand measured 7.69±0.72mm in length and 3.58±0.25mm in width. The puparia measured 4.47±0.64mm in length and 2.69±0.16 mm in width. The duration of egg hatching and the larval, pre-pupal and pupal period was 1.61±0.51days, and 9.97±2.25 days, 2.07±0.86 days and 8.52±0.88 days respectively. The average pre-oviposition period was 13.55±1.33, whereas the oviposition period was 21.46±1.42 days. Fecundity varied from 14-15 eggs per day. Sex ratio (♂**:** ♀) was 1:3. When adult fed on different diets such as on mango (natural diet) longevity extended from 20-30 days to males and 25-45 days for females, and when fed on water only 4-5 days male and female 5-8, while when they were fed both on (sugar and water) 13-25 days male and 15-30 days female, but without food the longevity was survived to only for 1-2 days. The longevity of fruit flies on different diets was significantly different ($P<0.05$). Developmental time duration of life stages of *Bactrocera dorsalis* was recorded on Chunsa, Sindhri, Beganpali varieties, incubation of egg (1.5±0.02 days, 2.3±0.16 days, 2.39±0.01 days, respectively), Larval developmental period (6.51±0.2 days, 7.5±0.16 days, 7.4±0.16 days, respectively), Pupal period (8.8±0.46 days, 8.9±0.40 days, 8.5±0.16 days, respectively), total life cycle period (16.8±0.086 days, 18.72±0.72, 18.29±0.33 days, respectively), There were no significant difference ($P>0.05$) between the developmental time of life stages of *Bactrocera dorsalis* on three varieties of mango .

Key words: Biology, Morphometric, Oriental fruit fly, Mango varieties.

INTRODUCTION

The Fruit flies belong to family Tephritidae. They are agriculture pests, which mostly attack fruits and vegetables, found all over world. (White and Elson-Harris, 1992). Fruit flies are economically important; their favorable hosts are mango, guava, peach as well as other fruits (Gafoor *et al.,* 2010). Their annual loss of fruits and vegetables by fruit flies are about 144.4 million US dollars (Stone house *et al.,* 2002). The production of mangoes is reduced by certain pre-harvesting problems, especially insect pest. Mango fruit flies *Bactrocera dorsalis* are major pest of mangoes, (Ishaq *et al.,* 2004; Iqbal *et al.,* 2004). In Pakistan two species of genus *Bactrocera* were found, *B.dorsalis* and *B.zonata,* which attack on mango orchards, (Stone house *et al*., 2002). The *Bactrocera dorsalis* H. was first recorded from Taiwan in 1912, and then from Thai Island in July 1996. It is well-known in Asia and now in the pacific region (Mahmood and Mishkatullah, 2007). *Bactrocera dorsalis* H. is polyphagous in nature, it has about 250 host plants. They mostly attack ripened as well as ripened fruits like commercial fruits such as citreous, mango and peach, At the same time it also infest vegetables and many other fruits of tropical and subtropical regions (Hui and Lui, 2005). *Bactrocera dorsalis* H. is mostly a destructive pest and causes about 10% losses to the mangoes (Sayed *et al.,* 1970). This infestation of fruit fly is increasing day by day; due to this its market value reduces and cannot be exported in international market (Stone House *et al*., 1998). The Oriental fruit fly mostly preferred mango as host (Jayanthi and Varghese *et al*., 2002). During 2013 the highest attack of fruit flies was observed in sindh on different mango varieties like Sindhri, Chunsa and Sonara cultivators (Leghari, 2013). The annihilation of fruit flies is the major source of control program for Integrated Pest Management (IPM); it is one of the effective methods to achieve sustainable agricultural production with less damage to the environment (Kogan *et al*., 1999). The Purpose of this study was to increase awareness about pest and to provide knowledge about different life stages of insect pest and to give accurate information of morph

metrics of various life stages and their developmental time duration, The monitoring of adult life span (Longevity), pre-oviposition, oviposition periods, fecundity and the effect of diet on adult longevity, which is helpful for the professional management strategies that will keep mangoes away from poor effect of drugs and insecticides. This investigation will be also help in the identification of pest and shall draw attention towards the commercial varieties of Sindh, which are undermined by the fruit fly pest at the present time. This whole study was conducted under controlled laboratory conditions.

MATERIALS AND METHODS

For culture of *Bactrocera dorsalis* initially the infested mangoes of three varieties Chunsa, Sindhri and Beganpali were collected from the mango research field (garden) of Dr. Muhammad Hassan Pawaner and market of Hyderabad. Infested mangoes were kept in separated plastic trays (10 × 10 × 8cm) on a 4-5cm thick layer of sieved moist sand for pupation. After 1-2 days the pupae were collected from trays. About 50 pupae were kept in 12cm-diam Petri dishes lined with moist filter paper. The newly emerged adult flies were collected and placed inside the rearing cages (35 × 30 × 35cm). Each rearing cage had plastic glass walls on three sides, Plastic glass on the top and a round trap door was provided in the muslin mesh sleeve on one side just like a door so as to facilitate entrance of adult flies and to provide food and water. The male and female flies were identified by the presence of their external morphs according to (Kapoor, 2005). The bottom of each cage had a 2cm thick layer of sterilized sand with 5% moisture. The moisture was maintained by spraying water daily. The sucrose solution (10%) was provided inside the cage for adult feeding. This sucrose solution was kept in a 50ml beaker. A water-soaked cotton swab was laid in such a manner that half of it was submerged in sucrose solution, while the remaining half stayed above the beaker so as to keep the solution in reach of adult fruit flies in cages. Four to five mango fruits of each variety were also provided in cages for oviposition and their longevity. These mangoes were replaced by fresh ones daily to avoid decay. The entire fruit culture was maintained at mean temperature 26◦c±1◦c with mean relative humidities 60%±65% and also provided photoperiod 10-12hrs by 100w bulb. The eggs were observed under the binocular microscope and the number of eggs laid was counted until female died. About 50 eggs collected were placed in (12cm diam) Petri dishes with moist filter paper at the bottom to avoid aridity of eggs. After egg hatching a slice of 5gms fresh mangoes of each variety, were kept in Petri-dish separately for feeding the young Larvae. After 24 hrs mangoes were replaced by fresh varieties until all the eggs had incubated. Morphometric study of different life stages of *B.dorsalis* was carried out by taking ten replicates of each stage, viz., egg, 1st instar, 2nd instar, fully grown 3rd instar, pupa and adults for linear measurements; in addition to the above parameters, color, shape, size, hatching and developmental time of eggs, larvae, pupae and adults were also noted by following the techniques of (Ekesi *et al.,* 2007; Mir *et al*., 2014). The study was conduct from June-August (2013).

RESULTS

The biology of *Bactrocera dorsalis* was observed on three most common varieties of mangoes, in which morphological characters, developmental time and morphometric characteristics of eggs, larvae, and pupae were observed. Furthermore, the developmental time of life stages on artificial diet (water, water+ sugar, no food & water and natural mango varieties) under laboratory controlled conditions (temp. 26-31◦c, Humidity 55-65%) was also monitored. Eggs were in the form of cluster (6-7 eggs / cluster); While three larval instars were found (1st, 2nd and 3rd instar), which not only have difference in size and color, but the visibility of alimentary canal in 2nd instar and presence of black moles on anterior and caudal side of 3rd instar are very distinguish characteristics of the *Bactrocera dorsalis* larvae.

Egg: The eggs of *Bactrocera dorsalis* were white, shiny, rice shaped, slightly curved in to elongate tapering at anterior and posterior end. The eggs lay in cluster form, which are embedded in the pulp of fruit vertically or slightly angled and twisting with each other (fig.1c). Morphometric study exposed that the size of egg, such as length of egg, varied from 0.5mm-0.6mm with mean of 0.54±0.11mm and width 0.1-0.3mm with mean 0.19±0.08mm (Table-1). Egg laying and hatching period is 1-2 days with average 1.61 ± 0.51 days (Table-2).

Larvae (1st, 2nd, 3rd instars): The larvae of *B.dorsalis* passes three instars, The 1st instar was emerged transparent and creamy/ white in color (fig.1d), with length 1-2mm (2.6±0.75mm) and width 0.2-0.4mm (0.27±0.82mm) (Table.1). The 2nd instar was elongated in shape and creamy in color, the distinguishing characteristic of 2nd instar was the presence of externally visible alimentary canal (fig.1e), The 2nd instar's length was 5-6mm (0.55±5.88), and width 2-3 (2.34±0.78mm) (Table.1). The fully grown 3rd instar larvae had some visible characters; the head was pointed anteriorly with well-developed mandibles, hooks, spiracles on both anterior and posterior side of body and black mole on anterior and caudal side (fig.1f). The 3rd instars' length 7-8mm (7.68±0.72 mm) and width 3-4mm (3.58±0.25mm) (Table.1). According to my findings the development period was 8-10 days with mean values 9.97±2.25 days, respectively (Table.2).

Pre-Pupae: The mature larvae of 3rd instar was slightly curved in position, became slothful and stopped feeding and remain inactive for different activities except metabolically. According to morph these are yellowish in color and ring structure appearance (fig.1g). The size of pre-pupae was 2-3mm as average 2.93±0.49m and Width 2-4mm with average 3.89±0.20mm (Table.1). Developmental period was 1-2 days (Table.2).

Pupae: The Pupae emerged segmented and cylindrical hard, dark brown capsule (fig.1h), The length 4-5mm with average (4.47±0.64mm) and width were 2-3mm with average (2.69±0.16 mm) (Table-1). Duration of pupae stage varied from 8 to 9 days with mean 8.52±0.88 (Table-2).

Adult: The Adult flies were emerged from pupae within 8.9 days in early morning from 7.00 to 10.00 A.M. The females were easily recognized by its head with black dot, scutilium was black and yellow stripe, wings with black color , brown black thorax, the abdomen tapered shape have yellow stripes, distinct black 'T'-shaped mark and most distinguish morph character pointed pin like ovipositor (fig.1b), length 8-9mm(9.79±0.53mm**)** and width 12-13mm (13.98±0.27mm**) (**Table-1). The male is dark brown with black stripes on abdomen. The scutelium is whole black. Male is slightly shorter than female (fig.9a), length 7-8mm (8.65±0.58mm) and width 9-11mm with average (10.34±1.18mm) (Table-1).

Mating Period: Usually the mating time of the *Bactrocera dorsalis* started from sunset; males expanded their wings and amassed and pursued their females one hour before the sunset, Males fen their wings and come close to the female at once as detected by them in their territory. Mating couples remain paired throughout the night and are separated at sunrise. The mating time of *Bactrocera dorsalis* is about 9-10 hr with average (9.27±0.82 hr) (Table-2).

Pre-oviposition and oviposition period: The pre-oviposition period varied from 13-14 days (13.55±1.33days) (Table. 2). The oviposition period ranged from 20-22 days (21.46±1.42 days) (Table 2).

Fecundity: The egg laying capacity of a sexually mature adult female is 14-15 per day (14.32±2.27) (Table.2). Hatching percentage was 75%-80% with average (77.31±1.97) (Table.2). The variations in oviposition rate may be affected due to different cultivars of mango, or due to some physical parameters such as ripened and ripened fruits as well as the size of peel thickness and color.

Sex ratio: The newly emerged adults were examined by presence and absence of ovipositor. The sex ratio was 1-3 average 1.17±2.07 (male: female) (Table.2).

Adult Longevity: At the time of emergence, if adults were provided neither food nor water, male died after 1-2 days (1.56±0.46) and female died after 2-3 days (2.34±1.01) (Table -3). If only water was provided to the adults, longevity of adults was extended up to, was 4-5 (4.69±0.48) for male and 5-8 (6.96±0.18) days for female (Table -3). When water and sugar solution provided, male adult flies lived 13-25 days **(**19.2±4.1days) and female lived 15-30 days (21.34±3.1days) (Table -3). When they were provided natural diet (mango of different varieties), the longevity increase to from 30-45 days with average (38.63±7.6**)** (Table -3).

Developmental characteristic of *Bactrocera dorsalis* on three varieties of mango (Chunsa, Sindhri and Beganpali): The time between eggs laying and hatching (incubation period) varied from variety to variety. On Chunsa variety 1.5±0.02 day; this is minimum as compared to other two varieties like sindhri 2.32±0.16 and Beganpali 2.39±0.01 (Table.4). The larval development duration on Chunsa variety was 6.51±0.2 days, as Beganpali and Sindhri variety developmental time was almost same 7.5±1.6 (Table.4) with little variation. This variation in development of larvae on Chunsa variety was due to rich nutrients. Moreover, it was the most sugary in all varieties, which provided the favorable medium of development to the larvae. During the present study, pupae period ranged from 8-9 days (Table.4) on three varieties of mango. Results indicate that the life cycle on three varieties varies from 16-18 days of mango (Table.4).

Table 1. The morphometric characteristics of different life stages of *Bactrocera dorsalis* on mango varieties.

Development stages	Length (mm)		Width (mm)	
	Range	Mean±Sd	Range	Mean±Sd
Egg	0.5-0.6	0.54±0.11	0.1-0.3	0.19±0.08
1st Instar	1-2	2.6±0.75	0.2-0.4	0.27±0.82
2nd instar	5-6	0.55±5.88	2-3	2.34±0.78
3rd instar	7-8	7.69±0.72	3-4	3.58±0.25
Pre-Pupae	2-3	2.93±0.49	2-4	3.89±0.20
Pupae	4-5	4.47±0.64	2-3	2.69±0.16
Male ♂	7-8	8.65±0.58	9-11	10.34±1.18
Female ♀	8-9	9.79±0.53	12-13	13.98±0.27

**All measurements was concluded to 10 specimens

Table 2. The time period of different developmental stages of *Bactrocera dorsalis* on mango varieties.

Developmental Stage	Jun- August 20013	
	Range	Mean±Sd
Egg (incubation period)(days)	1-2	1.61±0.51
1st instar (days)	2-3	2.69±0.48
2nd instar (days)	4-5	4.5±0.55
3rd instar (days)	3-2	2.75±0.54
Complete larval period (days)	8-10	9.97±2.25
Pre-pupae (days)	1-2	2.07±0.86
Pupae period (days)	8-9	8.52±0.88
Mating Period (Hours)	9-10	9.27±0.82
Pre-oviposition (days)	13-14	13.55±1.33
oviposition (days)	20-22	21.46±1.42
Fecundity	14-15	14.32±2.27
Hatching %	75-80	77.31±1.97
Sex ratio(♂ : ♀)	1-3	1.17±2.07
Temperature °c	26-31	29.56±1.83
Humidity %	55-65	58.69±4.1

Table 3. Adult longevities (days) of the oriental fruit fly, *Bactrocera dorsalis* on various food sources.

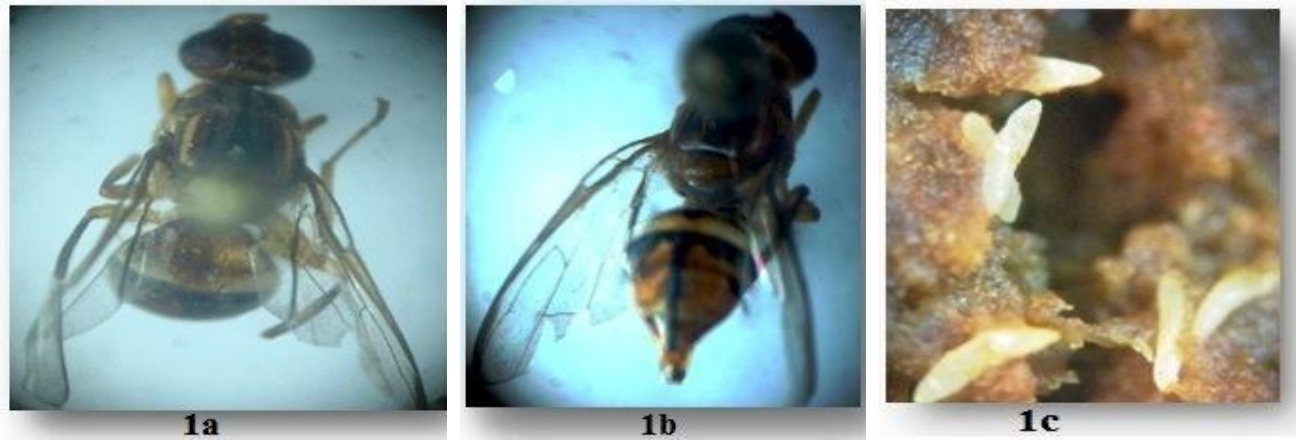
	Adults Longevities			
Treatment	Male		Female	
	Range	Mean±Sd	Range	Mean±Sd
Water	4-5 a	4.69±0.48 a	5-8a	6.96±0.18a
Water+ Sugar	13-25 b	19.2±4.1 b	15-30b	21.34±3.1b
Natural diet (mango)	20-30 c	24.3±3.5 c	25-45c	38.63±7.6c
No food and Water	1-2 d	1.56±0.46d	2-3d	2.34±1.01d
Temperature °c		26-31	29.56±1.83	
Humidity %		55-65	58.69±4.1	

Values (mean±Sd) followed by the different letter within the column are significantly different
(**P<0.05)

Tables 4. Effect of different varieties on developmental attributes and life cycle of *B.dorsalis*

Varieties name	Incubation period (days±SE)	Larval period (days± SE)	Pupal period (days± SE)	Total lifecycle (days± SE)	Sex ratio ♂ : ♀
Chunsa	1.5±0.02	6.51±0.2	8.8±0.46	16.8±0.086	1:3
Sindhri	2.32±0.16	7.5±0. 16	8.9±0.40	18.72±0.72	1:3
Beganpali	2.39±0.01	7.4±0.16	8.5±0.16	18.29±0.33	1:3

n.s = not significant (P>0.05)



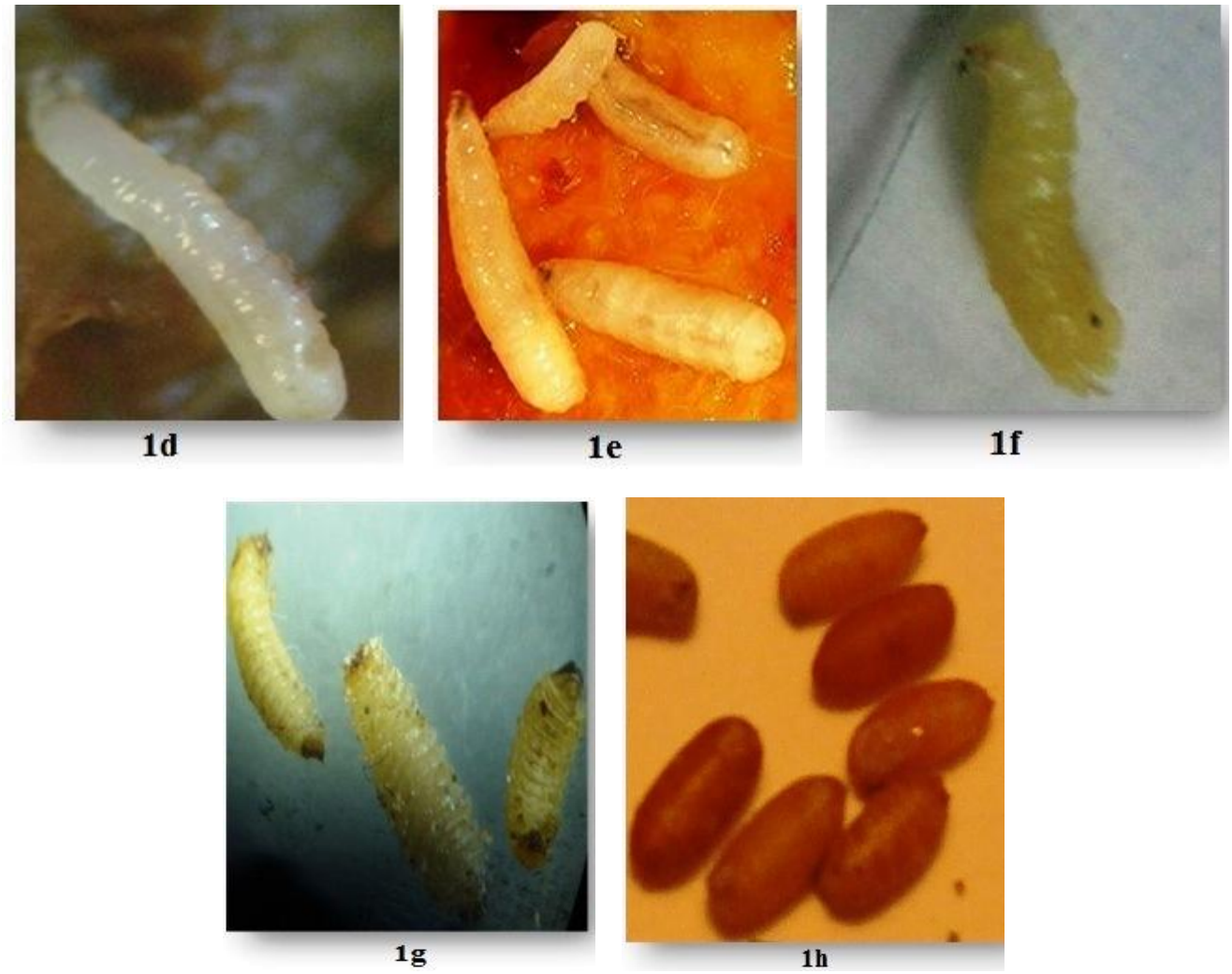


Figure 1. ***Bactrocera dorsalis*** **(Hendel); 1a, male;1b, female; 1c, eggs; 1d, 1st instar; 1e, 2nd instar; 1f, 3rd instar; 1g, Pre-pupal; 1h, Pupae.**

DISCUSSION

Bactrocera dorsalis is serious pest of mango over all the world especially in tropic and subtropical world. The present study shows the biology, morphometric characteristics and developmental rate on different diets. The present results shows the morphometric study such as that the size of egg (0.5mm-0.6mm) (Table.1) laying and hatching period, similar result was reported by (Mir *et al.,* 2014) for *B.cucurbitae* and (Kalia, 2015) on *B.dorsalis*. They observed that the eggs hatched within 12 to 24 hrs (Leghari, 2013) reported similar results on *B.dorsalis* and *B.zonata* which shows that in general hatching period of *Bactrocera* species is 1-2 days on different fruits. The larvae of *B.dorsalis* pass three instars with different size and morphology. The 1st instar was inactive and small in size as compare to two other instars. The 2nd instar had a distinguishing characteristic i.e. presence of externally visible alimentary canal, which was not described by (Christenson and Foot, 1960; Weems *et al.,* 2015). The fully grown 3rd instar larvae had visible characters. Third instar feed rapidly in the pulp of mango, formed the tunnels and holes in the fruit pulp and peel, come outside the fruit by holes of peel, fast move and Jump. Black mole on anterior and caudal side, this distinguishing character was not noted by (Christenson and Foot, 1960; Weems *et. al.,* 2015). According to my findings the development period was 8-10 days, but different workers (Renjhan, 1949) recorded 5 to 22 days, (Singh and Teotia, 1975) 5 to 11day, (Doharey, 1983) 3 to 8 days and (Shivarker and Dumber, 1985) 15days. This variation in larval developmental time may be because nature of variety of mango. The morphometric characters like color, length and width of all developmental stages of *Bactrocera dorsalis* were same of three varieties, Chunsa, Sindhri and Beganpali, as we have observed in 10 specimens of each developmental stage randomly, small

variation may be occurred but no significant changes were found in measurement and physical appearance in each developmental stage. Present results were very close to earlier study work of (Christenson and Foot, 1960) on *Bactrocera dorsalis.* Pupal period recorded on melon fruit fly by (Narayan and Batra, 1960, Agarwal *et al.,* 1987, Langar *et al.,* 2013). (Mir *et. al.*, 2014) worked on pupae of the *B.cucurbitae,* As he mentioned that the black dot on the posterior portion was the distinguishing characteristic of *B.cucurbitae*, However according to my findings, that same black dot was on the posterior side of pupae of *Bactrocera dorsalis,* also present*,* there is not a distinguishing characteristic which shows cucurbit but in fact a generic characteristic. Results were found by (Kalia, 1992) on different varieties of mango and also (Kalia, 2015) work generally on mango as compared to other fruits, noted that adult size (8.1mm) was maximum length of adults. Same observation way made under laboratory conditions, and similar findings were observed by (Koul and Bhagat, 1994). According to (Vishva, 2005; Shivyya *et al.,* 2007) copulation period was prolonged. Very close results were found by (Kalia, 2015) rearing the pre-oviposition period of *Bactrocera dorsalis* 18-22 days and same findings were also recorded on fruit flies by (Meats, 1981; Saeki *et al.,*1980; Qureshi *et al.,* 1993); Shehata, 2008). The variations in oviposition rate may be affected due to different cultivars of mango, or due to some physical parameters such as ripened and ripened fruits as well as the size of peel thickness and color. Related results were also found by (Kalia, 2015) fecundity rate of *Bactrocera dorsalis* per day, per female were 15.0-16.0 eggs. Sex ratio may be affected by environmental fluctuations or food availability, and may be its natural phenomena. Sex ratio (♂: ♀) of *Bactrocera dorsalis* on different host such as Bannana, guava, papaya and sapota and mango 1:1.22, 1:1.1 and 1:1.06 and 1:1 respectively was reported by (Kalia, 2015). These results related with the (Mir *et al*., 2014) and also close to (Shivyya *et al.,* 2007; Waseem *et al*., 2012) who reported that sugar (carbohydrates) nutrient extended the longevity of melon fruit fly. The longevity on different diets were significant (P<0.002), but the longevity of *Bactrocera dorsalis* on mango varieties were almost same there is no variation occurred. The time between eggs laying and hatching (incubation period) varied from variety to variety. On Chunsa variety 1.5±0.02 day; this is minimum as compared to other two varieties like sindhri 2.32±0.16 and Beganpali 2.39±0.01.The (Kalia, 1992) observed average incubation duration of *B. dorsalis* was 2.0 to 3.25 days on different varieties of mango. Similarly (Kalia *et. al.,* 2015) found that the incubation period of *Bactrocera dorsalis* on mango was 1.5 days as compare to other fruits such as (Sapeto, Bannana, Papaya). The larval development duration on Chunsa variety was 6.51±0.2 days, as Beganpali and Sindhri variety developmental time was almost same (7.5±1.6days) with little variation. This variation in development of larvae on Chunsa variety was due to rich nutrients. Moreover, it was the most sugary in all varieties, which provided the favorable medium of development to the larvae. Similarly (Kalia *et al*., 1992) defined the maximum time 7.75 days on different varieties of mango, and (Kalia 2015) reported 6.0 days larval minimum time on mango as compared to with other fruits like papaya, Banana and Sapeto. Other workers also observed 3 to 21 days larval development duration on different fruits and vegetables because every fruit has different medium for the larval development like high fiber content, sweetness and nutrient and at the same time environmental conditions like temperature and humidity play a major role in larval development time.(Renjhan *et.al.,* 1949; Narayanan and Batra, 1960; Hollingsworth *et al.,* 1997). During the present study, pupae period ranged from 8-9 days on three varieties of mango (Kalia *et. al.,* 1992) reported pupae period 8-11 days and also longest pupae period are 12 days on mango as compared to other fruits (Kalia, 2015; Jayanthi and Varghese, 2002) reported same pupae period on Bannana, (Kalia, 1992) reported that size of pupae varied not only between different fruits like guava and mango but also among cultivars of mango. Results indicate that the life cycle on three varieties varies from 16-18 days of mango (Table.4); similar findings were observed by (Kalia *et al.,* 2015), as the minimum time duration of life cycle of *Bactrocera dorsalis* are 18 days on mango. (Doharey, 1983) reported shorter life cycle on mango 19.8 days and (Jayanthi and Verghese, 2002) reported longest life cycle on mango 25.0 day. It may depend on varietal contents because some varieties of mango are very juicy and more sugary, therefore, life cycle rapidly developed, but some were fibrous and less sweet in test, the life cycle of *B.dorsalis* took some time for development in such cases. But there is no any significant difference (P>0.05) in all developmental stages of *Bactrocera dorsalis* in these three varieties of mango. Sex ratio (♂: ♀) among all the cultivars of mango was almost same, as in mango, sex ratio was 1:3. Similar findings were also reported by (Shimada *et al.,* 1981; Batra, 1964; Doharey, 1983; Kalia, 1992; Jayanthi and Verghese 2002).

Conclusion: The present study explored the morphometric characters as well as all the developmental stages of the fruit fly *Bactrocera dorsalis* on three varieties of mango namely Chunsa, sindhri, and Beganpali. The morph measurements of all developmental stages of *Bactrocera dorsalis* were same on each variety of mangos, but little bit variation in the developmental time duration, hatching, fecundity and life span of adults on these varieties, caused by some physical characters, chemical nutrients and dietary fibers of each variety. Longevity of adults was significantly varied with different diets such as provided water, water and sugar,

natural diet of mango and no food and water provided to adults. This study is beneficial for the IPM and for the betterment of fruits, especially for mango exporting cash varieties.

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