Chapter - 1

Rearing and Host Preference of *Sitophilus oryzae* (Coleptera: Curculionidae) in the laboratory conditions

1.1 Introduction

Economically important cereals consumed in the world are maize, rice, wheat, barley, sorghum, millet, and oat. Rice, *Oryza sativa* (Linn.), is one of the economically important cereal and is the most important staple food for half of the world's population which is grown in over 100 countries of the world (Oko *et al.*, 2012; Ranganathan *et al.*, 2018). More than 90% of the world's rice is produced and consumed in Asia. Every year nearly 25 to 30% crop yields are destroyed both infield and stores by different insect pests and post-harvest losses of food grains in India is estimated at 12 to 16 million MT/year (Danho *et al.*, 2002; Singh, 2007; Singh, 2010; Akhter *et al.*, 2017) and pests devour about 6.5% of total grains stored in India (Akhtar *et al.*, 2009; Mohale *et al.*, 2010).

In India, farmers are facing an annual loss of Rs 92,651 crore per year the main causes of which are inadequate storage and transportation facilities (Pandey, 2018). Gujarat is one of the leading states in terms of agriculture products considered as the backbone of the economy for the country. However, these high yield agricultural products also have a major problem of insect pest which not only reduce the weight of these grains but also the quality, commercial value and its seed viability (Singh 2010; Alexandratos and Bruinsma, 2012; Nagpal and Kumar, 2012; Ragunath *et al.*, 2014; Ranganathan *et al.*, 2018).

There are several sources of infestation; they are carried directly from the fields/ carried-over commodities/ waste and rejects/ agricultural machinery/ processing plants farm grain stores and re-used sacks/ means of

transportation/ alternative hibernation sites and hosts. Among the coleopteran pests of stored grains Sitotroga cerealella, Sitophilus spp., and Rhyzopertha dominica are known to be the major pests (Neupane, 2002; Follett et al., 2013). The rice weevil, Sitophilus oryzae L. (Coleoptera: Curculionidae), is one of the most destructive primary pests attacking many common stored cereals including rice, wheat, maize and split peas and has a worldwide distribution (Mohale et al., 2010; Shahidi and Chandrasekara, 2013; Ragunath et al., 2014; Schuster and Torero, 2016; Ranganathan et al., 2018). S. oryzae is an economically important pest, which grows by making bores in it and the grubs hollow out the remaining infested grain (Srivastava and Sabtharishi, 2016). As they are the primary pest of stored grains their larvae prefer to feed on the germ of the grains, which results in loss of germination. Stored and milled rice grains are prone to attack by S. oryzae and the latter grains are mostly preferred causing heavy economic losses and both the adults and larvae feed on the carbohydrates in rice grains causing weight loss and contamination (Park et al., 2003). In the absence of control, the stored grains can be destroyed even up to 100% (Dhaliwal et al., 2007; Derera et al., 2014). Enhancing the temperature and humidity of the infested grains, S. oryzae activity also induces accelerated growth of the secondary pests and creates the most favourable conditions for pathogens and further infestation (Hill, 2002; Srivastava and Subramanian, 2016).

To combat this problem effective chemical fumigants and contact synthetic insecticides are commonly used to prevent the loss of stored products throughout the world. Chemical insecticides are the most effective against insect infestation, but their repeated use has disrupted biological control by natural enemies and has led to outbreaks of other insect species and sometimes resulted in the development of pest resistance (Mishra *et al.*, 2013, b; Kim *et al.*, 2015). Besides these chemical insecticides also play a negative role in food quantity and quality, human health, serious environmental

problems and affects the non-target organisms (Palmer *et al.*, 2007; Jardim and Caldas, 2012; Lozowicka *et al.*, 2014; Skretteberg *et al.*, 2015; Liu *et al.*, 2016; Özkara *et al.*, 2016). The environmental problems caused by the overuse of pesticides have been a matter of concern for both scientists and the public in recent years. The estimated report shows that about 2.5 million tons of pesticides are used on crops each year and the worldwide damage caused by pesticides reaches \$100 billion annually (Koul *et al.*, 2008; Mohan *et al.*, 2011; Nguyen *et al.*, 2015; Dijiwati and Kaushik, 2019). The reasons for this are two folds: (1) the high toxicity and non-biodegradable properties of chemical pesticides and (2) the residues in soil, water resources, and food that affect public health.

To overcome these, insect-specific insecticide needs to be developed. For checking the efficacy of such insect-specific insecticide mass rearing and pure breed of the pest is a prerequisite under controlled conditions to reduce stochastic variation as there cannot be a real success in pest monitoring and management without a better understanding of the dynamics of insects' life cycle (Merville *et al.*, 2014). Many workers have delved into the life cycle of *Sitophilus oryzae* (Barbhuiya *et al.*, 2002; Choudhury and Chakraborty, 2014; Akhter *et al.*, 2017) with reference to the generations completed per year (Barbhuiya *et al.*, 2002), type of food grain (Singh *et al.*, 2013; Ojo and Omoloye, 2012; 2015 and 2016) and with different agro-climatic conditions (Abass *et al.*, 2014; Farrell, 2018; Ndemera *et al.*, 2018).

Host preference is a significant feature of any organism's life history. Indeed, for some species 'the host grain is not merely something fed on, it is something lived on' (Dent *et al.*, 2003). This is certainly the case for rice weevil *S. oryzae* where, in addition to feeding on their host grain, adults meet members of the opposite sex and mate, females oviposit and a larva then feeds. The entire life cycle is therefore dependent on the suitability of the host. Host preference and host suitability have been studied in *S. oryzae* using

different stored grain by Sharma et al., 2016, where they have focused on different varieties of maize with reference to the stored products. Zunjare et al., 2015 have unravelled the genetics of weevil resistance in maize for adopting a suitable breeding strategy for the development of S. oryzae resistant cultivars. Athanassiou et al., (2017) in their studies have proved that the coexistence of the species leads to competition for host preference in laboratory conditions. Previous studies have also reported that the host preference varies based on seed size, germ layer of the seed, moisture and nutritional values (Ojo and Omoloye, 2016; Akhter et al., 2017). Seasonal variation in the rate of infestation of S. oryzae has been studied by Okram and Hath (2019). Thus from the ongoing literature survey, it can be seen that there is plethora of information on biology and ecology of Sitophilus sp. with different approach, however, concerning the present inventory where the main focus was rearing of S. oryzae for the development of the cell line and to have an abundant number of adult S. oryzae the host preference was also investigated for four stored grains (Maize, Rice, Chickpea, and Millet) under laboratory conditions.

Hence, the present study was undertaken to investigate a suitable rearing protocol, life cycle, fecundity rate, and sex ratio as well as the preference of different stored grains by S. oryzae in control laboratory conditions.

1.2 Materials and Methodology

Collection of Insect pest

Infested grains were collected from the different granaries of Vadodara. Insect pests were separated from the grains and identified by using standard identifying keys. On keen observation, there were few major insect pests found i.e. *Sithophilus oryzae*, *Sitophilus granaries*, *Trogaderma granarium*, *Rhyzopertha dominica*, *Oryzaephilus surinamensis*, *Sitotraga cerealella* and *Tribolium castaneum*. Among these pests, *S. oryzae* were found on wide range of host grains and was reported as most dominant and hence, *S. oryzae* were selected as insect model for the further study. Identification of *S. oryzae* was done by standard reference key by Halstead *et al.*, 1969.

Rearing of S. oryzae

Stock Insect Culture preparation

The collected pest species were first acclimatised to the laboratory conditions. Twenty five pairs of adult *S. oryzae* were introduced into 500 gm grains of rice in plastic jars covered with cotton mesh lids and were allowed to feed, mate, and oviposit. Throughout the experiment the average temperature (minimum and maximum) as well as relative humidity was maintained from 26 ± 2 °C and $65 \pm 5\%$ respectively. *S. oryzae* were allowed to feed, mate, and oviposit for 7 days and then removed. Culture arena was observed daily until new progenies emerged; they were removed and sexed using morphological characters described by Halstead *et al.*, 1969. The whole set up was replicated three times and were maintained, monitored till the third generation so as to obtain the fresh pest population. The pure breeds from the fourth generation were removed and sexed using morphological characters described by Halstead *et al.*, 1969. This stock culture was used as source of *S. oryzae* throughout the period of the study.

Development and morphometry analysis of S. oryzae

Ten pairs of adult *S. oryzae* from the stock were sorted and transferred to vials having different grains weighing around 200 gm till they laid eggs. Grains with holes (2–3 mm diameter) were sorted out and individually placed in small plastic vials and checked daily till adult emergence. Further, the development time, number of F_1 progeny, total body length, grain loss was observed. Fecundity and longevity were measured by following the method of Ojo and Omoloye, 2016 with slight modification where one adult pair of the pest was taken into consideration and was introduced into individual vials having 20 gm of all the four grains and replicated three times. The grains were replaced every seven days and the eggs laid were determined following standard procedure and adult longevity was determined when all the pests exhibited morbidity.

Estimation of nutrition in the grains due to infestation

1. Estimation of Carbohydrates: The Carbohydrates were determined by DNSA method.

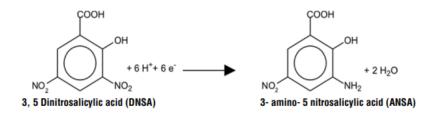


Figure 1.1: Chemical Reaction for DNSA Method

Procedure

Reducing sugars were determined in uninfected and infested grains by employing DNS Method. The underlying principle is that reducing sugars present in wheat reduce 3,5-dinitrosalicylic acid (DNS) to 3amino 5 nitro salicylic acid which absorbs light strongly at 540nm (Patil and Muskan, 2009). The amount of reducing sugar in the samples was calculated using a standard graph prepared from working standard of glucose solution (0 to 500μ g/ml).

2. Estimation of Protein

Procedure

Protein content was determined by Bradford method i.e. a colorimetric protein assay, based on an absorbance shift of the dye Coomassie Brilliant Blue G-250 in which under acidic conditions the red form of the dye is converted into its bluer form at 595nm, to bind to the protein being assayed (Pandey and Budhathoki, 2007). The standard curve was prepared by dissolving 25mg of bovine serum albumin in 0.15 NaCl and the volume was made up to 25ml of 1mg/ml.

Statistical Analysis

The difference in mean number of emerged adults, average grain loss, mean body length and sex ratios were compared by analysis of variance (One-Way ANOVA) using Graph pad Prism software version 6. Further, the significant level of the tests was set at 5% (p<0.05). The significance was tested at α =0.05 and noted *p<0.05 using Graph pad Prism software version 6.

1.3 Results

Identification of S. oryzae

Adult Size: The adult *S. oryzae* was found to be in the range of 2-3 mm in length. There was a slight difference in the size of male and female *S. oryzae*. The average length of adult male and female was 2.9 ± 0.6 mm and 2.8 ± 0.6 mm respectively.

Colour: The colour varied from reddish brown to black

Morphological identifying characteristics

Presence of well developed rostrum and four orange/ red spots on the corners of the elytra which fade inward to the middle point of the elytra. The prothorax is highly pitted and the elytra have rows of pits within the longitudinal grooves. Females are larger than males. Head is projected forward into a snout like rostrum. A pair of stout mandibular jaws is present at the extremity of the rostrum (Fig 1.3a and Fig 1.3b).

Feeble sexual dimorphism exists in *S. oryzae*, male and female *S. oryzae* has a distinct shape of rostrum as well as the arrangement of punctations on rostrum (Ikonomou *et al.*, 2003). Females were found to have slender and longer rostrum than male. Males were found to have thick-stout rostrum, widened at middle as compared to female with more prominent punctures. Two rows of punctures were found to extend backward and meet individually at interocular region forming two distinct grooves. Grooves were found to be more prominent in males and outer groove was seen extending deep in interocular region. (Fig 1.3a) Both male and female adults were dissected to identify the species of *S. oryzae* using their genitalia –through the examination of the aedeagi in males and Y-shaped sclerites in females (Fig: 1.4a).

Life cycle

The total life span of S. oryzae was found to differ with different hosts However, the average life span of the adult S. oryzae was found to be of 5-6 months. After copulation, adult female was found to bores a hole in the grain with the help of its powerful jaws and deposit a single egg in the grain cavity. The egg was oval in shape. There was fractional change in the size of the egg of different grains, each egg was found to be tiny, white, measuring 0.7 mm long and 0.3 mm broad (Fig 1.2). The egg hatched into larvae in 3-5 days. The tiny, white, fleshy, legless grubs with yellowish- brown head and biting jaws lasted for 20-35 days (Fig 1.2). A fully matured grub was then found to pupate (Fig 1.2). The pupal stage lasted for 3-6 days. The adults formed after pupation was found to bore its way out of the grains. The size of the newly bored adult S. oryzae was found to be directly proportional to the size of the grain in which the larval period has been spent. As the size of the maize grain was largest amongst all the grain tested, size of the Adult S. oryzae was accordingly largest (4.2-4.6mm), followed by chickpea (4.02 - 4.1mm), rice (3.1 -3.4mm) and the smallest size of *S. oryzae* were found to be in millet (2.6-2.8mm). The body lengths of females were observed to be significantly bigger as compared to male in all four cultures. And the overall body size of S. oryzae was largest in maize and smallest in millet (Fig 1.9) (Table 1).

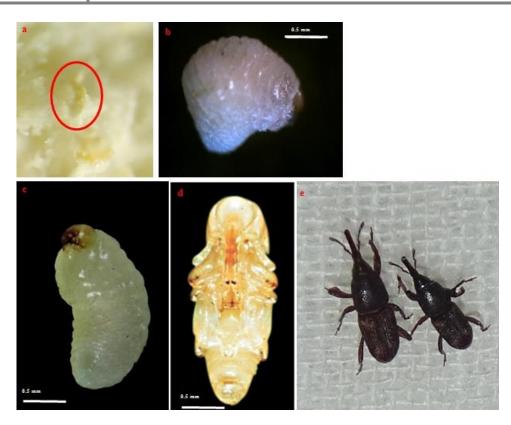


Figure 1.2: Life cycle of S. oryzae: a) egg, b) &c) larvae, d) pupa and e) adults



Figure 1.3a: Female Identification Characters

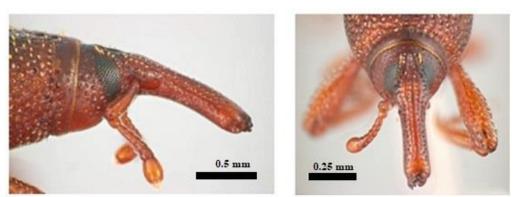


Figure 1.3b: Male Identification Characters

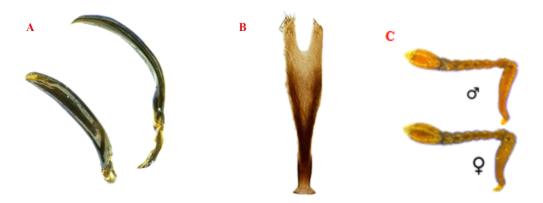


Figure 1.4a:A. Aedeagus of male genitalia B. Y-sclerite of female genitalia c. Antennae of *S. oryzae*

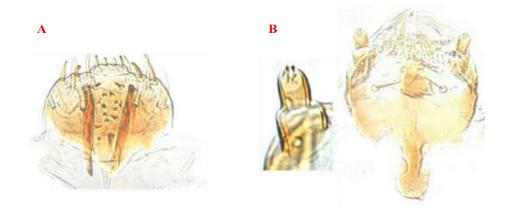


Figure 1.4b: Epipharyngeal rod of larva. B. Apical sensory organs on the labial palp of larva

Table 1.1 summarizes the incubation period, number of adults emerged, grain loss and variation in male and female body length. The incubation period and progeny size showed a significant (p < 0.001) difference in the developmental period where, the least development period (from egg to emergence of adult) was observed with maize (32.5 days) followed by chick pea, rice and millet by 35, 39 and 42 days respectively (Fig 1.6). Along with the developmental time the progeny size of S. oryzae was also found to be significantly different (p< 0.01). The mean emergence of F_1 progeny was found to be highest (P < 0.01) in maize (226 adults) and was least (126 adults) in millet (Fig 1.7). The average grain loss was found to be highest in maize (59.77 gm), followed by chick pea (44.04 gm) and was 53.08 gm in rice followed by least loss in millet (32.12 gm) (Fig 1.8). The consumption of the grains by individual pest was also observed and was found to maximum in rice and least in millet. The highest average losses of grain on daily basis were also calculated and they were found to be 1.12 gm in maize, whereas, with rice, chick pea and millet the loss grains per day was found to be 0.83, 1.0 and 0.6grams respectively with an average consumption rate by single larvae of 5.97, 5.68 and 4.76 mg per day respectively (Table 4).

There was a significant (p<0.001) decrease of reducing sugars of infested grains as sugars (Table 2). The loss of carbohydrates was highest observed from the maize (37.97%) (p<0.001), followed by chickpea (30. 27%) (p<0.001), Rice (26.96%) (p<0.01) and least in millet (23. 98%) (p<0.05) (Fig 1.10). It was noted that even protein content exhibited a downfall with 13.96% in maize, 11.6% in chickpea, 9.93% in rice and 8.03% in millet (Fig 1.11). But the effect was less pronounced as compared to reducing sugars in infested grains owing to difference in distribution of protein and carbohydrates in the different grains.

Table 1.3 summarizes the fecundity, longevity and gender ratio. *S. oryzae* grown on maize noted the highest fecundity (453 adults) as compare to

other three cultures (Fig 1.12). Furthermore, the culture of *S. oryzae* grown on rice, chickpea and millet showed a decline in the fecundity rate by 52.75%, 33.99% and 68.65% as compared to the culture grown on maize. In addition to this the gender ratio were also found to be affected with respect to different grain used. Among the four grains the highest male: female ratio was found to be in maize followed by chickpea, rice and the least in millet (Fig 1.14). As far as the longevity is concern, the highest longevity of *S. oryzae* was found to be 184 days in maize and least was with millet for 157 days(Fig1.13). However, the longevity was not significantly affected by different grains.



Figure 1.5: Infestation of S. oryzae in all four selected host grain

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Type of	Incubation	No of adult emerge	Grain Weight Loss	Body Length (in mm)		
Grain	period	± SE (Range)	± SE (in gm)(Range)	Male ± SE (Range)	Female ± SE (Range)	
Maize	32.5**	226±5.77**	59.77±3.02**	4.28±0.76**	4.6±0.41**	
		(202-241)	(49-65)	(4.17-4.31)	(4.51-4.87)	
Rice	39**	139±3.98**	44.04±2.53**	3.18±0.26**	3.43±0.17**	
		(122-158)	(41-49)	(3.02-3.28)	(3.37-3.49)	
Chick	35**	176±4.43**	53.08±2.94 **	4.02±0.54**	4.14±0.24**	
pea		(163-186)	(51-68)	(3.82-4.07)	(4.10-4.26)	
Millet	42**	126±3.07**	32.21±1.83 **	2.67±0.06**	2.84±0.03**	
		(116-132)	(27-39)	(2.64-2.69)	(2.79-2.91)	

 Table 1.1: Incubation period, Number of adult emerged, Grain weight loss and Body length of S. oryzae in different host grains

 Each value represents the mean ± SEM. (n=3)

 Significant level indicated by ^{ns}= non significant, *= (p<0.05); **=(p<0.01); ***=(p<0.001)</td>

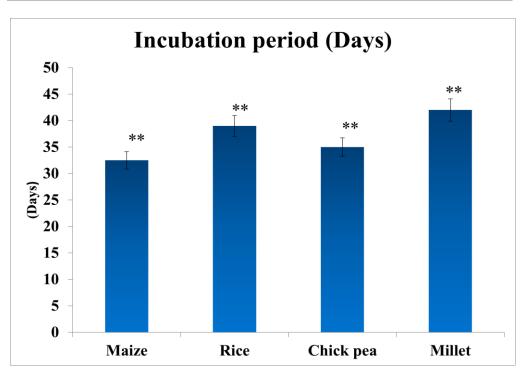


Figure 1.6 : Incubation time taken by *S. oryzae* in different grains Significant level indicated by *= (p<0.05); **=(p<0.01); ***=(p<0.001)

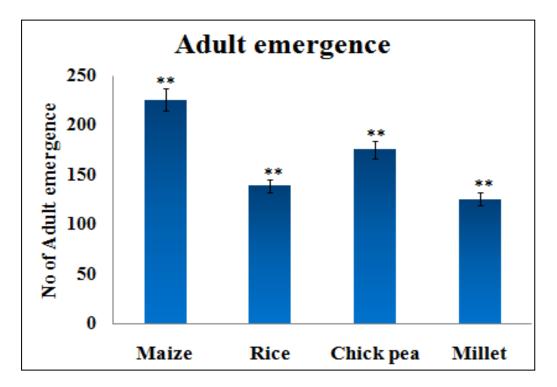


Figure 1.7: No. of Adult *S. oryzae* emerge in different grains Significant level indicated by *= (p<0.05); **=(p<0.01); ***=(p<0.001)

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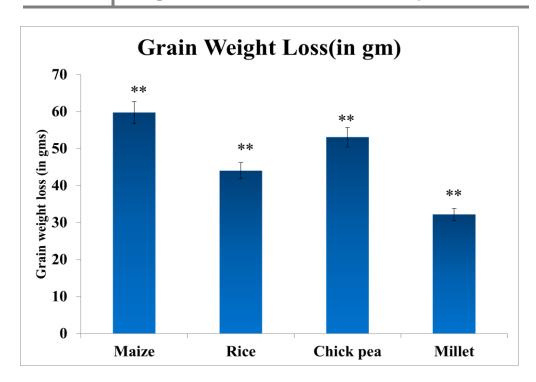


Figure 1.8: Amount of grain weight loss by *S. oryzae* Significant level indicated by *= (p<0.05); **=(p<0.01);***=(p<0.001)

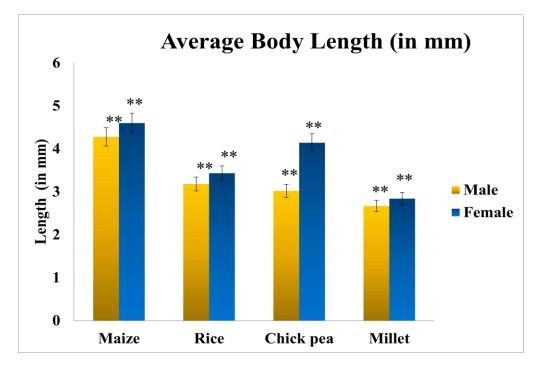


Figure 1.9: Body length of male and female in different grains Significant level indicated by *= (p<0.05); **=(p<0.01); ***=(p<0.001)

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Sr No.	Sample of grain	% Protein			% Carbohydrates		
		Uninfected (mg/g)	Infested (mg/g)	% loss of protein	Uninfected (mg/g)	Infested (mg/g)	% loss of protein
1	Maize	11.1±0.05	9.56±0.05***	13.96%	68.2±0.32	42.3±0.17***	37.97%
2	Rice	5.4±0.38	4.87±0.25**	9.93%	72.4±0.48	52.88±0.24***	26.96%
3	Chikpea	19±0.42	16.8±0.36***	11.60%	58±0.28	40.44±0.09***	30.27%
4	Millet	7.3±0.03	6.72±0.64*	8.03%	60.9±0.16	46.29±0.11***	23.98%

 Table 1.2: Effect of Insect Infestation on Quality Parameters of Grains

Each value represents the mean ± SEM.(n=3)

Significant level indicated by ns = non significant, s = (p<0.05); s = (p<0.01); s = (p<0.001)

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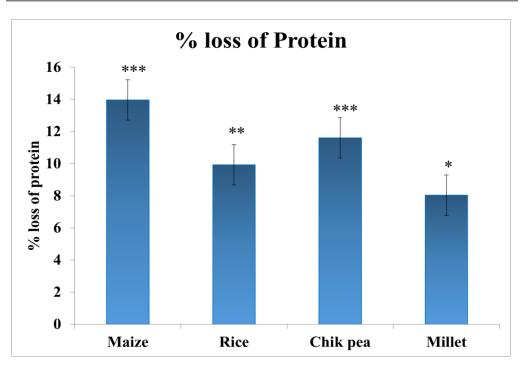


Figure 1.10: Effect of *S. oryzae* infestation on protein content of the different grains Significant level indicated by *= (p<0.05); **=(p<0.01);***=(p<0.001)

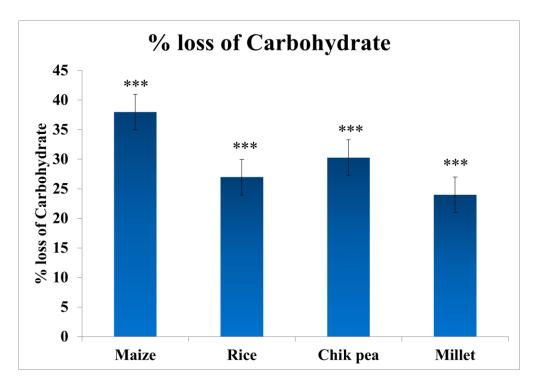


Figure 1.11: Effect of *S. oryzae* infestation on Carbohydrate content of the different grains Significant level indicated by *= (p<0.05); **=(p<0.01);***=(p<0.001)

Type of Grain	Fecundity	Female: Male	Adult Longevity
Maize	453±2.37**	2.36:1**	184±1.52 ^{ns}
	(422-494)	2.30.1	(179-198)
Rice	214±1.52**	2.35:1**	175±2.01 ^{ns}
	(201-236)	2,33,1	(167-183)
Chick pea	299±1.75**	1.98:1**	180±2.57 ^{ns}
	(271-315)	1.70.1	(169-192)
Millet	142±1.13**	2.21:1**	157±2.01**
	(135-152)	2,21,1	(145-169)

Table 1.3: Fecundity, Female: Male and longevity periods (±SEM) of S. oryzae on storedgrains (24–30°C; 60 ± 10%RH; 12 h photophase).

Each value represents the mean \pm SEM (n=3). Significant level indicated by ^{ns}= non significant, *= (p<0.05);**=(p<0.01);***=(p<0.001)

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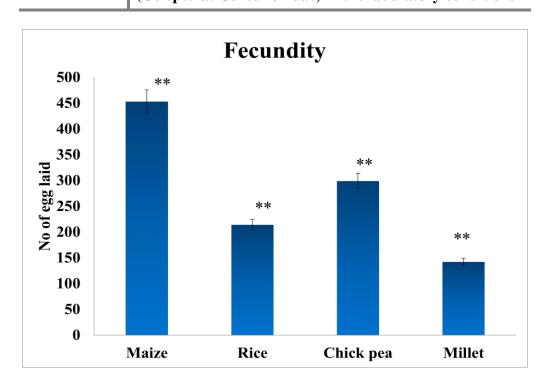


Figure 1.12: Effect of different grains on fecundity rate of *S. oryzae* Significant level indicated by *= (p<0.05); **=(p<0.01);***=(p<0.001)

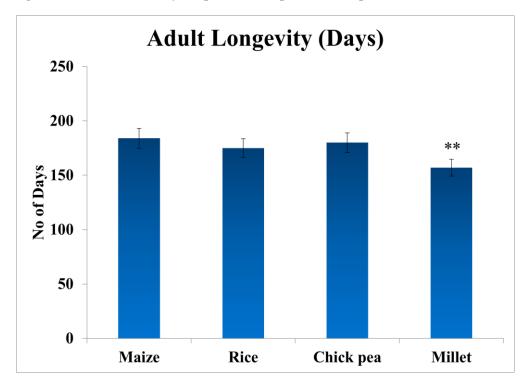


Figure 1.13: Effect of different grains on longevity of *S. oryzae* Significant level indicated by *= (p<0.05); **=(p<0.01);***=(p<0.001)

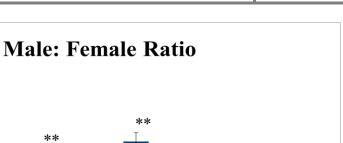
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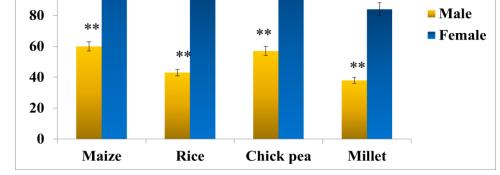


Figure 1.14: Male: Female ratio in newly emerge S. oryzae in different grains Significant level indicated by *= (P<0.05); **=(P<0.01); ***=(P<0.001)

Type of Grain	Grain loss (gm)/day	Average grain consumption by single larvae/day(mg)
Maize	1.12 ±1.53**	4.95±2.75*
Rice	0.83±2.43*	5.97±2.96*
Chickpea	1.0±1.09**	5.68±1.53**
Millet	0.6±3.12 ^{ns}	4.76±3.24 ^{ns}

Table 1.4: Grain loss/ day and Average grain consumption by single larvae/day (mg) of S. oryzae on stored grains

Each value represents the mean \pm SEM (n=3).

Significant level indicated by ns = non significant, *= (p < 0.05); ** =(p<0.01); ***=(p<0.001)

1.4 Discussion

The results revealed that S. oryzae illustrated a definite host preference for the grains viz. maize, rice, chickpea and millet. Throughout the infestation period, in none of the grains the S. oryzae was seen to come out of the grain till it was adult as they are internal feeders, and the entire development cycle occurs within the kernel and do not leave the kernel till their cuticle hardens (Mason and McDonough, 2012; Ranganathan et al., 2016; Kumari et al., 2017). The number of larvae that transformed into adults and emerged out from the respective grain culture was taken as a total incubation time. The lowest time taken by S. oryzae to develop from an egg to adult was 32.5 days with maize, followed by 35 days with chickpea, 39 days with rice and 43 days for millet. Food is the primary factor that determines the length of the life cycle, difference in the total length of life cycle from egg to adult may be because of the richness of the food, nutrients content and the size of the grain (Campbell, 2002; Danho et al., 2002; Subedi et al., 2009; Singh et al., 2013). The least incubation time taken in maize grains can thus be attributed to the freely availability of high nutrient from the germ layer of the maize grain. On other hand the highest time taken for maturation with millets grain suggests the least selectivity of this grain due to its size. Our study is in accordance with the earlier reported work (Keskin and Ozkaya, 2015; Danho et al., 2015; Ojo and Omoloye, 2016; Akhter et al., 2017; Oloyede-Kamiyo and Adetumbi, 2017) where they have proved the importance of the grain size and of oviposition of S. oryzae with different grains. Thus the larger size of maize compared to chickpea, rice and millet probably offers greater fitness benefits as well as huge probability of larval survival, larger progeny size, and support for larger numbers of progeny than smaller seeds.

As reported by Akhter *et al.*, (2017) the development of the larvae is better on seeds which are preferred for oviposition, in the present study a significant variation in size of adult of *S. oryzae* was observed and more so over, females were found to be larger than their male counterpart regardless of the food host. However, when a comparison of all the four grains was made, S. oryzae bred on maize was bigger (4.28 mm long for male and 4.60 mm long for female) than the other grains followed by chickpea (4.02 mm long for male and 4.14 mm long for female), rice (3.18 mm long for male and 3.43 mm long for female) and a relatively smallest was for millet (2.67 mm long for male and 2.84 mm long for female). The developmental biology of *Sitophilus* sp. is influenced by its fecundity and oviposition, shorter larval period, and ability to breed easily on any cereal crop (Ojo and Omoloye, 2016). Hence, as in the present study highest fecundity and ovisposition was reported for maize, larger size of adults is not a surprise. A distinct difference in the size of male and female Sitophilus has been reported earlier (Singh et al., 2018) females have slender and long rostrum compared to thick-stout rostrum of males. Thus in addition to the fecundity and oviposition the larger size of adults as well as size difference in male and females is in agreement with the previous work (Plarre, 2010; Choubey, 2013; Ojo and Omoloye, 2012 and 2016; Athanassiou et al., 2017).

The effect of grain size on the biology of *S. granarius* (L.) with respect to its oviposition, distribution of eggs and adult emergence has been reported by Stejskal and Kucerova way back in 1996. Earlier workers have also reported the oviposition strategy in relation to the con-specific species (Mathias *et al.*, 2015) with reference to different varieties of grains (Subedi *et al.*, 2009; Yadav *et al.*, 2018). In the present study F_1 progeny size of *S. oryzae* was highest for maize and least in millet. The reduction in adult emergence in millet could probably be due to low hatchability of eggs (Padmasri *et al.*, 2017). Furthermore, as they are known to spend a considerable part of their life inside the grain kernel which decrease the amount of endosperm (Keskin and Ozkaya, 2015) as reported earlier and is proved to be directly related to the size of the grain and hence variation in the hatchability which was found to

be directly proportional to the size of the grain is in accordance of the earlier work reported by Niewiada *et al.*, 2005 and Nawrot *et al.*, 2006.

The percentage loss of grain weight by S. oryzae were observed highest in maize by 29.88%, followed by chick pea, rice and millet by 26.54%, 22.02% and 16.10% respectively. Variation in grain loss by S. oryzae indicates its preference of particular grain on which they can lay large amount of eggs for the healthy upcoming F_1 progeny. Various studies show that the annual loss of stored grain by these pests from 15% to 57% with preference of different grains to be dependent on the size as well as the nutrient content (Shivakoti and Manandhar, 2000; Upadhyay et al., 2001; Bhandari et al., 2015). Nawrocka et al., 2012 also explained that the degree of damage is directly related to the infestation rate. Our results are in accord with the work of Ansari in (2003) on varietal screening of rice weevil on wheat and maize. The grain weight loss has also been correlated with the susceptibility and resistance of the grains seed damaged which is more in susceptible inbreeds, while it is less in resistant ones (Dari et al., 2010; Masasa et al., 2013; Derera et al., 2014; Garcia-Lara and Bergvinson, 2014; and Zunjare et al., 2016), probably in our study the maize is more susceptible compared to millet, chickpea and rice. Further, endosperm of susceptible inbreed promotes growth of the larvae much faster and in turn results into more grain weight loss as compared to resistant genotypes (Castro-Álvarez et al., 2015). Grain hardness has been identified as an important mechanism of resistance against the maize weevil (Garcia-Lara and Bergvinson, 2014), and according to Kelvin (2002) the snout penetration by the weevils into the grain depends on the hardness of the kernel thus the rate of penetration of the pest will be dependent on the hardness. In the present study of all the grains the millet is the hardest of the grain and thus the reduction in the adult emergence is well justified. Keskin and Ozkaya, 2015 has also said from his study that comparisons of wheat varieties indicated that Eser (soft wheat variety) was less resistant to the

insect attacks than Ceyhan-99 (hard wheat variety), as shown by the higher insect populations. This was attributed to differences in grain hardness, which is an important factor in insect infestation. Generally, insects develop more rapidly on soft grains compared to hard grain varieties (Ozkaya and Ozkaya, 2005; Ozkaya *et al.*, 2009; Mebarkia *et al.*, 2009).

Insects, moulds, and mites that attack the stored grains bring about big-deterioration, and low moisture or water activity further produces in situ undesirable metabolities, apart from depletion of calorie and selective nutrilites. (Srivastava and Subramanian, 2016). Further, cereals and millets carry only marginal amounts of proteins, however, they are rich in total carbohydrates and in calorie contents (Shewry, 2007). Depletion of different seed tissues such as germ, bran and endosperm has an individual effect on food quality and calorie supply. In the present study, the nutritional content of the infected grains was also measured and a phenomenal decrease in sugars of infested grains was observed. Sugars are mainly confined to the endosperm of the crop (Singh et al., 2013), the reduction in carbohydrates observed in the maize (37.97%), followed by chickpea (30. 27%), rice (26.96%) and least in millet (23. 98%) is parallel with the rate of infestation. It was noted that even protein content exhibited a downfall with 13.96% in maize, 11.6% in chickpea, 9.93% in rice and 8.03% in millet. However, the effect was less pronounced compared to carbohydrates which might be because of S. oryzae consumes exclusively endosperm, and often leaves the bran intact (Singh and Raghuvanshi, 2012). These observations are in agreement of previous studies by Bamaiyi et al., (2006) where they have reported Callosobruchus maculates infestation on nutritional loss on stored cowpea grains. Keskin and Ozkaya (2013) have also observed the effect of infestation had negatively affected the physical, chemical, and physicochemical properties of wheat and flour.

As reported by Campbell (2002) seed size and competition among larvae can impact offspring survival and fecundity, to maximize fitness, females need to make decisions about which seeds to use, how many eggs to lay, and whether to lay eggs in hosts already parasitized. In the present study as the main focus was rearing and obtaining a good number of adults for establishing the cell line fecundity for a single pair of S. oryzae was taken into consideration. Highest fecundity was observed for maize along with the higher number of females compared to males. This increase in the fecundity as well as the female ratio with maize may be attributed to the preference of grains by S. oryzae (Thakare et al., 2009; Ojo and Omoloye, 2016 and Akhter et al., 2017). Body size also affects the reproductive fitness of S. oryzae in numerous ways (Niewiada et al., 2005; Mebarkia et al., 2010; Volker et al., 2014). Thus large size of females has results them to produce more fecund off-springs. Parallel changes in longevity were also observed with respect to the fecundity rate of S. oryzae reared on different stored grains. The males lived for slightly shorter period as compared to females and the significant difference was recorded in the longevity of adult S. oryzae of both sexes reared on different varieties of grains. The present findings are in conformity with Jakhar et al., (2006) and Riyad (2009) who found significant variation in the longevity of adult of T. granarium in different varieties of stored grain pests. Similarly, Chander (2003) also found variation in the length of life of *R. dominica* in different varieties of barley. Yadav et al., (2018) have also reported difference in the longevity of S. oryzae with different varieties of wheat. Thus from the present study one can conclude that the maize grains were found to be the most suitable host rearing of S. oryzae compared to other grains viz. chickpea, rice and millet.

1.5 Conclusion

Thus, the study on rearing and host preference of *S. oryzae* concludes, among all the grains tested; maize is the most preferred host for rearing of *S. oryzae* in the laboratory conditions. All the stages of the life cycle, starting form egg laying to the emergence were found to be highest in the maize. The study also suggests that maize was rich in carbohydrate that has increased the size, fecundity and longevity of the insects. Hence, maize is the best suitable grain for rearing of *S. oryzae* insects.