
Assessment of maize stem borer damage on hybrid maize varieties in Chitwan, Nepal

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ABSTRACT

Maize is the second most important cereal crop in Nepal. However, national figure of grain production still remains below than the world's average grain production per unit area. Thus, this experiment was designed to determine the suitable time of maize planting, and to assess the peak period of one of the major insects, maize stem borer, in Chitwan condition. The results showed that plant damage percentage as per the maize planting month varies significantly, and the average plant damage percentage by stem borer was up to 18.11%. Length of the feeding tunnel in maize stem was significantly higher in January than July. In case of exit holes made by borer counted more than four holes per plant that were planted in the month of January. All in all, except the tunnel length measurement per plant, we observed similar pattern in other borer damage parameters such as exit whole counts and plant damage percentage within the tested varieties. Stem borer damage was not significantly affect on grain yield.

INTRODUCTION

In spite of diverse cultivation areas and seasons of maize, *Zea mays* L, in Nepal, the productivity maize is 2.3 ton/ha (MoAD, 2013); both biotic and abiotic constraints have played a major role in limiting grain production per unit area as compared to other developed nations. Among the biotic factors, maize stem borer complex is one of the most important insects in maize field (Neupane, Chapman, & Coppel, 1984; Jyoti & Shivakoti, 1992; Sharma & Gautam, 2010). About 20 to 80% of plants damaged due to maize stem borer were recorded in various studies (Thakur, Shrestha, Bhandari, & Achhami, 2013; Neupane, Coppel, & Chapman, 1984a). Similarly, Sharma & Gautam (2010) recorded more than 28% of grain harvested from stem borer protected field as compared to borer unprotected field. Mainly, the borer complex associated species in Chitwan condition were *Chilo partellus*, *Sesamia inferens*, *Chilo suppressalis* (Jyoti & Shivakoti, 1992). Female stem borer lay eggs in the leaves of both host and non-host plants. Newly hatched larvae migrate to search for host leaf and started to feed on the leaf that was enclose in whorl or was not fully exposed leaf. As a result of the leaf damage remarkable symmetrical holes are visible

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once the leaf fully exposed. Similarly, depends upon the size of the hole made by larvae on leaves, the damages are categorized as pin hole and window panes, the small (=pin head size) and large size hole (=larger than pin head to tends to leaf tearing). After having leaf damage, the larvae move inside the collar region of the plant, and started feeding on stem leading to develop a feeding tunnel. After feeding, some larvae pupate inside the stem while others migrate out of the stem. The objective of larval migration, by making a circular hole on the stem, from the stem is to become a pupa. Roughly, up to five generations of *C. partellus* recorded in the Chitwan valley of Nepal (Neupane, Chapman, & Coppel, 1986). Various biotic and abiotic factors including micro-climatic and edaphic factors play significant role in overall production of maize. In Chitwan, the month of June and July the maximum temperature is increased by 0.03°C to 0.04°C per year (Nayava & Gurung, 2010). In this slow but gradual changing scenario of temperature, crop cycles tend to shift from its original cycle. Consequently, many studies reported that changing cropping pattern as well as crop cycle and associated insect pest biology is shifting accordingly (Malla, 2008; Nayava & Gurung, 2012; Paudel, Acharya, Ghimire, Dahal & Bista, 2014). Thus, we may need to manipulate our crops and cropping pattern to mitigate the possible consequences of changing scenario of climates and insect-pest biology. Thus, objectives of the study are, 1) to find the best planting time for hybrid maize varieties; 2) to determine peak activities of maize stem borer in hybrid maize varieties in Chitwan condition.

MATERIALS AND METHOD

Plant materials: The research was carried-out at the research farm of National Maize Research Program, Rampur, Chitwan, Nepal. The geography of the experimental site is latitude $27^{\circ} 40' \text{N}$ and longitude $84^{\circ} 19' \text{E}$, and 228 m mean sea level. At field, four maize genotypes, two hybrids: RML32/RML17 and RML4/RML17 and two open pollinated varieties: Across 9942/Across9944 and S99TLYQ-B were planted at every Wednesday from July, 2012 to July 2014. For each genotype, two rows of five meter long, and the crop geometry $75 \times 25 \text{cm}$ were maintained. All the agronomic practices such as fertilizer application, weeding, side-dressing, and other necessary management practices were done as per the standard protocol to grow a good crop stand except any application of plant protection measure.

Data collection: Plant damage percentage, stem tunneling, exit hole count, number of kernel rows, number kernels per row, cob diameter, and grain yield were taken from the tested varieties and each date of planting. However, we have included only two hybrid varieties: RML32/RML17, and RML4/RML17, was considered for the following result.

Plant sampling: For exit hole and tunnel length measurement, five plants from each tested variety and each date of planting were collected. Similarly, five cobs from each date of planting and each tested variety were taken to measure cob diameter, number of kernel rows, kernels rows per row, and cob diameter.

Plant damage parameters: Plant damage percentage was assessed during the vegetative stage just before tasselling stage visually by counting healthy and damage plants of all tested varieties and each date of planting. However, the exit holes made by stem borer from the five sampled plants were counted visually after removing the intact leaves on stem, and then proceed for tunnel length measurement. Thereafter, the sampled plants were dissects longitudinally, and then measure the groove made by stem borer by using simple measuring scale in centimeter.

Yield attributes: Cob diameter, number of kernel per row, and number of kernels rows per cob were measured after harvesting five sample cobs from each date of planting and each tested varieties. For cob diameter was performed by using Vernier caliper, and counting the kernel rows and number of kernels per row by shelling individual cob each count. In case of grain yield estimation, we harvested all cobs and converted it into ton per hectare by using the formula: $\text{field weight (kg)} \times (100 - \text{moisture content}) \times (10,000 \times 0.8) / (\text{net plot area} \times \text{shelling percentage} \times 1000)$

Data analysis: All the collected data were analyzed by using statistical software R.3.2. Analysis of variance, regression analysis and clustering of maize planting months according to borer damage parameters, and yield attributes.

RESULTS

1) Plant damage percentage

The Plant damage percentage is presented in Figure 1 and Table 1. The highest plant damage percentage (22%) caused by maize stem borer was recorded in the maize planting month of June followed February and March. The lowest damage percentage was recorded in the planting month of October. Within the tested varieties, year of planting, and interactions among these factors were not found significantly differences in case of plant damage percentage (data not shown).

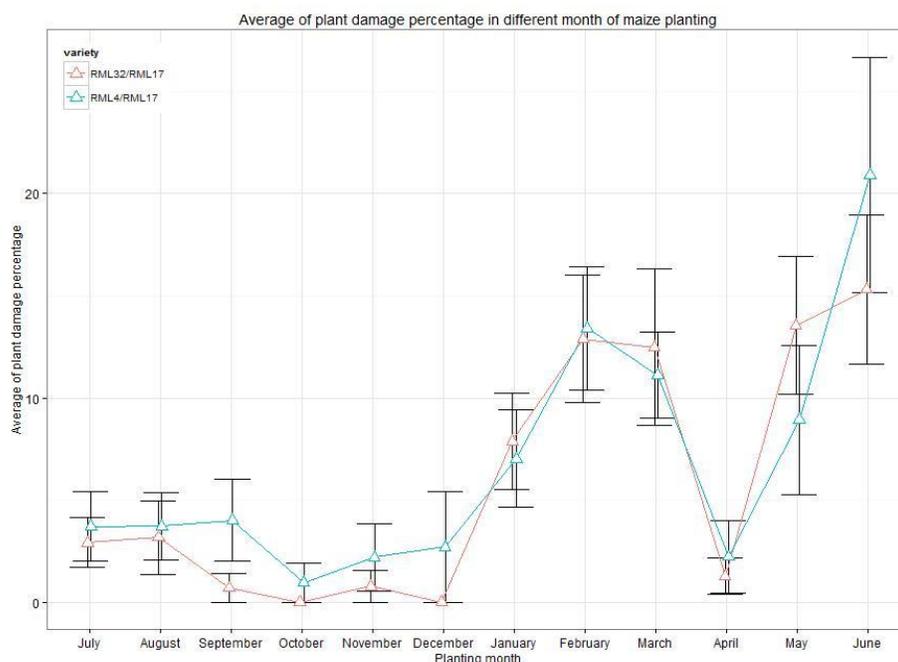


Figure1: Average of plant damage percentage with error bar in different month of maize planting in two varieties. The average is calculated based on data from two year of observations.

2) Tunnel length measurement

Average of stem tunneling caused by maize stem borer was measured in centimeter. The expression of measured length represented in figure 2 and table 1. The highest (16.82 cm per plant) in the plant that were planted in the month of January. And the lowest (0.94 cm per plant) measurement was recorded those plants which were planted in the month of June. Similarly, the second year (2013/14) of planting was relatively higher stem tunneling per plant than first year (2012/13), and RML32/ RML17 was more susceptible to stem damage than the RML4/RML17.

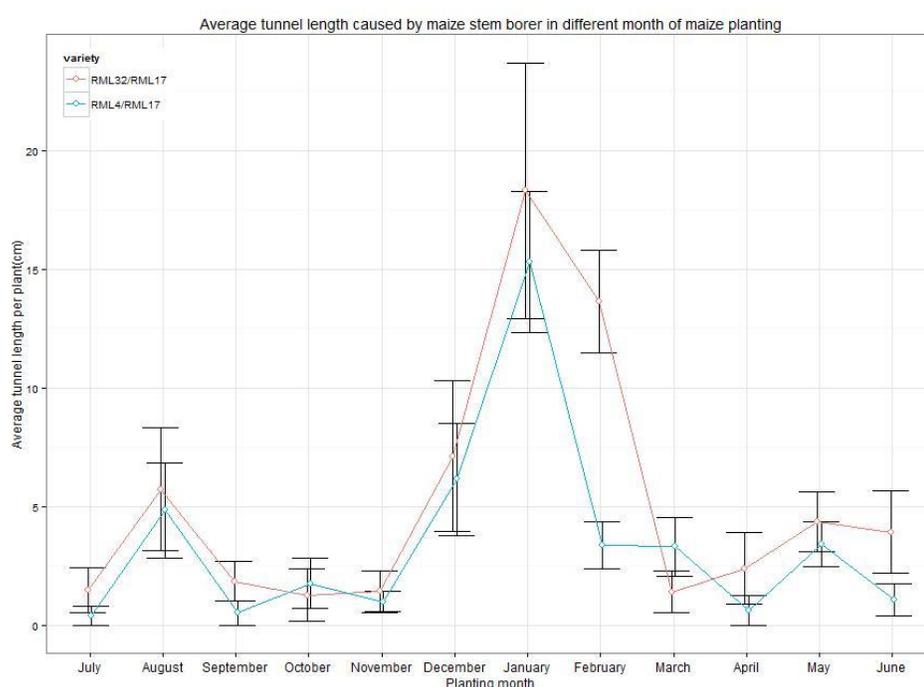


Figure 2: Average of tunnel length measurement (in centimeter) with error bar in different month of maize planting. The average is calculated based on data from two year of observations.

3) Exit hole counts

Plants that were planted in January and February had the highest average number (5.4 holes) of exit holes per plant as compared to rest of the planting months of maize (figure 3 and table 1). Within the tested varieties and year of maize planting, and their interactions were not found significantly different in terms of number of exit holes counted per plant (data not shown).

Table 1: Mean of stem borer damage parameters and yield attribute traits. Data represents mean of two hybrid varieties (RML4/RML17 and RML32/RML17), and over two years period

Planting month	Stem borer damage parameters			Yield components			
	Tunnel length per plant (cm)	Exit hole per plant	Plant damage percentage	Number of kernel row diameter (cm)	Number of kernels per row	Grain yield kg ha ⁻¹	
January	16.83 ^a	4.16 ^a	7.45 ^{cd}	12.95 ^b	4.36 ^a	33.34 ^{ab}	5050.11 ^{abcd}
February	8.51 ^b	5.42 ^{ab}	13.15 ^{ab}	12.67 ^b	4.29 ^a	36.39 ^a	6045.20 ^{ab}
March	2.36 ^{bc}	0.84 ^{bc}	11.80 ^{bc}	13.36 ^a	4.37 ^a	35.97 ^a	6264.16 ^{ab}
April	1.51 ^{bcd}	0.60 ^c	1.75 ^e	20.08 ^b	4.18 ^a	26.80 ^{bc}	5836.21 ^{abc}
May	3.89 ^{cde}	1.91 ^{bc}	11.23 ^{bc}	21.88 ^a	4.05 ^a	24.59 ^c	4869.14 ^{abcd}
June	3.04 ^{ae}	1.28 ^{bc}	19.00 ^a	23.38 ^a	4.05 ^a	23.45 ^c	4748.64 ^{abcd}
July	0.94 ^{ae}	0.31 ^c	3.32 ^{ae}	13.01 ^d	4.07 ^a	34.79 ^a	6471.44 ^a
August	5.28 ^{ae}	0.55 ^c	3.45 ^{ae}	12.79 ^d	4.22 ^a	33.69 ^{ab}	5812.84 ^{abc}
September	1.18 ^{ae}	0.30 ^c	2.35 ^{ae}	12.63 ^d	4.37 ^a	29.15 ^{abc}	6776.35 ^a
October	1.51 ^{ae}	0.17 ^c	0.48 ^e	12.36 ^d	4.32 ^a	26.98 ^{bc}	3733.02 ^a
November	1.21 ^{ae}	0.33 ^c	1.50 ^e	12.69 ^d	4.18 ^a	24.81 ^c	3878.69 ^{cd}
December	6.64 ^e	1.06 ^{bc}	1.36 ^e	12.58 ^b	4.16 ^a	30.03 ^{abc}	4451.50 ^{bcd}
Year	**	ns	ns	**	*	*	*
Planting month	**	**	**	**	ns	*	*
Variety	*	ns	ns	ns	*	ns	ns

(*P < 0.05; **P < 0.01; ns: not significant) Means with same letter are not significantly different at 0.05 level of significance

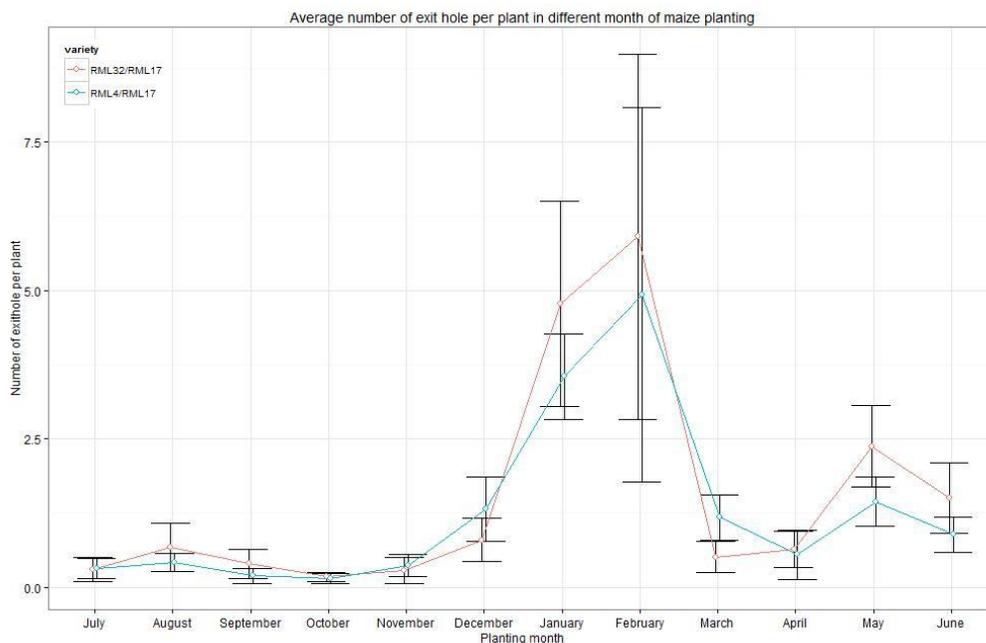
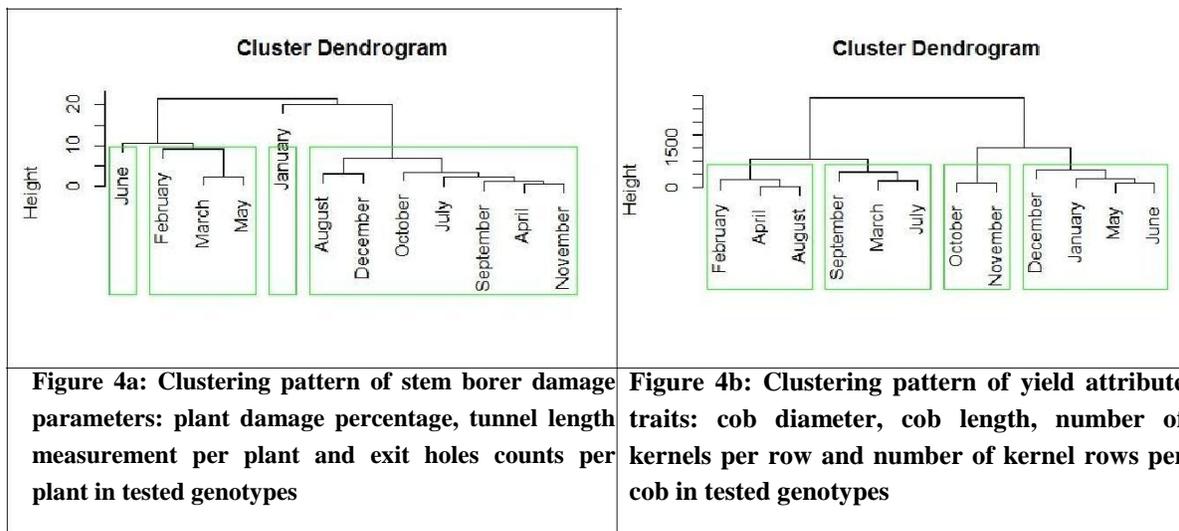


Figure 3: Average number of exit hole with error bars different month of maize planting. The average is calculated based on data from two year of observations.

4) Clustering of borer damage parameters and yield traits

Percent plant damage, tunnel length, and exit holes were categorized as stem borer damage parameters. By considering these parameters, we made a dendrogram according to the planting months of maize (fig 4a). The borer damage parameters were similar in August, September, October, July and November. Similarly, February, March and May were in one category; while the month of June and January each has made a separate cluster clusters based on borer damage parameters.



The figure 4b represented the yield attribute traits, which were included plant height, number of kernel rows per cob, kernels numbers per row. According to the cluster dendrogram, there are four categories of maize planting months. February, April and August in one group; September, March and July in second group; October and November in third group; and December, January, May and June are in the fourth category.

All the observed damage parameters of maize stem borers had not exert a significant impact on grain yield of both tested varieties; however, cob length and number of kernels per cob had a significant positive role for grain production (figure 5).

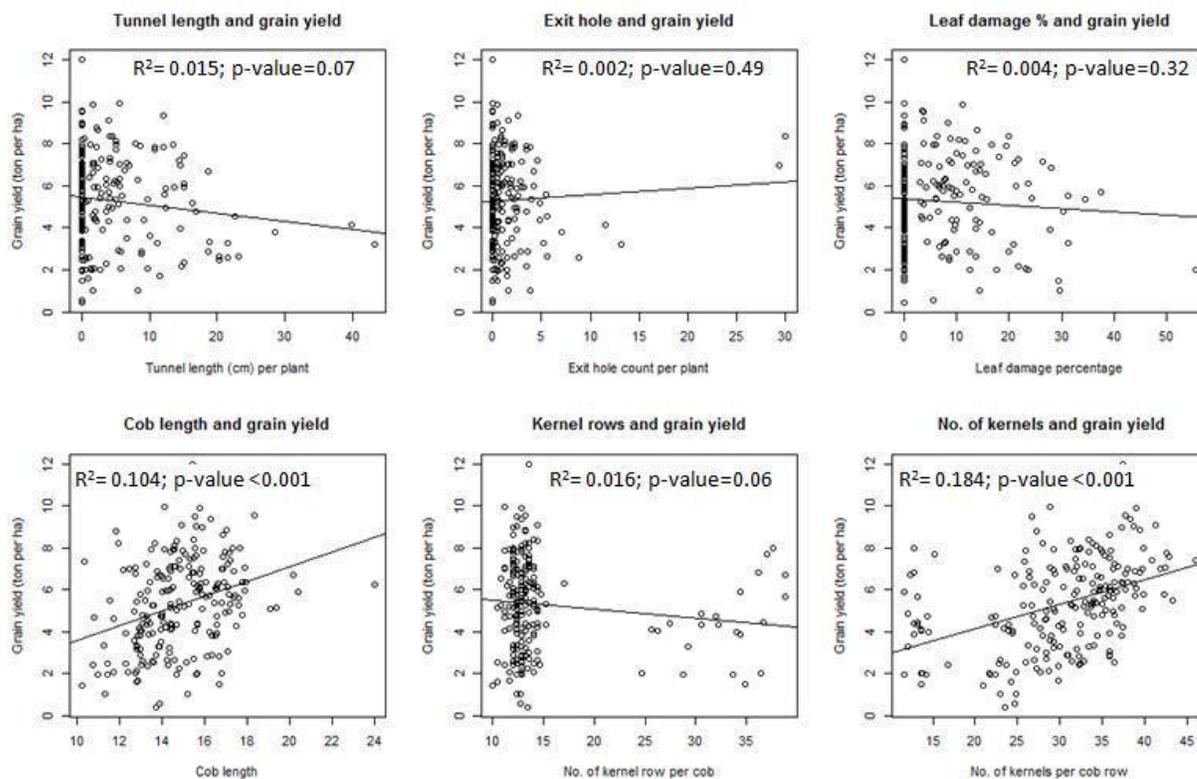


Figure 5 Relationship of stem borer damage parameters (tunnel length measurement, leaf damage percentage, and exit hole count per plant) with grain yield; and relationship of yield measurement traits (cob length, kernel rows per cob, and number of kernels per cob row) with grain yield

DISCUSSIONS

Two tested genotypes, promising hybrids: RML4/RML17 and RML32/RML17 have a common male parent, RML 17. Thus, the damage parameters, except tunnel length measurements, were recorded significantly different each other. In hybrid (F1) has a common male parent means share at least a 50% character of each so that a similar level of resistance mechanism against to maize stem borer damage. Similarly, other experiments have shown these two varieties showed moderate resistance against maize stem borer in terms of borer damage parameters (Anonymous, 2014). It would be possible that both varieties have capacity to recover the plant damage at early stage of the plant growth; thus showed no difference statistically on grain yield, although there was significant difference in plant damage parameters. Alternatively, in maize, one of the important herbivore-induced volatiles is terpene synthase TPS10, which is strongly expressed after the damage of lepidopteran pest, and it recruits natural enemies of the pest (Schnee *et al.*, 2006). Furthermore, secondary metabolites in maize such as C-glycosyl flavones maysin and the phenylpropanoid product chlorogenic acid in silk are detrimental to growth and development to certain lepidopteran insects, *Helicoverpa zea* and *Ostrinia nubilalis* respectively (McMullen, Frey & Degenhardt, 2009). Thus, it would be one of the possible reasons that the above mentioned secondary metabolites sufficiently produced by plants in our experimental field to recruit

natural enemies of the maize stem borer, as a result of that we were unable to observe the significant variation in plant damage percentage between the tested varieties.

In general, larvae of stem borer started to migrate from leaf to collar region, and thereafter initiate devouring in the stem. In this experiment, we observed similar level of preference by maize stem borer larvae in order to cause plant damage percentage on both of the tested genotypes at early stage of the plant growth; however, per plant stem tunneling measurement was higher (5.20 cm) in RML32/RML17 than per plant stem tunneling, 4.20 cm, of RML4/RML17 (Table 1). In this case, the female parent, RML32, of the tested genotype might contain at least some chemical cues that would enough to attract to maize stem borer adult. On the other hand, female parent, RML 4, of other tested hybrid may have contained certain specific metabolites that deter the growth and development of maize stem borer so that we observed lower stem tunneling data on it. Young leaves of maize contain maysin, which would deterrent chemical compounds to many herbivores, more than 300 µg per gram fresh weight of leaves (Maag, Bernal, Wolfender, Turlings, & Glauser, 2015). However, we need to conduct series of experiments to measure the available secondary metabolites in these inbred lines.

Borer damage parameter, exit holes count per plant, was not observed significantly different between the tested varieties (Table 1). Similarly, 4 and 3 number of exit holes per plant was on RML32/RML17 and RML4/RML17 respectively (Anonymous, 2013/14). In general, each of the exit holes on plant was made by stem borer larva, which was migrated from the stem somewhere to soil or base of the maize stem to spend its pupal stage. Thus, the reduced number of exit hole per stem means either the limited number of larvae were migrated from the stem where it fed; or even the larvae were inside the stem, the larvae were captive by the hardness of the stem. It provides sufficient ground for further investigation.

Different months of maize planting, from January to December, had significantly different on the measured borer damage parameters. It is obvious that almost each month had a wide range of minimum temperatures, maximum temperatures, humidity levels, and rainfall amount. Similarly, each organism has own level of requirements for their growth and development. Thus, an influence of these weather parameters to regulate growth and development of the larvae as well as adult of maize stem borers would not be beyond the

expectation. As adult population in the field fluctuates in response to ambient weather condition, the degree of damage by larvae may also be fluctuated. In this case, we clustered the all months of maize planting. Based on the observed data of borer damage parameters, we found a clear figure that August, December, October, July, September, April and November in one group; February, March and May is second large group; June in third group; and January in the fourth group (Figure 4 a, and 4 b). It is clearly observed from figure 1, 2, and 3 that the highest plant damage percentage was recorded in the planting month of June; the highest tunnel length measurement found on the maize plants that were planted on January; and the highest number of exit hole per plant found on the maize plants, which were planted in the month of February. It would be possible that during the planting month of January and February had lower number of plant damage because during this time period the average temperature was recorded less than 15 °C (Appendix 1) which would not be an ideal condition for the maize stem borer growth and development. In support of that Tamiru, Jembere, & Bruce (2012) suggested that *Chilo partellus* (Swinhoe), one of the important insect species of maize borers, takes longer duration to complete its life cycle at temperatures 22 °C as compared to higher 30 °C. Similarly, Neupane, Chapman, & Coppel (1986) reported that a 30 °C temperature range was an optimum for *C. partellus* development, and the subsequent threshold temperatures for eggs, larval, Pupal and entire life development were 13.2, 12.5, 12.7 and 13 °C respectively. Plants that were planted during the months of January and February had greater exit holes and longer tunnel lengths compared to other months, but lower plant damage percentage (Table 1). Here, temperature might have played a significant role for both maize growth and stem borer damage to maize plant. Additionally, longer tunnel length and greater counts of exit holes indicated increase level of borer activities. This could be possible that the growth and development of maize increased with increasing temperature and the duration of the maize growth in these two months of planting periods extended up to April and May (=fourth week of Jestha and Ashad), one of the peak period the maize stem borer dynamics (Gyawali, 1978). In agreement with that two open pollinated varieties, Across9942/Across9944 and S99TLYQ-B, had the highest tunnel length measurement as well as the highest number of exit holes count per plant in January planting than the measurements of any other months of maize planting (Achhami, Bhandari, Thakur, & B.K., 2014). Similarly, Thakur, Shrestha, Bhandari & Achhami (2013) also found similar pattern in tunnel length measurement and exit holes count in Chitwan condition but with different tested varieties than that is reported in our study. We observed that stem borer damage parameters did not show significant negative impact on grain yield. However, in a separate experiment, Odiyi, (2007) showed moderate to high correlations of leaf feeding damage, number of broken stems, and stems tunneling with grain yield. Similarly, 4-5 t ha⁻¹ yield was reported by Neupane, Coppel, & Chapman (1984a) in Arun-2 maize variety despite the high stem borer damage (46.7% damage; 4.4 cm stem tunneling per plant). In conclusion,

January was the most damage prone planting month of varieties: RML32/RML17 and RML4/RML17 followed by June, whereas October and November were the lowest grain yield as compared to other month of plantings on these tested varieties in Chitwan condition. Thus, we recommend further studies related to interaction between maize varieties with secondary plant metabolites, potentially by using molecular tools to better understand maize stem borer biology and population dynamics in Chitwan condition.

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Appendix 1: Weather parameters during the maize planting months from July 2012 to July 2014 at Rampur, Chitwan

