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Assessment of seed production potential of Teosinte (*Euchlaena mexicana*) under varying agronomic management practices in the central region of Nepal

A thesis
submitted in partial fulfilment
of the requirements for the Degree of
Doctor of Philosophy

at
Lincoln University
by
Sunita Sanjyal

Lincoln University
2021

Abstract of a thesis submitted in partial fulfilment of the
requirements for the Degree of Doctor of Philosophy

Abstract

Assessment of seed production potential of Teosinte (*Euchlaena mexicana*) under varying agronomic management practices in the central region of Nepal

by

Sunita Sanjyal

Teosinte (*Euchlaena mexicana*) is a popular summer herbage crop in Nepal. While it has good seed production potential, the management for seed production is unknown. A two year study was undertaken in order to investigate teosinte seed yield and seed quality for different sowing dates, seed sowing rates and cutting management in the Terai region of Nepal. A seed development, a genotypic diversity and an economic study were also conducted. There were four different sowing dates (30 March, 30 April, 30 May and 30 June), four seed rates (20, 40, 60 and 80 kg ha⁻¹) and three cutting management treatments (uncut, once cut and twice cut) arranged in a split split plot design. The only certified variety of teosinte in Nepal, Sirsa was used for the study.

Herbage yield of teosinte was affected by sowing date, seed rate and cutting management. Maximum herbage yield (HY) and dry matter yield (DMY) from a teosinte crop grown for seed production was obtained from the 30 April sowing together with the 60 kg ha⁻¹ seed rate and two cuts. There was a positive correlation of plant height, tiller number, leaf number and leaf area index (LAI) with DMY.

The effect of the environment on both teosinte herbage and seed yield was studied. Higher herbage and seed yield were produced from early sown teosinte because the longer growing season allowed the accumulation of higher growing degree days (GDD). Five critical growth stages of teosinte were identified. The temperature and GDD requirements for each growth stage were 26.1°C (135°C days), 26.0°C (2189°C days), 24.1°C (2442°C days), 20.4°C (3049°C days) and 17.2°C (3150°C days) for emergence stage (GS1), vegetative stage (GS2), flowering stage (GS3), seed development stage (GS4) and seed maturity stage (GS5) respectively.

Maximum seed yield (kg ha⁻¹) was obtained from the two earlier sowings (30 March and 30 April) in both years because early sown plants were taller, and had higher LAI and more tillers and cobs per plant, ears per cob and seeds per ear than later sown plants. In 2017, there was a non-insignificant

effect of seed rate on seed yield because of natural thinning of plants due to heavy rainfall and wind which caused lodging in the early vegetative stage, but in 2018 the two lower seed rates (20 and 40 kg ha⁻¹) produced the highest seed yield. For cutting management, seed yield was higher for uncut plants in both years.

Seed quality was tested for seeds harvested from different sowing dates, seed rates, cutting management and cob position on the plant. Over the two seasons of trials, seed harvested from the 30 March sowing at the two lower seed rates (20 and 40 kg ha⁻¹) and uncut plants resulted in higher germination percentage and thousand seed weight (TSW) in both years. There was a significant negative correlation between the sowing dates and germination percentage and a negative correlation between sowing dates and TSW because germination percentage and TSW were reduced with each delay in sowing. There was a positive correlation between TSW and germination percentage for different sowing dates for all cobs in 2017 ($R^2 = 0.77$) ($P > 0.05$) and 2018 ($R^2 = 0.80$) ($P > 0.05$). Seed quality was also affected by the cob position on the plant. When seeds were hand harvested separately from top, middle and bottom positioned cobs, higher quality seeds (germination percentage and TSW) were obtained from seed harvested from the top positioned cobs. Teosinte seeds physiological maturity (PM) was attained at 59 days after anthesis. Harvesting teosinte seeds from the top and middle positioned cobs on the plant recovered 78% of the total seed yield, while that from the middle and the bottom positioned cobs recovered 57% of the total seed yield. Therefore harvesting mature seeds from the top and middle positioned cobs is recommended to minimize loss of quality seed from shattering which would occur if harvesting was delayed until seeds from the bottom cobs were mature.

A diversity study of teosinte was conducted for 18 teosinte accessions, 17 from CYMMIT Mexico and Sirsa, to identify if any of the introduced accessions could perform better than Sirsa in terms of herbage yield, seed yield and time to seed maturation. Out of 17 accessions, accessions 5, 7 and 12 out yielded Sirsa in terms of herbage yield, seed yield and were earlier to maturity. This preliminary result suggest a possible source of material for developing new teosinte varieties in Nepal better suited for farmer's needs, particularly to reduce the length of time required to grow a seed crop. A separate study was conducted on seven seed lots of teosinte collected from different regions of Nepal. Hierarchical cluster analysis based using morphological characteristics gave two distinct clusters; cluster I (Makwanpur) from the midhills and cluster II (Sarlahi, Mohattari, Bara, Chitwan, Gaughat and Tikapur) from across the southern Terai. These two clusters suggest an agro ecological differentiation for teosinte genotypes grown in Nepal.

An economic analysis conducted based on the total costs and income from the different management in this research study showed that the highest gross margin was obtained from the March and the

April sowings at the 20-60 kg ha⁻¹ seed rates and none or one cuts. Taking one herbage cut was not detrimental to a farmer's gross margin for seed production.

Keywords: Teosinte, sowing date, seed rate, cutting management, herbage yield (HY), dry matter yield (DMY), seed yield, cob position, harvesting, germination, thousand seed weight, dormancy, seed development, temperature, growing degree days (GDD), diversity, accessions, genotypes, economics, gross margin.

Acknowledgements

It is a great pleasure to take this opportunity to thank all those who have helped me with my PhD project. Firstly, I would like to express my sincere and cordial thank you to my supervisor, Professor John Hampton for persevering as my Ph.D. supervisor. I cannot thank him enough for his rigorous support and guidance; both academic and for my personal circumstances during this entire journey. It was a matter of pride and a great honour to get this opportunity to work under the mentorship of a world renowned seed scientist. The knowledge and wisdom he has imparted to me will be of great assistance in my career. I express my gratitude for his invaluable support, motivation and patience as I completed my thesis. My sincere thank you to my associate supervisor, Professor Phil Rollston for his specialist knowledge, guidance and reassurance through out my PhD career. I would also like to express my gratitude to my advisor Dr Santosh Maranhattha for all the help during my field experiments in Nepal and being available whenever needed. I would also like to thank Keith Armstrong and Gill Armstrong for their love, continuous support and encouragement throughout this journey. I am also thankful to my former chiefs in Nepal, Dinesh Pariyar and Kishore Kumar Shrestha for their constant support and encouragement during my professional and academic career.

I am grateful to Manaaki New Zealand (Ministry of Foreign Affairs and Trade) for awarding me a scholarship to pursue my doctoral degree at Lincoln University. I am also thankful to the National Agricultural Research Council (NARC), Nepal for providing me with study leave for my PhD studies at Lincoln University, New Zealand. Many thanks to the project of Prof. John Hampton, Cool Season Crop Improvement Program (CSCIP) for providing funds for my field research in Nepal. Many thanks to Agriculture and Forestry University (AFU), Chitwan, Nepal for providing land for research trials and to staff for their hands-on assistance in seed laboratory work. Special thanks to seed laboratory technicians, Suraj Karki and Dinesh Marasaini for making sure that assistance and equipment were available, where needed. Also thanks to National Cattle Research Centre (NCRP), Rampur, Chitwan for providing land for my research trial. I am also thankful to the staff of the Bio-Protection Research Centre, particularly Dr Andrew Holyoake and Mr Brian Kwan. I am grateful to Dave Saville for his expertise and support with his statistical help. I would also like to thank two technicians of NCRP, Sanjit Niroula and Lilaram Pathak for their support during data recording in the field.

I am also very appreciative to my office mates, Nghia Thi Nguyen, Sylvester Atijegbe and Ali Kakhiki for all the happy moments we shared. Special thanks also to my friends particularly, Tauseef Khan Babaa Khan, Hossein Alizadeh, Huong Tam Thi Pham, Sundar Tiwari, Nirajan Bhattarai, Nisha Bhattarai and Sachin Upadhyay for their joyful company during my stressful period of research and writing. I would like to thank my family especially my parents, brothers, sisters, nephew and niece for always being there for me and standing by my major decisions. Special thanks to my mother for all the effort she

has made in my upbringing. I would also like to express my special gratitude to my parents in law and sister in law for being supportive and taking care of my kids while I was away for my study. I wouldn't have gotten this far without their constant belief and support. My special thanks to my best companion, my husband Saroj Sapkota for being very helpful and compassionate throughout this journey. My special love and thanks to my two sons Shubham Sapkota and Sauhard Sapkota who grew up with my PhD and for their patience and understanding as to why their mother was away at their very young age.

Thank you all!

Sunita Sanjyal

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Chapter 1

Literature Review

1.1 Introduction

This thesis includes a set of experiments designed to investigate agronomic management for seed production, a seed quality assessment, a diversity study and the economics of growing teosinte (*Euchlaena mexicana*) in the plains of Nepal.

In this chapter management factors affecting herbage and seed production of teosinte are reviewed; sowing date, seed rate and cutting management were known factors, maximizing herbage and seed yield are identified. This PhD research programme include six objectives covering sowing time, seed rate and cutting management, phenological growth stages and also seed development. The scope of the study was extended by examining the seeds for quality and conducting a diversity study along with an economic analysis of the management factors on herbage yield, seed production and quality. Teosinte is grown as a popular summer forage crop around the tropical world. Being the progenitor of maize, it is surprising that apart from some archaeological studies, very little is known about its agronomic management and seed production. In this review therefore, examples from maize are used where there is no information for teosinte.

1.1.1 World scenario of teosinte crop

Just like maize, teosinte has a varied genetic diversity. Agronomically, it has several useful characteristics, including its capacity to produce high biomass, withstand multi cutting, produce more tillers than maize, and a good capacity to resist biotic and abiotic stresses. Therefore, teosinte has been suggested as a possible source for maize improvement. The higher leaf ratio in the total biomass compared to maize means teosinte has higher digestibility. Further, the nutritional content of the foliage is also higher than maize forage. Teosinte has been domesticated for herbage purposes, and it can be a continuous source of herbage during the hot periods (Niazi, Rauf, da Silva, & Munir, 2015).

The teosinte crop is important as herbage for milking animals in Asia, either as fresh for herbage or as a silage crop. Teosinte is commonly called “Makchari” and “Makiya” in India and numerous varieties are available. The total teosinte cultivated area in India is about 10,000 ha (NDDDB, 2012; Kundu, 2015) while in Nepal the total area under teosinte cultivation is 27,232 ha which is higher

than all other herbage crops grown in the country. The annual teosinte seed production in Nepal is 870 tonnes which means the seed deficit in the country is 25 percent (NPAFC, 2018).

Teosinte is popularly called “Guatemalan teosinte”, “Venezuelan grass”, “Imperial grass”, “Teosinto” or “Dente de burro” by farmers of Santa Catarina state, in the south of Brazil. Although the presence of teosinte in maize fields was officially recorded in Santa Catarina in 2011, the local farmers conveyed that it had been present since 1949. Further, it has become a noxious weed in maize fields of most farmers in the Toluca and Chalco valley of Mexico. However for many other agrarian communities in the world with hot summers and where dairying is one of the major source of economic gains, teosinte has become a valuable herbage source for dairy cattle, leading to improved food safety (Silva, Vidal, Costa, Vaio, & Ogliari, 2015). Teosinte is called Rayana in Egypt and contributes about 20% of the total herbage crops. It is valued for its multicut production in the warm regions of Egypt where the area under teosinte is about 21,000 ha (El-Nahrawy, 2011). In Spain and France teosinte is considered a weed of maize crops (EFSA, 2016) and control measures are being investigated (Pardo et al., 2015; 2016). Teosinte can adapt well to abiotic stress. Teosinte contains distinct and favorable genes that could be transmitted to novel maize varieties for improving the adaptation of maize to a changing climate (Sanchez et al., 2018).

1.1.2 Origin and history of teosinte (*Euchlaena mexicana*)

Euchlaena mexicana has been coined as a universal name for the progenitor of the wild taxa *Zea*. It belongs to the Poaceae family (subfamily *Panicoideae*) and is monocotyledonous in nature. Other agriculturally important members of the Poaceae family are maize, sorghum, rice, barley, wheat, bamboo, and grasses. Mangelsdorf and Reeves (1939) first suggested maize was domesticated from teosinte by human selection. The *Zea* class is separated into four sub species, three of which are in the wild state (*ssp. huehuetenangensis*, *ssp. mexicana*, and *ssp. parviglumis*) and the one considered to be in the cultivated form is *Zea mays* (Matsuoka & Yoshida, 2005). Along with variations in morphology, these sub species also have a different geographical distribution. The sub species *huehuetenangensis* is a native of western Guatemala while sub species *mexicana* and *parviglumis* are native to Mexico. *Euchlaena mexicana*, commonly called teosinte, belongs to the sub species *mexicana*. The name teosinte, comes from the Indian word Nahuatl meaning “grain of the gods” (Doebley, 1990).

Some species of teosinte differ phenotypically from maize both genetically and taxonomically, and they in no way appear to be associated with the origin of maize. However, one form, *Z. mays ssp. parviglumis*, shares a particularly close genetic relationship with maize and available evidence indicates that it is the direct ancestor of maize (Doebley, 1990; Matsuoka et al., 2002). It

is believed that teosinte originated in the valleys of south-western Mexico, where, it grew commonly as a wild plant along streams and on hillsides, and often invaded the cultivated fields as a weed. It is most common in the Balsas river drainage area of southwest Mexico and hence is also known as Balsas's teosinte (Doebley, 1990; Matsuoka et al., 2002). Diversification within the major maize races in Mexico today is believed to be the result of introgression from teosinte (Sanchez et al., 2018).

1.1.3 The teosinte hypothesis

The term teosinte communally represents all taxa inside the genus *Zea* except for maize (de Lange, Balmer, Mauch-Mani, & Turlings, 2014). There have been two schools of thought regarding teosinte being the progenitor of maize. The first view opines that cryptogenic studies of maize and teosinte determine that they belong to the same biological species, although transformation exists. It states that the large ears of maize were the result of drastic changes that took place over many years to the small ears of teosinte (Beadle, 1939). The second school of thought was led by Mangelsdorf and Reeves (1938, 1939), who were reluctant to accept the so called tripartite hypothesis or wild maize hypothesis. Since great variation was observed morphologically, they considered that the progenitor of maize had to be looked for outside the *Zea* family for an ancestor that no longer existed.

Beadle (1939, 1978, and 1980) proposed the wild maize hypothesis using evidence from cytological, archaeological, anthropological, geographical, and linguistic research findings which showed teosinte as a sole progenitor of maize. To prove this, he conducted an experiment where he grew 50,000 maize – teosinte F2 plants. During the experiment he found that 500 F2 individuals had almost half maize or teosinte ear types, suggesting strongly that teosinte was the progenitor of maize. Only a few gene changes would have accounted for the difference in the ear morphology. Later QTL mapping conducted by Doebley and Stec (1991, 1993) to assess morphological difference in a maize –teosinte F2 population showed that five to six regions of the genome have a robust influence on genome group. They identified a candidate gene named Teosinte branched 1 (*tb1*), which controls the apical dominance between maize and its progenitor. This gene was identified as a major one influencing apical dominance resulting in the long lateral branches ending with tassels. Further, DNA marker based and isozyme related studies have shown that *Zea mays spp. parviglumis* is very similar at the molecular level while at the morphological level *Zea mays spp. mexicana* has a more maize like appearance than *Zea mays spp. parviglumis* (Matsuoka, 2002).

1.1.4 Teosinte in Nepal

The place of teosinte in Nepal in terms of herbage and seed production is reported in Chapter 2 and 4 respectively. Figure 1.1 shows the early vegetative growth stage of teosinte. Figure 1.2 demonstrates the overnight regrowth after cutting. Figure 1.3 shows the plants under different cutting managements, where uncut, once cut and twice cut plants can be seen, figure 1.4 demonstrates the vegetative growth of teosinte and figure 1.5 shows farmers harvesting teosinte herbage using a reaper.



Figure 1.1 Early vegetative growth



Figure 1.2 Regrowth of teosinte overnight



Figure 1.3 Teosinte plants under different cutting management



Figure 1.4 Full grown teosinte plant



Figure 1.5 Farmer harvesting teosinte herbage using a reaper

1.1.5 Reproductive development of maize and teosinte

Both maize and teosinte look very similar morphologically. They both are tall plants and have broad leaves. However teosinte has much longer lateral branches and produces more tillers than maize. Further, they differ in their reproductive behaviour as well (Doebley, 1990). Both maize and teosinte are monoecious in nature and bear male and female flowers within the same plant as shown in Figure 1.6. However, there are morphological differences between the inflorescences of maize and teosinte. The seeds of maize are arranged in 3-12 rows that adhere so strongly to the rachis that they need human interference to detach for dispersal and propagation. Maize has a large cob containing hundreds of seeds (Matsuoka & Vigorous, 2002). The female inflorescence of teosinte consists of a capitulated fruit case. The fruit case is a rachis internode and the spikelet is attached with it. The rachis is strongly invaginated into rows such that seeds fit well into it. A series of bracts holds the spikelet which consists of the female inflorescence. The female inflorescence, called a silk, is produced in each leaf axil of each branch. The silks are white to purple in colour and are produced prior to or after the tassel emergence. The silks turn brown in colour and detach from the fruit after receiving the pollen, leading to the maturation of fruit in a descending order. The bract consists of the outer glume which closes the opening of the spikelet to protect from predators. At maturity the rachis and outer glume form a very indurate structure called a cob, also known as fruit cases, which disarticulate to allow the rows to shed seeds naturally (Iltis, 1987). Teosinte has a cluster of fruit cases with 3-5 rows in them and each row bears 5-10 small hard seeds enclosed in a stone hard fruit case/cob. The seeds are single rowed, light to dark brown when fully mature as shown in Figure 1.9. The teosinte seeds disarticulate as soon as they attain

physiological maturity (Benz, 2001; Iltis, 1987). However the seeds mature at different time with in the same plant (Figure 1.8).

For the male inflorescence, teosinte has several lateral branches which bear a multiple terminal flower called a tassel. The primary tassel appears on the tip of the main stem, secondary tassels are produced at the tip of each tiller and tertiary tassels are produced on secondary tillers. In some plants tassels appear in leaf axils as well. The tassels are usually green in colour before maturation. Later a purple coloured pollen bag appears on the tassel which turns brown after maturation. Despite the profound variation in ear morphology of both, some species of teosinte and maize can hybridize with each other (Collins, 1919; Emerson & Beadle, 1932).



Figure 1.6 Teosinte inflorescence



Figure 1.7 Teosinte plants ready for seed harvest



Figure 1.8 Seeds from different cob position



Figure 1.9 Fresh seeds

1.1.6 Temperature and photo period

Temperature and photoperiod are two factors that play major roles in determining the rate of development of crops grown under optimal conditions of moisture and nutrient supply (Warrington & Kanemasu, 1983a). The pace of plant growth is determined by the quantity of heat dispersal and the availability of heat to the plants (Tollenaar & Bruulsema, 1988; Muchow, Sinclair, & Bennett, 1990). Thus crop growth is directly influenced by temperature (Brown, 1977; Hardacre & Turnbull, 1986; Warrington & Kanemasu, 1983a). Temperature plays a vital role in the growth and development of maize by regulating the time within each developmental phase (Hatfield & Prueger, 2015; Tsimba, Edmeades, Millner, & Kemp, 2013). Maize crops respond to photoperiod only after the juvenile stage, which is followed by a photo period sensitive stage termed the tassel Initiation stage. The duration of both of these stages is determined by thermal time (Daynard, 1972; Major et al., 1983; Muchow & Carberry, 1989; Cutforth & Shaykewich, 1990).

Temperature also affects the total leaf number (Hesketh, Chase, & Nanda, 1969; Tollenaar & Hunter, 1983; Stevenson & Goodman, 1972) and leaf canopy expansion (Tollenaar, Daynard, & Hunter, 1979; Hesketh & Warrington, 1989; Warrington & Kanemasu, 1983b) and ultimately the leaf area index. Studies conducted by Eberhart (1971), Acosta and Crane (1972), and Hallauer and Sears (1972) showed a negative correlation between maize vegetative growth and floral initiation during long hot days. Maize is a periodically sensitive short day plant (Garner & Allard, 1923; McClelland, 1928; Kuleshov, 1932, 1933; Thomas, 1948; Kiesselbach, 1950). However, Birch, Hammer, and Rickert (1998) have reported that temperature response in maize depends on the cultivar.

A study by Stevenson and Goodman (1972) demonstrated that reduced temperature would delay flower initiation along with the growth rate, thus reducing the apical dominance of the crop. Hallauer and Sears (1972) showed a negative correlation between floral initiation and vegetative growth in the long hot days during maize growth.

Temperature variation because of different sowing dates of forage maize regulated the heat availability to the plants during their growth, especially during the period from planting to silking (Birch et al., 1998; Omafra, 2009). The heat accumulated during this time is recorded as Growing Degree Days (GDD) which is interpreted by two equations: The first is, if the daily mean temperature is less than the base, the daily mean temperature is set equal to the base temperature. The second is if T_{max} or $T_{min} < T_{base}$ they are reset equal to T_{base} . Thus GDD is calculated as $GDD = (T_{max} + T_{min})/2 - T_{base}$ temperature, where T_{max} , T_{min} and T_{base} are the maximum, minimum temperature and base temperature respectively. MacAdam and Nelseon

(2003) also proposed that the base temperature for C₄ grasses like maize is 10⁰C. Teosinte varieties are sensitive to photoperiod and there is variation among the species in the strength of their response. Mexican teosintes are known to flower much earlier as compared to Guatemalan teosintes (Emerson, 1924; Langham, 1940). Similarly, Emerson (1924) and Langham (1940) found that the F1 hybrid of Mexican teosinte-maize flowered at the same time as that of the maize parent, showing that maize is almost completely dominant in this photoperiodic response. However, the Guatemalan teosinte-maize F1 hybrids bloomed slightly later than the maize parent. The F2 hybrids on average bloomed slightly later than the F1 hybrids. The (teosinte × maize) × teosinte progenies encompassing the Mexican teosintes were somewhere in between the F1 and the teosinte parent, but the Guatemalan teosinte backcrosses were more close to the teosinte parent.

1.1.7 Temperature and seed development

Successful crop production relies on the availability of good quality seeds (Hampton, Boelt, Rolston, & Chastain, 2013). Yield in cereals depends on the meristematic activity of leaves, ears, tassels, and tillers, which may compete with each other at various stages of plant development. Indeed, much of the art of cereal growing depends on management of this competition (Bunting & Dretnan, 1966).

Normal seed development is interrupted during the seed filling stage if the temperature goes beyond the optimum (Spears, Tekrony, & Egli, 1997). The ability of seeds to produce assimilates required during plant growth depends on temperature (Dornbos & McDonald, 1986). A small variation in the temperature during seed development and maturation can affect the quality of the seed (Gutterman, 2000). High temperature causes heat stress which causes physiological damage to the seeds (McDonald & Nelson, 1986; Coolbear, 1995; Powell, 2006) and the seeds may lose the ability to germinate. Seed vigour loss due to high temperature has been reported by Powell (1988), Spears et al. (1997); Egli, Tekrony, Heitholt, and Rupe (2005). Rashid, Hampton, Rolston, Tretheway, and Saville (2018) reported that the loss of seed vigour is the result of seed deterioration which is because high temperature impairs equilibrium between reactive oxygen species (ROS) production and ROS scavenging enzymes in seeds which disturbs the metabolic activity of seed. At low temperature, the ability of the seeds to store assimilates is reduced (Didonet, Rodrigues, Mario, Ide, & Tissot, 2001).

1.1.8 Seed position on plant and shattering of seeds

Seed size, structure and quality can be affected by the position of seed on the plant (Gutterman, 2000; Hampton, 2000). This can be a result of variation in assimilates obtained by the seed during development (Deleuran, Olesen, & Boelt, 2013). Both position of the inflorescence on the mother plants or seed position in the inflorescence and even seed position inside the fruit may affect the germination of seeds (Datta, Evenari, & Gutterman, 1970; Grey & Thomas, 1982). Teosinte is an indeterminate plant; seed maturity begins from the top of the plant and proceeds towards the base. The fruit of teosinte (seeds) are encapsulated in rows within a thick layered cob. The cobs are branched into three to five rows and each row bears 3-7 seeds attached as a single row. These seeds separate easily from each other when mature and the ripe seeds shatter readily. Teosinte produces a large number of seeds, many more than can be successfully harvested. Any delay in harvesting these cobs will result in progressively lower seed yield due to shattering. Fairey and Smith (1999) and Garcia-Diaz and Steiner (2000) have reported that indeterminate flowering, pod dehiscence and seed shattering results in considerable loss in seed yield. Gray, Steckel, and Ward (1985) have reported that the ideal time to harvest mature seeds in indeterminate plants is when the quantity of seeds yet to reach maturity is less than the mature seeds which could be lost due to shattering.

1.1.9 Seed germination

ISTA (2019) defines germination as the emergence and growth of the seedling to a stage where its essential structures indicate if the seedling can develop further into a satisfactory plant under favourable conditions in the field. Germination of a seed begins with the water uptake and is physiologically completed when the radicle emerges from the seed coat. Seed germination is completed in three phases (Figure 1.10). In phase I, rapid imbibition occur until all the cell contents are fully hydrated. In phase II water uptake is limited but the metabolic activity is high. In phase III there is only a slight increase in water intake, radicle emergence starts and ends with radicle elongation (Bewley & Black, 1994; Nonogaki, Bassel, & Bewley, 2010). Water imbibition is an important step in seed germination because it allows the activation of metabolic process that occur in the second phase and the emergence of the radicle in the third phase (Wolny et al., 2018).

Germination is a physiological process and is affected by moisture, light oxygen and temperature (Baskin & Baskin 1998). It is also highly affected by the environmental condition during seed development, harvesting, cleaning, drying and storage (Dornbos, 1995). Maize germination is affected by various climatic components, however temperature is directly related to the emergence of maize seedlings in the lowland tropics (Nielsen, 2017; Tozzi et al., 2014). The minimum temperature requirement for maize germination is 10⁰C and the optimum temperature is

30°C (Bosci & Kovacs, 1990; Birch, Vosb, & van der Puttenb, 2003). The ability of maize to germinate at high temperature (>37°C) is dependent on the temperature sensitivity of the embryo because protein synthesis is temperature sensitive (Riley, 1981). The thermal time for emergence of a maize is 115-120 GDD after planting. Thermal time can be estimated using soil temperature (Nielsen, 2017). The number of calendar days to emerge will be site specific and dependent on soil temperature.

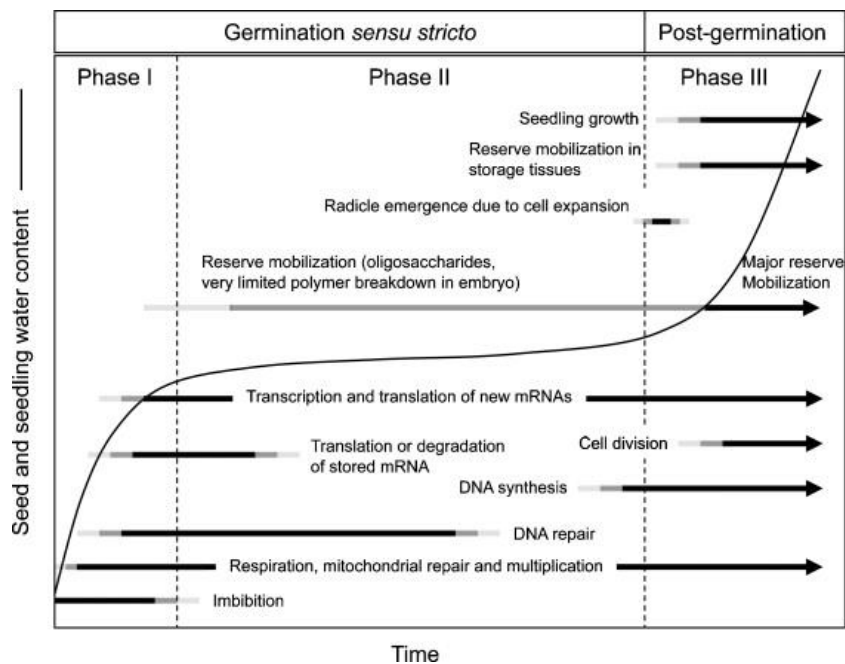


Figure 1.8 Time course of physical and metabolic events occurring during germination (Phases I and II) and early seedling growth (Phase III). The time taken for these events to occur varies between species and is influenced by germination conditions. The curve shows a stylized time course of water uptake (Adapted from <https://sciencedirect.com/science/article/abs/pii/S0168945210000403>, modified by Nonogaki, Chen, & Bradford, 2008; Bewley, 1997)

1.1.10 Thousand seed weight

Seed weight is usually signified as thousand seed weight (TSW) in the seed industry. TSW is a component of seed yield but more importantly it has an impact on seedling vigour and growth that indirectly affects the seed yield, and is important for evaluating crop variety breeding (Botwright, Condon, Rebetzke, & Richards, 2002). It is an important seed measurement indicator in crop research (Afshari, Eftekhari, Faraji, Ebadi, & Ghanbarimalidareh, 2011; Li, Thomson, & McCouch, 2004). Low TSW is an indicator of poor seed quality and is a result of poor seed filling and unfavourable environmental conditions during crop growth (Deivasigamani & Swaminathan, 2018). TSW varies with the size of the embryo and reserved nutrients (Ebadi & Hisoriev, 2011; Cao, Zhang, Chen, Wu, & Cui, 2011). Seed weight is affected by assimilate availability, water availability and

temperature (Brooks, Jenner, & Aspinall, 1982; Blum, 1998; Castro, Hodar, & Gomez, 2006). Genetics have been reported to be the main cause of variations in seed weight in maize (Reddy & Daynard, 1983). However, high temperature (Hampton et al., 2013) and low temperature (Shim, Lee, Koo, Shin, & Yoon, 2019) can reduce seed weight by affecting the seed filling period. At higher temperature, low seed weight is due to a shorter seed filling duration and at low temperature, it is due to smaller volume of assimilates available for seed filling (Deivasigamani & Swaminathan, 2018).

1.1.11 Seed dormancy

A seed is dormant when germination does not proceed even though all the conditions required for germination are available. It is governed by both factors inside the seed and external to it (Cabej, 2019). The phenomenon of dormancy is quite common for seeds even under favourable conditions (Graeber, Nakabayashi, Miatton, Leubner-Metzger, & Soppe, 2012). Baskin and Baskin (2014) recognised seven kinds of seed dormancy, based on the earlier classification by Nikolaeva (1969), whose system relied on the grounds of dormancy and the conditions required for breaking it. These are i) Morphological dormancy (MD); freshly matured embryos underdeveloped upon dispersal and require some time to continue development before they can germinate (the dormancy period); ii) Physiological dormancy (PD) broken through specific physiological responses to environmental cues, such as warm and/or cold temperatures or through dry after-ripening; iii) Physical dormancy (PYD), seeds are surrounded by a water-impermeable palisade layer, and dormancy is broken by physically making this layer water-permeable; iv) Morpho physiological dormancy (MPD); a combination of MD and PD; and v) Physio physical dormancy (PYPD); a combination of PY and PD (Nikolaeva, 1969, 1999; Baskin & Baskin, 2004a, 2014); vi) ND, when there is no dormancy on freshly matured seeds (Nikolaeva, 1969), and vii) dust seeds (DUST), which are small in size (mostly ≤ 1.0 mm in length) and have undifferentiated embryos with as few as two to three cells (Leake, 1994; Eriksson & Kainulainen, 2011). Non-dormant seeds germinate under the widest range of conditions possible immediately after dispersal without any dormancy-breaking treatments (Baskin & Baskin, 2004; 2014).

Villiers and Wareing (1964) suggested that chemical promoters and inhibitors inside the embryo are associated with dormancy and breaking of dormancy. The inhibitory substance present in the embryo is abscisic acid (ABA). Growth promoter Gibberellic acid (GA), are synthesised over time in response to environmental factors. It has been proposed that two kinds of dormancy exist in dormant warm season grasses. The first is dormancy based within structures external to the embryo, and the second is the mechanism based within the embryo. GA may be involved in the release of dormancy through its growth promoting activity on hydrolytic and proteolytic enzymes

that act to mobilise the food reserves in the cotyledons or endosperm. In warm season grasses, although the endogenous balances between germination promoters and inhibitors have not been studied, it has been reported that GA application in grass seed overcomes dormancy (Adkins, Bellairs, & Loch 2002).

Finch-Savage and Leubner-Metzger (2006) broadly divided dormancy into distinct dormancy 'classes' through a wide range of different physiological and structural mechanisms. They noted that the prevalence of dormancy can differ among habitats, climatic zones, and taxonomic lineages. Seed dormancy is prevalent in major grass species being grazed by animals around the world. Grasses from temperate, sub-tropical and tropical climates can exhibit seed dormancy although it has been said that seed dormancy in grasses is general rather than exceptional (Simpson, 1990). In some species of warm season grasses, at the time of shedding/shattering the embryo is not completely developed and sometime is required to allow the full development of the embryo before it can germinate. Embryo dormancy is a mechanism for many temperate grass species but, there are very few in-depth studies for embryo dormancy in warm season grass species (Simpson, 1990). In today's maize cultivars, dormancy does not occur (McCarty, 1995; Avendano Lopez et al., 2011). However, Avendano et al. (2011) reported that dormancy exists in more than 90 % of teosinte populations. Experiment conducted by Mondrus-Engle (1981) showed that teosinte seeds were still viable 3-7 years after harvesting, and the germination was higher than for freshly harvested seeds. GA pre-treatment was found to be the most effective among all other methods in breaking dormancy where germination was enhanced to 95 % when treated with 1000 ppm GA. Absence of natural gibberellins in new seeds of tetraploid teosinte was suggested as the cause of this dormancy (McDonough, 1977; Mayer & Poljakoff- Mayber, 1975; Mondrus-Engle, 1981). Osborne (1965) and Mondrus-Engle (1981) found that GA induces seed germination by depressing genes that stimulate enzyme synthesis.

1.1.12 Sowing date

Optimum sowing date is an important plant production practice to allow the best utilization of weather components in plant growth and development (Choi et al.,2017; Abbas et al., 2019). Changes in sowing dates can change the crop growth rate (Hussain et al., 2016; Abbas et al. ,2019). Both early and delayed sowing are detrimental for crop growth and production as leaf area development is low for early sowings because the intercepted photosynthetically active radiation (PAR) is lower, while in delayed sowings higher minimum and maximum temperature also reduces PAR thus reducing herbage yield (Liu et al., 2013; Tariq et al., 2018). Yield reductions due to early or late planting have been well documented in the literature (Abendroth, Woli, Myers, & Elmore,

2017, Lauer, 1999 and Nafziger, 1994). Early sowing has been reported to enhance reproductive growth, while delayed sowing has been reported to favour vegetative growth (Cirilo & Andrade, 1994). Similarly, sowing date also affects seed yield and quality. Seed yield increases with early planting but decreases with the delay in planting date (Dahmardeh, 2012). Killi and Altanbay (2005) reported a significant variation in seed weight under different sowing dates, because of a longer growth duration due to lower temperature during early growth phases while plants planted late had a shorter growth period and lower seed weight. Variation in sowing date may lead to high temperature during seed development and maturation, which can reduce the storage compounds in seeds leading to low germination (Dornbos & McDonald, 1986) and also may cause physiological damage (Coolbear, 1995; Powell, 2006).

1.1.13 Seed rate

Plant density is an important management practice for seed yield, and other significant agronomic characteristics. Plant density influences the growth and development pattern of the plant. A high population increases interplant competition for light, water and nutrients, which may encourage apical dominance leading to decreases in the number of ears produced per plant and seed set per ear thus reducing the final yield (Sangoi, 2001; Ali, Khalil, & Raza, 2003; Abuzar, et al., 2011). According to Gonzalo, Vyn, Holland, and McIntyre (2006) plant density responses are not defined by a single factor, rather it is the combination of various agronomic and environmental factors such as soil fertility, moisture supply, genotype, planting date, planting pattern, plant population and harvest time of all which can have a significant effect on seed yield, plant height, number of seeds per ear, number of cobs per plant, cob length, ear diameter and stem diameter. However, genetic predisposition is associated with higher plant populations of maize hybrids (regardless of the reproductive plasticity) to cope with the various environmental conditions and stresses associated with production of extra yield (Fromme, Spivey, & Grichar, 2019). At higher plant density, grain yield per plant is decreased (Luque, Cirilo, & Otegui, 2006) in response to decreasing light and other environmental resources available to each plant (Ali et al., 2003). Although affected by various biotic and abiotic factors, among all the species of grass family, maize yield is most affected by plant density (Vega et al., 2001). The dry matter yield of maize plant shows an asymptotic response to plant density (Bunting, 1971), but the grain yield response is parabolic (Tollenaar, 1989). As a result, the optimum plant density for forage production is higher than that for seed production. Abuzar et al. (2011) revealed that the leaf area index showed a linear response with increased planting density while plant height had an inverse relation with the density of plants.

1.1.14 Cutting management

Growth of leaves after cutting is restricted to their basal part: extension of leaves is a result of cell division, cell elongation, and cell maturation (Allard & Nelson, 1991). The exposure of the first leaf tissue above the cutting height is because of the expansion of cells located at the bottom of the growing leaves which already existed at the time of cutting which merges from the whorl of the older sheaths (Wilhelm & Nelson, 1978). Cutting reduces plant leaf number which is directly related to the photosynthetic activity of the plant. Therefore the leaf growth after herbage cutting uses either pre- defoliation reserves or post-defoliation assimilates (Alberda, 1957; Danckwerts & Gordon, 1987; Gonzalez, Boucaud, Salette, Langlois, & Duyme, 1989).

Hartt, Korschark, and Burr (1964) and Ryle and Powell (1975) reported that defoliation or cutting the herbage immediately reduces the supply of assimilate to the roots because of higher carbon allocation to the shoot meristems and also due to the reduced assimilates as a result of lower leaves. The quality and quantity of regrowth's is determined by the height at which the stems are cut because energy for the regrowth is supplied by the assimilate synthesized by the remaining photosynthetic tissue and non-structural carbohydrates stored in the lower stem (Morvan-Bertrand, Boucaud, & Prudhomme, 1999; Donaghy, Turner, & Adamczewski, 2008). However harvesting the plant at a lower height will remove many of the photosynthetic tissues which reduces the energy required for the regrowth (Ong, Marshall, & Saoar, 1978).

Temperature influences the energy status of regrowing grasses following defoliation because temperatures above optimum lower the photosynthetic activity because of reduced activity of Rubisco enzyme due to lower Photosystem II reactions and ATP synthesis on the thylakoid membrane (Havaux, 1996; Bukhov, Wiese, Neimanis, & Heber, 1999; Allakhverdiev et al., 2008). Since respiration is high at higher temperature (Yamori, Hikosaka, & Way, 2014), carbohydrate consumption can be higher than that being produced from photosynthesis (Brown & Blaser, 1970; Youngner & Nudge, 1976; Slack, Fulkerson, & Scott, 2000).

1.1.15 Diversity

Both traditional and modern agricultural systems utilize agrobiodiversity. Both farmers and breeders together promote biodiversity by recognizing traditional and improved crop varieties adapted to diverse environments (Brauner et al., 2019) and biotic and abiotic stresses (Akem, Ceccarelli, Erskine, & Lenne, 2000; Dwivedi et al., 2016). Agro biodiversity is conserved either by ex situ (i.e., in genebanks) or in situ (i.e., in farmers' fields) conservation practices that signify harmonize efforts to protect the biodiversity. Phenotypic identification is the primary step in documenting and classifying germplasm. It is easy to record, cost effective and dependable for

estimating heritability (Govindaraj, Vetriventhan, & Srinivasan, 2015) in developing countries, where cheap labour is readily available. Phenotypic assessments are good indicators to analyse diversity in many cereal crops, including maize (Salazar, Correa, Araya, Mendez, & Carrasco 2017; Sattler et al., 2018; Tiwari, Tripathi, Khatri, & Bastola, 2019). Genetic diversity is the basis for crop improvement, and it plays a significant role in breeding programs (Ali et al., 2008). Maize is gifted with amazing diversity, and its genome harbours a huge phenotypic and molecular diversity as a result of prolonged selection (Buckler, Gaut, & McMullen, 2006; Matsuoka et al., 2002). Maize has a molecular diversity five times higher than other domesticated crops (Ali et al., 2008; Whitt, Wilson, Tenaillon, Gaut, & Buckler, 2002) and is a model crop for cereals (Prasanna, 2012). Mexican farmers have harnessed maize diversity through management decisions that take into account a complex interplay between environmental and cultural factors (Orozco-Ramirez, Ross-Ibarra, Santacruz-Varela, & Brush, 2016; Pressoir & Berthaud, 2004). At present, native Mexican maize landraces represent 59 of the 219 maize races designated and characterized in Latin America (Sanchez, Goodman, & Stuber, 2000), forming two of the four main diversity groups identified among New World maize populations (Vigouroux et al., 2008). Teosinte is the wild progenitor of maize and has greater genetic diversity than maize inbreds and landraces (Karn, 2017).

1.1.16 Economics

Economics play a main role in every business. In current agriculture they lead to narrowing the mixture of crops grown, to a simple cropping pattern, and an increase in the area under crops where minimum technology can be used to achieve reasonable yields. The return rate of grown crops is influenced by the achieved yield, production costs, and the sale price of the product (Homolka & Bubenikova, 2013). Yields and costs are of primary importance when making an economic decision for growing a crop. Cost of cultivation of any crop is the sum total of several components. Variable costs vary directly with the production. Variable costs may be either cash costs or non-cash costs. Cost of seed, farm yard manure, fertiliser, plant protection measures, hired irrigation, hired human labour / machine labour etc. are cash costs. Unpaid family labour, machine labour (own) and interest on working capital are considered as a non-cash cost with the assumption that alternative employment opportunities were available to the labour force. Fixed costs are the rental value of land, depreciation of implements, interest on fixed capital, land revenue etc. (Shah, Makwana, Sharma, & Vidyanagar, 2011). Profit is derived in terms of gross return. Gross margin is calculated by the difference between gross return and total variable cost. The following equation is used to calculate gross margin (GM).

$$GM = GR - \Sigma Cv$$

Where,

GR = Gross return (NRs/ha); and

ΣC_v = Total variable cost (NRs/ha).

1.2 Research Objectives

The main objective of this study was to examine seed yield potential of teosinte along with seed quality. However, herbage yield was taken from the crop that is grown for seed because Nepalese farmers are used to taking a herbage harvest once or twice from the same crop grown for seed. Apart from this, a diversity study, the climatic influences on herbage and seed development and an economic analysis were also conducted involving the major effect of sowing date, seed rate and cutting management. There is negligible published information on seed yield and quality of teosinte. This research aimed to study the seed production potential of teosinte in the central plains of Nepal which was done through the following six objectives:

The objectives of this research were to:

- Determine the effect of sowing date, seed rate, and cutting management on herbage yield of teosinte.
- Assess the influence of the environment on growth and yield of teosinte.
- Identify the appropriate sowing date and seed rate for maximizing seed production of teosinte under different cutting regimes in Nepal.
- Assess the effect of sowing date, seed rate and cutting management on seed quality of teosinte harvested on the basis of cob positions.
- Evaluate different genotypes of teosinte in Nepal for herbage and seed production.
- Conduct an economic analysis of teosinte cultivation for herbage and seed production.

Chapter 2

Effect of sowing dates, seed rates, and cutting management on herbage yield of Teosinte

2.1 Introduction

The economic sustainability of the livestock industry in Nepal is basically dependent on the breed, feed and health management of the herd, where green herbage plays a vital role. However, the situation of herbage production in the country is not encouraging. Profitable livestock production requires sources of fresh quality feed and herbage, because the contribution of feed cost in this business amounts to 65-70% of the total cost of the production (Premy, Pandey, & Pudasaini, 2016). Thus, scanty herbage availability with the associated poor nutrition, health and fertility, are the biggest constraints to milk production and a major weakness to improving rural livelihoods (Armstrong et al., 2011). However, the country has reduced the deficit feed balance (TDN) at national level from 31% in 1980 to 20% in 2016/17 (Singh & Singh, 2019). One of the ways to improve livestock productivity is through increasing the production of green herbage, but this is hindered due to a shortage in supply of quality seeds and planting materials in Nepal (Sanjyal, Shrestha, Upreti, & Pandey, 2016). Nepalese crop production is low for herbage crops, which limits the availability of feed, and poses a serious threat to the expansion of commercial dairy enterprises (Paudel, Pokharel, & Shrestha, 2019). Therefore, high quality herbage aided by development of best agronomic management production practices is required to better provide the nutritional requirement of dairy animals (Radzi & Droege, 2014).

Teosinte, a tall and vigorously growing tropical crop produces high-quality herbage from multi-cuts in mid to late summer when cool-season perennials have low production. It is appreciated for its profuse tillering and good biomass production, especially during the dry summer season in Nepal (Sharma, 2018). It is an important livestock feed, often fed fresh in a cut and carry system, or used to produce silage or hay during wet summers when an adequate supply of teosinte is available. It has a good capacity to resist biotic and abiotic stresses (Warburton et al., 2011; Wang, Wang, Yuan, & Xu, 2004). Because it has a higher leaf ratio in the total biomass than maize, teosinte has a higher digestibility (Baumont, Prache, Meuret, & Morand-Fehr, 2000). It can tolerate acidic as well as waterlogged conditions whereas other forage crops like maize and sorghum cannot survive in these conditions. Its versatile nature allows it to be grown under an intensive herbage production system

for good biomass production (Mohan, Dar, & Singh, 2017). Teosinte is popular within Nepal, but also has a good scope in neighbouring countries as well (Hampton & Armstrong, 2019).

Teosinte can tolerate a very high temperature (36-45°C) and is able to produce larger amounts of fresh herbage than maize (27% and 55% higher yield than maize) under non-stressed and stress conditions respectively (Niazi, Rauf, Silva, & Munir, 2015). As for many other agrarian communities in the world with hot summers, and where dairying is one of the major sources of economic gains, teosinte has become a valuable herbage source for dairy cattle, leading to improved food security (Silva, Vidal, Costa, Vaio, & Ogliari, 2015). It is considered to be better than maize because of its higher nutrient content and easier agronomic management (Kundu, Hedayetullah, Bera, Biswas, & Chatterjee, 2015).

Teosinte in India is used for herbage but also used commonly as a silage crop (NDDDB, 2012). Teosinte is also grown as a herbage crop in several USA southern states (Louisiana, Mississippi, Georgia, Florida, Texas and Kansas) (Vasey, 1887; Lamson-Scribner, 1899). Similarly, it is popularly called “Guatemalan teosinte” in the South of Brazil. Teosinte which is called “Rayana” in Egypt, contributes about 20% of total herbage production in that country, providing quality herbage during the summer hot periods (Niazi et al., 2015).

Teosinte is one of the most popular summer herbage crops in Nepal as well. Pasture and Forage Division (2016) released a cultivar of teosinte locally known as “*Makaichari*” which was developed through mass selection from the improved variety *Sirsa*. It produces 35-45 tonnes green herbage per hectare and 1-1.5 tonnes of seed per hectare. Teosinte has a high metabolizable energy and a crude protein of 13.7 % (Devkota, Pokharel, Paudel, Upreti, & Joshi, 2015; Osti, Bhandary, Shrestha, & Pradhan, 2001; Upreti & Shrestha, 2006). Though teosinte is a tropical crop, its cultivation can be extended up to an elevation of 1500 meters above sea level (Hampton & Armstrong, 2019). It has the highest herbage production potential among all the summer herbage species grown in the sub-tropical regions of Nepal under a cut and carry system. As compared to other cereal herbage crops grown in the summer season, teosinte stays green for a longer period of time, ensuring an extended availability of green herbage for livestock during periods of herbage scarcity (ICAR, 2011). The area under teosinte cultivation in Nepal is 27,232 ha and the seed production is 870 tonnes. However, there is still a seed deficit of 220 tonnes per annum, which is 25% of total seed requirement in the country (NPAFC, 2018).

Realizing the value of this crop, teosinte was one of the important component of summer herbage crops for the Nepal Government’s ‘Forage Mission’ Program, which was implemented in 49 dairy

pocket districts of Nepal to mitigate the scarcity of green herbage for dairy animals (Devkota et al., 2015) during 2013 to 2016.

2.1.1 Effect of sowing date on herbage yield

Sowing date can impact crop productivity worldwide (Sieling, Bottcher, & Kage, 2017; Aiken, Baltensperger, Krall, Pavlista, & Johnson, 2015). It is one of the primary agronomic management practices for farmers because it primarily influences crop productivity as determined by temperature during growth, heat units, phenological events and physiological maturity (Ahmad et al., 2017; Pavlista, Isbell, Baltensperger, & Hergert, 2011). Sowing date has a great impact on the herbage production because it regulates the interaction between growth, development and any stress during the growth period of herbage (Gururanjan, 1993).

Among the different agronomic practices, sowing date is one of the most important factors influencing teosinte herbage production because optimal sowing date is important to determine yield, especially under rain fed conditions. Average teosinte herbage yield is quite low in Nepal. Thus, alteration in sowing date modifies the radiative and thermal conditions during growth and the proportion of incident light intercepted by the crop which directly determines crop growth rate and consequently the time during which incident radiation can be intercepted (Muchow, Sinclair, & Bennett, 1990; Tollenaar & Bruulsema, 1988) to improve the herbage yield. Information on the optimum sowing date helps to regulate the best time required for germination, crop establishment, canopy development, etc. (Bussmann, Eladib, Fayyad, & Ribbe, 2016). Delay in sowing after the optimal date consistently reduces yield because it reduces individual plant growth and tiller production (Darwinkel, Ten Hag, & Kuizenga, 1977; Fielder, 1998; Gooding & Davies, 1997).

2.1.2 Effect of seed rate on herbage yield

Population density, as influenced by seed rate is an important factor influencing yield (Meyer, 1970). Competition for space and nutrition among plants increases as distance between plants decreases (Duncan, 1984). Olson and Sander (1988) reported that for maize grain and herbage production, higher plant densities favoured herbage rather than seed yield.

Good plant spacing gives the right plant density, which is the number of plants required on a given unit of land for optimum yield (Vafias, Goulas, Lolas, & Ipsilandis, 2007). Seed rates influence the interplant competition for space, light, water and nutrients (Malik, Ahmad, & Hussain, 2009). Lower seed rate decreases the interplant competition during vegetative growth (Ozturk, Caglar, & Bulut, 2006). Previous studies have revealed increasing maize plant density lead to increased yield until an optimum density is reached (Ipsilandis & Vafias, 2005).

Optimum seed rate changes with the accessibility of environmental resources (Arduini, Masoni, Ecoli, & Mariotti, 2006; Gooding, Pinyosinwat, & Ellis, 2002), as a dense planting does not always have a positive effect on the yield due to the influence of seed rate on inter-plant competition (Holliday, 1960; Park, Benjamin, & Watkinson, 2003), pathogens, soil moisture, and nitrogen availability (Fischer, Aguilar, Maurer, & Rivas, 1976; Read & Warder, 1982). Increased Herbage yields (HY) and Dry Matter Yields (DMY) under increased planting density of forage maize has been reported by Mandic et al. (2015). Plant density exerts a strong influence on maize growth because of its competitive effect both on vegetative and reproductive development (Onasanya et al., 2009; Singh & Choudhary, 2008). Similarly, a positive influence of seed rate on the herbage yield of maize was observed in research conducted by Anders et al. (2020) who found two varieties of maize produced the highest herbage yield at the highest seed rate.

2.1.3 Effect of cutting management on herbage yield

Time of cutting intervals and frequency of cutting is a very important agronomic practice (Kumar, Channakeshava, Belavadi, Shivprakash, & Siddaraju, 2017) as it impacts on quality and quantity of herbage crops. Cutting management also plays a crucial role in the recovery of the pasture quality. Cutting permits the regeneration of crops, and an ability to withstand cutting is a desirable character for successful herbage crop production (Khair, 1999) as it reduces the cost of production in terms of land preparation and seed cost (Mohamed & Khair, 2011). Time of first cutting is very critical, as it governs the number of cuts and green herbage yield at each cutting (Kallenbach, Nelson, & Coutts, 2002; Kumar, et al., 2017). Cutting allows the production of new leaves during the regrowth. These new leaves have improved quality, expressed in the form of gross protein, allowing greater acceptance and better digestion of the plant (Schneider, Caron, Elli, Schwerz, & Engroff, 2019). In herbage crops with a high fibre content, the high concentration of cell wall constituents makes a low quality herbage, especially for tall growing plants, resulting in lower digestibility (Pinho, Vasconcelos, Borges, & Rezende, 2006). The effect of cutting frequency on quantity and quality of crop (legumes and non-legumes) depends on the seasons of the year and on the species of herbage crop (Enoh, Kijora, Peters, & Yonkeu, 2005; Njarui & Wandera, 2004). Teosinte has a good regeneration capacity; cutting has a positive effect on the herbage yield because it encourages the production of more tillers. Therefore, cutting practice influences the total herbage yield. However, inappropriate cutting may result in lower yield.

The major constraint in teosinte cultivation in Nepal is the lack of production technology. Very little work has been done regarding the impacts of sowing date, planting density as determined by seed rate, and cutting management for teosinte production in Nepal or abroad. Despite its popularity,

there is a scarcity of information regarding the optimum planting time to maximize herbage yield under different seed rates and cutting management. This is a serious concern for teosinte cultivation and its expansion in use across the Terai and inner-Terai regions of Nepal.

2.2 Objective

The ultimate objective of the experiments and treatments described in this chapter was to determine the effect of sowing date, seed rate and cutting management on teosinte seed production, the results of which are presented in Chapter 4. However, Nepalese farmers are accustomed to taking at least one, and sometimes two forage cuts from a teosinte seed crop, the first at around 45 days after sowing (DAS) and the second at around 75 DAS. This chapter is therefore not a study of the full forage potential of teosinte in response to sowing date, seed rate and cutting management. The objective was to determine the effects of these factors on teosinte herbage yield within a crop being managed for seed production.

2.3 Methodology

2.3.1 Site description

Experiments were carried out from March 30 to September 15, in 2017 and March 30 to August 30, in 2018 at the forage experimental plot of the National Cattle Research Program (NCRP), Nepal Agricultural Research Council (NARC), Bharatpur Metropolitan 15, Rampur, Chitwan, Nepal. The farm is situated in the central region of Nepal at 27°40' N Latitude and 84°35' E longitude with an altitude of 228 m above mean sea level.

2.3.2 Description of soil properties

A soil sample was taken on February 15, 2017 and sent for testing at the laboratory of the Agriculture Technology Center, Kathmandu. Similarly, in 2018, a soil sample was taken and sent for testing to the laboratory of the National Soil Research Center (NSRC), NARC, Lalitpur. The soil was slightly acidic, light textured sandy loam. The samples were sent to two different laboratories because of a high number of soil samples at NSRC laboratory in 2017. The details of N, P, K and OM available in the soil of the experimental field are presented in Table 2.1a and Table 2.1b.

Table 2.1 a Soil analysis report of the experimental field at NCRP, Rampur, Chitwan, 2017

Soil	pH	Total N %	Available P ₂ O ₅ kg ha ⁻¹	Available K ₂ O kg ha ⁻¹	OM%
Depth of soil					
Top soil (0-5 cm)	6.90	0.12	51	300	2.50
Sub soil (20cm-5cm)	6.70	0.15	31	261	3.15

Note: N% - Nitrogen percentage, P₂O₅ - Phosphorus, K₂O - Potassium, kg ha⁻¹ - kilogram per hectare, OM% - Organic Matter percentage

Table 2.1 b Soil analysis report of the experimental field at NCRP, Rampur, Chitwan, 2018

Soil	pH	Total N %	Available P ₂ O ₅ kg ha ⁻¹	Available K ₂ O kg ha ⁻¹	OM%
Depth of soil					
Top soil (0cm-5cm)	6.70	0.15	35	277	2.78
Sub soil (20cm-25cm)	6.69	0.16	25	255	3.23

Note: N%- Nitrogen percentage, P₂O₅ - Phosphorus, K₂O - Potassium, kg ha⁻¹- kilogram per hectare, OM% - Organic Matter percentage

2.3.3 Climatological variation during the study

Daily weather data (precipitation, maximum, minimum, mean temperature, and humidity) were obtained from the agro meteorological station installed at National Maize Research Programme (NMRP), NARC, Rampur which was 200 meters away from the research field. The experimental site during the two years had a humid and subtropical climate with a cool winter (7.8-21.4°C) and hot summer (22.1-33.8°C). The annual average rainfall was 1453 mm with a distinct monsoon period (>75% of annual rainfall) from mid-June to mid-September (Figure 2.1a and Figure 2.1b) (NMRP, 2017; NMRP, 2018).

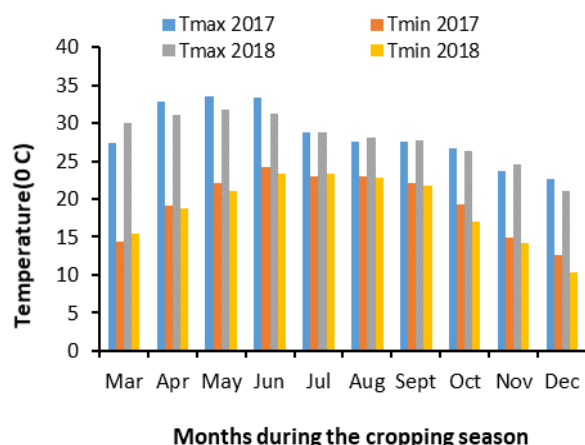


Figure 2.1a Minimum and maximum monthly temperature during 2017-2018

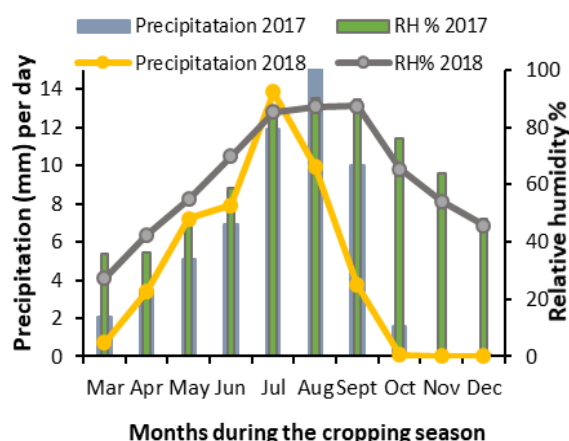


Figure 2.1b Average monthly precipitation and relative humidity during 2017-2018

Data Source: NMRP, 2017; NMRP, 2018

2.3.4 Cropping history

The three years of cropping history in the experimental field is presented in Table 2.2.

Table 2.2 Cropping history of the experimental field for three years from 2014/15 to 2016/17

Year	Crops
2014-15	Oat, Teosinte, Sorghum, Millet, Vetch
2015-16	Oat, Teosinte, Sorghum, Millet, Vetch, Cabbage, Cauliflower
2016-17	Teosinte, Oat

Source: NCRP, 2017

2.3.5 Land preparation and experimental lay-out

The experimental plot was ploughed twice and a total of 192 experimental plots were laid out for the first year and 144 experimental plots for the second year, with each plot being 3.2x2 square meter. The fertilizers Urea, Diammonium Phosphate (DAP), and Muriate of Potash (MoP) were applied at the rate of 120:60:40 kg per hectare. Half the amount of nitrogen along with all the phosphate and potash were applied as a basal dressing at the time of field preparation and the remaining nitrogen was applied as a top dressing just before flowering. For herbage purposes, farmers would prefer to have a higher plant density, and thus a row to row spacing of 40 cm and plant to plant spacing ranging from 10 cm to 2.5 cm was maintained to produce different plant densities.

2.3.6 Varietal selection

The *Sirsa* variety of teosinte, popular both in Nepal and India was used for the experiments. This is the only certified variety of teosinte grown in Nepal. It is mostly a self-pollinated (90%) crop (Pariyar & Shrestha, 2016).

2.3.7 Experimental design

This experiment used a Split-Split Plot Design with four sowing dates, four seed rates, and 0, 1 and 2 herbage cuts, all replicated four times. Sowing dates were main plots, seed rate sub plots, and cutting management sub-sub plots which were randomised in the sowing plots. The sowing plots were randomised within the field. The first sowing was done on March 30, 2017 followed by three consecutive sowings at 30-day intervals in 2017. Based on the performance of teosinte in the June sowing of 2017, in 2018 the June sowing was omitted, and only three sowing dates were used (March 30, April 30 and May 30). The seed rates used for each sowing were 20, 40, 60 and 80 kg seed per hectare. These were arranged under three cutting managements, with no cut, one cut at 45 DAS and a second cut at 75 DAS for each seed rate. The seeds were sown manually in rows at a depth of 5 cm. The seeds were planted untreated. The seed rate used and the density maintained in each sowing and seed rate is given in Table 2.3.

Table 2.3 Seed rate and plant density for each sowing

Seed rate (kg ha ⁻¹)	Weight of seed for each plot (g)	No of seeds in each plot (g)	Spacing between each row (cm)	Spacing between plants (cm)	No of plants in a row of 2 meters length	Plant population per hectare
20	12.8	158	40	10	20	222,222
40	25.6	316	40	5	40	444,444
60	38.4	474	40	3.4	59	666,667
80	51.2	632	40	2.5	79	888,889

Note: 1000 seed weight - 90 grams; germination - 90%

2.3.8 Inter-cultural operations

Flood irrigation was done on the seventh day after the first sowing but for the other three sowings in both years no irrigation was required as it rained regularly after sowing was done (Figure 2.1 and 2.2). Weeding was done manually every 10 days, as the weed *Cyperus rotundus* (Nut grass) was a serious problem in the research field in both years throughout the earlier stage of plant growth. However, during the later stage, the weeds were suppressed by teosinte. *Cyperus rotundus*, coined

as the world's worst weed, has a wide distribution in 90 countries (Holm, Plucknett, Pancho, & Herberger, 1977). Its presence in a field reduces crop yield significantly as it releases a chemical which is harmful to other crops and also it exhibits strong competition with the crops for ground surface area (Darmanti, Santosa, Dewi, & Nugroho, 2015).

2.3.9 Plant selection, tagging and data recording

Ten plants from the central row of each plot were randomly selected and tagged to record Herbage Yield (HY) and Dry Matter Yield (DMY) at 45 and 75 days after sowing (DAS). At 45 DAS, all plants in both cutting treatments including the 10 tagged plants were cut at a height of 15 cm above ground to record herbage yield. After harvesting plants at 45 DAS, the plants were allowed to regenerate for the two cut treatments, 10 plants from each sowing were tagged again. At 75 DAS, after recording the data, the whole plot was harvested to record the herbage yield at 75 DAS. The data recorded from the 10 tagged plants were:

Plant height

Plant height of the ten randomly selected tagged plants in the central row of each experimental plot was recorded in centimetres from ground level to the tip of the fully opened leaves at both the first and second vegetative harvest.

Number of leaves per plant

The total number of leaves was recorded at both 45 and 75 DAS before harvesting for herbage from the tagged ten plants in the central row.

Leaf Area Index (LAI)

The length and breadth of a mature leaf that was fully opened and physiologically active was measured. The length was taken from the base of the lemma and the width from the centre of the leaf for 10 selected plants at both 45 and 75 DAS. Based on the above information LAI was calculated for each combination of sowing date, sowing rate and cutting management using the following formula as suggested by Pal and Murari (1985); and Tanko and Hassan (2016). The leaf area was calculated by using the formula:

$LA = L \times W \times K$, Where,

LA: Leaf area (cm²)

L: Length of leaf (cm)

W: Width of leaf (cm)

K: Factor (0.75)

$LAI = \text{Leaf area (LA) (cm}^2\text{) / Land area per plant (sq. cm.)}$

Number of tillers per plant

The number of tillers per plant was recorded at the first and second harvests by counting all the tillers of each of the 10 tagged plants separately at 45 days and 75 DAS and then averaging the value of the 10 plants.

Herbage yield / Dry matter yield (kg ha⁻¹)

The first herbage harvest was taken at 45 DAS by cutting the whole plot 15 cm above the ground surface and the second cutting was also taken similarly at 75 DAS. Herbage mass was weighed to record green matter and then placed in a hot air oven for 72 h at 65°C until a constant weight was obtained to record the dry matter for each harvest. Dry matter percentage was calculated by subtracting the dry weight from the fresh weight and dividing by the fresh weight multiplied by 100 (Devkota et al., 2015). The dry matter yield was calculated by using the formula:

$$\text{DMY (kg ha}^{-1}\text{)} = \text{Herbage yield (kg ha}^{-1}\text{)} \times \text{DM (\%)} \text{ content}$$

2.3.10 Statistical analysis

Analyses of Variance (ANOVA) to determine the effect of sowing date, sowing rate and cutting management on herbage yield and associated yield components of teosinte and their interactions among the factors were performed using General Linear Model (GLM) of Genstat 19th Edition (VSN International, 2019). Accordingly, replication was considered as the block, whole plots as sowing plot, seed rate as a sub-plot and cutting management as a sub-sub-plot factor, respectively. For presenting the significance of different factors on yield and its attributing characters, $P < 0.05$, $P < 0.01$, and $P < 0.001$ were used for 5, 1, and 0.1 percent level of significance respectively. Significantly different means of each level of factors considered were compared using Fisher's unprotected test of least significant difference (LSD). The relationship between the sowing date, seed rate and cutting managements with the yield components was determined by regression analysis.

2.4 Results

One of the objectives of this study was to generate information on optimum sowing date, seed rate and cutting management to get maximum herbage productivity per unit area from a seed crop. Herbage and dry matter yield (kg ha⁻¹) is a function of various growth and yield attributing parameters like plant height, number of leaves plant per plant, Leaf Area Index (LAI), and number of tillers plant per plant, and their results and discussion are presented hereunder. Similarly, the correlation and regression analyses between dry matter with yield attributing parameters are also presented in this section.

2.4.1 Effect of sowing dates, seed rates and cutting management on plant height (cm)

Sowing date

There was no significant difference among the sowing dates for plant height at 45 DAS in both years ($P>0.001$)(Table 2.4). During 2017, plant height at 75 DAS was significantly higher for the April sowing than the June sowing (Table 2.4). In contrast plant height did not vary among the sowing dates in 2018 (Table 2.4). There was a negative correlation between sowing dates and plant height at 45 DAS ($R^2=0.67$) ($P>0.05$) and 75 DAS ($R^2=0.42$) ($P>0.05$) because plant height reduced as the sowing date was delayed (Figure 2.2 and 2.3).

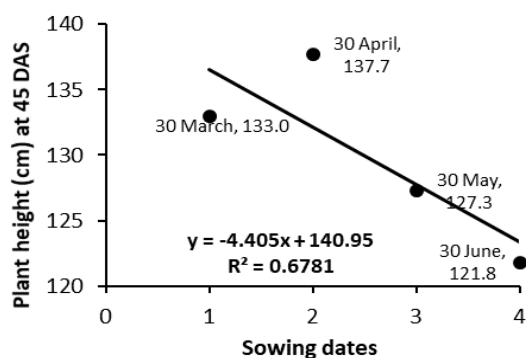


Figure 2.2 Correlation between sowing date and plant height at 45 DAS; data for March, April and May are means for 2017 and 2018

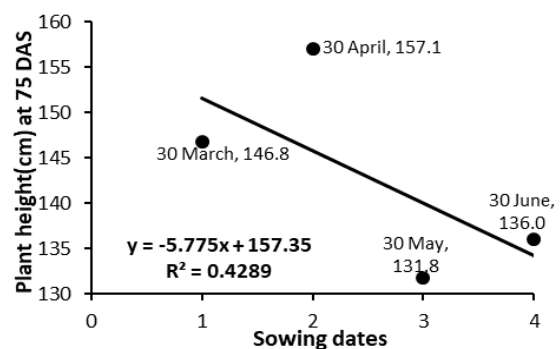


Figure 2.3 Correlation between sowing date and plant height at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

In 2017 plant height for the 40 kg ha⁻¹ seed rate did not vary from the 60 kg ha⁻¹ and 80 kg ha⁻¹ seed rate but it was significantly taller than the 20 kg ha⁻¹ seed rate at 45 DAS. In 2018, the tallest plants recorded at 80 kg ha⁻¹ did not differ from 60 kg ha⁻¹ but varied significantly from 20 kg ha⁻¹ ($P<0.001$) (Table 2.4). At 75 DAS, plant height was greater for the 40 kg ha⁻¹ seed rate than the two higher seed rates in 2017 ($P<0.05$) but in 2018 the lower three seed rates were similar to each other and the lowest plant height was from the 20 kg ha⁻¹ seed rate (Table 2.4). There was a positive correlation between seed rate and plant height at 45 DAS ($R^2=0.61$) ($P>0.05$) because plant height increased as the seed rate was increased (Figure 2.4). At 75 DAS, a significant negative correlation was recorded because plant height decreased as the seed rate was increased ($R^2=0.95$) ($P<0.05$) (Figure 2.5).

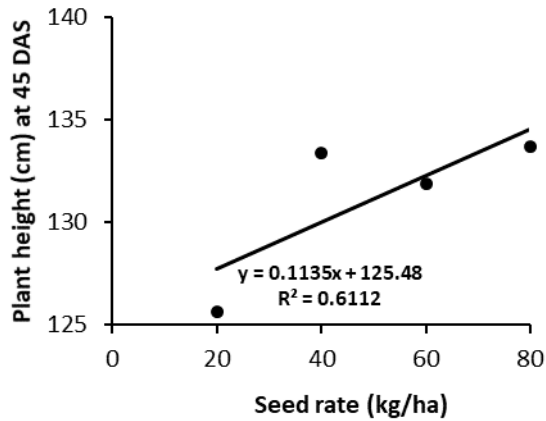


Figure 2.4 Correlation between sowing date and plant height at 45 DAS; data for March, April and May are means for 2017 and 2018

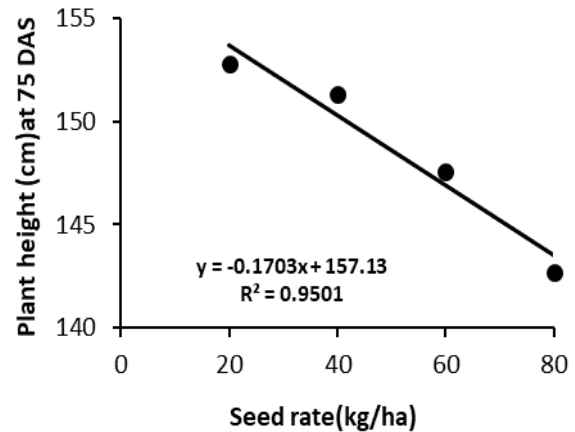


Figure 2.5 Correlation between seed rate and plant height at 75 DAS; data are means for 2017 and 2018

Cutting Management

At 75 DAS, cutting had significantly reduced plant height but there were no differences between the single and double cuts in either year ($P < 0.001$) (Table 2.4). A negative correlation was recorded for plant height under different cutting management at 75 DAS which is because plant height decreased as the cutting frequency increased ($R^2 = 0.79$) (Figure 2.6).

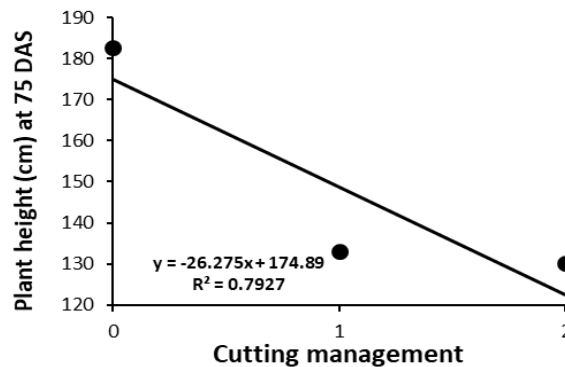


Figure 2.6 Correlation between cutting management and plant height at 75 DAS; data are means for 2017 and 2018

Interactions

In 2017 at 75 DAS, a significant interaction between seed rate and cutting management occurred because cutting reduced the plant height for the 20 kg ha⁻¹ and 40 kg ha⁻¹ seed rates ($P < 0.05$) (Table 2.5). In 2018, there was a significant interaction between sowing date and seed rate at 75 DAS because the lower three seed rates produced the tallest plants for all the sowing dates ($P < 0.05$) (Table 2.6).

Table 2.4 Main effect means of sowing date, seed rate and cutting management on plant height (cm) of teosinte at 45 and 75 DAS in 2017 and 2018 at NCRP, Chitwan, Nepal

Main effect means of:	Plant height - 45 DAS (cm)		Plant height - 75 DAS (cm)	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	136a	130a	145ab	148a
30-Apr	139a	136a	159a	155a
30-May	127a	128a	144ab	150a
30-Jun	122a	-	136b	-
Significance of linear trend (<i>p</i> value)	0.153	0.554	0.209	0.806
LSD (0.05)	25	10	23	19
CV%	12	4	10	7
Seed rate (SR)				
20 kg ha ⁻¹	128b	123c	148ab	158a
40 kg ha ⁻¹	136a	131b	154a	149ab
60 kg ha ⁻¹	130ab	134ab	140b	156a
80 kg ha ⁻¹	129ab	138a	142b	143b
Significance of linear trend (<i>p</i> value)	0.786	<0.001	0.026	0.059
LSD (0.05)	7	7	9	12
CV%	7	6	9	9
Cut management (CM)				
No cut	133a	130a	181a	184a
One cut	131a	134a	132b	135b
Two cut	128a	131a	125b	135b
Significance of linear trend (<i>p</i> value)	0.122	0.806	<0.001	<0.001
LSD (0.05)	7	15	7	11
CV%	15	10	13	18
Significance of interactions of linear contrasts (<i>p</i> value)				
SD (lin) x SR	0.323	0.593	0.228	0.032
SD (lin) x CM	-	-	0.239	0.950
SR (lin) x CM	-	-	0.002	0.621
SD x SR x CM	-	-	0.620	0.582

Note: DAS = Days after sowing; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($p < 0.05$). SD=sowing date; SR=seed rate; CM=cut management; The first cut at 45 DAS occurred after the data was recorded; Data were collected at 45 DAS before the herbage was harvested, therefore there will be no interaction of cutting management at 45 DAS on any vegetative yield components.

Table 2.5 Interaction between seed rates and cutting management for plant height (cm) at 75 DAS in 2017

Seed rates (kg ha ⁻¹)	Cutting management		
	0	1	2
20	185	135	125
40	203	137	123
60	168	126	124
80	169	129	129

LSD (5%) (Comparisons within a sowing date) 14
LSD (5%) (Other comparisons) 13.75

Note: 0 = No cut; 1 = one cut; 2 = two cut

Table 2.6 Interaction between sowing dates and cutting management for plant height (cm) at 75 DAS in 2018

Sowing dates	Seed rates (kg ha ⁻¹)			
	20	40	60	80
30 March	149	144	159	143
30 April	151	157	165	147
30 May	173	145	143	140

LSD (5%) (Comparisons within a sowing date) 20.42
LSD (5%) (Other comparisons) 23.7

2.4.2 Effect of sowing date, seed rate and cutting management on tillers per plant

Sowing date

Sowing date was an important source of variation with respect to tiller number in 2017 and 2018. At 45 DAS, tiller number was greater for the April sowing in both years ($P < 0.001$) (Table 2.7). At 75 DAS, tiller numbers for the first two sowings were significantly higher than for the last two sowings in 2017 ($P < 0.001$) (Table 2.7). In 2018 tillers were highest for the March sowing and the April and the May sowing did not differ with each other ($P < 0.001$) (Table 2.7). There was a negative correlation between sowing dates and tiller numbers at both 45 ($R^2 = 0.69$) ($P > 0.05$) and 75 DAS ($R^2 = 0.53$) ($P > 0.05$) because delayed sowing reduced the tiller numbers (Figure 2.7 and 2.8).

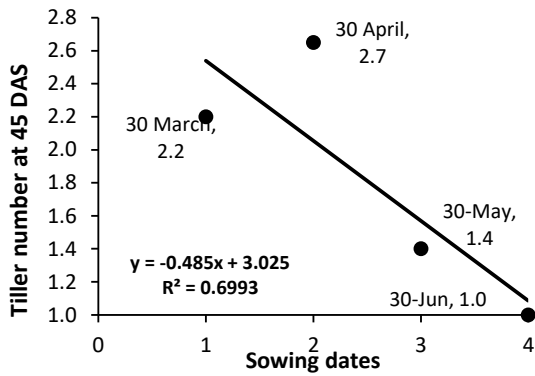


Figure 2.7 Correlation between sowing date and tillers per plant at 45 DAS; data for March, April and May are means for 2017 and 2018

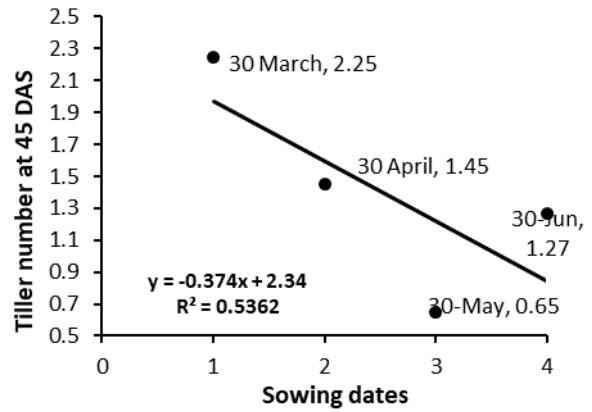


Figure 2.8 Correlation between sowing date and tillers per plant at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

In 2017, tiller number for the 80 kg ha⁻¹ seed rate was significantly lower than for the other three seed rates, while in 2018 the 20 kg ha⁻¹ had the highest tiller number ($P < 0.001$) (Table 2.7). At 75 DAS, tiller number was not significantly affected by seed rate in 2017 but in 2018, the 80 kg ha⁻¹ seed rate had a lower tiller number which did not vary from the 40 kg ha⁻¹ seed rate ($P < 0.005$) (Table 2.7). There was a significant negative correlation between seed rate and tiller number at 45 DAS because tiller number was reduced with increased plant density ($R^2 = 0.91$) ($P < 0.05$). There was a negative correlation between seed rate and tiller number at 75 DAS ($R^2 = 0.77$) ($P > 0.05$) for the same reason as of 45 DAS (Figure 2.9 and 2.10).

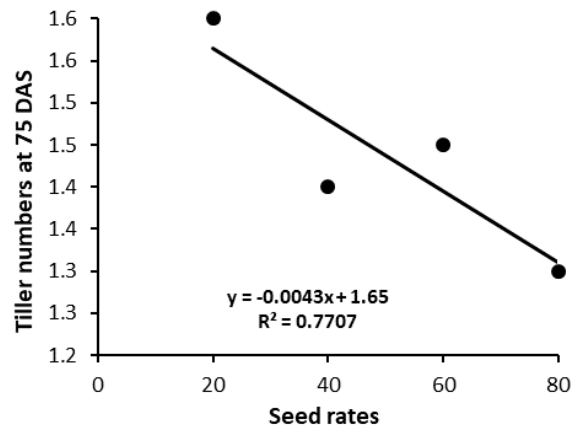
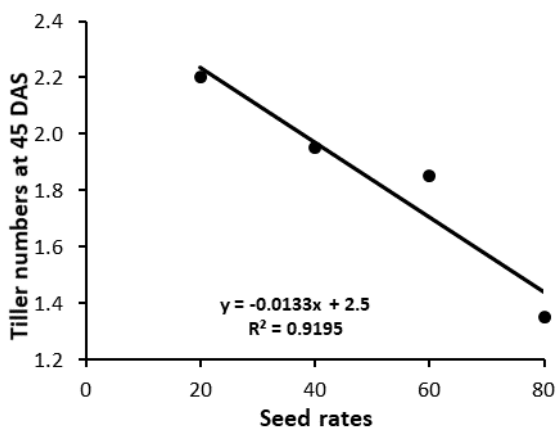


Figure 2.9 Correlation between seed rate and tiller number at 45 DAS; data are means for 2017 and 2018

Figure 2.10 Correlation between seed rate and tiller number at 75 DAS; data for are means for 2017 and 2018

Cutting management

At 75 DAS in 2017, cutting did not affect the tiller numbers but in 2018, cutting increased tiller numbers at 75 DAS ($P < 0.001$) (Table 2.7).

Interaction

In 2018 at 75 DAS, a significant interaction ($P < 0.01$) between sowing date and cutting management occurred because cutting increased tillers in the March sowing (Table 2.8).

Table 2.7 Main effect means of sowing date, sowing rate and cut management on the tillers per plant at 45 and 75 DAS in 2017 and 2018

Main effect means of:	Tillers per plant - 45 DAS		Tillers per plant -75 DAS	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	2.1b	2.3b	1.9a	2.6a
30-Apr	2.6a	2.7a	1.9a	1.0b
30-May	1.3c	1.5c	0.5c	0.8b
30-Jun	1.0d	-	1.3b	-
Significance of linear trend (<i>p</i> value)	<0.001	<0.001	<0.001	<0.001
LSD (0.05)	0.4	0.1	0.4	0.5
CV%	15.2	14.1	17.5	20
Seed rate (SR)				
20 kg ha ⁻¹	1.9a	2.5a	1.5a	1.7a
40 kg ha ⁻¹	1.8a	2.1b	1.4a	1.4bc
60 kg ha ⁻¹	1.7a	2.0b	1.4a	1.5ab
80 kg ha ⁻¹	1.5b	1.2b	1.4a	1.2c
Significance of linear trend (<i>p</i> value)	<0.001	<0.001	0.929	0.002
LSD (0.05)	0.2	0.1	0.17	0.2
CV%	16.0	20.1	17.6	17.5
Cutting management (CM)				
No cut	1.6a	2.1a	1.4a	1.1b
One cut	-	-	1.3a	1.6a
Two cut	-	-	1.5a	1.7a
Significance of linear trend (<i>p</i> value)	-	-	0.166	<0.001
LSD (0.05)	-	-	0.2	0.2
CV%	-	-	40.8	26.6
Significance of interactions of linear contrasts (p value)				
SD (lin) x SR	0.013	0.608	0.772	0.244
SD (lin) x CM	-	-	0.234	<0.001
SR (lin) x CM	-	-	0.806	0.132
SD x SR x CM	-	-	0.962	0.233

Note: DAS = Days after Sowing; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($p < 0.05$). SD=sowing date; SR=seed rate; CM=cutting management.

Table 2.8 Means for the interaction between sowing dates and seed rate on tillers per plant at 75 DAS in 2018

Sowing date	Cutting management		
	0	1	2
30-Mar	1.7	3.0	3.1
30-Apr	0.9	0.9	0.8
30-May	0.7	0.9	0.8
LSD (5%) (Comparisons within a sowing date) 0.3			
LSD (5%) (Other comparisons) 0.5			

Note: 0 = No cut; 1 = one cut; 2 = two cut

2.4.3 Effect of sowing dates, seed rates and cutting management on Leaf Area Index (LAI)

Sowing date

At 45 DAS, LAI for the April sowing was greater for both 2017 ($P < 0.001$) and 2018 ($P = 0.057$) (Table 2.9). At 75 DAS, the first two sowings had higher LAI than the later two sowings in 2017 ($P < 0.005$). In 2018, the March sowing had greater LAI than the April and the May sowings at 75 DAS ($P < 0.001$) (Table 2.9). A negative correlation between sowing date and LAI occurred at 45 ($R^2 = 0.38$) ($P > 0.05$) and 75 ($R^2 = 0.73$) ($P > 0.05$) DAS because LAI reduced with delay in sowing (Figure 2.11 and 2.12).

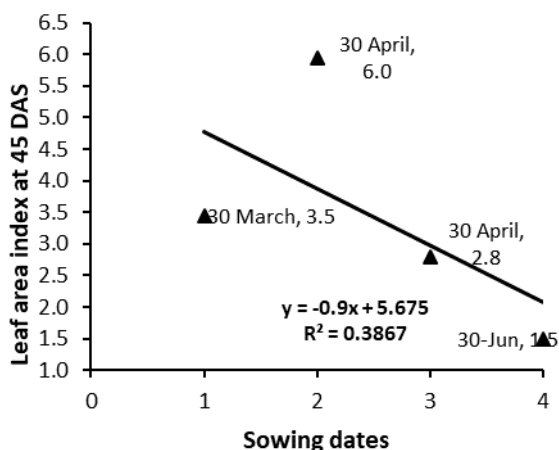


Figure 2.11 Correlation between sowing dates and leaf area index at 45 DAS; data for March, April and May are means for 2017 and 2018

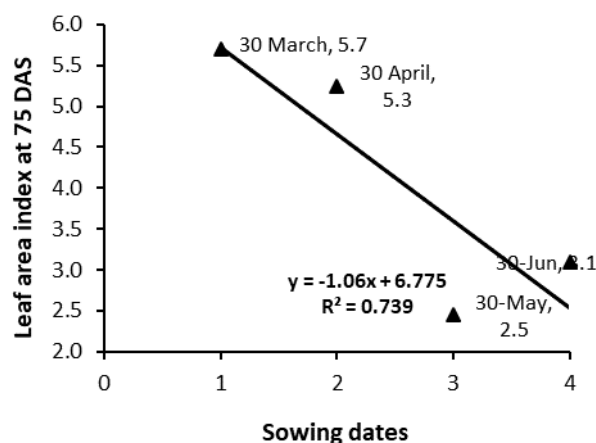


Figure 2.12 Correlation between sowing dates and leaf area index at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

In 2017 at 45 DAS, the 40 kg ha⁻¹ seed rate had a LAI greater than the 20 kg ha⁻¹, 60 kg ha⁻¹ and 80 kg ha⁻¹ seed rates respectively (Table 2.9). In contrast, in 2018 the 20 kg ha⁻¹ seed rate had the highest LAI and the 80 kg ha⁻¹ seed rate had the lowest LAI (P<0.001) (Table 2.9). At 75 DAS in 2017, the highest LAI was observed in the 20 kg ha⁻¹ seed rate which was significantly higher than the other three seed rates (P<0.05) (Table 2.9). However, the highest LAI at 75 DAS in 2018 occurred in the 20 kg ha⁻¹ seed rate and the lowest LAI was recorded for 80 kg ha⁻¹ seed rate (P<0.001) (Table 2.9). There was a negative correlation (R²=0.62) (P>0.05) between seed rate and LAI at 45 DAS. At 75 DAS, there was a significant positive correlation (P<0.05) (R²=0.95) between seed rates and LAI (Figure 2.13 and 2.14).

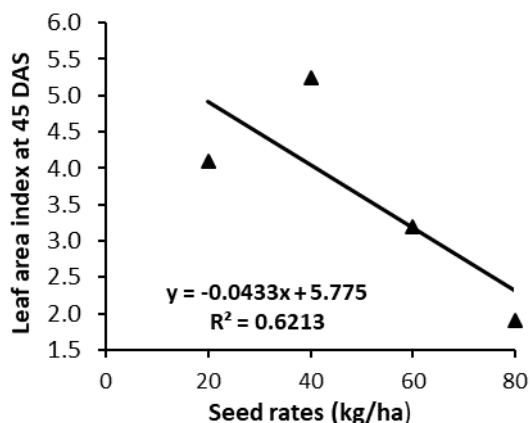


Figure 2.13 Correlation between seed rates and leaf area index at 45 DAS; data are means for 2017 and 2018

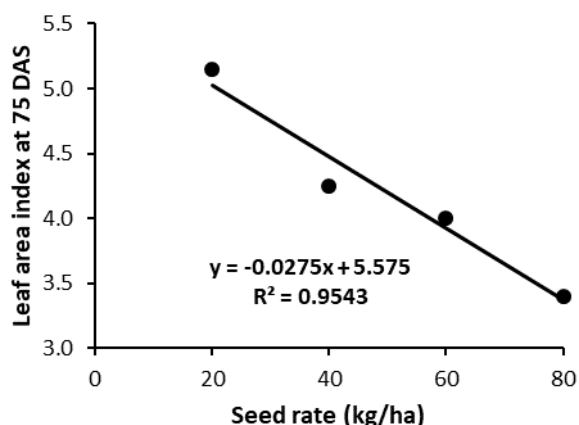


Figure 2.14 Correlation between seed rates and leaf area index at 75 DAS; data are means for 2017 and 2018

Cutting Management

At 75 DAS, cutting reduced the LAI in 2017 ($P < 0.001$) (Table 2.9). However, in 2018, LAI for uncut and twice cut plants was not different (Table 2.9). A negative correlation ($R^2 = 0.80$) ($P > 0.05$) occurred between cutting management and LAI at 75 DAS because in 2017 LAI was reduced as the cutting frequency increased (Figure 2.15).

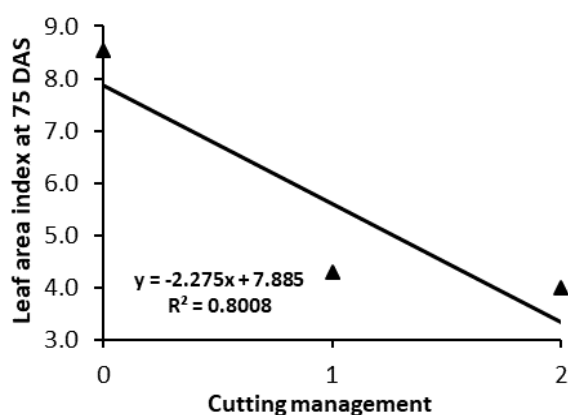


Figure 2.15 Correlation between cutting management and leaf area index at 45 DAS; data are means for 2017 and 2018

Interaction

At 75 DAS in 2017, there was a significant interaction between sowing date and cutting management because for the two later sowings, cutting reduced LAI but this did not occur for the first two sowings ($P < 0.05$) (Table 2.10). The significant interaction between sowing date and seed

rates occurred at 75 DAS in 2018 because the LAI for the 30 March sowing was greater at the lowest seed rate ($P<0.05$) (Table 2.11). A significant interaction occurred between sowing date and cutting management at 75 DAS in 2018, because for the March sowing, LAI was similar for uncut and twice cut plants which did not occur for the two later sowings ($P<0.001$) (Table 2.12). The significant interaction between seed rate and cutting management at 75 DAS in 2018 occurred because the lowest seed rate had the higher LAI for both uncut and twice cut plants ($P<0.05$) (Table 2.13). Similarly, there was also a significant interaction among sowing date, seed rate and cutting management in 2018 which is because the 30 March sowing produced higher LAI at the lowest seed rate when uncut ($P<0.05$) (Table 2.14).

Table 2.9 Main effect means of sowing date, sowing rate and cutting management on teosinte Leaf Area Index (LAI) at 45 days and 75 DAS in 2017 and 2018

Main effect means of:	LAI - 45 DAS		LAI - 75 DAS	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	4.4b	2.5b	5.9a	5.5a
30-Apr	7.4a	4.5a	7.5a	3.0b
30-May	3.6b	2.0b	2.7b	2.2b
30-Jun	1.5c	-	3.1b	-
Significance of linear trend (<i>p</i> value)	<0.001	0.057	0.004	<0.001
LSD (0.05)	1.0	0.5	2.4	1.3
CV%	15.3	9.3	31.5	20.7
Seed rate (SR)				
20 kg ha ⁻¹	4.4b	3.8a	5.6a	4.7a
40 kg ha ⁻¹	7.4a	3.1b	4.8b	3.7b
60 kg ha ⁻¹	3.6b	2.8b	4.5b	3.5b
80 kg ha ⁻¹	1.5c	2.3c	4.5b	2.3c
Significance of linear trend (<i>p</i> value)	<0.001	<0.001	0.002	<0.001
LSD (0.05)	0.3	0.4	0.7	1.0
CV%	11.3	15.3	21.2	30.5
Cutting management (CM)				
No cut	4.0a	3.1a	6.9a	4.0a
One cut	-	-	3.7b	3.0b
Two cut	-	-	4.0b	3.7a
Significance of linear trend (<i>p</i> value)	-	-	<0.001	0.397
LSD (0.05)	-	-	0.900	0.600
CV%	-	-	53	42.5
Significance of interactions of linear contrasts (<i>p</i> value)				
SD(lin) x SR	0.260	0.433	0.918	0.030
SD(lin) x CM	-	-	0.019	<0.001
SR(lin) x CM	-	-	0.083	0.002
SD x SR x CM	-	-	0.876	0.002

Note: LAI = Leaf Area Index; DAS = Days after Sowing; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($p < 0.05$). SD=sowing date; SR=seed rate; CM=cutting management.

Table 2.10 Means for the interaction between sowing dates and cutting management on LAI at 75 DAS in 2017

Sowing dates	Cutting management		
	0	1	2
30 March	7.2	5.3	5.3
30 April	13.0	4.8	4.8
30 May	2.9	2.4	2.8
30 June	4.3	2.2	2.8
LSD (5%) (Comparisons within a sowing date) 1.8			
LSD (5%) (Other comparisons) 2.7			

Note: 0 = No cut; 1 = one cut; 2 = two cut

Table 2.11 Means for the interaction between sowing dates and seed rates on LAI at 75 DAS in 2018

Sowing dates	Seed rates (kg/ha)			
	20	40	60	80
30 March	7.7	5.7	5.4	3.0
30 April	2.8	3.5	3.1	2.4
30 June	3.6	1.8	2.2	1.3
LSD (5%) (Comparisons within a sowing date) 1.5				
LSD (5%) (Other comparisons) 1.7				

Table 2.12 Table of means for the interaction between sowing dates and cutting management on LAI at 75 DAS in 2018

Sowing dates	Cutting management		
	0	1	2
30 March	3.4	5.2	7.7
30 April	5.5	1.8	1.6
30 May	3.1	1.8	1.8
LSD (5%) (Comparisons within a sowing date) 1.1			
LSD (5%) (Other comparisons) 1.4			

Note: 0 = No cut; 1 = one cut; 2 = two cut

Table 2.13 Table of means for the interaction between seed rates and cutting management on LAI at 75 DAS in 2018

Seed rates (kg/ha)	Cutting management		
	0	1	2
20	4.2	4.7	5.2
40	4.1	2.6	4.3
60	4.3	2.7	3.6
80	3.3	1.8	1.7

LSD (5%) (Comparisons within a sowing date) 1.2
LSD (5%) (Other comparisons) 1.2

Note: 0 = No cut; 1 = one cut; 2 = two cut

Table 2.14 Table of means for the interaction between sowing dates, seed rates and cutting management on LAI at 75 DAS in 2018

Sowing dates	Seed rates (kg/ha)	Cutting management		
		0	1	2
30 march	20	4.0	8.5	10.7
	40	3.6	4.5	9.1
	60	3.6	4.4	8.1
	80	2.5	3.5	3.1
30 April	20	3.8	2.5	2.1
	40	6.3	1.9	2.3
	60	6.4	1.7	1.1
	80	5.2	1.2	0.8
30 May	20	4.8	3.2	2.9
	40	2.4	1.4	1.6
	60	3.0	1.8	1.8
	80	2.0	1.0	1.0

LSD (5%) (Comparisons within a sowing date) 2.3
LSD (5%) (Other comparisons) 2.4

Note: 0 = No cut; 1 = one cut; 2 = two cut

2.4.4 Effect of sowing dates, seed rates and cutting management on leaf number per plant

Sowing date

At 45 DAS, the number of leaves per plant for the April sowing was significantly higher than for the March and the May sowings in both years ($P < 0.001$) (Table 2.15). At 75 DAS, sowing date had no effect on leaf number in either year (Table 2.15). A negative correlation between sowing date and leaf number occurred at 45 DAS ($R^2 = 0.69$) ($P > 0.05$) and 75 DAS ($R^2 = 0.45$) ($P > 0.05$) because leaf number reduced with delay in sowing (Figure 2.16 and 2.17).

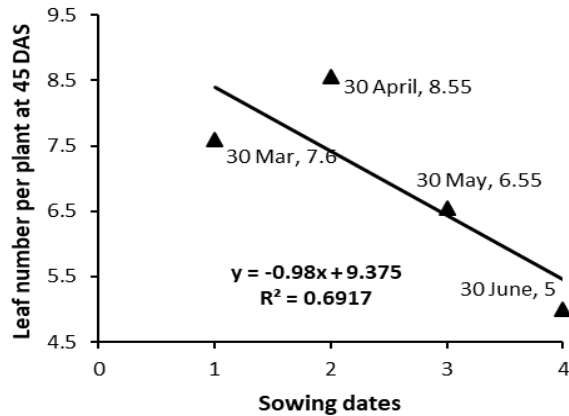


Figure 2.16 Correlation between sowing dates and leaf number at 45 DAS; data for March, April and May are means for 2017 and 2018

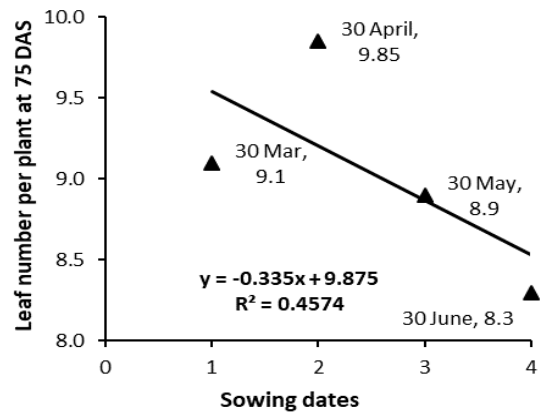


Figure 2.17 Correlation between sowing dates and leaf number at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

Seed rate had no effect on the leaf number at 45 DAS in 2017 (Table 2.15). But in 2018, the 20 kg ha⁻¹ seed rate had the highest leaf number followed by the 40 kg ha⁻¹ and 60kg ha⁻¹. The 80 kg ha⁻¹ seed rate produced the lowest number of leaves ($P < 0.001$) (Table 2.15). At 75 DAS leaf number differed only between the 20 kg ha⁻¹ and 80 kg ha⁻¹ seed rates in 2017 ($P < 0.05$). But in 2018, leaf number at the 20 kg ha⁻¹ seed rate was significantly higher than the 80 kg ha⁻¹ seed rate ($P < 0.001$) (Table 2.15). There was a significant negative correlation between seed rate and leaf number per plant at 45 DAS ($R^2 = 0.98$) ($P < 0.01$) and 75 DAS ($R^2 = 0.99$) ($P < 0.01$) because leaf number had decreased as seed rate increased for both 45 and 75 DAS (Figure 2.18 and 2.19).

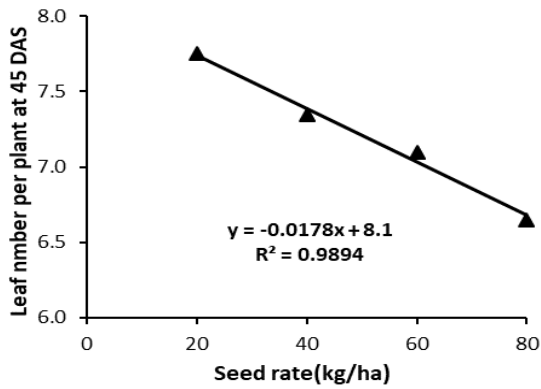


Figure 2.18 Correlation between seed rates and leaf number at 45 DAS; data are means for 2017 and 2018

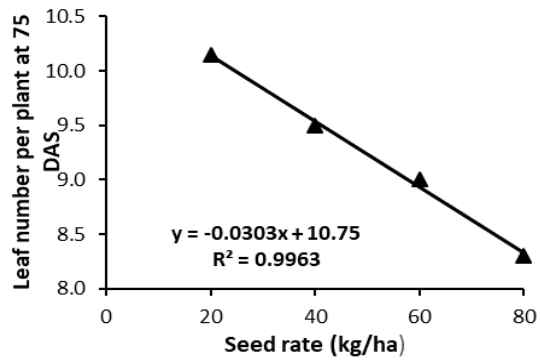


Figure 2.19 Correlation between seed rates and number of leaf at 75 DAS; data are means for 2017 and 2018

Cutting Management

At 75 DAS, cutting had significantly reduced leaf numbers in both years ($P < 0.001$) (Table 2.15). There was a negative correlation between leaf number and cutting management at 75 DAS ($R^2 = 0.73$) ($P > 0.005$) because cutting reduced leaf number (Figure 2.20).

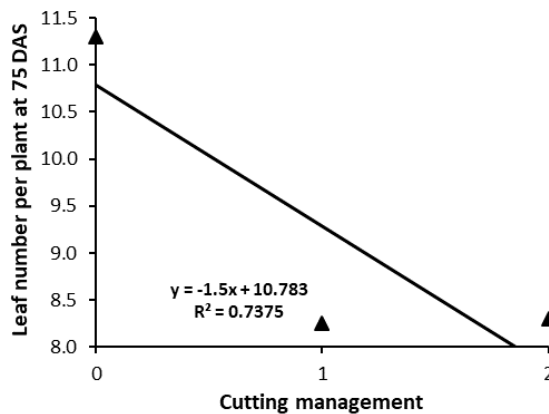


Figure 2.20 Correlation between sowing dates and leaf number at 75 DAS; data are means for 2017 and 2018

Interactions

There was a significant interaction between sowing date and cutting management at 75 DAS in 2018 because cutting significantly reduced leaf number in all sowings ($P < 0.001$) (Table 2.16).

Table 2.15 Main effect means of sowing date, sowing rate and cutting management on leaf number per plant at 45 and 75 DAS in 2017 and 2018

Main effect means of :	Leaf No per plant- 45 DAS		Leaf No per plant -75 DAS	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	7.6b	7.6b	8.5a	9.7a
30-Apr	8.9a	8.2a	8.8a	10.9a
30-May	7.6b	5.5c	8.2a	9.6a
30-Jun	5.0c	-	8.3a	-
Significance of linear trend(<i>p</i> value)	0.<001	<0.001	0.325	0.865
LSD (0.05)	0.6	0.3	0.8	2.0
CV%	5.1	2.5	6.1	11.7
Seed rate (SR)				
20 kg ha ⁻¹	7.2a	8.3a	8.7a	11.6a
40 kg ha ⁻¹	7.4a	7.3b	8.4ab	10.6b
60 kg ha ⁻¹	7.2a	7.0b	8.4ab	9.6b
80 kg ha ⁻¹	7.3a	6.0c	8.2b	8.4c
Significance of linear trend(<i>p</i> value)	0.703	<0.001	0.013	<0.001
LSD (0.05)	0.3	0.8	0.4	1.1
CV%	5.1	13.6	7.0	13.5
Cutting management (CM)				
No cut	7.3a	7.3a	9.2a	13.4a
One cut	-	-	8.0b	8.5b
Two cut	-	-	8.0b	8.6b
Significance of linear trend(<i>p</i> value)	-	-	<0.001	<0.001
LSD (0.05)	-	-	0.3	0.8
CV%	-	-	9.1	21.6
Significance of interactions of linear contrasts (p value)				
SD (lin) x SR	0.116	0.15	0.069	0.586
SD (lin) x CM	-	-	0.370	<0.001
SR (lin) x CM	-	-	0.657	0.119
SD x SR x CM	-	-	0.313	0.218

Note: No = Number; DAS = Days after Sowing; t/ha = tonnes ha⁻¹; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (*p*<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

Table 2.16 Table of means for the interaction between sowing dates and cutting management on leaf number per plant at 75 DAS in 2018

Sowing dates	Cutting management		
	0	1	2
30 March	9.0	10.1	10.1
30 April	18.1	7.2	7.4
30 May	12.4	8.0	8.3

LSD (5%) (Comparisons within a sowing date) 1.5
LSD (5%) (Other comparisons) 2.2

Note: 0 = No cut; 1 = one cut; 2 = two cut

2.4.5 Effect of sowing dates, seed rates and cutting management on fresh herbage yield (HY), tonnes ha⁻¹

Sowing date

Sowing date was an important source of variation with respect to HY in both seasons (Table 2.17). At 45 DAS, the April sowing produced the greatest HY and this also occurred at 75DAS in 2017(P<0.001). In 2018, the greatest HY was also recorded from the April sowing at 45 DAS and the March sowing produced the greatest HY at 75 DAS (P<0.001) (Table 2.17). A negative correlation (R²=0.60) (P>0.05) between sowing date and HY occurred at 45 DAS while there was a significant negative correlation between sowing dates and HY at 75 DAS (R²=0.98) because HY was reduced with delay in sowing (Figure 2.21 and 2.22).

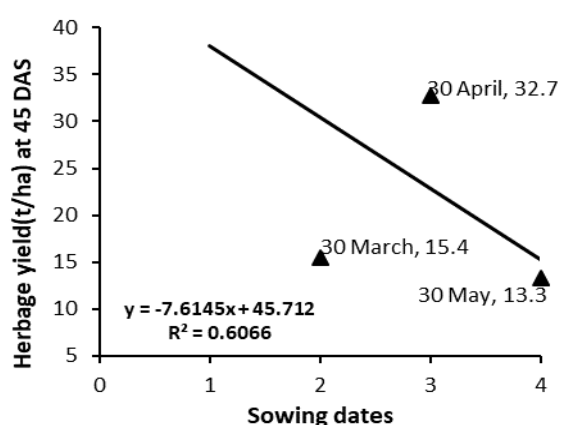


Figure 2.21 Correlation between sowing dates and herbage yield at 45 DAS; data for March, April and May are means for 2017 and 2018

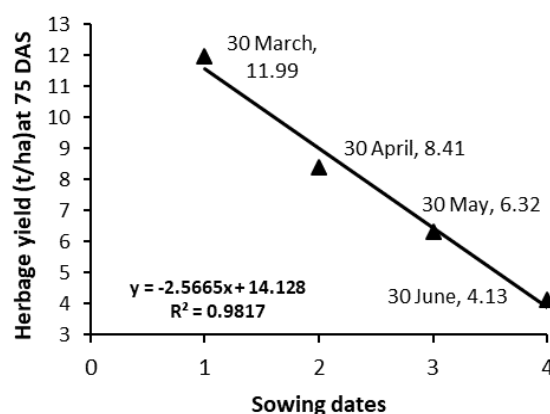


Figure 2.22 Correlation between sowing dates and herbage yield at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

In 2017, the HY did not differ markedly with seed rate at 45 DAS, although it did differ between the 40 kg ha⁻¹ and 80 kg ha⁻¹ seed rate (Table 2.17). In contrast, in 2018 HY was significantly greater for the two higher seed rates ($P < 0.001$). However at 75 DAS, HY did not differ among the seed rates in either year (Table 2.17). A positive correlation between seed rate and HY occurred at 45 DAS ($R^2 = 0.90$) ($P > 0.05$) because HY increased with the increase in seed rate. But at 75 DAS, there was a negative correlation between seed rate and herbage yield ($R^2 = 0.44$) ($P > 0.05$) because HY reduced with increase in seed rate (Figure 2.23 and 2.24).

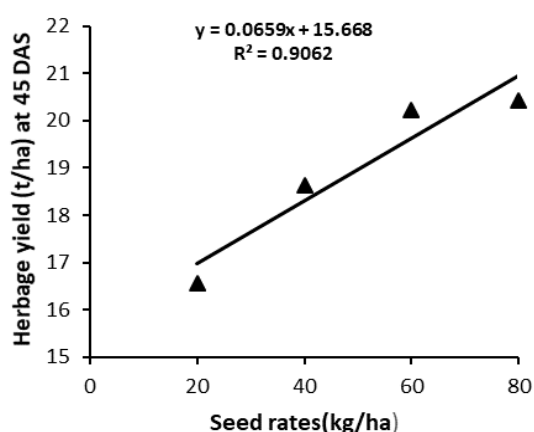


Figure 2.23 Correlation between seed rates and HY at 45 DAS; data are means for 2017 and 2018

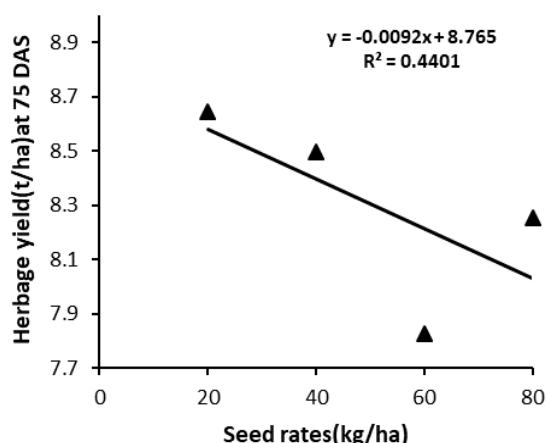


Figure 2.24 Correlation between seed rates and HY at 75 DAS; data are means for 2017 and 2018

Cutting

At 45 DAS, HY was not influenced by cutting management in either year. There was no mean comparison for HY at 75 DAS because only plants in the two cut treatment were harvested (Table 2.17).

Interaction

During 2017 and 2018, there were no significant interactions among sowing date, seed rate and cutting management for fresh HY at both 45 and 75 DAS (Table 2.17).

Table 2.17 Main effect means of sowing date, sowing rate and cutting management on HY of teosinte at 45 and 75 DAS in 2017 and 2018

Main effect means of:	HY (t/ha) - 45 DAS	HY (t/ha) - 75 DAS
-----------------------	--------------------	--------------------

	2017	2018	2017	2018
Sowing date(SD)				
30-Mar	19.5b	11.3b	8.1b	15.9a
30-Apr	29.8a	35.6a	12.5a	4.3b
30-May	10.3c	16.2b	6.2c	6.5b
30-Jun	8.0c	-	4.1d	-
Significance of linear trend(<i>p</i> value)	<0.001	0.165	<0.001	<0.001
LSD (0.05)	3.1	7.7	1.02	3.3
CV%	11.5	21.2	8.3	21.4
Seed rate (SR)				
20 kg ha ⁻¹	17.0ab	16.1c	8.0a	9.3a
40 kg ha ⁻¹	17.6a	19.7b	7.9a	9.0a
60 kg ha ⁻¹	16.6ab	23.8a	7.5a	8.2a
80 kg ha ⁻¹	16.4b	24.5a	7.5a	9.1a
Significance of linear trend(<i>p</i> value)	0.140	<.001	0.056	0.719
LSD (0.05)	1.2	3.0	0.7	2.4
CV%	10.0	17.1	13.5	32.6
Cutting management (CM)				
No cut	-	-	-	-
One cut	17.1a	20.7a	-	-
Two cuts	16.7a	21.5a	7.7	8.9
Significance of linear trend(<i>p</i> value)	0.504	0.555	-	-
LSD (0.05)	1.12	2.24	-	-
CV%	18.6	25.7	-	-
Significance of interactions of linear contrasts (p value)				
SD(lin) x SR	0.807	0.074	0.122	0.429
SD(lin) x CM	0.615	0.723	-	-
SR(lin) x CM	0.290	0.893	-	-
SD x SR x CM	0.284	0.666	-	-

Note: HY = Herbage Yield; DAS = Days after Sowing; t/ha = tonnes ha⁻¹; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (*p*<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

2.4.6 Effect of sowing dates, seed rates and cutting management on dry matter yield (DMY), tonnes ha⁻¹

Sowing date

In 2017, there was a significant difference ($P < 0.001$) among the sowing dates for DMY at 45 DAS ($P < 0.001$). In both years, the greatest DMY was produced from the 30 April sowing. In 2017, the 30 March sowing produced more DMY than the 30 May and 30 June sowings, but this did not occur in 2018 (Table 2.18). Similarly, at 75 DAS, DMY differed significantly among the sowing dates in both years being highest at the April sowing in 2017 and at the March sowing in 2018 (Table 2.18). A significant negative correlation between sowing date and DMY occurred at 75 DAS ($R^2 = 0.96$) ($P < 0.05$) because DMY reduced with delay in sowing (Figure 2.25).

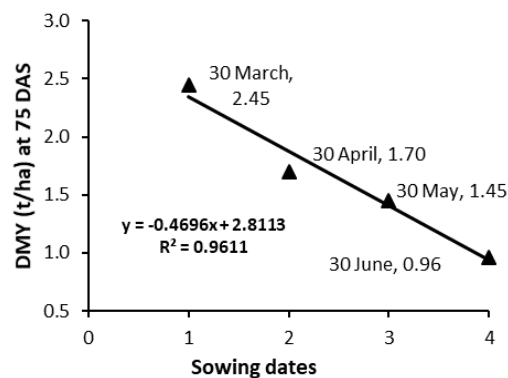


Figure 2.25 Correlation between sowing dates and DMY at 75 DAS; data for March, April and May are means for 2017 and 2018

Seed rate

The effect of seed rate on DMY was non-significant ($P > 0.05$) in 2017 but was significant ($P < 0.001$) in 2018 at 45 DAS only (Table 2.18). DMY for the 20 kg ha⁻¹ seed rate was lower than for the higher three seed rates at 45 DAS (Table 2.18). A positive correlation between seed rate date and DMY occurred at 45 DAS ($R^2 = 0.72$) ($P > 0.05$) because DMY increased with increased seed rate at 45 DAS (Figure 2.26).

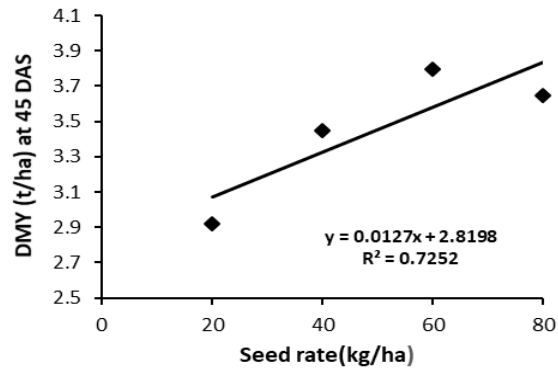


Figure 2.26 Correlation between seed rates and DMY at 45 DAS; data are means for 2017 and 2018

Cutting management

There was no significant effect of cutting management on DMY of teosinte during 2017 and 2018 at 45 and 75 DAS (Table 2.18).

Interactions

There were no significant interactions for DMY at either 45 DAS or 75 DAS between or among treatments in either year (Table 2.18).

Table 2.18 Main effect means of sowing date, sowing rate and cutting management on DMY of teosinte at 45 and 75 DAs in 2017 and 2018

Main effect means of:	DMY (t ha ⁻¹) - 45 DAS		DMY (t ha ⁻¹) - 75 DAS	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	2.6b	1.2b	1.6b	3.3a
30-Apr	6.2a	7.8a	2.5a	0.8b
30-May	1.8c	2.7b	1.4c	1.5b
30-Jun	1.5c	-	0.96d	-
Significance of linear trend (p value)	<0.001	0.062	<0.001	0.001
LSD (0.05)	0.8	1.6	0.2	0.7
CV%	17.2	24.4	7.1	22.6
Seed rate (SR)				
20 kg ha ⁻¹	3.0a	2.8b	1.6a	1.8a
40 kg ha ⁻¹	3.1a	3.8ab	1.7a	1.9a
60 kg ha ⁻¹	3.2a	4.4a	1.6a	1.7a
80 kg ha ⁻¹	2.8a	4.5a	1.6a	2.0a
Significance of linear trend (p value)	0.52	0.001	0.925	0.659
LSD (0.05)	0.4	1.0	0.2	0.5
CV%	19.7	29.8	17.5	33.6
Cutting management (CM)				
No cut	-	-	-	-
One cut	3.0a	3.9a	-	-
Two cut	3.1a	3.9a	1.6	1.9
Significance of linear trend (p value)	0.767	0.938	-	-
LSD (0.05)	0.3	0.6	-	-
CV%	31.7	34.6	-	-
Significance of interactions of linear contrasts (p value)				
SD(lin) x SR	0.700	0.110	0.791	0.831
SD(lin) x CM	0.938	0.771	-	-
SR(lin) x CM	0.777	0.617	-	-
SD x SR x CM	0.618	0.539	-	-

Note: DMY- Dry Matter Yield; DAS = Days after sowing; t/ha = tonnes ha⁻¹; LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

Note*: For the sowing date and seed rate main effect means, DMY at 45 DAS and DMY at 75 DAS do not add up to Total DMY. This is because the main effect means for DMY at 45 DAS are averages of two cutting treatments, while the main effect means for DMY at 75 DAS are based on only one cutting treatment. Further, there are no values (LSD, P value and mean) at 75 DAS because of only one mean resulting from one cut at 75 DAS.

2.4.7 Association of vegetative yield components with dry matter yield in teosinte

Plant height, number of tillers per plant, LAI and number of leaves per plant were the major contributors to DMY of teosinte at 45 and 75 DAS for differ sowing dates. DMY was positively correlated with the plant height. The correlation was positive at 45 DAS ($R^2= 0.61$) but it was very weak at 75 DAS ($R^2= 0.25$) (Figure 2.27). Similarly tiller numbers were positively correlated with the DMY at 45 DAS ($R^2= 0.58$) and at 75 DAS ($R^2 = 0.56$) (Figure 2.28).The association between the LAI and DMY at 75 DAS was positive ($R^2=0.63$) (Figure 2.29). Similarly leaf number per plant was positively correlated with the DMY at 45 DAS ($R^2=0.57$) (Figure 2.30). Likewise for different seed rates, LAI showed positive and a highly significant correlations for DMY at 45 DAS ($R^2=0.65$) and 75 DAS ($R^2=0.99$) (Figure 2.31) respectively. Tiller number at 45 DAS was significantly correlated with plant height ($R^2=0.98$) (Figure 2.32) but is weakly correlated at 75 DAS ($R^2=0.34$) (Figure 2.32). There was a positive correlation between LAI and plant height at both 45 ($R^2=0.85$) (Figure 2.33) and 75 DAS ($R^2=0.77$) (Figure 2.33). A significant positive correlation occurred between plant height and leaf number per plant at 45 DAS ($R^2=0.98$) (Figure 2.34), while it was positive at 75 DAS ($R^2=0.71$) (Figure 2.34).

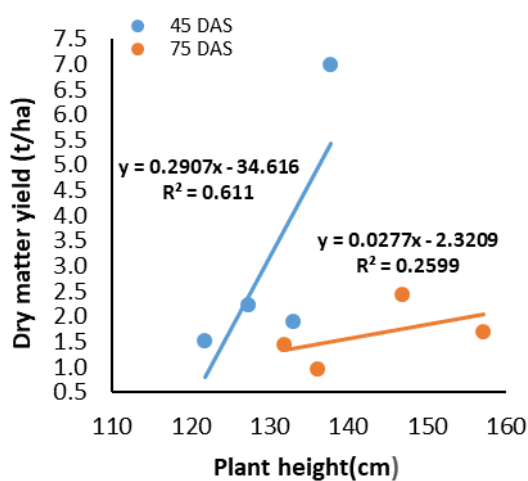


Figure 2.27 Correlation between plant height and DMY using main effect means of different sowing dates

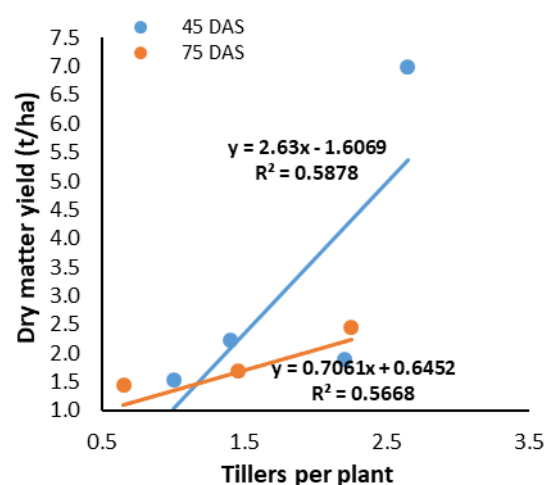


Figure 2.28 Correlation between tiller numbers and DMY using main effect means of different sowing dates

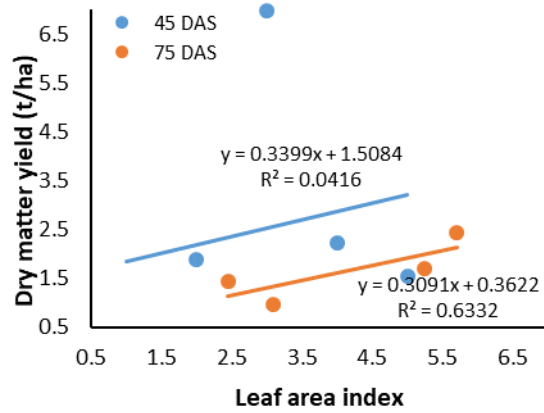


Figure 2.29 Correlation between LAI and DMY using main effect means of different sowing dates

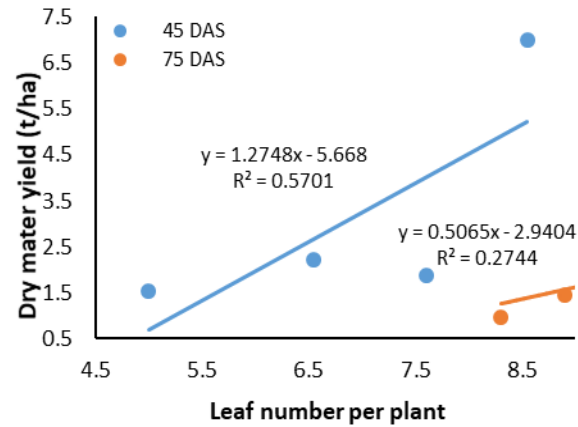


Figure 2.30 Correlation between leaf number per plant and DMY using main effect means of different sowing dates

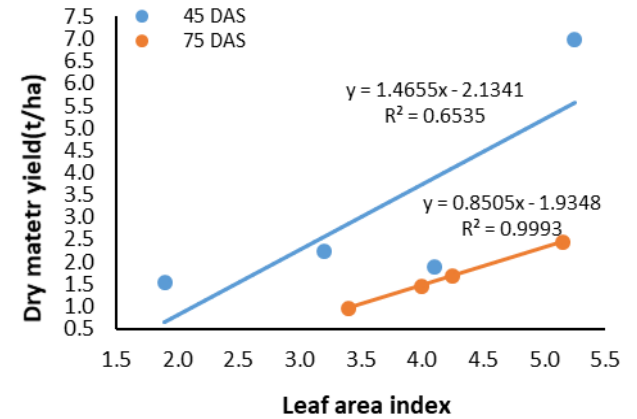


Figure 2.31 Correlation between LAI and DMY using main effect means of different seed rates

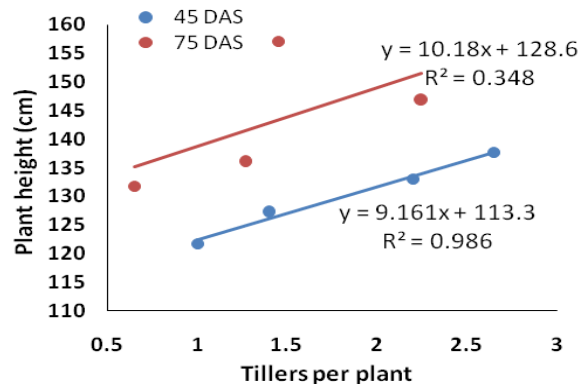


Figure 2.32 Correlation between plant height and tiller number using main effect means of different sowing dates

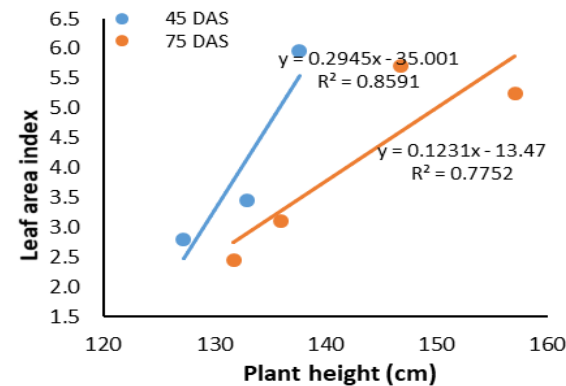


Figure 2.33 Correlation between plant height and leaf area index using main effect means of different sowing dates

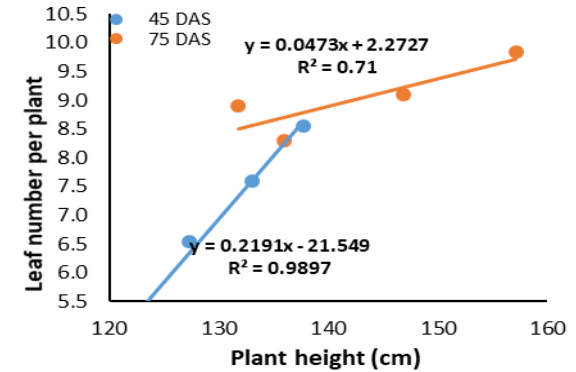


Figure 2.34 Correlation between plant height and leaf number per plant using main effect means of different sowing dates

2.5 Discussion

2.5.1 Effect of sowing dates, seed rates and cutting management on plant height (cm)

Sowing date

Plant growth as determined by the individual component development rate and the plant's progress through its various ontogenetic stages is quantitatively dependent on solar radiation and temperature (Sinclair, 1994). In the present study, sowing date did not affect plant height at 45 DAS in both years. At 75 DAS, June sowing in 2017 had the lowest plant height. Lower temperature reduced GDD accumulation (Table 3.1-3.4, Chapter 4), and higher rainfall during July and August (Figure 2.2) broke stems and damaged the regrowth produced following the 45 days harvest, resulting in lower plant height for the June sowing. Al-Darby and Lowery (1987) reported a reduced plant height in maize at lower temperature. Under excess water supply from monsoon rain (June, July, August and September, 2017) there was poor root development which caused reduced aeration that impaired plant growth. Similar results have been reported by Hoogenboom (2000) and Sprague & Dudley (1988) in various crops. The plant height of the March and May sowing did not vary with that of the June sowing because of the breakage of regrowth due to heavy wind and rain in the month of July and August respectively. Higher temperature (Figure 2.1) and ambient rainfall (Figure 2.2) for the April sowing resulted in taller plants. At higher temperature, plant height increases due to greater cell elongation (Erwin, Velguth, & Heins, 1994). The variation in results between the two years can be explained by the temperature (-1.9°C in March, 1.1°C in April, and 1.4°C in May) and rainfall difference (1.3mm in March, April 0.1,-2.2 mm and May -1.0mm) in both years (Figure 2.1a and 2.1b).

Seed rate

Seed rate had no major effect on teosinte plant height at 45 DAS in 2017, which might be because even the highest seed rate was not sufficient to create competition among the plants at the earliest stage of growth. This result was similar to several plant density studies in maize (Abzur et al, 2011; Iptas & Acar, 2003). At 45 DAS in 2018, the lowest seed rate produced the smallest plants. Lashkari, Madani, Golzardi, and Zargari (2011) found that in maize the higher seed rates produced the tallest plants. At higher plant density plants are closer to each other which improves the quantity of FR reflected by competing seedlings, leading ultimately to a higher FR/R ratio which stimulates apical dominance as a result of prioritizing the allocation of assimilates to the main stem (Kasperbauer & Karlen, 1994; Zhang, Dong, Wang, Hu & Liu, 2006) in maize. Increased plant height at higher plant density has also been reported by Donald (1963) and Sangoi, Gracietti, Rampazzo, and Bianchetti (2002) in maize, and Bayu, Rethman, and Hammes (2005) in

sorghum. Lowest plant height at the highest seed rate at 75 DAS in both years might be because of interplant competition for resources which limited the supply of assimilates to individual plants. This result is similar to the findings of Mukhopadhy and Sen (1997) who reported that decreased plant height at higher densities is due to competition for resources.

Cutting management

Cutting at 45 DAS significantly reduced plant height at 75 DAS in both years. As opposed to non-cut plants, cutting immediately removes photosynthetic capability and resources required to support regrowth (Arif et al., 2012; Noy-Meir & Briske, 2002). The fact that approximately one month after cutting, the cut plants were only around one third shorter than the uncut plants indicates the rapid potential for growth of teosinte in the Nepalese environment (Devkota et al., 2015).

Interactions

At 75 DAS in 2017, cutting reduced the plant height because uncut plants were taller for the two lower seed rates. A contrasting result was reported by Naveed et al. (2014) who found no interaction between seed rate and cutting in wheat. In 2018, significant interactions between sowing dates and seed rates occurred because the highest seed rate did not change the plant height for any sowing. The result of 2018 was similar to the findings of Kumar et al. (2017) in lucerne.

2.5.2 Effect of sowing dates, seed rates and cutting management on tillers per plant

Sowing date

In the present study, the April sown plants produced the highest number of tillers at 45 DAS in both years. This was most probably a temperature and growth period effect, because temperature in the March and later sown treatments was lower between 45 DAS and 75 DAS. The higher temperature in the April sowing (Figure 2.1) allowed the accumulation of a higher GDD (Table 3.1-3.4, Chapter 4) than for the other sowings. Similar results have been reported by Friend (1965) and Longnecker, Kirby, and Robson (1993) in wheat where the tiller number increased with increasing temperature and light intensity. The result of this study is similar to the findings of Devkota et al. (2015) who reported that teosinte sown at the end of April produced a higher number of tillers than other sowing dates at the same study site. The reduction in tiller numbers at 75 DAS is the result of heavy rainfall in July and August in both years (Figure 2.2). Regrowth tillers following the cutting at 45 DAS were weak and broken off/damaged by the rain. Lower food reserves stored in the crown and less leaf area was therefore available on the cut plants to

support the re-growth. Similar results have been reported by Fribourg (1995) and Stephenson and Posler (1984). The variation in results for the two year reflects the difference in climatic factors between the years (Figure 2.1 and 2.2).

Seed rate

Tiller numbers per plant were significantly affected by seed rate at 45 DAS in 2017. The low number of tillers from the highest seed rate in 2017 is likely due to competition among the plants for light and nutrients that resulted in a lower supply of assimilate to individual plants. At 45 DAS in 2018, the lowest seed rate produced the highest number of tillers because more assimilates was obtained by an individual plant due to less competition for resources. At 75 DAS in 2017, seed rate had no major effect on tiller number per plant of teosinte which is similar to results reported by Jan, Hamid and Muhammad (2000) and Nazir, Ahmed, Siddiq, and Ahmed, (1987) in wheat. Although data were not recorded, this lack of difference in tiller number per plant is probably a result of the monsoonal rain between 45 and 75 DAS which broke /severely damaged more tillers in the higher density plots. This result is in line with the findings of Turki, Al-Namazi, and Masrahi (2019) in sorghum. At 75 DAS in 2018, the lowest tillers at the highest plant density once again is a competition effect and similar to the findings of Skalova and Krahulec (1992) in *Festuca rubra*. The reason why the tillers in the 80kg ha^{-1} seed rate did not differ with the 40kg ha^{-1} seed rate is unknown.

Cutting management

Tillers, the key components of plant structure, are affected by cutting management (Kuraparthi, Sood, & Gill, 2008). At 75 DAS in 2018, uncut plants had fewer tillers than the cut plants, as cutting removed apical dominance and allowed tiller buds at the base of the plant to produce new tillers because more light was available than in the non-cut canopy. Further, cutting also results in stimulation and restructuring of carbon assimilates available to support the early growth of young tillers (Xia, Hodgson, Matthew, & Chu, 1990). This result is in line with the findings of Schneider, Caron, Elli, Schwerz, and Engroff (2019) and Abuelgasim and Abusuwar (2001) in sorghum and in lucerne respectively. However, Chapman et al. (1983), Korte (1986) and Tallowin (1981) reported that tillering is not influenced by cutting frequency as occurred in 2017. The 2017 result at 75 DAS might be the temperature effect where higher temperature accompanied by higher rainfall in 2017 allowed higher GDD accumulation in uncut plants and quick regeneration of cut plants under the more favourable temperature.

Interactions

At 75 DAS in 2018, there was a significant interaction between sowing date and cutting management because the March sowing produced more tillers due to the removal of apical dominance. Tiller buds were exposed to a better photoperiod and an extended growth period for the early sowing. However, Kumar et al. (2017) reported a non-significant effect of sowing date and cutting intervals on tillers per plant which might be because, as the number of cuttings increases, the normal growth of the plants is disturbed, causing slow regrowth, resulting in poor tiller numbers.

2.5.3 Effect of sowing dates, seed rates and cutting management on Leaf Area Index (LAI)

Sowing date

LAI is the ratio of total area occupied by a plant's leaves per unit total area of land (Watson, 1997). It is an important measure to know rate of plant growth and development status (Fortin, Pierch, & Edwards, 1994; Steward & Dwyer, 1999). Plant growth activities like photosynthesis, transpiration and accumulation of dry matter mainly depend upon the total area occupied by the plant canopy and the distribution of leaves which regulate the interception of solar radiation, gaseous exchange and maintain temperature around the plant canopy.

At 45 DAS, the highest LAI was recorded for the April sowing in both years, as a result of the combination of higher temperature and regular precipitation during the early vegetative growth. Higher temperature and precipitation lead to expansion of leaves because of an increase in the gas flow between the plant and the atmosphere which expands the plant tissue and shoot development, increasing the LAI (Ferraz et al, 2012; Tsimba, Edmeades, Millner, & Kemp, 2013). The lower temperature for the March sowing reduced the LAI which is in line with the findings of Muchow and Carberry (1989). Lower LAI in May and June sowings was from reduced solar radiation interception. Similar findings have been reported by Tsimba, Edmeades, Millner, Kemp, and Morris (2014) in maize. Higher LAI at 75 DAS for the March sowing in both years is because the higher temperature and extended growth period allowed a greater LAI after the harvest at 45 DAS. Similar results have been reported by Tsimba et al. (2013) in maize. But why there was no difference in LAI for the March and the April sowing in 2017 is not clear. The differences in LAI between the two years is possibly due to the temperature differences where the first year had higher temperature than the second year.

Seed rate

At 45 DAS, the LAI was lowest for the highest seed rate in both years as a result of inter-plant competition for resources that reduced the assimilate supply to individual plants reducing their growth. A similar response was reported by Crozier, Gehl, Hardy and Heiniger (2014) in maize. However, LAI at 75 DAS was higher for the lowest seed rate in both years, because the wider plant spacing would have improved assimilates availability to individual plants as a result of less competition between the plants. Similar results have been reported by Tollenaar, Aguilera & Nissanka (1997) in maize.

Cutting management

At 75 DAS, cutting had reduced LAI in both years because of the removal of photosynthetic tissues. Similar results have been reported by Donaghy, Turner, and Adamczewski (2008) in tall fescue. Surprisingly LAI for twice cut plants at 75 DAS in 2018 did not vary with that of uncut plants but the reason for this is unknown. Contrasting results were reported by Jardel et al. (2017) in sorghum where LAI increased with cutting.

Interactions

There was a significant interaction between sowing date and cutting management at 75 DAS in both years because longer exposure to light and better carbohydrate reserves allowed the production of a greater LAI in uncut plants. Similar results to the present study were reported by Koireng, Ansar Ul-Haq, and Devi (2018) and Tahir, Ahmad, Khaliq and Cheema (2019) in maize. Further in 2018, the significant interaction between sowing date and seed rate was because delayed sowing decreased the LAI at higher plant density due to interplant competition. A similar result was reported by Yarnia (2010) in amaranth where leaf area decreased at higher plant density. The interaction between seed rate and cutting management in 2018 was because more leaves in uncut plants intercepted more light at the higher seed rates. The significant interaction among sowing dates, seed rate and cutting management occurred because there were more leaves in uncut plants at the lowest seed rate due to lower inter plant competition and a longer photoperiod for the earliest sowing.

2.5.4 Effect of sowing dates, seed rates and cutting management on leaf number per plant

Sowing date

In the present study, the April sowing produced the highest number of leaves at 45 DAS in both years. The April sowing allowed a higher GDD accumulation (Table 3.1-3.4, Chapter 4), as March

had an average mean temperature of 3.6°C lower than April (Figure 2.1). Fewer leaves reflects the impact of temperature differences, because leaf number depends on temperature, day length and soil moisture (Al-Darby & Lowery, 1987; Bonaparte, 1975; Duncan & Hesketh, 1968; Hesketh, Chase, & Nanda, 1969). However at 75 DAS, the leaf numbers did not differ among sowing dates, which is similar to the findings of Eik and Hanway (1965) in maize. The difference in the two years result can be explained by the variation in weather parameters.

Seed rate

There was a significant effect of seed rate on leaf number at 45 DAS in 2018 where increasing plant density decreased leaf number because of increasing interplant competition. This result is in line with the findings of Bonaparte and Brawn (1976b) and Eik and Hanway (1965) who reported a decrease in leaf number with increases in plant density in maize. The non-significant effect of seed rates at 45 DAS in 2017 might be because in that season there was little competition between the plants. This is similar to the findings of Dogan et al. (1997) and Iptas and Acar (2003) who reported no effect of plant density on leaf number in maize. However by 75 DAS, leaf number was lowest at the highest seed rate in both seasons, again a plant density response (Cuomo et al., 1998; Iptas & Acar, 2006).

Cutting management

Cutting significantly reduced the leaf number in teosinte at 75 DAS in both years which is because cut plants did not have enough reserves to support more leaves. Ansa and Garjila (2019) reported a strong negative relationship for number of leaves and cutting in Pennisetum grass.

Interactions

In 2018, the interaction between sowing date and cutting management occurred at 75 DAS because cutting reduced the leaf number for all sowings.

2.5.5 Effect of sowing dates, seed rates and cutting management on HY and DMY

Sowing date

Sowing date can change the plant environment because of differences in climatic parameters; sunshine hours, soil moisture, temperature and relative humidity. Thus sowing date is important because it influences the growth and development of plants (Sharma, Singh, & Sharma 2017).

In the present study April sowing produced the highest HY and DMY at 45 DAS in both years which as already explained was because the temperature allowed a longer growth period that

accumulated higher GDD and allowed greater plant growth (height, tillers and leaves). More tillers, leaves, and higher LAI allows better photosynthetic activity that enhances the HY and DMY. Similar results have been reported in fodder oats where higher HY is reported following increases in plant height, leaf number and tillers per plant (Dubey et al., 1995). Low soil moisture and delayed emergence in the March sowing as a result of low temperature meant a greater use of seed reserves for respiration and hypocotyl growth, which reduces the size and weight of cotyledons resulting in decreased autotrophic growth (Durr, Boiffin, Fleury, & Coulomb, 1992). Further, low temperature delays activation of the amylase enzyme which is critical for seed emergence (Zhanda, 2017). Higher temperature and higher rainfall during the May and June sowings hampered early plant growth resulting in lower HY which is similar to the findings of Park, Kang, Moon and Jong (1987). DMY in forage crops is directly dependent on the climatic conditions, with precipitation being the most important (Jardel et al., 2017). Koireng, Ansar-Ul-Haq, and Devi (2018) reported decreased HY of forage maize when sowing was delayed. Similar results of higher HY in April sowing has been reported by Devkota, Pokharel, Paudel, Upreti, and Joshi (2015) in Chitwan, Nepal. In both seasons HY from the harvest at 45 DAS was higher than that at 75 DAS, which might be as a result of excessive rainfall after the first harvest at 45 DAS. Excess water can delay plant growth by damaging the regrowth and also results in poor root development due to aeration problems (Sprague & Dudley, 1988). Reduction in the DMY with increased cutting frequency is attributed to a reduced ability of the plant to provide resources for regrowth as has been explained in Sudangrass-sorghum hybrids by Beuerlein et al. (1968) and summer annuals. Further lower HY and DMY at 75 DAS can also be explained by climatic variations that affect the productivity of grasses. Temperature after the harvest at 45 DAS in March was higher than the temperature after 45 DAS for April sown plants which allowed better regrowth of March sown plants in 2018. This is in line with the findings of Jehangir et al. (2013) who reported variations in HY in the Poaceae family under different sowing dates. Similarly at 75 DAS in 2018, the March sowing produced the highest herbage for the same reason. The temperature for the March sowing at 75 days was 27.3°C which was higher than all the other sowings for the second cut. Lower HY for the April sowing at 75 DAS was a result of higher rainfall during the regrowth stage. This regrowth was damaged badly by the rain which reduced the HY. As teosinte is a thermophilic species, high biomass is accumulated at a relatively high temperature (Niazi et al., 2015). The difference between sowing date responses for HY and DMY between the two years can be ascribed to the differences in temperature and precipitation (Figure 2.1 and 2.2).

Seed rate

Seed rate is an important agronomic attribute as plant density exerts a strong influence on crop growth, because of its competitive effect on both vegetative and reproductive development (Farnia & Mansouri, 2014; Singh & Choudhary, 2008).

In the present study, HY and DMY at 45 DAS did not differ markedly amongst the seed rates in 2017, but in 2018 the two highest seed rates had a greater vegetative yield than the two lower seed rates. The reason for this is not clear, but is likely a population effect on leaves per tiller. While the HY and DMY per plant were higher for the two highest seed rates (Appendix A2.2), they had more tillers and leaves per square meter (Appendix A2.3) than the two lower seed rates. However this does not explain why no major response to seed rate was obtained in 2017. What is apparent is there were bigger plants at the 80kg ha^{-1} seed rate at 45 DAS in 2018 than 2017 (Appendix A2.2), perhaps because for some reason there was less competition for resources during early vegetative growth in 2018 than 2017. However by 75 DAS any initial effects of seed rate on vegetative production had disappeared, as the interrelationships between plant density and plant size (Appendix A2.2) produced a similar vegetative yield response irrespective of plant density. The reduced HY and DMY at 75 DAS in both years is because for all the plant populations, tillers and leaves per square meter were reduced by cutting. Higher herbage and DMY yield at higher plant population due to higher numbers of vegetative shoots have been reported by Cirilo and Andrade (1994), Grasybill, Cox, and Oti (1991), Graybill et al. (1991), Cox and Cherney (2001) and Koireng et al. (2018) in maize and Springer, Dewald, Sims, and Gillen (2003) in gamagrass. Similar results have been reported by Ipsilandis and Vafias (2005) and Walli and Relwani (1970). Environmental variability at 75 DAS in both years did not result in differences among the seed rates for HY and DMY.

Cutting management

Cutting is one of the main factors to influence HY and DMY (Patel et al., 2013) as it has a major effect on biomass synthesis. Cutting management is important from the perspective of regeneration potential of the crop as well as peak growth and yield. HY and DMY is strongly influenced by growth stage and cutting intervals during the life span of plants which are suitable for multi cutting during the season. The first cutting also determines the number of cuts and HY at each cutting, determining the herbage quality (Kumar, Singh, Kumar, Kumar, & Tripathi, 2017).

In the present study, one cut at 45 DAS produced more HY and DMY than the second cut at 75 DAS in both years. This is because plants at the first cut had 45 days of growth which provided enough time for photosynthetic activity by the plant compared to only 30 days of growth for the second cut. A similar response was reported by Demetrio et al. (2012), Jehangir et al. (2013) and

Singh and Dubey (2007) in oat, Kallenbach et al. (2002) in alfalfa, Singh, Sharma, Verma, and Joshi (1988) in teosinte and Guha, Sharangi and Debnath (2014) in coriander. Further, the regrowth after the first cut was thin and weak and broken more easily by rain and wind. Generally, more frequent cuttings decrease HY (Ahmad et al., 2016; Ventroni et al., 2010). In addition, Machicek, Blaser, Darapuneni, and Rhoades (2019) reported that while a shorter harvesting interval produces better forage quality, HY is lower.

Interactions

There were no interaction among the sowing date, seed rate and cutting management for HY and DMY at 45 and 75 DAS in either year.

2.5.6 Association of vegetative yield components with dry matter yield in teosinte

Plant height, tillers per plant, LAI and leaf number per plant were all important contributors for determining the total DMY of teosinte harvested at 45 and 75 DAS from different sowing dates in the current study. This response was similar to the response in silage maize and sorghum where there was a positive relationship between plant height and DMY (Goes, Rodrigues, Arf, Arruda, & Vilela, 2011; Jing, Christensen, Sorensen, Christensen, & Rubaek, 2019). However the difference was negligible at 75 DAS which might be because the competition effect was not as strong. Similarly, a positive correlation between DMY and tiller number observed in this study is similar to the findings of Vinutha, Kumar, Blummel, and Rao (2017) in sorghum. However a contrasting results was revealed by Silungwe (2011) where DMY had no association with the tiller numbers in various grasses. LAI is a reliable parameter for HY and DMY of herbage crops as an indicator of radiation and precipitation interception, energy conversion, and water balance. Leaf area is a main source of light interception in plants that determines plant efficiency in producing higher photosynthates and higher productivity (Gifford, Thorne, Hitz, & Giaquinta, 1984; Koester, Skoneczka, Cary, Diers, & Ainsworth, 2014 ; Singh, 1991). A positive linear relationship between LAI and fresh HY was observed in sorghum (Iqbal, Iqbal, Siddiqui & Maqbool 2018) and a positive relation between LAI and DMY was reported by Ayisi and Poswall (1997), Al-Suhaibani (2011) and Carpici and Celik (2010) in maize because of increased light interception by the crop canopy which is similar to the findings of the current study. Lower association between LAI and dry herbage yield ($r = 0.698$) reported by Moosavi, Seghatoleslami, and Moazeni (2012) is at par with the association between LAI and DM at 75 DAS ($r = 0.63$) in this study. In the study by Carpici and Celik (2010) there was a moderately positive association ($r=0.414$) between the leaf number and dry forage yield of maize which was lower than coefficient between leaf number and DMY at 45 DAS ($r=0.57$)

and higher as compared to the coefficient between leaf number and dry matter content of teosinte at 75 DAS ($r=0.27$) in the present study. As the number of leaves in each tiller increase, the dry matter production increases. Trimble (2019) reported that nearly 90% of the herbage (or dry matter) of a plant is contributed by leaves. This is because leaves boosts the photosynthetic activity, increasing the dry matter production through greater sunlight capture (Bingham & Lupton, 1987; Duncan, 1971). LAI was a significant contributor to DMY at 45 and 75 DAS in both years at the different seed rates in the present study. This response was comparable to the response in maize, which had a positive relationship between LAI and DMY (Li, Peng, Yu, Zhang & Fang, 2011; Song et al., 1998). The relationship between leaf number, LAI and dry matter in this study was also in line with the findings of Moosavi, Seghatoleslami, and Moazeni (2012) who reported a correlation coefficient between leaf number and dry herbage yield ($r = 0.431$) which was lower than the present findings.

In line with the findings of the present study, the relationship between the number of tillers and LAI was also well described by San-Oh, Mano, Ookawa, and Hirasawa (2004), who reported an increased LAI with increase in the number of tillers. However, Simon and Lemaire (1987) reported a negative association between tillering and LAI of perennial ryegrass and Italian ryegrass, and suggested that the tillering rate slowed down as soon as the LAI increased, which corresponds with almost complete blockage of solar radiation at the level of the tiller buds. Tiller number and plant height are two major components of a cereal crop that contribute to DMY. A positive correlation between plant height and tiller number has been recorded in this study. A similar relationship has been reported by Belay et al. (1993), Eunos et al. (1986) and Sandhu & Mangat (1985) in wheat. However a contrasting response between plant height and tiller number has been reported by Cui et al. (2004) and Richards (1988) in rice because of competition for limited nutrients required between tiller generation and stem elongation. Similarly, a positive correlation was recorded between plant height and LAI at both 45 ($r=0.859$) and 75($r=0.775$) DAS, which was similar to the findings in *Brachiaria decumbens* Stapf ($r^2=0.59$) by Coelho et al. (2020) and Galzerano et al. (2010) for *Panicum maximum* cv. Aries and *Cynodon nlemfuensis* ($r^2=0.88$). Engel et al. (1987) found that in a cool season grass, *Bromus inermis*, the LAI and light interception had a strong correlation with season of the year and fertilizer level, because herbage mass per unit area is highly affected by the light interception through higher LAI which elongates the tillers and thus the canopy became more erect. Further, a positive correlation between plant height and leaf numbers was recorded in this study, which is likely because plant height was partitioned into leaf number through increased internode length. As internodes increased leaf number increased as reported for maize (Rood & Major, 1981) and sorghum (Sinha, & Khanna, 1975) respectively.

Similar results have also been reported by Hesketh, Chase, and Nanda (1969) in maize, sorghum and Hungarian millet with a positive correlation between number of leaves on a plant and the plant size.

2.6 Conclusions

In these field trials, the effects of different sowing dates, seed rates and cutting management on herbage yield of teosinte was explored. The results showed that sowing date, seed rate, cutting management and their interactions significantly influenced plant growth and dry matter yield of teosinte. The study showed that at this site:

- Early sowing of teosinte (April sowing) at 60 kg ha⁻¹ and cutting twice produced 31.9 tonnes green herbage per hectare, suggesting this management should be used in the Terai and inner Terai regions of Nepal.
- The current average herbage yield from teosinte in Nepal is 40-45 mt/ha which is similar to the herbage harvest from the present study on seed crop (31.9t/ha). Under this agronomic management, higher herbage yield can be achieved under multiple harvests, if teosinte is grown separately as a herbage crop.
- This level of production could help reduce the herbage and energy requirement deficits of livestock across the southern lower plains of Nepal.
- However this herbage is from a crop established for seed production, and the crop management which produced the greatest green herbage yield per hectare will not necessarily be that which produces the greatest seed yield.

Chapter 3

Assessing the influence of the environment on growth and yield of teosinte (*Euchlaena mexicana*)

3.1 Introduction

Teosinte is considered the progenitor of maize, and its physiology closely resembles that of maize. However, as there are no reports on environmental effects on the growth and developments of teosinte, most of the literature cited in this chapter is that for maize.

More than 80% of the variation in agriculture productivity is due to inconsistency in weather conditions, especially for areas dependent on rainfall (Petr, 1991; Fageria, 1992). The major important agrometeorological components linked with agriculture are solar radiation, air temperature, growing degree days (GDD), precipitation and relative humidity (Hoogenboom, 2000). The relationship between climatic influences and crop features are a significant part of agro-climatological zoning. Maize growth is highly affected by temperature, photoperiod, water availability and radiation, providing nutrients are not a limiting factor (Tsimba, Edmeades, Millner, & Kemp, 2013). Maize is a warm climate crop and can grow under variable climates. It has a very quick growth with best results under modest temperatures and abundant water supply (Aldrich, Scott, & Leng, 1978). The flowering and maturity of maize depend on the sunshine hours and temperature at a specific location (Bonhaomme et al., 1994). By comparing and relating the agro climatic requirement of a particular crop, predictions can be made on the cropping pattern for a particular location (Moeletsi, 2004) that satisfy the different phases of crop development (Todorov, 1981). Sowing time is an important factor due to the variability in the environmental conditions which affect the vegetative and the reproductive period of a crop, thereby influencing the yield, yield components and seed quality (Bhuiyan, Mondol, Bahaman, Alam, & Faisal, 2008). Teosinte and maize are C₄ photosynthesis plants. C₄ plants have a higher efficiency in capturing environmental atmospheric carbon dioxide and converting it to biomass (Irving, 2015). The thermophilic nature of teosinte allows it to survive at high temperature and produce larger amounts of fresh herbage than maize (27% and 55% higher yield than maize) under non-stressed and stress conditions respectively (Niazi, Rauf, Silva, & Munir, 2015). Along with higher herbage yield, teosinte also has several other advantages like lower input requirements than maize and

better adaptability (flooding and drought) to different climates (Schmidt, Bowles, & Gaudin, 2016).

3.1.1 Temperature

Temperature regulates the expansion rate of plants and the commencement and end of certain developmental process is also determined by temperature (FAO, 1978). High temperatures throughout the growing season accelerate crop development, resulting in less herbage production and lower seed set and seed yield (Otegui & Melon, 1997). Low temperatures result in the predominance of vegetative development over reproductive development and limit the seed set and growth as a result of reduced photosynthesis. Thus the ratio between dry matter at silking and final seed number at lower temperature (Cirilo & Andrade, 1994) results in lower seed weight due to the decline of both radiation use efficiency and biomass partitioning to the seeds. For maize, optimum temperature ranges from 28°C-32°C for the complete growth period (Arnold, 1974; Yin et al., 1995). Minimum temperature for an entire season basis is 6.2°C (Olsen et al., 1993; Shaykewich, 1994) and maximum temperature is 42°C, beyond which maize growth stops (Yin et al., 1995). Uprety and Reddy (2016) reported that photosynthetic activity in C₄ crops like maize can tolerate higher temperature because of the lack of photorespiration that increases as the temperature increases and because of higher evapotranspiration during heat stress. The Rubisco enzyme in maize enables the conservation of intercellular CO₂ among the atmosphere and the location of carboxylation allows the maize plant to maintain optimal internal CO₂ concentration at minimum stomatal conductance, so that the rate of increase in evapotranspiration with the rise in temperature is much reduced (Uprety & Reddy, 2016). However, the maximum temperature threshold for photosynthesis is around 35°C.

3.1.2 Effect of temperature on plant height

Plant height depends upon the accumulation of nodes and internodes (Berghage, 1998). The initial internode elongation and the whole shoot development of maize are highly susceptible to high temperature, but plant height also decreases with a reduction in temperature during vegetative growth (Uprety, & Reddy, 2016). At elevated temperatures, the number of nodes increases with increasing temperature but the length of individual nodes is reduced by higher night temperatures and lower day temperature (Allen, Zhang, Boote, & Hauser, 2018; Shukla, Felderhoff, Saballos, & Vermerris, 2017) which is due to decrease in cell elongation rather than decrease in cell number (Erwin, Velguth, & Heins, 1994). The nodes increase as a result of leaves unfolding more quickly at higher temperature. A decrease in temperature to 10°C results in a rapid reduction of maize growth, which might be due to slow rate of cell division, or may partly be

due to a reduced translocation rate of food materials at low temperatures (Berghage, 1998). Seedling root and shoot biomass is decreased by 10% at soil temperature above 26°C for every degree rise until 35°C, where plant growth is severely delayed (Walker, 1969) which is due to poor reserve mobilisation and reduced protein synthesis (Riley, 1981). Heat stress in maize is accompanied by a shortened life cycle, increased respiration, reduced photosynthesis, reduced light interception and pollen sterility (Uprety & Reddy, 2016). Lower temperatures induce earlier flowering, fewer leaves, fewer nodes, and shorter plants (Bonaparte, 1975).

3.1.3 Effect of temperature on tiller number

The growth of tillers is affected by temperature, but at a constant temperature between 7°C to 32°C (Mitchell, 1956), this however depends on the species. In wheat, increase in temperature from 10° to 25°C increased the relative rate of tillering (Friend, 1965). Very high temperature results in failure of tiller bud emergence which results in low tillers in wheat because at temperature above 10°C to 20°C, the cell division process accelerates which increases the assimilate requirements of the plants main axis rather than the tiller due to a surge in the level of leaf primordium formation (Friend, Helson, & Fisher, 1962). Higher supply of assimilates to the main axis limits the supply of assimilates to tiller buds and root development which increases the apical dominance in plants. Inversely at low temperature, rate of tiller growth decreases which increases the duration of tiller growth leading to a delay in flowering of the plant (Friend, Helson, & Fisher, 1963). In some grasses, lower day temperature reduces the development of an individual tiller more than lower night temperatures (Lucanus, Mitchell, Pritchard, & Calder, 1960).

3.1.4 Effect of temperature on leaf number

Leaf numbers per plant are also affected by temperature (Tollenaar & Hunter, 1983; Stevenson & Goodman, 1972). High temperature affects leaf canopy expansion (Tollenaar, Daynard, & Hunter, 1979; Warrington & Kanemasu, 1983) and ultimately affects the leaf area index. A rise in leaf number with increasing mean daily temperature from 15- 30°C has been reported by Hesketh, Chase, and Nanda (1969). Allen, McKee & McGahen (1973) have suggested that night temperatures exert a higher influence on leaf development than day temperatures. Longer photoperiod increases the number of leaves (Warrington, & Kanemasu, 1983). Newton (1958) reported that higher temperature expands the leaf area through cell expansion and cell division where cell division plays a major role. Further high temperatures activate enzymes and increase the rate of leaves unfolding (Milthorpe, 1959). Bonaparte (1975) showed that mean leaf number increase did not differ at temperature regimes of 18/10°C to 26/10°C, but significantly increased

when temperature was increased from 26/10 to 34/10°C. However, leaf numbers have an inverse relationship with flower induction (Duncan & Hesketh, 1968). Bonaparte (1975) reported that the earlier the flower induction, the fewer leaves will be developed. For maize leaf initiation minimum temperature on a whole season basis is 7.3°C, optimum temperature is 31.1°C, and maximum temperature is 41.3°C, beyond which leaf initiation stops (Sanchez, Rasmussen, & Porter, 2014).

3.1.5 Effect of temperature on tassel emergence and seed filling

With an elevation in temperature during the crop reproductive phase, the growth of plants is accelerated but the seed mass is reduced due to the shorter seed filling duration (Hampton, Boelt, Rolston & Chastain, 2013). Temperature during tassel emergence is very important to maize yield because the seed number per cob is determined during this phase (Tollenaar & Brulsema, 1988). A negative correlation has been reported between maize vegetative growth and floral initiation during long hot days (Acosta & Crane, 1972; Hallauer & Sears, 1972). Warrington and Kamemasu (1983) reported that minimum temperature for tassel emergence for maize was 8°C and the optimum temperature was 28°C. Ellis, Summerfield, Edmeades, and Roberts (1992) in an experiment on 12 cultivars of maize found optimum temperature for tassel initiation ranged from 19-22°C for cultivars adapted to cool environments, but was 31°C for a tropical low land cultivar adapted to warm conditions. The influence of high temperature is more evident before and during anthesis (Sanchez, Rasmussen & Porter, 2014) in maize. Bonhomme, Dereux, and Edmeades (1994), Coligado and Brown (1975), and Cutforth and Shaykewich (1990) all have reported that the temperature range between optimum and maximum temperature from maize seedling emergence to tassel initiation may vary by 6°C to 8°C. Muchow, Sinclair, and Bennett (1990) reported an increase in temperature reduces the length of the seed filling period in maize which is because at high temperature the ability of seeds to store the available photo-assimilates is reduced (Magalhaes & Jones, 1990a, 1990b). Aitken (1980); Kiniry, Ritchie, Musser, Flint, and Iwig (1983) reported that maize is a short-day plant which responds inversely to increasing temperature for tassel initiation. Diurnal temperature variation modifies the period of tassel emergence. The rate of tassel emergence increases with an increase in temperature but the number of days to complete tassel emergence is substantially reduced at higher temperature regimes of 18/10°C and 34/10°C (Bonaparte, 1975). High temperature affects pollination seriously. Temperatures beyond 30°C result in pollen shedding earlier than silk emergence as silking is delayed. Reduced pollen viability and poor silk acceptability results in decreased seed set and reduced yield (Shrestha, Gurung, & Dhital, 2018). Further at high temperature pollen viability and silks receptiveness is abridged which leads to a major decline in the rate of seed setting, reducing seed yield (Waqas et al., 2021). At temperatures 32°C and above the percentage of non-

germinated pollen can be up to 51% in maize (Schoper, Lambert, & Vasilas, 1987). High temperatures reduces the pollen water potential where pollen grains drop in the pollen tube and germination occurs in the tube (Dupuis & Dumas, 1990). Moderate temperatures at pollination allow active transmission of pollen to the silks but elevated temperature causes stress resulting in flower abortion and lower seed yield (Zhanda, 2017). For maize tassel initiation, minimum temperature on a whole season basis is 9.3°C, optimum temperature is 28.3°C, and maximum temperature is 39.2°C beyond which the tassel will die. For seed filling, maize has a minimum temperature of 8°C, optimum temperature of 26.4°C and maximum temperature of 36°C (Sanchez, Rasmussen, & Porter, 2014).

3.1.6 Effect of temperature on seed dry weight

Temperature also affects the seed dry weight at physiological maturity, because the increased seed growth rate under increased temperature is related to a decline in the duration of seed growth (Badu-Apraku, Hunter, & Tollenaar, 1983). The duration of seed growth or seed filling is delayed at lower temperature because assimilate supply for seed growth is less (Tollenaar & Daynard, 1978). When the air temperature is higher than 25°C, a subsequent decline in seed growth rate in maize has been reported (Badu-Apraku, Hunter & Tollenaar, 1983). A reduction to 3 to 4% in seed yield has been reported by Shaw (1983) with every 1°C increase in temperature above the optimum (25°C) in maize. Duncan, Shaver, and Williams (1973) reported that increased night temperature reduces seed yield in maize. Brooking (1993) reported a linear increasing response of seed filling rate between 13°C and 32°C, but it decreased below 13.5°C.

3.1.7 Effect of weather components on seed quality

Seed quality play a major role in crop production. High viability and good vigour can only be obtained from healthy seeds. Seed quality is influenced by various environmental components like temperature, rainfall, humidity, solar radiation and GDD (Krishna, & Surya Rao, 2005). Higher temperature during seed development may affect the germination percentage of seeds (Begcy, Sandhu, & Walia, 2018). High temperature diminishes the capacity of the plant to supply the essential assimilates to produce the storage compounds crucial for later germination due to a shortened seed filling duration (Hampton, Boelt, Rolston & Chastain, 2013). Sometimes, higher temperature might also damage the seed physiologically resulting in loss of germination (Dornbos & McDonald, 1986). This is because high temperature during seed development and maturation reduces the ability to produce the storage compounds that will be required later in the germination process (Dornbos & McDonald, 1986). High temperature during seed development lowers the shelling percentage, reduces the thousand seed weight and oil content, but improved

the seed protein content in maize (Uprety & Reddy, 2016). Starch deposition in the seeds is reduced following exposure to higher temperature during the 1-10 days after pollination resulting in lower weights of seed (Lu et al., 2014). In *Paspalum dilatatum* (a C₄ grass) high temperature affected the floret fertility. Above 27/22°C, rapid flowering reduced the panicle and raceme elongation, reducing the seed yield and quality (Pearson & Shah, 1981).

High temperature effects on seed germination can be explained by the reduced enzyme activity and higher respiration at higher temperature. At 41°C seeds will still be normally imbibing and have ATP levels that are adequate for germination. But the specific activities of some vital enzymes are lower, along with a reduced rate of protein synthesis as compared to seeds imbibing at 28°C. This reduced rate of protein synthesis in the embryos of tropical seed is called temperature sensitivity (Riley, 1981). Plants under drought stress produce smaller or medium sized seeds which have lower germination and decreased seed vigor (AA-germination) (Alqudah, Samarah & Mullen, 2010). Smiciklas et al. (1992) stated that drought at the beginning of seed filling reduces seed germination, seedling dry weight, and vigor (Alqudah, Samarah & Mullen, 2010) of seed because drought results in shift of enzymatic activity of seed which change the metabolic pathways in seed germination (Almas, Bagherikia, & Mashaki, 2013; Botia, Carvajal, Cerda, & Martinez, 1998).

3.1.8 Effect of solar radiation on growth and development

Photosynthesis, a process required by all plants is governed by solar radiation for carbohydrate partitioning and biomass development of each plant component (Hoogenboom, 2000). Photosynthetic photon flux density affects plant biomass production (Bergamaschi et al., 2010), and conversion of solar radiation to chemical energy by the photosynthetic process. Solar radiation is lowered by environmental factors like cloud cover and rainfall during the early growth phase, flowering and seed development. In maize, lower solar intensity during early seed development and seed filling results in lower yield due to fewer seeds (Lindquist et al., 2005), lower seed weight and premature plant death (Arora, Swami, & Bhan, 2017). Further, duration of solar radiation is also responsible for the growth and development of an individual leaf and the entire canopy (Hatfield, 1977). The higher leaf area index of a full canopy allows a greater amount of sunlight capture for photosynthesis. Higher solar radiation enhances herbage and seed quality. Longer solar radiation exposure of plants during stem elongation and seed filling results in higher thousand seed weight while exposure to lower intensity of solar radiation negatively influences seed yield (Lekes, 1984). Rolling up of leaves due to a moisture deficit in maize plants reduces solar radiation interception (Müller & Bergamaschi, 2005). Prolonged water deficit also reduces

leaf size and number, reducing the solar radiation interception (Collinson et al., 1999). Higher solar radiation causes increased transpiration from the stomata, so stomata close to conserve the water, and if they are closed for a prolonged period of time, the plant dies. In environments with high temperature, leaf anatomy and biochemistry of C₄ plants allows the binding of CO₂ when it enters the leaf producing a four compound carbon. This compound then transfers and concentrate the carbon dioxide in specific cells around the rubisco enzyme in C₄ plants. This enzyme increases the photosynthetic and water use efficiency making C₄ plants more productive at higher temperature and solar radiation (Zhu, Long, & Ort, 2008).

The higher intensity of radiation increases the photosynthetic activity in C₄ plants. Tiller production is highly affected by light intensity; the higher the light intensity during plant growth, the higher will be the rate of growth and rate of tillering (Aspinall, & Paleg, 1964; Khalil, 1956). Tiller numbers decrease with decreasing light intensity. Similarly, tiller numbers were reduced by exposure to long days and lower light intensity (9 hour photoperiod to 18 hrs) in orchard grass (Auda, Blaser & Brown, 1966).

3.1.9 Effect of precipitation on plant growth and development

Precipitation modifies the environment in which the plant grows but does not affect any of the plant growth and developmental process directly (Moeletsi, 2004). Both low and high precipitation is harmful to the crop. Water requirement depends on the species and stage of growth of the plant. During a drought period, stomata on the leaves close to prevent transpiration, causing reduced gross carbon assimilation, which modifies the biomass partitioning to the different plant parts (Hoogenboom, 2000). Maize can tolerate water deficits during the vegetative and ripening periods (Doorenbos & Kassam, 1979) but is susceptible to water stress during the seed filling period which reduces the grain yields (Cakir, 2004; Zaidi, Rafique, Rai, Singh, & Srinivasan 2004). However, adequate water is required for optimum production during the entire growing season (Neild & Newman, 1990). In early vegetative growth, excess water retards the vegetative growth by injuring the plants and also results in poor root development due to aeration nutrition problems (Sprague & Dudley, 1988). However moderate water stress at the early vegetative stage might be beneficial because such stress boosts early season root growth which might be beneficial later in the season when there may be water stress (Shaw & Newman, 1991). A prolonged period of water logging closes the stomata, reduces leaf area, and causes chlorosis, reduced root growth and ultimately plant death. During water logging, anaerobic conditions occur which damage the root mainly due to the accumulation of toxic products like lactic acid in the plant (Srinivasan, Zaidi, Singh, & Sanchez, 2004). Similarly, excessive water causes a deficiency of

oxygen in the soil which affects the root activities causing senescence of roots (Hoogenboom, 2000; Lauer, 1998). Further Kramer (1963) reported that water stress during vegetative growth will result in loss of turgidity in cells which stops cell enlargement resulting in smaller plants. Reduced turgidity might also change the pattern of plant growth. However, water requirements in the later vegetative stage will be high because evapotranspiration from crops will be higher than the evaporation from the soil surface (Sprague & Dudley, 1988). Plant height and leaf area development were reduced by water stress in maize. Water deficits during rapid vegetative growth have been reported to reduce plant height and leaf numbers. It also results in seed yield losses of up to 28-32% in maize (Cakir, 2004).

Moisture stress is also related to a reduced number of days through any phenological stage and the number of days to crop growth indices (Ihsan, Nakhlawy, Ismail, Fahad, & Daur, 2016). Decreasing water availability reduces the biomass production in plants as it affects the crop during the vegetative growth eventually resulting in a reduction in final biomass (Turner et al., 1986). A delay in flowering results from water stress (Wopereis, Kropff, Maligaya, & Tuong, 1996; Winkel, Renno, & Payne, 1997). Moisture stress adversely affects flower induction and pollen production, which consequently leads to reduced fertilization and hence seed set (Samarah, Alqudah, Amayreh, & McAndrews, 2009; Sheoran & Saini, 1996). Decreased pollen grain availability (Trueman & Wallace, 1999) and increased pollen grain sterility (Al-Ghzawi, Zaitoun, Gosheh, & Alqudah, 2009) result from water deficits. Further, it results in drying of the silk and reduced seed yield (Sprague & Dudley, 1988). After seed filling the water requirements of crops reduces greatly because the seeds start desiccating after physiological maturity (Hoogenboom, 2000). Low moisture can result in seed abortion which decreases the seed number (Zinselmeier, Jeong, & Boyer, 1999). Further, it can reduce the level of assimilates available to the emerging ear which are essential to ensure seed formation and growth (Boyer & Westgate, 2004; Schussler & Westgate, 1995) resulting in yield loss due to lower seed numbers (Herrero & Johnson 1981; Kamara, Menkir, Badu-Apraku, & Ibikunle, 2003; Westgate & Boyer 1985). Between 45-72% reductions in seeds per ear in maize due to water stress have been reported by Schussler and Westgate (1994).

3.1.10 Effect of relative humidity (RH) on growth and development

Relative humidity is also an important agro meteorological aspect as it determines the quantity of water present in the air which can affect plant growth and development (Hirai, Okumura, Takeuchi, Tanaka, & Chujo, 2000; Tibbitts, 1979). Relative humidity mainly influences the transpiration water loss and stomatal opening of plants. These two physiological activities affect

the plant temperature, water potential, nutrient translocation, photosynthesis, and moisture condensation (Tibbitts, 1979). The vapour pressure of air inside the leaf surface is believed to be saturated (100 % RH) but any decrease in the atmospheric moisture increases the vapour pressure in the leaves, resulting in transpiration loss by the leaves. However, if the plants are exposed to a higher vapour pressure deficit, they will transpire more water. Transpiration is proportional to temperature; the higher the temperature, the higher will be the transpiration, causing excessive water loss resulting in water stress (Mitchell, Robotham, & Warrington, 1976; Forde, Mitchell, & Edge, 1977; Schulze et al., 1974). Response to vapour pressure deficit is different in individual species but in maize the stomatal opening has been reported to decrease with an increase in transpiration (Tibbitts, 1979). A dry atmosphere lowers the plant water potential and if this situation persists during fertilization and early embryo enlargement, there will be a severe decline in seed yield (Hoffman & Rawlins, 1970). Photosynthesis increases with increasing humidity (Bunce, 1984) because the stomatal conductivity is accelerated at higher humidity. Lower humidity results in lower carbon dioxide exchange by the plants as a result of excessive water loss, resulting in stomatal closure. A single leaf may not show the significant effect of relative humidity, but the effect is visible in a whole plant because the water uptake by the plant is never satisfied by the transpiration loss at lower humidity as reported in barley (Rawson et al., 1977). Higher humidity is reported to increase the leaf area per unit of plant dry weight although the assimilatory ability of the leaf area (net assimilation rate) was reduced (Acock, 1974). However this differs with species. Ford and Thorne (1974) reported a slight increase in plant growth by increasing the humidity from 0.7 k Pa vpd to 0.23 kPa vpd (at 20°C) and growth was decreased at lower humidity. Singh, Kunze, and Satyanarayana (2011) have reported that with low humidity accompanied by high-temperature, silk and pollen grains can wither when released from anthers due to their thin outer membrane. At harvest maturity, both air and dew point temperature affect the drying period of the harvested product (Sheriff, 1977). Humidity lower than 55-90% RH results in lower growth rate as a result of plant water stress but higher RH provides a favourable conditions for disease and disorders in plant growth and development (Grange & Hand, 1987).

3.1.11 Effect of sowing date on phenology of plants

Maize is a periodically sensitive short day plant (Garner & Allard, 1923; Kuleshov, 1933; Kiesselbach, 1950; McClelland, 1928) and it responds to photoperiod only after the juvenile stage, which is followed by a photoperiod sensitive stage termed the tassel initiation stage. The duration of both of these stages is determined by thermal time. Temperature variation during various sowing dates of forage maize regulates the heat availability to the plants during their growth period, especially during the period from planting to silking (Birch, Hammer & Rickert 1998;

Omafra, 2009). Sowing time is one of the important factors that oversees the crop phenological development (Dar, Brar & Yousuf, 2018). Environmental changes related to various sowing dates (sunshine, temperature) modify the growth and expansion of the maize plant. Early sowing as well as late sowing hampers maize sowing, as it will face a lower temperature and higher humidity, and the vegetative growth will be poor due to longer days from sowing to emergence (Wann, 1986). Further, sowing dates alter the thermal and radiative environments during growth (Cirilo, & Andrade, 1994) and the temperature influence on plant growth along with the developmental process (FAO, 1978). Reduced grain yield in early or late sowing of maize has been reported by Lauer et al. (1999). Yield reduces as a result of poor emergence and early plant growth for an early sowing and late planting might hamper the yield due to lower temperature reducing the seed filling period (Maddonni, Otegui, & Bonhomme, 1998; Shim, Lee, Koo, Shin & Yoon, 2019). The cumulative heat during the growth period is termed Growing Degree Days (GDD). It is the daily mean temperature, beyond a base temperature appropriate for plant growth (Shaykewich, 1995). GDD is used as a method for predicting the phenological stages of crops. A crop requires a certain number of units of heat to complete a phenological stage of its growth (Crane, Goldsworthy, Cuany, Zuber, & Francis, 1977; Rao, Singh & Singh, 1999) and this can be used to measure the growth stages of plants (Sunil & Sarma, 2005). GDD are used as an indicator for crop growth estimation (Dar, Brar & Yousuf, 2018) and change with growth stage (McMaster & Wilhelm, 1997). MacAdam and Nelseon (2003) proposed that the base temperature for C4 grasses like maize is 10° C. The base temperature is the lowest commencement temperature at which growth of plant starts (Borras, Maddonni, & Otegui, 2003).

The climatic variable effect on the growth and development of teosinte is now a research area. Establishing a direct link between one of the climatic components with the plant yield is very difficult as the herbage and seed yield depend on a great number of other factors. Further, photoperiodic response of teosinte limits its production and distribution in temperate climates. Therefore altering sowing date may overshadow the effect of one climatic component on the growth and yield of teosinte.

3.1.12 Estimating the Growing Degree Days (GDD) for critical phenological growth stages of teosinte

As a change in climatic conditions can impact crop production (eg. due to a rise in temperature), therefore a precise estimate of growth and development of a crop is a key to determine the crop adaptation and yield potential (Kumudini et al., 2014). Increasing temperature has a major effect on crop phenology which is usually expressed as a rate or duration and determines its adaptability

to a particular location, its ability to mature and also set seed within a growing season (Craufurd, & Wheeler, 2009). Although teosinte phenological growth responses may differ with the variety used, measuring the effect of temperature on each phenological growth of teosinte and predicting the time and length of critical periods of its growth is necessary. This will allow the prediction of stages of crop development for efficient management decisions and assessment of climate change impacts (Tonnang, Makumbi, & Craufurd, 2018). This estimate is based on the postulation that growth rate slows down when the temperature decreases, within certain ranges, and if the temperature declines enough, the growth will stop at the plant's lowest growing threshold; which is frequently called the base temperature. As temperature increases, growth rate accelerates until temperature reaches the optimum, beyond which the growth rate declines and ultimately stops at a point called maximum temperature (Tonnang, Makumbi, & Craufurd, 2018). This led to the addition of the mean daily air temperature for an identical number of months in a location, which headed to the evolution of the degree-day concept as the values of the growth rate of plants attained from year to year were roughly constant. This GDD concept is regarded as the functions of plant development rate (Montagnes, Kimmance, & Atkinson, 2003) and is used to predict crop development, which is the function of temperature at each critical growth stage of the plant on a daily basis (McMaster & Wilhelm, 1997; Yang, Logan, & Coffey, 1995).

3.2 Objectives

This objective for this study was to determine the influence of environmental factors on the phenological growths and vegetative yield of teosinte as influenced by different sowing dates. The effects on reproductive yield will be reported in Chapter 4.

3.3 Methodology

3.3.1 Weather records

As reported in Chapter 2, the experiments were conducted in 2017 and 2018 at the forage experimental site of the National Cattle Research Program (NCRP), NARC Bharatpur Metropolitan -15, Rampur, Chitwan, Nepal. The farm is situated in the central region of Nepal at 27° 65' N Latitude and 84° 35' E Longitude with an altitude of 228 m above mean sea level at Rampur, Chitwan District which is 157 km from Kathmandu, the capital city and lies southwest from the city of Narayangarh (DDC profile, 2018).

Daily weather data (precipitation, maximum and minimum temperature, and humidity) were obtained from the agro meteorological station installed at the National Maize Research

Programme (NMRP), NARC, Rampur located 200 meters away from the research field (NMRP, 2017; NMRP, 2018). Sunlight radiation data were adapted from NASA Power 2017 and 2018 and converted to photosynthetically active radiation (PAR) by multiplying the solar radiation by 0.48 (Andrade, Uhart, & Cirilo, 1993; Monteith, 1972).

3.3.2 Experimental design

The experimental design has been described in Chapter 2.

3.3.3 Growth and development stages of teosinte

Teosinte was divided into five development stages from seedling emergence to seed maturity. The phenological development of teosinte across these five stages was studied on the basis of heat accumulation (GDD) during each of these stages of plant growth. The phenological stages were determined using the recorded dates.

The first stage (Stage 1) of teosinte development was from sowing to seedling emergence. The time required for emergence depends on moisture conditions, soil temperature, depth of planting, and seed vigour (Diawara, 2012). Stage 1 was achieved when 80 % of the seeds sown had emerged from the soil. The second stage (Stage 2) is the vegetative growth stage, defined as the number of days from seedling emergence to when 5% of the plants had started flowering. The third stage is the flowering stage (Stage 3) from when 5 % of plants in each plot had one open flower until there was no further increase in flower number. The fourth stage (Stage 4) was the seed filling stage. This stage was from the end of flowering to Physiological Maturity (PM) of seed. Seed PM was defined as that date when more than 70 percent of the seeds had attained maximum dry weight (Ghassemi- Golezani, Tajbakhsh, & Yaeghoob, 2011) and the colour of the teosinte seed had changed from white to shiny brown. The fifth (Stage 5) and last stage is the maturity stage, the time from physiological maturity to harvest maturity. Harvest maturity was when the individual seed inside the ear could be easily separated from each other but still were inside the covering. Each of these five stages were defined on the basis of GDD which was calculated using the McMaster and Wilhelm (1997) methods. McMaster and Wilhelm (1997) gave two methods to interpret the equation for calculating GDD: The first is, if the daily mean temperature is less than the base, the daily mean temperature is set equal to the base temperature. The second is if T_{max} or $T_{min} < T_{base}$ they are reset equal to T_{base} . Thus GDD is calculated using the formula $(T_{max} + T_{min})/2 - T_{base}$ temperature, where T_{max} , T_{min} and T_{base} are the maximum, minimum temperature and base temperature respectively. Because T_{base} for

teosinte was not known and therefore the base temperature of maize (10°C) was used (Andrew, Ferwerda, & Strommen, 1956).

3.3.4 Influence of sowing date on vegetative growth of teosinte

The influence of sowing date on vegetative growth was studied by examining the relationship of sowing dates with vegetative growth parameters (plant height, LAI and DMY) in 2017 and 2018. These three yield components were used as a means of comparing growth rate across the sowing dates. Measurements were taken at 45 DAS and 75 DAS during both years at the time that vegetative cuts were taken (see Chapter 2). The data for these three growth attributes were then plotted as a function of GDD.

3.3.5 Estimation of GDD for phenological growth stages of teosinte

The phenological development of the teosinte crop from emergence through flowering to maturity was recorded. The sowing dates of 30th March, 30th April and 30th May and 30th June in 2017 and 30th March, 30th April, 30th May 2018 were used in this study. Temperature and time during each phenological growth stage were recorded as these two components are the major factors affecting the teosinte growth. The overall growth rate for each phenological stage was calculated on the basis of cumulative GDD and the estimate was made by averaging the value for each growth stage in both years across all sowing dates. The growth assessment of each phenological stage of teosinte was done as described in the growth and development stages of teosinte section above.

3.3.6 Statistical Analysis

The daily mean, maximum and minimum monthly temperature were calculated by averaging the daily temperature recorded for each sowing date. Similarly, total rainfall was a sum of all days in each month and monthly average was calculated by taking the average for all days in each month. Average humidity and solar radiation inception were also averaged for all the days each month. GDD was calculated by adding the temperature each day from seedling emergence to seed harvest date.

3.4 Results

3.4.1 Environmental conditions during the growing season

Environmental variables were recorded during the experimental period from March 30, 2017 to December 30, 2019. In the 2017 -2018 growing season, monthly mean temperature ranged from a

high of 20.9°C in March to a low of 17.6°C in December but for 2018, the monthly mean temperature ranged from a high in March of 22.8°C and a low of 15.7°C in December. Monthly maximum temperature ranged from 27.4°C in March to 22.6°C in December and the monthly minimum temperature ranged from 14.4°C in March to 12.7°C for December in 2017. Monthly maximum temperature in 2018 was higher in March by 2.7°C and it was lower in December by 1.4°C. Monthly minimum temperature in March 2017 was 1°C lower than in March 2018 and 2.7 °C higher than in December 2018. Events of very high day temperature mostly occurred during early vegetative growth in the months of May (33.5°C) to September (27.6°C) in 2017 and from May (31.8°C) to September (27.6°C) in 2018. Total rainfall received during 2017 was 1447.8mm and monthly rainfall ranged from 52.2 mm in March to 1.4mm in November. Out of the total rainfall in 2017, 88% of this rainfall was concentrated between March to September. Similarly, total rainfall received during 2018 was 1191.3 mm and monthly rainfall ranged from 17.0 mm in March to 0.7 mm in November. Out of the total rainfall received 87 % of it was concentrated between April to September. The monthly mean relative humidity (% RH) ranged from 36% in March to 90 % in August in 2017. Similarly, in 2018 the lowest RH occurred in March (27%) and the highest in August and September (87%). The average photosynthetically active radiation (PAR) was lowest (21.4 MJ/m²/day) in December and highest (40.7 MJ/m²/day) in June (Figure 3.1 and 3.2).

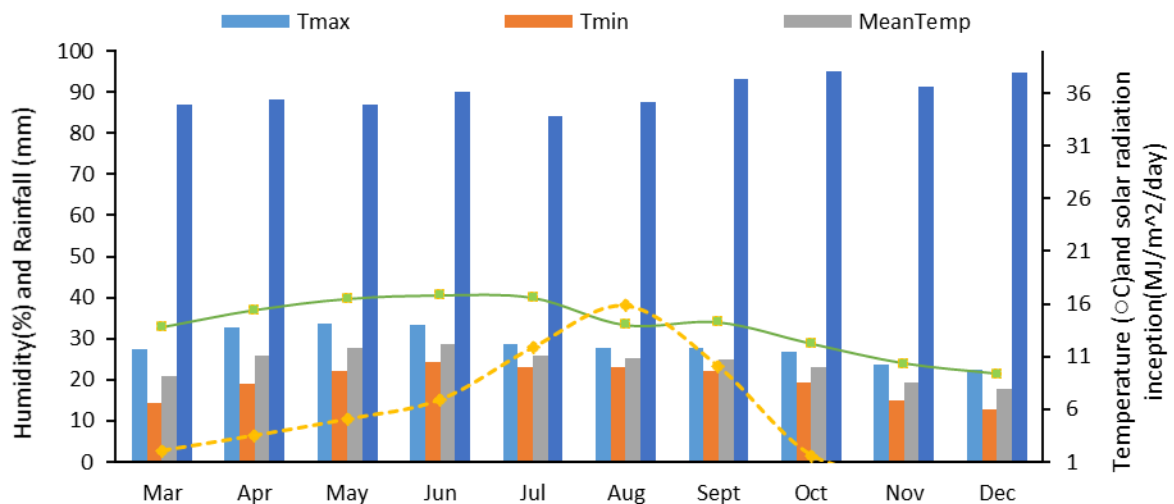


Figure 3.1 Weather data during teosinte growth period (March 2017-December 2017)

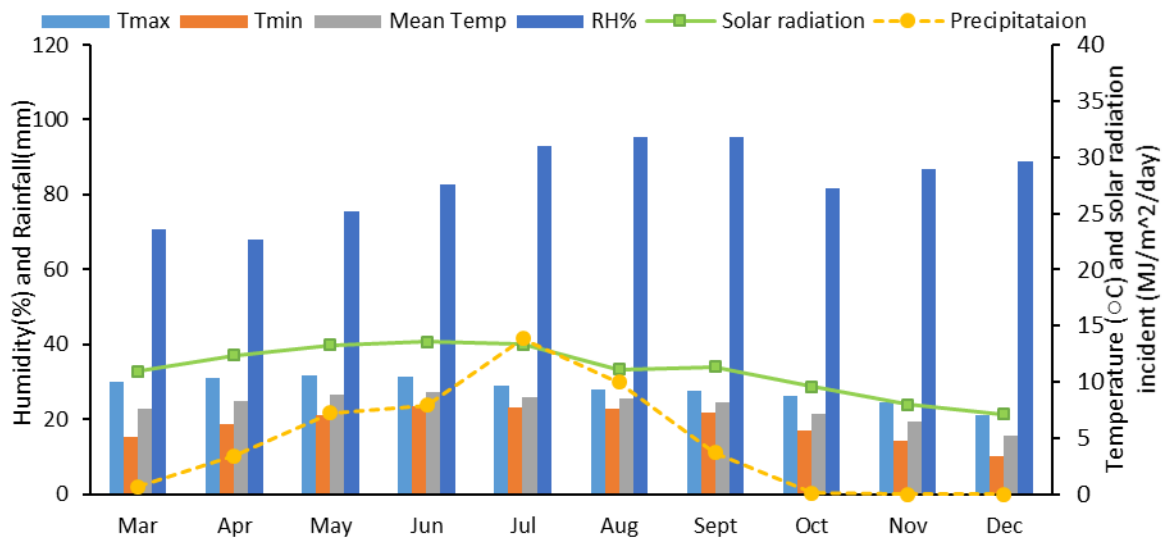


Figure 3.2 Weather data during teosinte growth period (March 2018-December 2018)

3.4.1.1 Environmental conditions during sowing to emergence (Stage 1)

The mean temperature in 2018 during stage 1 was slightly lower than that of 2017. The mean temperature varied from 24.5 -25.7°C for the March sowing, 25.0 -26.4°C for the April sowing and 26.5 -29.1°C for the May sowing in 2017 and 2018 respectively. It was 26.2°C for June 2017. The mean maximum temperature during stage 1 in 2017 was 32.7°C, 32.6°C, 29.5°C and 28.9°C for the March, April, May and June sowings. In 2018 it was 30.5°C, 30.1°C and 30.5°C for the March, April and May sowings. The mean minimum temperature during 2017 for each sowing was 18.7°C for the March, 20.1°C for April, 24.7°C for May and 23.4°C for June. Similarly, the mean minimum temperature in 2018 was 18.4°C, 19.9°C and 22.4°C for the March, April and May sowings. The total precipitation during stage 1 in 2017 and 2018 was 56 mm, and 36 mm for the March sowing; 35mm and 47mm for the April sowing and 55 mm and 90 mm for the May sowing respectively. The June sowing in 2017 had a precipitation of 104 mm. The average precipitation for the first, second and third sowing in 2017 and 2018 was 5.6 mm and 3.5 mm, 4.5 mm and 5.9 mm and 6.9mm, and 11.3 mm respectively. June sowing in 2017 had an average precipitation of 14.9mm. Average relative humidity during this stage was 37 % and 39% for the March sowing; 39 % and 57% for the April sowing, 51% and 71 % for the May sowing in 2017 and 2018 respectively. The average relative humidity in June 2017 was 85%. PAR in 2017 was 17.2, 18.7, 19.5 and 19.5 MJ/m²/day in March, through June sowing. For 2018, it was 17.0, 18.7, 19.5 MJ/m²/day for the March, April and May sowings (Figure 3.3 and 3.4).

3.4.1.2 Environmental conditions during vegetative growth (Stage 2)

The mean temperature during stage 2 varied from 26.0 -26.7°C for the March sowing, from 26.1-26.2°C for the April sowing, from 26.0 -26.1°C for the May sowing and was 25.2°C for the June sowing. The mean maximum temperature during stage 2 in 2017 was 30.9°C, 30.3°C, 29.2°C and 28.0°C for the March, April, May and June sowings. In 2018 it was 30.0°C, 29.6°C and 29.03°C for the March, April and May sowings. The mean minimum temperature during 2017 for each sowing was 22.2°C, for March, 21.5°C for April, 21.6°C for May and 15.4°C for the June. Similarly, the mean minimum temperature in 2018 was 22.8°C, 23.1°C and 23.1°C for the March, April and May sowings respectively. The total precipitation during this stage in 2017 and 2018 was 1483 mm, and 1364 mm for the March sowing, 1433mm and 1255mm for the April sowing and 1271 mm and 1039 mm for the May sowing respectively. The June sowing in 2017 had a precipitation of 1036 mm. The average precipitation for the first, second and third sowing in 2017 and 2018 was 9.4 mm and 8.5 mm; 10.7 mm and 9.22 mm and 12.1mm, and 9.19mm respectively. June sowing in 2017 had an average precipitation of 13.1mm. Average relative humidity during this stage was 67 % and 72% for the March sowing, 75 % and 77% for the April sowing, 82% and 82 % for the May 2017 in 2017 and 2018 respectively. The average relative humidity in June 2017 was 88.6%. PAR in 2017 was 18.3, 18.2, 17.9 and 17.2 MJ/m²/ in March, through June sowing. For 2018, it was 19, 18.2 and 18.5 MJ/m²/day for the March, April and May sowings (Figure 3.3 and 3.4).

3.4.1.3 Environmental conditions during the time of first flowering to the end of flowering (Stage 3)

The mean temperature during stage 3 in 2017 varied from 24.7 -24.9°C for the March sowing, from 23.2 -24.6°C for the April sowing, 24.5°C, for the May sowings, 24.1 -25.2°C for the June sowing. Similarly, the mean temperature in 2018 was 24.2°C for the March sowing, from 23.4 -23.8°C for the April, from 23.4- 24.1°C for the May sowing which was slightly lower than that of 2017. The mean maximum temperature during stage 3 in 2017 was 27.7°C, 27.7°C, 27.7°C and 27.8°C for the March, April, May and June sowings. In 2018 it was 27.5°C, 26.9°C and 26.9°C for the March, April and May sowings. The mean minimum temperature during 2017 for each sowing was 22.2°C, for the March, 21.5°C for the April, 21.6°C for the May and 15.4°C for the June. Similarly, the mean minimum temperature in 2018 was 20.9°C, 19.8°C and 19.7°C for the March, April and May sowings. The total precipitation during this stage in 2017 and 2018 was 78.4 mm, and 16.3 mm for the March sowing, 70.6 mm and 12.1mm for the April sowing and 76.6 mm and 2.8 mm for the May sowing respectively. The June sowing in 2017 had a precipitation of 57.7 mm. The average precipitation for the first, second and third sowing in 2017 and 2018 was 4.6 mm and 1.0 mm; 3.9 mm and 0.7 mm and 3.8mm, and 0.1mm respectively. June sowing in 2017 had

an average precipitation of 2.9mm. Average relative humidity during the 2017 flowering period was 88 %, 87%, 86% and 83% in March, April, May and June respectively. Relative humidity in 2018 was 86%, 83 % and 81% in March, April and May sowings respectively. PAR during this stage in 2017 was 16, 15.4, 15.3 and 14.8 MJ/m²/day and it was 16, 15.3 and 15.2 MJ/m²/day in 2018 (Figure 3.3 and 3.4).

3.4.1.4 Environmental conditions from end of flowering to physiological maturity (seed filling)(Stage 4)

The mean temperature during stage 4 in 2017 and 2018 was 21.5°C and 20.5°C, 20.9°C and 20.7°C, 20.5°C and 19.8°C for the March, April and May sowings respectively. The mean temperature in June 2017 was 19.4°C. The mean maximum temperature during stage 4 in 2017 was 25.5°C, 24.7°C, 24.5°C and 24.1°C for the March, April, May and June sowings. In 2018 it was 25.4°C, 25.4°C and 24.8°C for the March, April and May sowings. The mean minimum temperature during 2017 for each sowing was 17.5°C, for the March, 16.3°C for April; 16°C for May and 15.4°C for June. Similarly, the mean minimum temperature in 2018 was 15.5°C, 15.9°C and 14.68°C for the March, April and May sowings. The total precipitation during stage 4 in 2017 and 2018 was 60 mm, and 3.5 mm for the March sowing, 31.8mm and 0.9mm for the April sowing and 26.4mm and 0.9mm for the May sowing respectively. The June sowing had a total precipitation of 8.6 mm. The average precipitation for the first, second and third sowing in 2017 was 1.1mm and 0.5mm, 0.4mm and 0.2mm and that for 2018 was 0.1mm, and 0.01 mm and 0.01 for the March, April and May sowing respectively. Average relative humidity during stage 4 was 72%, 67%, 67%, and 64% in 2017 and it was 59%, 57% and 57% for the March, April, and May sowings respectively in 2018. PAR during stage 4 in 2017 was 12.8, 12.2, 12.1 and 11.7 MJ/m²/day for the first, second, third and fourth sowings. In 2018, incident solar radiation was 13, 12.2 and 12 MJ/m²/day for the first to third sowings respectively (Figure 3.3 and 3.4).

3.4.1.5 Environmental conditions during physiological maturity to harvest maturity (Stage 5)

The mean temperature during stage 5 in 2017 and 2018 varied from 17.5 - 17°C, 18.2 - 16.1°C, and 18.3 - 16.3°C in the March, April and May respectively. The mean temperature for the June sowing was 18°C in 2017. The mean maximum temperature during 2017 for each sowing during this stage was 22°C, for the March, 23.6°C for the April and 23.1°C for the May and 22.2°C for the June. Similarly, the mean maximum temperature during this time in 2018 was 22°C, 22.6°C and 21.4°C for the March, April and May sowings. The mean minimum temperature during 2017 was 12.4°C, 13.4°C, 13.0°C and 10.8°C for the March, April, May and June sowings. Similarly, the mean

minimum temperature during 2018 was 12°C, 10.6°C and 10.2°C for three sowings respectively. There was no precipitation in the first and second sowings during stage 5 but there was a very small amount of rain for the third sowing in 2018. The average relative humidity during 2017 was 58%, 51%, 53% and 42 % for the March to June sowings respectively. Likewise, the average relative humidity in 2018 was 53% 50% and 45% respectively for the three sowings. The average PAR was 10.5, 10.3, 10.2, and 10.2 MJ/m²/day for the first, second, third and fourth sowings in 2017. In 2018, PAR during this time was 10, 10.3 and 10.3 MJ/m²/day (Figure 3.3 and 3.4).

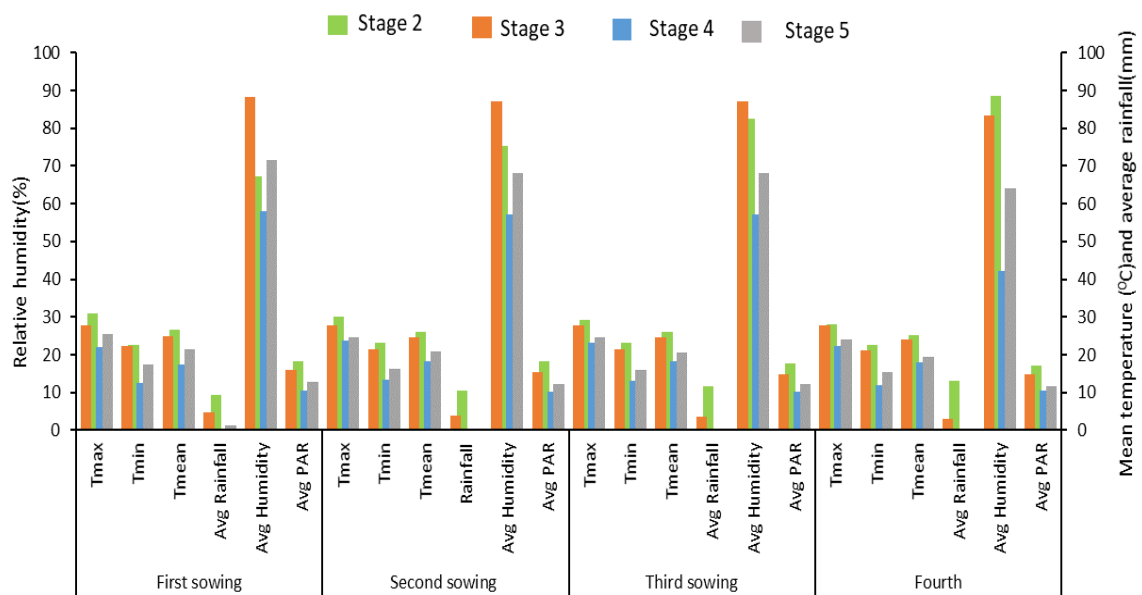


Figure 3.3 Post emergence environmental conditions during teosinte growth in 2017 for four sowing dates

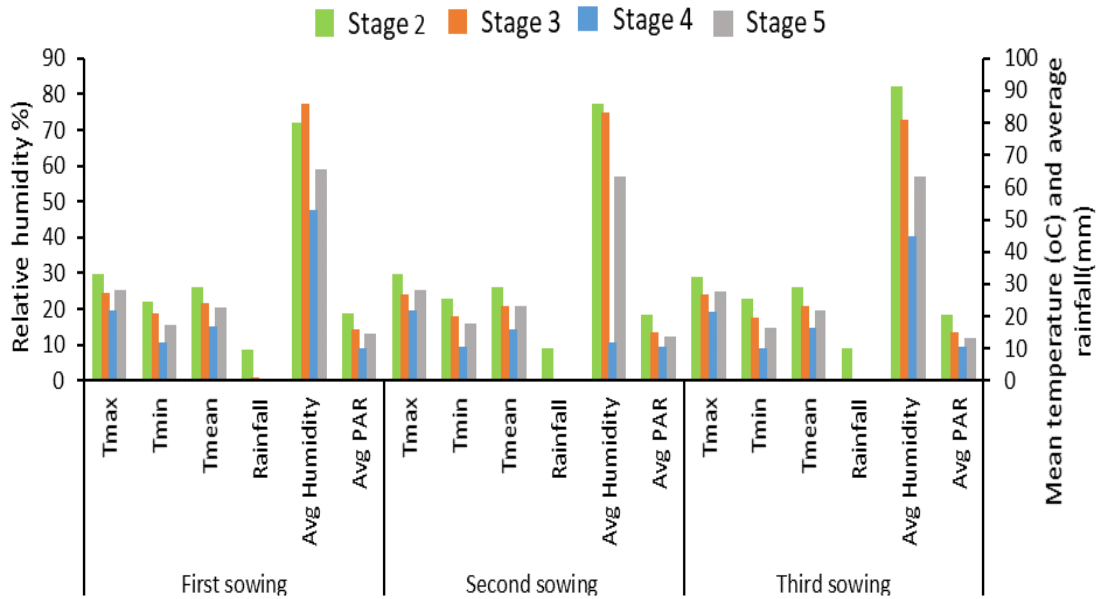


Figure 3.4 Post emergence environmental conditions during teosinte growth in 2018 for three sowing dates

3.4.2 Phenological growth stages and growing degree days (GDD)

Sowing date affected the growth of teosinte. The later the plants were sown, the shorter were the durations of the phenological growth stages, and the growing degree days were also lower than for the early sown plants. The total GDD accumulation was reduced with delayed sowing in both years. The GDD accumulation from sowing until harvest of teosinte seeds in 2017 was 3799, 3364, 2857 and 2330°C days for the March, April, May and June sowing respectively. For 2018, the GDD accumulation was lower than in 2017 with 3646, 3230 and 2761°C days for the March, April and May sowings respectively. The growing period for each sowing lasted for 255, 232, 206 and 183 days respectively in 2017. In 2018 the growing season lasted for 259, 235, and 209 days for each sowing respectively (Table 3.1-3.4, Figure 3.17-3.19).

3.4.2.1 Emergence (Stage 1)

The duration of stage 1 was affected by the sowing date in both years. The GDD accumulated for the stage 1 varied within sowing date and year. The accumulated GDD for emergence for the March sowing was 157°C days (10d) and 145°C days (10d) for 2017 and 2018 respectively, for the April sowing it was 131°C days (8d) and 120°C days (8d) GDD and for the May sowing it was 153°C days (7d) and 132°C days (8d) GDD, for both years respectively. The June sowing had 113°C days (7d) accumulated GDD in 2017 for stage 1 (Table 3.1-3.4, Figure 3.5).



Figure 3.5 Emergence of teosinte (Stage 1)

3.4.2.2 Vegetative phase (Stage 2)

The vegetative growth of teosinte was affected by the sowing date in both years. The GDD accumulated for the overall vegetative growth from seedling emergence to first flowering varied within the sowing date and year. The accumulated GDD for vegetative growth for the March sowing was 2781°C days (167d) and 2695°C days (170d) for 2017 and 2018 respectively, for the April sowing it was 2369°C days (142d) and 2320°C days (144d) GDD and for the May sowing it was 1878°C days (113) and 1844°C days (115d) GDD for both years respectively. The June sowing had 341°C (87d) accumulated GDD in 2017 for stage 2 (Table 3.1-3.4, Figure 3.6).

The LAI, plant height and dry matter yield (DMY) were measured based on the GDD accumulations at 45 and 75 DAS. The April sowing had a better vegetative growth as compared to other sowings in both years. April sowings produced the tallest plants at 45 DAS and 75 DAS in both years with higher LAI at 45 DAS in both years. The DMY in both years at 45 DAS was higher in the April sowing but at 75 DAS it was higher for the April sowing in 2017 and for the March sowing in 2018. The GDD accumulation for the April sowing at 45 DAS in 2017 and 2018 was 821°C days and 748°C days respectively and that for 75 DAS in 2017 and 2018 was 1338°C days and 1252°C days. The GDD accumulation for the March sowing in 2018 at 75 DAS was 1173°C days.

The generalized linear regression analysis among vegetative yield components (Figure 3.7-3.12) revealed important relationships between GDD and DMY. There was a positive correlation between plant height and GDD at 45 DAS in both years but in 2018 a significant negative correlation of GDD with plant height occurred at 75 DAS. LAI had a positive correlation with GDD at 45 DAS in 2017 and 2018 and at 75 DAS there was a positive correlation in 2017 but the correlation was significantly negative at 75 DAS in 2018. Similarly, the DMY and GDD also had

positive relationship at 45 DAS in both years. In 2017, DMY at 75 DAS had a significant correlation with GDD ($P < 0.05$), showing that DMY increased with increased GDD accumulation, but there was a highly significant negative correlation between DMY and GDD in 2018 at 75 DAS (Figure 3.7-3.12).



Figure 3.6 Vegetative stage (stage 2)

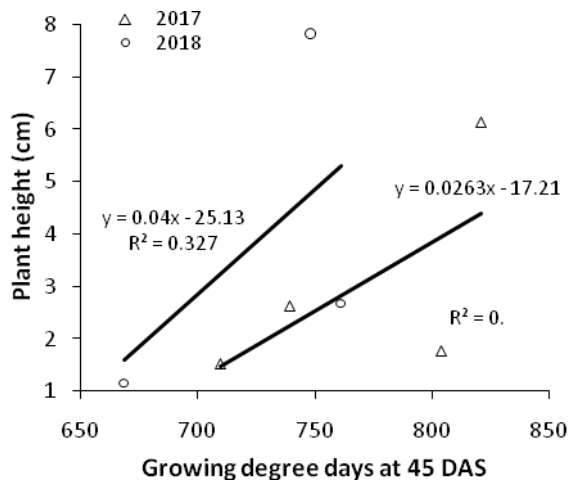


Figure 3.7 Correlation between GDD and plant height at 45 DAS in 2017 and 2018

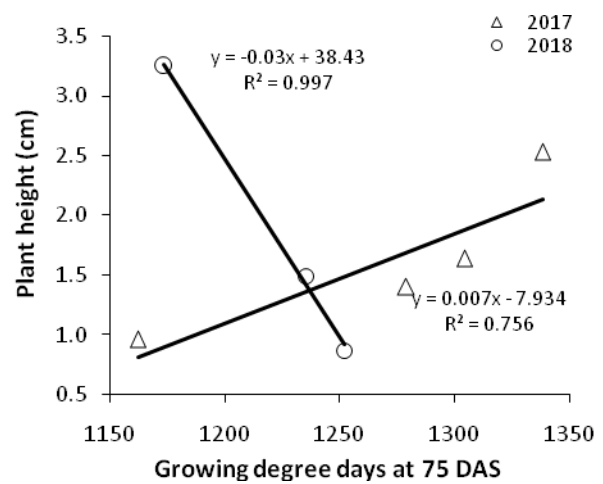


Figure 3.8 Correlation between GDD and plant height at 75 DAS in 2017 and 2018

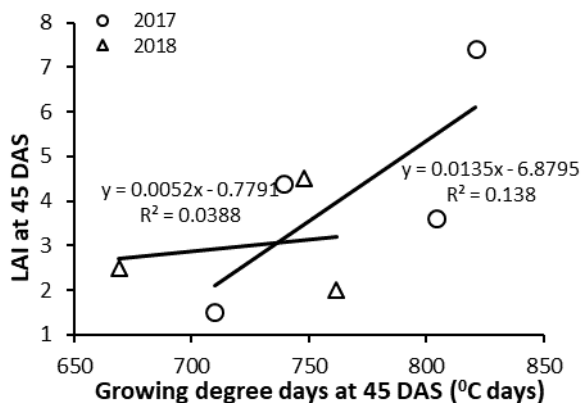


Figure 3.9 Correlation between GDD and LAI at 45 DAS in 2017 and 2018

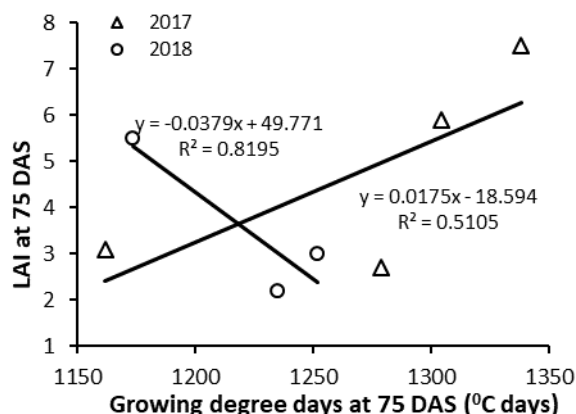


Figure 3.10 Correlation between GDD and LAI at 75 DAS in 2017 and 2018

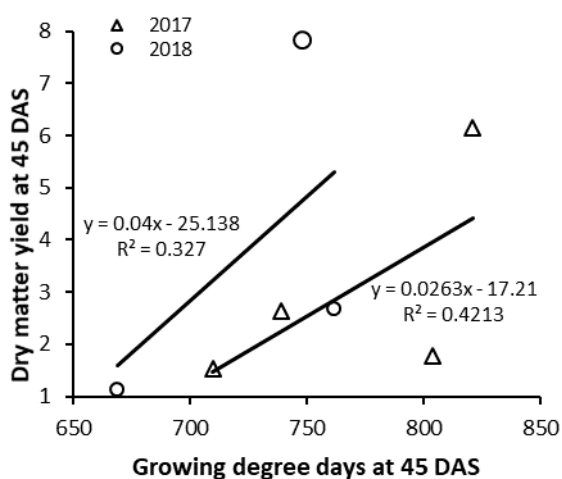


Figure 3.11 Correlation between GDD and DMY at 45 DAS in 2017 and 2018

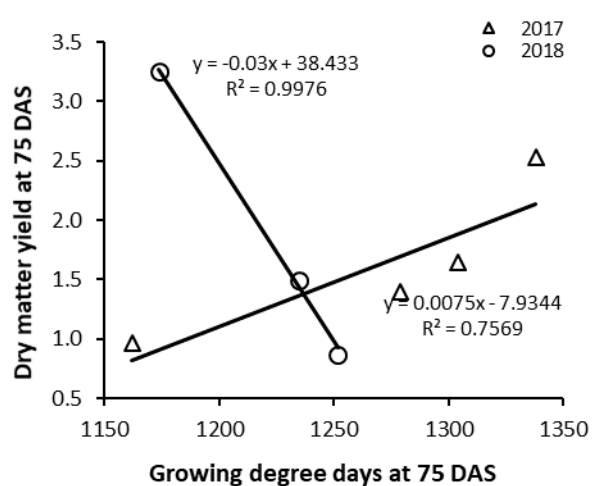


Figure 3.12 Correlation between GDD and DMY at 75 DAS in 2017 and 2018

3.4.2.3 Flowering phase (Stage 3)

The time of first flowering (beginning of stage 3) was affected by sowing date. Flowering began when the mean temperature ranged from 21°C minimum temperature to 28°C maximum in both years. However the GDD accumulated at stage 3 varied within the sowing date and year. The accumulated GDD for stage 3 for the March sowing was 3035°C days (184d) and 2937°C days (187d) for 2017 and 2018 respectively, for the April sowing it was 2631°C days (160) and 2551°C days (161d) GDD and for the May sowing it was 2143°C days (133d) and 2085°C days (133d) GDD for both years respectively. The June sowing had 1632°C days (107d) accumulated GDD in 2017 (Table 3.1-3.4, Figure 3.13).



Figure 3.13 Flowering stage (Stage 3)

3.4.2.4 Seed development phase (Stage 4)

Seed development period was influenced by the sowing date. A delay in sowing slightly extended the number of days for seed filling due to reduced temperature. Days to physiological maturity (PM) were 58, 58, 59, and 60 days for the March, April, May and June sowings in 2017. The cumulative GDD to PM was 3702°C days, 3253°C days, 2745°C days and 2218°C days for the first to fourth sowing respectively in 2017. In 2018 the days to PM were 58, 58 and 60 days for the March, April and May sowing. The GDD to PM was 3554°C days, 3137°C days and 2667°C days for the first, second and third sowing respectively in 2018. The time to reach PM was lengthened as sowing date was delayed (Table 3.1-3.4, Figure 3.14).



Figure 3.14 Seed development of teosinte (Stage 4)

3.4.2.5 Seed maturity phase (Stage 5)

The time from physiological maturity (PM) to harvest maturity (HM) increased (13, 13, 14, and 16 days in 2017) and (13, 15 and 16 days in 2018) as sowing date was delayed in both years. A decrease in mean temperature with delayed sowing during the maturity period was observed for each sowing in both years but the mean temperature in April sowing was slightly higher than other two sowings for this stage in 2018. The GDD accumulated for this stage in 2017 was 3799, 3364, 2857 and 2330°C days. For 2018, it was 3646, 3230 and 2761°C days accumulated (Table 3.1-3.4, Figure 3.15-3.16).



Figure 3.15 Mature plants (Stage 5)



Figure 3.16 Mature seeds

Table 3.1 Phenological growth stages of teosinte for the March sowing

Crop growth stages: First sowing (March)	Cumulative days to reach phenological stage		Phenological stage (days)		Mean daily temp (°C)		GDD accumulated (°C days)	
	2017	2018	2017	2018	2017	2018	2017	2018
Stage 1	10	10	10	10	25.7	24.5	157	145
Stage 2	167	170	157	160	26.7	26.0	2781	2695
Stage 3	184	187	16	16	24.9	24.2	3035	2937
Stage 4	242	246	58	59	21.5	20.5	3702	3554
Stage 5	255	259	13	13	17.5	17.0	3799	3646

Table 3.2 Phenological growth stages of teosinte for the April sowing

Crop growth stages: Third sowing (April)	Cumulative days to reach phenological stage		Phenological stage (days)		Mean daily Temp (°C)		GDD accumulated (°Cdays)	
	2017	2018	2017	2018	2017	2018	2017	2018
Year	2017	2018	2017	2018	2017	2018	2017	2018
Stage 1	8	8	8	8	26.4	25	131	120
Stage 2	142	144	136	136	26.2	26.1	2369	2320
Stage 3	160	161	16	16	24.6	23.4	2631	2551
Stage 4	219	220	58	59	20.9	20.7	3253	3137
Stage 5	232	235	13	15	18.2	16.1	3364	3230

Table 3.3 Phenological growth stages of teosinte for the May sowing

Crop growth stages: Third sowing (May)	Cumulative days to reach phenological stage		Phenological stage (days)		Mean daily Temp (°C)		GDD accumulated (°Cdays)	
	2017	2018	2017	2018	2017	2018	2017	2018
Year	2017	2018	2017	2018	2017	2018	2017	2018
Stage 1	8	8	8	8	29.1	26.5	153	132
Stage 2	113	115	107	107	26.1	26	1878	1844
Stage 3	133	133	17	17	24.5	23.4	2143	2085
Stage 4	192	193	59	60	20.5	19.8	2745	2667
Stage 5	206	209	14	16	18.3	16.3	2857	2761

Table 3.4 Phenological growth stages of teosinte for the June sowing (2017)

Crop growth stages: Third sowing (June)	Cumulative days to reach phenological stage	Phenological stage (days)	Mean daily Temp (°C)	GDD accumulated (°Cdays)
Stage 1	7	7	26.2	113
Stage 2	87	80	25.2	1341
Stage 3	107	19	24.1	1632
Stage 4	167	60	19.4	2218
Stage 5	183	16	18	2330

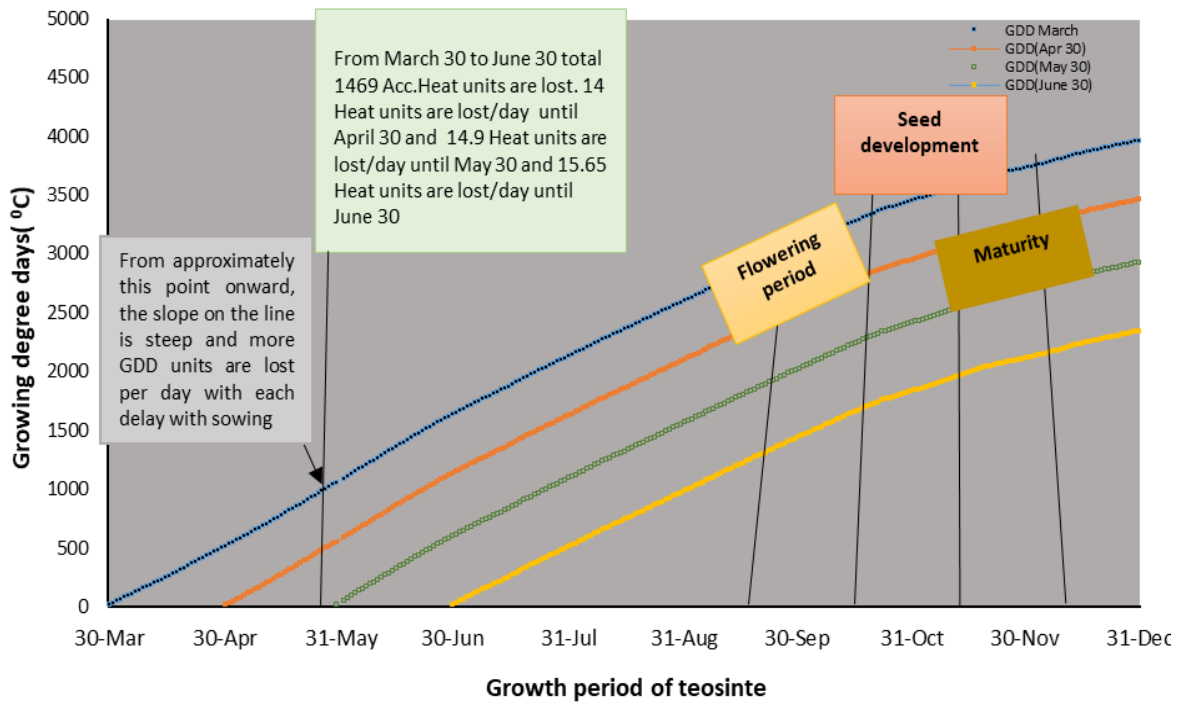


Figure 3.17 Accumulated growing degree days for teosinte in 2017

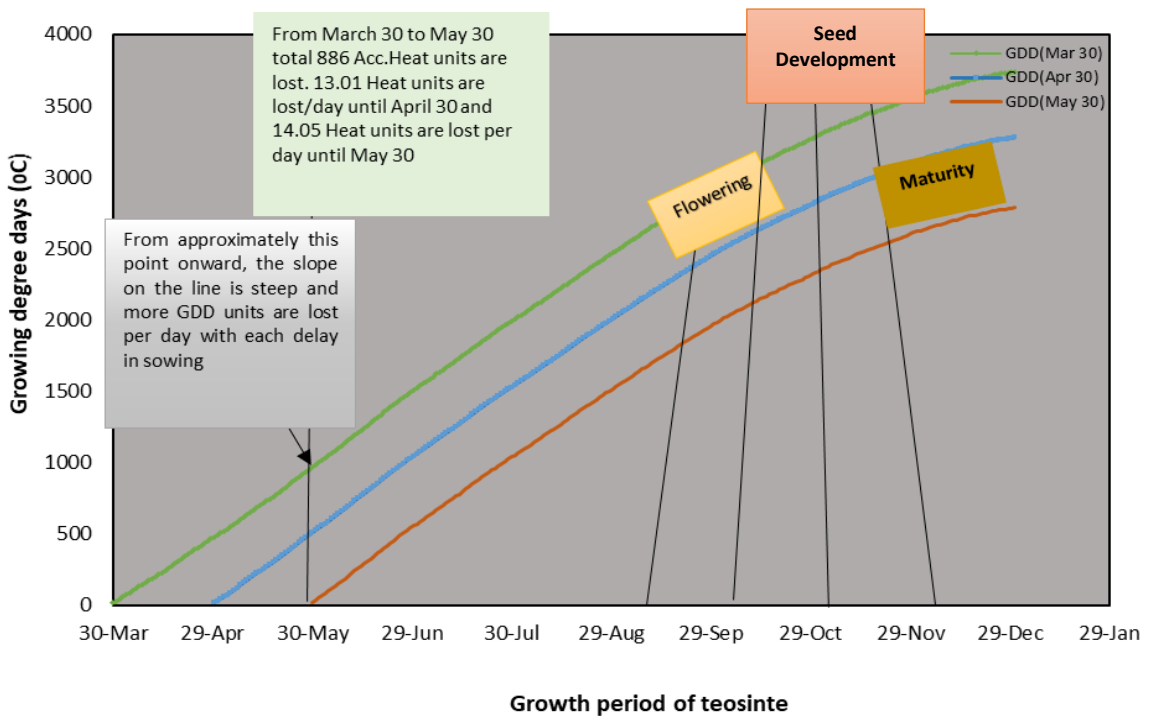


Figure 3.18 Accumulated Growing degree days for teosinte in 2018

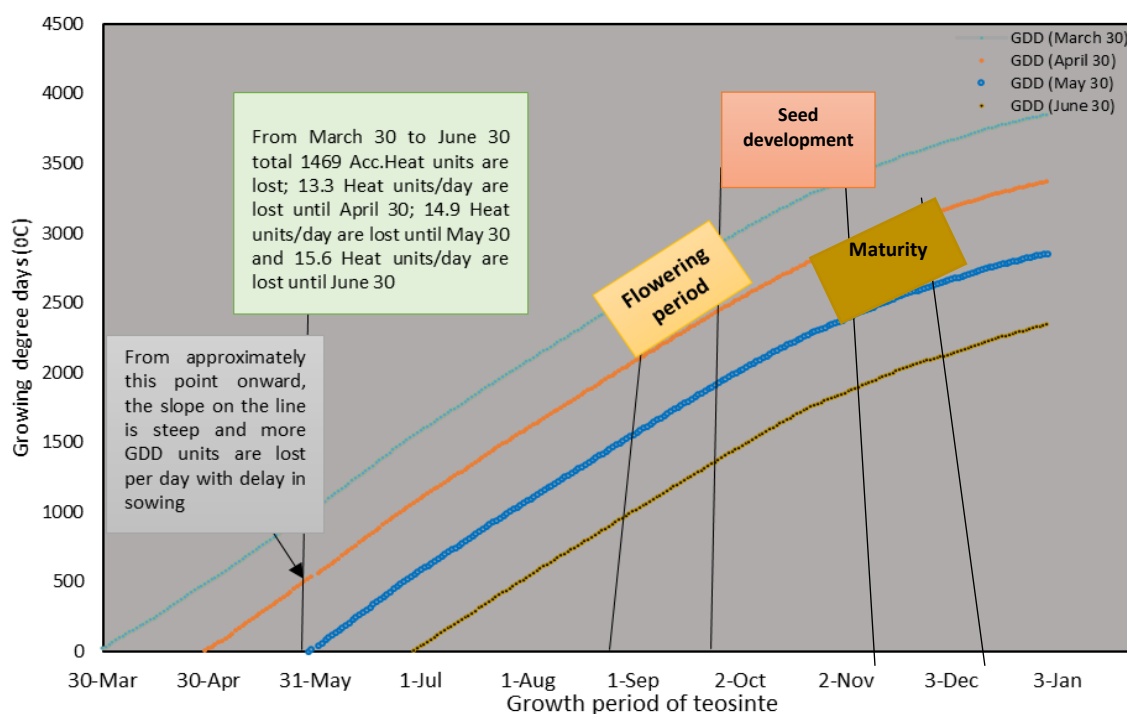


Figure 3.19 Average accumulated growing degree days for teosinte (2017 and 2018)

3.4.3 Estimation of GDD for phenological growth stages of teosinte

For each crop phenological stages, the GDD accumulation required estimate is shown in table 3.5. The GDD requirement for the five phenological stages were 135°C GDD (sowing to seedling emergence), 2189°C GDD (seedling emergence to completion of vegetative stage), 2442°C GDD (flower appearance to completion), 3049°C GDD (completion of flowering to seed physiological maturity) and 3150°C GDD (seed physiological maturity to harvest maturity). Figure 3.20 shows the GDD accumulation for the critical phenological growth stages of teosinte.

Table 3.5 Estimated growing degree days (GDD) for phenological growth stages of teosinte

Crop growth Stages	Phenological days	Days	Max temp(°C)	Min temp(°C)	Mean temp (°C)	GDD accumulated (°C)
Stage 1	9	9	30.7	21.0	26.1	135
Stage 2	135	133	29.6	22.7	26.0	2189
Stage 3	153	18	27.4	20.9	24.1	2442
Stage 4	212	59	25.0	15.8	20.4	3049
Stage 5	226	14	22.2	11.8	17.2	3150

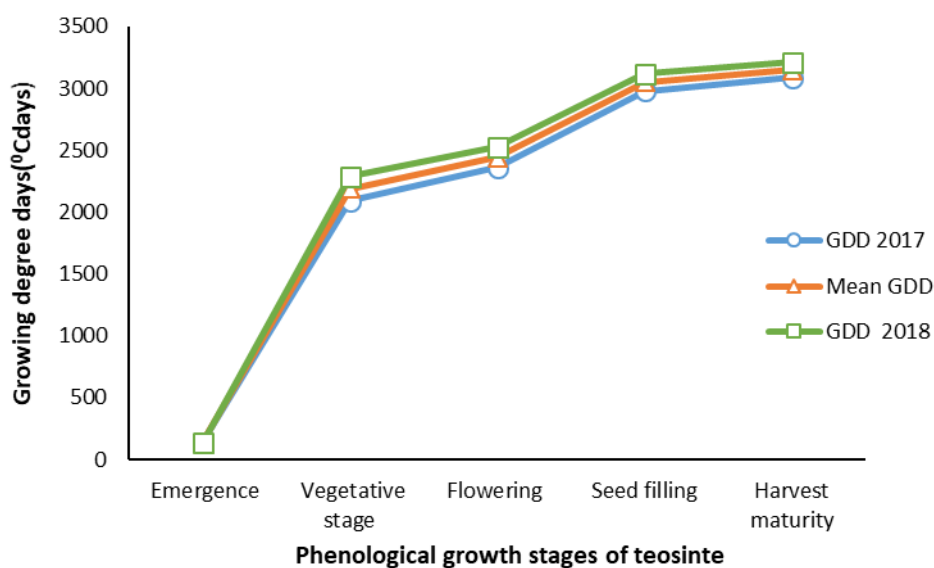


Figure 3.20 Growing degree days required for each growth stage of teosinte

Note: The relationship between GDD and seed yield will be presented in chapter 4.

3.5 Discussion

3.5.1 Effect of sowing date on the phenological growth stages of teosinte in terms of GDD

Sowing date selection and management can avoid severe environmental conditions during the critical growth stages of plants (Diawara, 2012; Huang et al., 2010). The plant growth, morphology, development and the period to reach maturity is highly influenced by temperature (Dar, Brar, & Yousuf, 2018; Gowda, Halikatti, & Manjunatha, 2013; Hoyos et al., 2012; Kingra & Kaur, 2012; Orduz-Rodríguez, Monroy, & Fischer, 2010) by stimulating photosynthesis, respiration, transpiration, transport and cell growth of the plant (Blum, 2011; Struik, 1983). The inter-relationship between temperature and plant development is indicated by GDD (Dar, Brar, & Yousuf, 2018). It provides the time required for each growth stage of a plant in a specific location and changes with growing stage (McMaster, Green, Erskine, Edmunds, & Ascough, 2012). Temperature variation in the field can be adjusted by varying the sowing dates of the plant (Dahmardeh, 2012). In the present study phenological growth stages of teosinte were affected by sowing dates and were strongly influenced by GDD. The growing period in each year was shortened by delay in sowing date where a progressive decrease of GDD was recorded in both years. The second year accumulated lower GDD than the first year, because of differences in temperature as the second year had a lower temperature during the phenological stages in each sowing. Higher GDD accumulation for the March sowing in both years is likely due to the higher number of days required to complete each phenological stage and the lower GDD for the later

sowing might be due to the decrease in the length of available growing period in both years. The reduced GDD for delayed sowing is because of a decrease in both temperature and PAR in both years. This result is in line with the findings of Dar, Brar, and Yousuf (2018); Nielsen et al. (2002); Singh Brar, Kumar Vashist, and Bedi (2016) in maize and Haider, Alam, Alam, and Paul (2003) and Ram, Singh, Mavi, and Sohu (2012) in wheat. The reduced GDD and longer duration to reach each phenological stage from emergence until harvest maturity in the second year might be related to lower temperature, and precipitation as compared to the first year. The mean temperature difference for each of the five phenological stages between 2017 and 2018 was 1.7°C, 0.3°C, 1.0°C, 0.6°C and 1.5°C and the difference in precipitation was 19.4mm, 391.2 mm, 63.0mm and 30.4mm for the first four phenological stages. There was no rainfall at the fifth stage of growth in either year. Low temperature increased the length of time for plant growth and development (Bhusal, & Timsina, 2010). This result was in line with the findings of Amgain (2011) and Gowda, Halikatti, and Manjunatha (2013) in maize and Ghosh, Nandi, and De (2000), Paul and Sarker (2000) and Sandhu, Sharma, and Sur (1999) in wheat. A difference in the GDD accumulation for the same crop in different years has been reported by Wang et al. (2021) in maize. Sowing date is crucial in the growth and development of teosinte herbage and seed yield. Identifying the phenological growth stage of teosinte through GDD accumulation will help to understand the phenostages of teosinte which will help in predicting the teosinte growth stages for agriculture practices.

3.5.1.1 Relation between GDD and seedling emergence

The emergence of seeds may differ from one location to another and from one year to another (Hampton & Hill, 2002). Maize development is basically driven by temperature from emergence to physiological maturity (Blacklow, 1972; Cutforth & Shaykewich, 1990; Grzesiak, Major, Rood, & Frhyman, 1981). This is further controlled by the soil water potential which determine the uptake of water by the seed (Sanchez, 1988; Vitheson, 1973). Emergence of seed (both imbibition and elongation) occurs only when seed imbibes sufficient water to activate growth (Bewley & Black, 1978). This study showed that the early sowing took longer for seedling emergence than the later sowings. Seedling emergence in the current research was delayed in the first sowing in both years because of lower soil temperature and lack of sufficient soil moisture at the time of sowing in March. The minimum and maximum soil temperature for March sowing was lower than the other sowing dates (Appendix B3.1). These results are in line with the findings of Blacklow (1972), Heiniger, Vanderlip, and Welch (1997) and Nielsen et al. (2002) who reported that early sowing may affect emergence due to an unfavourable soil environment which delays emergence and the rate of shoot elongation. Other studies also have reported the reduced efficiency of seeds to

emerge under water stress conditions (Li, Li, Zhang, Liu, & Guan, 2013; Van den Berg, & Zeng, 2006; and Wang et al., 2009). The earliest sowing took longer for seedling emergence but at later sowings seeds emerged quicker than the first sowing. The GDD at the first sowing was higher because of the longer time the seeds took to emergence. The GDD was reduced at the second sowing but again increased at the third sowing in both years, as a result of increased temperature (by 2.8°C in 2017 and by 1.4°C in 2018). The increase in GDD in the May sowing may be due to the fact that the temperature in May exceeded the optimum for teosinte germination. In May sowing in both years, the increase in temperature was accompanied by higher precipitation compared to the March and April sowings.

3.5.1.2 Relationship between GDD and vegetative growth (Plant height, LAI, leaves, total herbage yield and total dry matter yield)

GDD gives a reliable assessment of the thermal time essential for vegetative development of crops (Stewart, Dwyer, & Carrigan, 1998). The vegetative growth of the crop is determined by the duration of plant life cycle and it is further influenced by temperature (Liu et al., 2016). In the current study, for each month sowing was delayed, the GDD accumulation for vegetative growth was reduced in both years. Reduction in GDD as the planting was delayed in both years shortened the vegetative period that directly or indirectly interferes with the growth processes in plants (Baker & Reddy, 2001). The length of the vegetative period increases in earlier sowing which ultimately increases the yields (Bastidas et al., 2008; Nielsen et al., 2002; Wilcox & Frankenberger, 1987). Early sowing allows a longer growth duration, which therefore provides a longer growing period to accumulate more biomass in comparison to a late sowing (Singh & Pal, 2003). The period of photosynthesis is largely reduced with late sowing and less photosynthates are accumulated which results in less DMY (Wiggans, 1956). In the current study, a positive relationship between GDD and vegetative growth attributes (LAI, plant height and DMY) was revealed although not all were significant, when they were compared at 45 and 75 DAS. Positive relation occurred because GDD adopts a direct and linear relationship between growth and temperature (Nuttonson, 1955). Lower DMY despite the higher GDD at 75 DAS in April 2018, was because of the stem breakage in the regrowth of plants cut at 45 DAS. The stems of the cut plants were weak and were easily broken by heavy rain and wind in the month of July. The increase in GDD for the May sowing in 2018 might be due to higher mean temperature in May (higher by 4.2°C from March and 1.5°C from April) and the fact that the minimum temperature (23.5°C) in May was the optimum range for teosinte vegetative growth. The optimum temperature for vegetative growth of teosinte ranges from 23.5 -31.7°C, but this does differ with genotypes as reported by Aburto-Cansino, Ruiz-Corral, Sanchez Gonzalez, and Gonzalez Eguarte (2018). This

result is similar to the findings of Hatfield and Prueger (2015); Prasad and Jagadish (2015) and Zhao, Dai, Jing, Jiang, & Cao (2007) who reported that higher temperature reduces the growth period of plants by accelerating the growth rate, resulting in earlier crop maturity, lower biomass and lower crop yield.

3.5.1.3 Relationship between GDD and flowering

Along with temperature, photoperiod is also a major environmental influence determining time to flowering in plants (Daba, Taran, Bueckert, & Warkentin, 2016; McClung, Lou, Hermand, & Kim, 2016). Further, solar radiation also affects the flowering behaviour in plants (Mata, & Botto, 2011). In the current study, the GDD accumulation during flowering differed among the sowing dates, and the earliest sowing accumulated the highest GDD. Further, the duration of flowering was extended with delayed sowing, which might be the temperature and solar radiation effect. Hung (2012) reported that because teosinte is a tropical crop, flowering is delayed under long day lengths. Higher temperature in the first sowing accelerated the flowering of teosinte resulting in completion of flowering in a shorter duration than in the delayed sown plants. The result of this present study is in line with the findings of Craufurd and Wheeler (2009) who reported high temperature influences on earlier flowering and earlier crop maturity in maize. Stevenson and Goodman (1972) demonstrated that reduced temperature delayed flower initiation along with the growth rate, reducing the apical dominance of the crop.



Figure 3.21 Broken tiller of teosinte from first harvest plant



Figure 3.22 Broken tiller of teosinte from twice cut plant

3.5.1.4 Relationship between GDD and the seed filling period

Seed filling is an important stage of plant growth that regulates several biochemical procedures for the synthesis of carbohydrates, proteins, and lipids and their mobilization in the developing seeds (Barnabas, Jager, & Feher, 2008; Farooq et al., 2017). This process is highly sensitive to

environmental changes which will have an impact on quality and quantity of the final yield (Yang, & Zhang, 2006). The seed filling period is influenced by environmental conditions although genetic variation exists in most crops. It is determined by the source sink relationship, where the assimilate supplied by the leaves is used by the developing seeds for continued growth (Egli, 2004). In this study, the GDD during the seed filling period was lowered with delayed sowing, but the seed filling period was lengthened with delayed sowing. The possible reason for lower GDD accumulation with delayed sowing in both years might be due to reduced growing periods. Further lower temperature, precipitation and reduced solar radiation in later sowings could be a reason for the lengthened seed filling period. Low temperatures extends the cell cycle by reducing the cell division process (Francis & Barlow, 1988) thus extending the plant growth duration. This result is in line with the findings of Boden, Kavanova, Finnegan, and Wigge (2013); Dias and Lidon (2009); Gibson and Paulsen (1999) and Lobell, Sibley, and Ortiz-Monasterio (2012) who reported high temperatures significantly shorten the grain filling period. Gambin, Borrás, and Otegui (2007) reported an inverse relation of moisture loss and biomass during the seed filling process and this gets worse under temperature stress (Awasthi et al., 2017). Further, lack of moisture has been reported to decrease assimilate production and mobilization during seed filling (Leport, Turner, Davies, & Siddique, 2006; Zare, Ghahremaninejad, & Bazrafshan, 2012).

3.5.1.5 Relationship between GDD and seed maturity

Seed maturity is widely affected by temperature. High temperature during seed development produces smaller seeds while low temperature impedes seed growth and maturation (Bareke, 2018). Delayed sowing reduced the GDD accumulated from PM to harvest maturity in both years but the days to reach harvest maturity from physiological maturity increased. This is likely to happen because the delayed sowing shortened the duration from PM to harvest maturity and reduced temperature with delayed sowing during the seed maturing process lengthen the seed maturation period. Furthermore, delayed sowing exposes the crop to the occurrence of hot, dry periods during the flowering period, which risks the seeds damage from autumn frosts at maturity and also reduces the required GDD for that particular season (Menkir & Larter, 1985).

3.5.1.6 Estimation of GDD for phenological growth stages of teosinte

GDD provides a linear response to the development rate above a base temperature (Kiniry, Kim, & Tonnang, 2019,) where the basic assumptions on the development rate of plants are made in response to temperature irrespective of other favourable weather components (Kincer, 1992; Dar, Brar, & Yousuf, 2018; Wiggans, 1956). The growth stages of maize are estimated using a growing degree day (GDD) system (Kiniry, & Bonhomme, 1991). The current study demonstrates as a short day plant, teosinte growth is governed by temperature. Similar findings were conveyed

by Emerson (1924); Hung (2012) and Minow et al. (2018) who reported that teosinte is a short day plant and its flowering is affected by the day length. Rogers (1950) reported the physiological processes responsible for flowering in teosinte, with photoperiod and temperature the key factors affecting flowering time. Similar responses for short day plants were reported by Jackson (2009). Therefore, GDD will be an appropriate method to estimate the phenological growth stages of teosinte. This will enable teosinte growers to predict appropriate times for field activities, including the time for fertilizer application, irrigation and harvest. This will further help to improve the linkage between climate impacts on teosinte phenological stages and the development of adaptation strategies for future planning and management of the crop over the years. However contrasting results in terms of GDD accumulation by teosinte at flowering initiation were reported by Le Corre, Siol, Vigouroux, Tenaillon and Delye (2020) where GDD accumulation for mexicana genotypes was 1703°C days and for parviglumis genotypes it was 2221°C days in France. The GDD accumulation at flower initiation for *Euchlaena mexicana* in the current study was 2189°C days which is very close to the parviglumis genotypes. The variation from mexicana genotypes might be because of variations in temperature and the short period of data availability in this research. Therefore, estimation of teosinte phenology on the basis of GDD needs to be done from a larger set of data collected over a long period of time. While comparing the GDD of teosinte with that of maize, the total GDD required for maize seed maturity is a 2700°C day which is less than that for teosinte (Table 3.6). This is likely because maize is a day neutral crop and has a shorter growing season than that of teosinte. Further it also might be because maize has been selected for a shorter maturity to catch up with the season for places with late start of rain and to allow for more crop production in a rotation, particularly in areas with irrigation facilities as reported by Ado, Abubakar, and Mani (1999). A comparison of teosinte and maize growing degree days for different phenological growth stage is given below.

Table 3.6 GDD accumulation for critical growth stage of teosinte and maize

Growth stages	GDD accumulated (°C) teosinte	GDD* accumulated (°C) maize
Emergence	135	125
Vegetative stage	2189	475
Flowering stage	2442	1400
Seed filling stage	3049	2450
Seed maturity stage	3150	2700

Note: * GDD for maize is adapted from Hoeft, Aldrich, Nafziger, and Johnson (2000)

3.5.2 Comparative estimation of GDD at tropical and temperature areas of Nepal for Teosinte vegetative growth

Temperature is a key variable for the adaptation and distribution of teosinte (Aburto-Cansino et al., 2018). Adjustment by teosinte to various elevations (Ruiz, Sánchez, & Aguilar, 2001; Wilkes, 1967), has led to its development under various conditions of temperature, light and humidity under different geography (Sanchez et al., 2011). As reported by Aburto-Cansino et al. (2018), teosinte is tolerant to both high and low temperatures. Therefore the adaptability of teosinte in temperate areas for herbage yield might be an additional advantage for seed production, especially Nepal and India where the cut and carry system of livestock husbandry is practiced. Table 3.7 provides an estimate of the GDD requirement for teosinte at a higher altitude site of Nepal (Khumaltar) based on the GDD estimate provided in Table 3.5. The lower GDD accumulation in Khumaltar is because of the lower temperature across the growing season as compared to the Terai regions of Nepal. Looking at the GDD accumulated for each phenological stage, it is likely that teosinte will take longer for seed maturation due to higher seed moisture and also the colour of the seeds might not be shiny brown when crop matures late due to lower temperature during seed maturation. This finding is similar to the findings of Zanda (2017) who reported that lower temperature might not allow the translocation of enough assimilates from the mother plant to seeds, which is noticeable by the absence of black layer at the tip of the seeds. However, for GDD during the vegetative growth stage, it is likely that teosinte will grow well producing a good herbage yield, because of the mean temperature (20.4°C) of Khumaltar (Table 3.7), which is slightly lower than the mean minimum temperature (22.7°C) requirement for vegetative growth in GDD estimation as shown in Table 3.5 Therefore, the GDD based phenological models can be useful to make a practically accurate prediction of crop growth in geographic areas where some crops have not been previously grown (Kumudini et al., 2014). This will further make it easier for producers to decide on the timing of crop management aspects as reported in maize by Jame and Cutforth (1996).

Table 3.7 GDD accumulation for critical growth stage of teosinte at a tropical and a temperate sites in Nepal

Crop growth stages	Mean temperature (°C)	Tropical (Chitwan)	Mean temperature (°C)	Temperate (Khumaltar)
Sowing to seedling emergence	26.1	135	21.1	88
Seedling to completion of vegetative stage	26.0	2189	20.4	1624
Flower initiation to completion of flowering	24.1	2442	18.9	1800
Completion of flowering to seed physiological maturity	24.0	3049	13.2	2230
Seed physiological maturity to harvest maturity	17.2	3150	12.1	2154

Note : Chitwan: 228 masl; Khumaltar: 1340 masl

3.6 Conclusion

- In these field trials, the effects of different sowing dates on the phenological growth and vegetative yield of teosinte in Chitwan, Nepal were investigated.
- Teosinte growing period and phenological growth stages were divided into five critical growth stages which are seedling emergence stage (GS1), vegetative stage (GS2), flowering stage (GS3), seed development stage (GS4) and seed maturity stage (GS5). The growth stages were affected by sowing dates being 3799 GDD (255d) for the March, 3364 GDD (232d) for the April, 2857GDD (206d) for the May, and 2330 GDD (183d) days for the June in 2017 and 3646 GDD (259d) for the March, 3230 GDD (235d) for the April, and 2761 GDD (209d) for the May in 2018.
- The time to reach physiological maturity of seeds was lengthened with the delayed sowing which took 58d, 58d, and 59d and 60d days in 2017 for S1, S2, S3 and S4 respectively. In 2018, the time to reach physiological maturity of seeds was 59d, 59d and 60d days in S1, S2 and S3 respectively. This delay was accompanied by reduced temperature and solar radiation.
- The result from this study showed that at this site the mean maximum temperature from teosinte seedling emergence for the five critical growth stages were 31.0°C (GS1), 29.8°C (GS2), 26.9 - 27.4°C (GS3), 25.1°C (GS4) and 22.3°C (GS5). Similarly the mean minimum temperature for these five stages are 20.7°C (GS1), 22.8°C (GS2), and 21.0 -21.5°C (GS3), 16.0°C (GS4) and 12.0°C (GS5) and the mean temperature for the five critical stages are 26.1°C (GS1), 26.0°C (GS2), 24.1°C (GS3), 20.4°C (GS4) and 17.2°C (GS5). The mean GDD

for each critical growing stages were 135°C, 2189°C, 2442°C, 3049°C and 3150°C for GS1, GS2, GS3 GS4 and GS5 respectively.

- Vegetative yield attributes (plant height and LAI) and dry matter yield were higher in April sowings in both years because of higher GDD and ambient temperature and rainfall.
- An estimate of teosinte phenology has been prepared which will be helpful in planning the crop cultivation for teosinte growers in tropical as well as temperate regions.

Chapter 4

Identifying the appropriate sowing dates and seed rates for maximizing seed production of Teosinte under different cutting regimes in Chitwan, Nepal

4.1 Introduction

Teosinte is high yielding, energy rich, multi-cut in nature, versatile (Mohan, Dar & Singh, 2017) and the most popular herbage in tropical areas where dairying is a common business (Khanal, Devkota, Tiwari & Gorkhali, 2020). Teosinte can be grown successfully in areas with a hot and humid climate. There is an opportunity to utilize the crop for both herbage and seed, making it a dual purpose crop. For seed production, farmers commonly take a single cut of herbage from the standing crop and then leave it to produce seeds. However, this cut may reduce seed yield resulting in an economical loss. High seed yield and accessibility to quality seeds are important for herbage productivity (Patil & Merwade, 2016). Successful production of every crop relies on the availability of good quality seeds (Hampton, Boelt, Rolston & Chastain, 2013). Yield in cereals depends on the meristematic activity of tillers, leaves and ears, which may compete with each other at various stages of plant development. Indeed, much of the art of cereal growing depends on management of this competition (Bunting & Dretnan, 1966). Farmers use different sowing dates and seed rates depending on location and purpose of the crop. For herbage production, Nepalese farmers broadcast a large amount of seed without knowing anything about its economic returns. This is also currently the case for teosinte seed production. Considering the demand for and importance of teosinte seeds in Nepal, it is of the utmost necessity to have a new perspective of impacts of sowing dates, sowing rates and cutting management on sustainable seed production, to give farmers the best economic returns for growing this crop.

In addition, very little is known about the seed production potential of teosinte. The biggest challenge in producing teosinte seeds is identifying its planting window in terms of sowing date, seed rate and cutting management. Sowing date and plant density have a great influence on crop production. The ideal sowing date and plant density allows the plants to efficiently use time, light, temperature, precipitation and other factors (Johnson & Mulvaney, 1980; Kondra, 1977). Imbalance in the source and sink availability as affected by the sowing date may alter the seed yield of teosinte as reported for maize by Otegui, Andrade and Suero (1995) and Bolanos and

Edmeades (1996). The area currently under teosinte cultivation in Nepal is 27,232 ha and the seed production is 870 tonnes. However, there is still a seed deficit of 220 tonnes per annum, which is 25% of the total seed requirement (NPAFC, 2018).

For teosinte, there have been no reported studies on agronomic management for factors such as sowing time, planting density and cutting interval on seed yield. This study was conducted to investigate the effect of different sowing dates, seed rates and cutting management on seed yield and its components of teosinte *var. Sirsa*.

4.1.1 Effect of sowing date on seed yield

Teosinte is a short day plant and thus its seed production requires the full season of growth. This means like maize, it has to be sown early to fully exploit the thermal time available over the growing season (Wilson, Johnstone & Salinger, 1994). Seed production is much more impacted by variation in sowing date than herbage production. However, early sowing combined with low soil moisture can be conflicting for emergence and seedling establishment (Hayhoe, Dwyer, Stewart, White & Culley, 1996). With delayed sowing, growth occurs under higher temperatures, with simultaneous reductions in period of growing cycles which affects the cumulative incident photosynthetically active radiation (PAR) at silking, significantly reducing the yield (Millner & Toor, 2007). Sub-optimum temperature during sowing affects field emergence and crop early growth and development, resulting in poor seed set, forced maturation and low seed yield (Bhuker, Mor & Digamber, 2019). Therefore, it is necessary to provide a planting window to the farmers to maximize yields and profit from seed production (Tsimba, Edmeades, Millner & Kemp, 2013).

The ideal sowing date will allow the plant to interact with various weather components between seasons and within a climatic range, resulting in consistent yield (Alam, Mukta, Nahar, Haque, & Razib, 2020). However, an inappropriate sowing time may result in a seed yield penalty in teosinte. Increased seed yield is due to increased capacity of seeds to assimilate the supply (source) for seed filling accompanied by an enhanced capacity of the seeds (sink) to accommodate those assimilates. Variation in sowing date does affect seed yield (Hampton, Conner, Boelt, Chastain & Rolston, 2016). Early or late sowings alters the source-sink balance of the crop, where seed number is usually strongly associated with seed yield (Bolanos & Edmeades, 1996; Otegui, Andrade & Suero, 1995) and therefore defines sink size. Reduced maize yield following early and late sowing has been reported by several authors (Johnson & Mulvaney, 1980; Sorensen, Stone & Rogers, 2000). Maize growers are now aware that timely sowing is critical for maximizing yield for both seed and biomass (Van Roekel & Coulter, 2012). But the optimum sowing date will vary from area to area due to differences in climate and the length of the growing season where the crop is

produced (Bruns & Abbas, 2006). Maize requires warm soil to germinate and grow (Abendroth, Woli, Myers & Elmore, 2017; Bruns & Abbas, 2006) therefore early sowing could contribute to profitability in plant development and reduced costs from a reduced need to dry seed postharvest (Johnson & Mulvaney, 1980; Lauer et al., 1999). Higher temperature affects the phenological stages like pollination and seed development in most crops (Hatfield & Prueger, 2015), therefore sowing date may also impact the seed quality of teosinte if flowering or seed development occur during a high-temperature period for an early sowing. Therefore, sowing date should be timed to avoid the damaging effects of high temperatures at flowering and during seed set which have a negative impact on seed yield (Singh, Prasad & Reddy, 2013).

4.1.2 Effect of seed rate on seed yield

Each crop has its own optimum plant density. Seed rate is considered as one of the most important cultural practices to determine seed yield and other significant agronomic characteristics. Plant density influences the growth and development pattern of the plant. It also alters the carbohydrate production in plants (Sangoi, 2001). Gonzalo, Vyn, Holland, and McIntyre (2006) noted that while plant density does have a significant effect on seed yield, it is a combination of various agronomic and environmental factors such as soil fertility, moisture supply, genotype, planting date, planting pattern, plant population and harvest time which collectively determine final yield. Too few plants per unit area does not allow maximum usage of resources available and again, too many plants increase the competition between plants and reduce the yield (Moosavi, Seghatoleslami & Moazeni, 2012). With increased planting density, there is a decrease in light interception (Luque, Cirilo & Otegui, 2006) and resources available to each individual plant which leads to decreased seed yield (Abuzar et al., 2011; Ali, Khalil, Raza & Khan, 2003). Crop growth rate, leaf area index, and total dry weight decline at high plant density as compared to low density over all the crop growth season (Saberli, Sadatnoori, Hejazi, Zand & Baghestani, 2007). Under low plant density, the yield per unit area decreases but single-plant production increases (Gardner, Pearce & Mitchell, 1984; Ghanbari & Taheri, 2003). Therefore, optimising planting spacing improves the seed yield and quality, at the same time reducing the input cost (Miguel Frade & Valenciano, 2005).

4.1.3 Effect of cutting management on seed yield

For annual herbage seed crops, cutting vegetative growth is not a common practise. However in Nepal, farmers like to take at least one herbage cut from their seed crop. Cutting has a definite negative effect on the seed yield with both cutting interval and frequency being important (Kumar, Channakeshava, Belavadi, Shivprakash, & Siddaraju, 2017). Increased cutting frequency of

herbage will reduce the plant starch reserves and also adversely affect the seed setting. However, Makarana et al. (2018) reported that taking one harvest for herbage and then leaving for seed in pearl millet was productive as it allowed more opportunity for leaves and tiller development, which increased the seed yield. However other researchers have reported that any increased seed yield after cutting is generally a result of reduced lodging (Drou-shiotis, 1984). A dual purpose crop should be cut at an early stage of growth to allow more tillers to survive and provide a large photosynthetic area for reproductive growth (Miller, Joost & Harrison, 1993; Patil & Merwade, 2016). For Nepalese farmers growing teosinte for seed production, it is important to determine the economic impact of cutting on seed yield and quality.

4.2 Objective

This research was conducted with the objective of identifying the appropriate sowing dates and seed rates for seed production of teosinte under different cutting regimes.

4.3 Methodology

The detailed methodology is explained in Chapter 2.

4.3.1 Varietal selection

As explained in Chapter 2.

4.3.2 Experimental design

The experimental design is as described in Chapter 2.

4.3.3 Inter-cultural operations

The inter cultural operations are as explained in Chapter 2.

4.3.4 Plant selection, tagging and data recording

Ten plants from the central row of each plot were randomly selected and tagged after the herbage was harvested at 45 and 75 DAS. After taking both harvests for herbage yield, the plants were left for seed production.

4.3.4.1 Plant height

The final height was taken at full maturity stage after tasselling. For final measurement, height from the base of the plant to the node of the flag leaf was recorded in cm with a measuring tape (IBPGR, 1991). Average height of ten plants was calculated.

4.3.4.2 Leaf Area Index (LAI)

The length and breadth of a mature fully open and physiologically active leaf was measured after flowering. The length was taken from the base of the lemma and the width from the centre of the leaf from 10 selected plants after flowering. Based on the above information LAI was calculated for each combination of sowing date, seed rate and cutting management using the following formula as suggested by Pal and Murari (1985) and Musa and Usman (2016). The leaf area was calculated by:

$$LA = L \times W \times K$$

Where,

LA: Leaf area (cm²)

L: Length of leaf (cm)

W: Width of leaf (cm)

K: Factor (0.75)

$$LAI = \text{Leaf area (cm}^2\text{)}/\text{Land area per plant (cm}^2\text{)}$$

4.3.4.3 Number of tillers

The number of tillers per plant were recorded by counting all the tillers of each of the 10 tagged plants separately before seed harvest and then averaging the value of the 10 plants. Both primary and secondary tillers on each plant were counted after flowering.

4.3.4.4 Seed yield and its components

The number of cobs per plant (Figure 4.1), numbers of ears per cob (Figure 4.2), seeds per ear (Figure 4.3) and seed yield (Figure 4.4) at three different positions (top, middle and bottom) of a plant were recorded for 10 tagged plant in each plot and the average was determined. Seed yield per plant was calculated by multiplying the number of cobs with number of ears per cob and seeds per ear. Based on the position of cobs, seeds were harvested three times from each plant. The first harvest was taken from the top three cobs, the second harvest was taken from the middle three cobs and the remaining cobs at bottom were taken for the third harvest. Each time seeds were harvested at a moisture content of 25-27%, which was confirmed before seed harvest using a moisture meter. Seed production was recorded for uncut plants, once cut plants and twice cut plants. The secondary tillers present along the internodes of the main stem were also harvested together with the seeds from the main stem at each harvest. These cobs were then threshed manually by hand, and the seeds cleaned, weighed, placed in a paper bag, and then dried separately in shade to 14 % seed moisture content.



Figure 4.1 Cobs of teosinte



Figure 4.2 Ears on cob



Figure 4.3 Seeds inside a single ear



Figure 4.4 Fresh seeds

4.3.5 Statistical Analysis

Analysis of variance (ANOVA) to determine the effect of sowing date, seed rate and cutting management on seed yield and associated yield components of teosinte and the interactions among the factors was performed using General Linear Model (GLM) of Genstat 19th Edition (VSN International, 2019). Accordingly, replication was considered as the block, whole plots as sowing date plot, seed rate as a sub-plot and cutting management as a sub-sub-plot factor, respectively. For presenting the significance of different factors on yield and its attributing characters, $P < 0.05$, $P < 0.01$, and $P < 0.001$ were used for 5, 1, and 0.1 percent level of significance respectively. Significantly different means of each level of factors considered were compared using Fisher's Unprotected Test of Least Significant Difference (LSD).

4.4 Results

The major objective of this study was to generate information on optimum sowing date, seed rate and cutting management to get maximum seed production per unit area. Seed yield (kg ha^{-1}) is a function of various growth and yield attributing parameters like plant height, leaf area index, number of tillers plant^{-1} , cob number, number of ears per cob and seeds per ear; these results are presented hereunder.

4.4.1 Main effect means of sowing date, seed rate and cutting management on plant properties of teosinte

4.4.1.1. Final plant height (cm)

Sowing date

There was a significant difference in final plant height among the sowing dates in both years ($P < 0.05$) (Table 4.1). In 2017, final plant height for the first three sowings did not differ but all were significantly higher than the June sowing (Table 4.1). In 2018, the March and April sowing had significantly higher final plant height than the May sowing (Table 4.1). There was a significant negative correlation ($R^2=0.93$) ($P < 0.05$) between sowing dates and plant height because plant height decreased as sowing time was delayed (Figure 4.5).

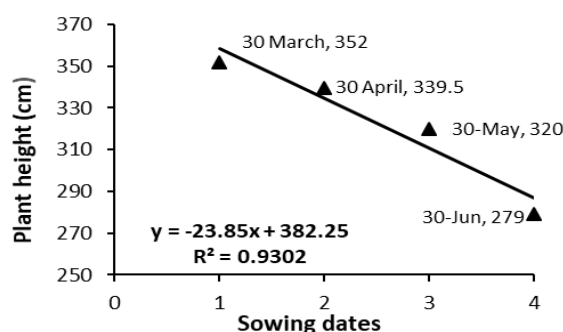


Figure 4.5 Correlation between sowing date and plant height; data for March, April and May sowings are means for 2017 and 2018

Seed rate

Seed rate had no significant effect on final plant height in either year (Table 4.1).

Cutting management

In both 2017 and 2018, cutting significantly reduced final plant height but there was no differences in height between the one and two cut plants ($P < 0.001$) (Table 4.1). There was a significant negative correlation ($R^2=0.90$) ($P < 0.05$) between cutting management and plant height because plant height decreased as the cutting frequency was increased (Figure 4.6).

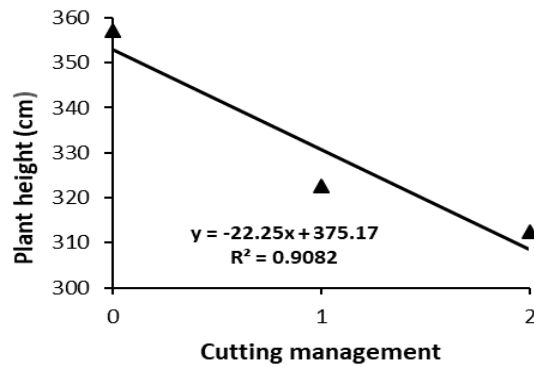


Figure 4.6 Correlation between number of herbage cuts and plant height; data are means for 2017 and 2018

Interactions

There was a significant interaction between sowing date and cutting management for final height in 2017 ($P < 0.001$) (Table 4.1) because of the impact of cutting on reducing plant height for the June sowing. In 2018, there was a significant interaction between seed rate and cutting management because cutting reduced final plant height irrespective of seed rates ($P < 0.05$) (Table 4.1).

4.4.1.2 Tiller numbers per plant

Sowing date

Sowing date was an important source of variation with respect to final tiller numbers per plant in both 2017 and 2018 ($P < 0.001$) (Table 4.1). In 2017, tiller number did not differ between the first and second sowing but both were significantly higher than the third and fourth sowing. In contrast, in 2018 tiller number was significantly lower for the second and third sowings respectively (Table 4.1). There was a negative correlation ($R^2 = 0.88$) ($P > 0.05$) between sowing dates and tiller numbers because delayed sowing reduced the tiller numbers (Figure 4.7).

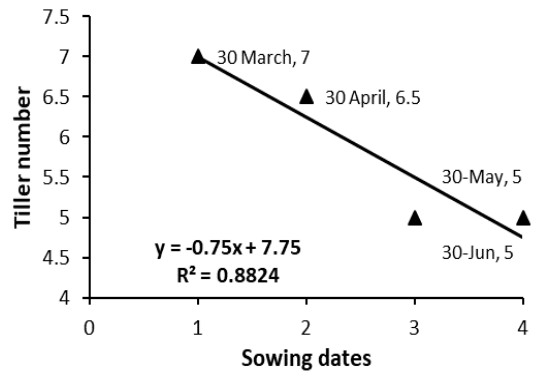


Figure 4.7 Correlation between sowing date and tiller numbers; data for March, April and May sowings are means for 2017 and 2018

Seed rate

There was no significant effect of seed rate on tiller numbers in either year (Table 4.1).

Cutting management

In 2017, cutting significantly reduced the tiller numbers per plant ($P < 0.001$) (Table 4.4). No difference was recorded in tiller numbers between uncut and single cut plants in 2018 but the second cutting reduced tiller numbers ($P < 0.05$) (Table 4.1). There was a significant negative correlation ($R^2 = 0.95$) ($P < 0.05$) between cutting management and tiller number because tiller number was reduced with increased cutting frequency (Figure 4.8).

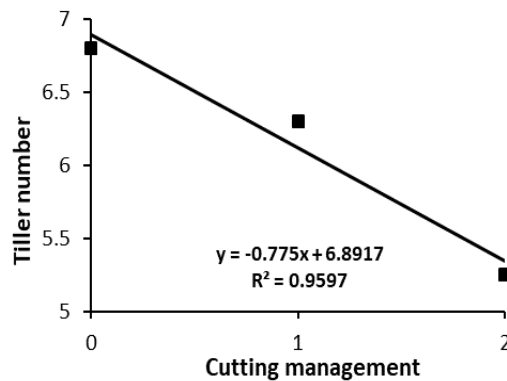


Figure 4.8 Correlation between number of herbage cuts and tiller numbers; data are means for 2017 and 2018

Interactions

No significant interactions occurred among sowing date, seed rate and cutting management for tiller number per plant in either year (Table 4.1).

4.4.1.3 Leaf Area Index (LAI)

Sowing date

In 2017, LAI for the March and April sowing did not differ but both were significantly greater than the May and June sowings ($P < 0.001$) (Table 4.1). In 2018, the March and April sowings also had a LAI significantly greater than the May sowing ($P < 0.001$) (Table 4.1). A significant negative correlation ($R^2 = 0.94$) ($P < 0.05$) between sowing date and LAI occurred because LAI reduced with delay in sowing (Figure 4.9).

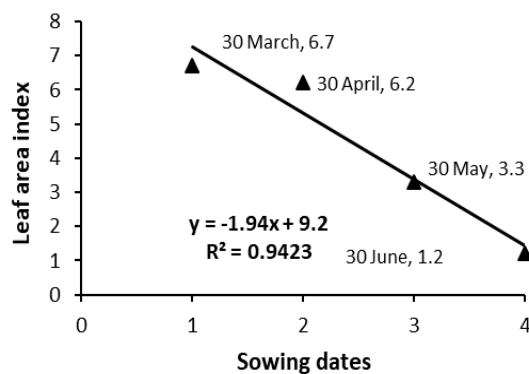


Figure 4.9 Correlation between sowing date and LAI; data for March, April and May sowings are means for 2017 and 2018

Seed rate

LAI was significantly affected by seed rates in 2017 and 2018 ($P < 0.01$) (Table 4.1). The lowest seed rate had the highest LAI in 2017 and the three higher seed rates did not differ with each other. In contrast, in 2018 LAI for the lower three seed rates did not differ but was significantly higher than the highest seed rate ($P < 0.001$) (Table 4.1). There was a significant negative correlation ($R^2 = 0.93$) ($P < 0.05$) between seed rate and LAI because LAI decreased as seed rate increased (Figure 4.10).

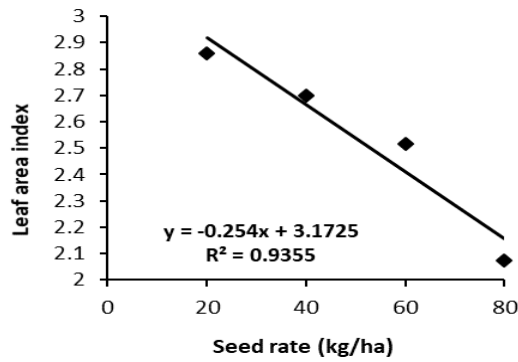


Figure 4.10 Correlation between seed rate and LAI; data are means for 2017 and 2018

Cutting Management

Cutting management was an important source of variation with respect to LAI ($P < 0.001$) (Table 4.1) in both years. Uncut plants had a significantly higher LAI than cut plants but the LAI did not differ between the two cutting treatments ($P < 0.001$) (Table 4.1). A negative correlation ($R^2 = 0.81$) ($P > 0.05$) between cutting management and LAI occurred because LAI reduced with increased cutting frequency (Figure 4.11).

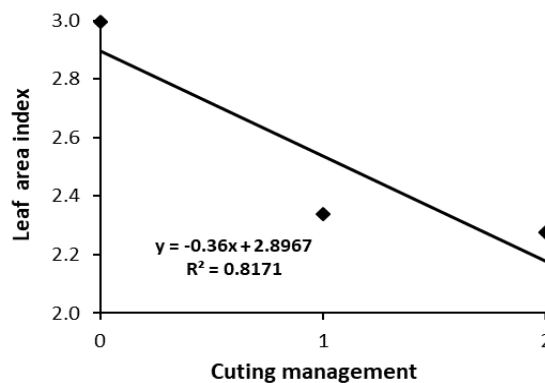


Figure 4.11 Correlation between number of herbage cuts and LAI; data are means for 2017 and 2018

Interactions

There was a significant interaction between sowing date and seed rate ($P < 0.05$), and seed rate and cutting management ($P < 0.05$) for LAI in 2017 (Table 4.1). This interaction between sowing date and seed rate was because of the higher LAI for the two earliest sowing dates at the lowest seed rate. Further interactions between seed rate and cutting management occurred because uncut plants recorded highest LAI for the lowest seed rate. In 2018, a significant interaction between sowing date and seed rate occurred because the earliest two sowings also recorded the highest LAI at the lowest seed rate ($P < 0.001$) (Table 4.1). The interaction tables are attached in Appendix C.

Table 4.1 Main effect means of sowing date, seed rate and cutting management on plant properties of teosinte in 2017 and 2018 at NCRP, Chitwan, Nepal

Main effect means of:	Final plant Height(cm)		Final tillers per plant		LAI	
	2017	2018	2017	2018	2017	2018
Sowing date(SD)						
30-Mar	340a	364a	8.1a	6.6a	3.2a	3.5a
30-Apr	331a	348a	7.8a	5.4b	3.0a	3.2a
30-May	321a	319b	6.1b	4.2c	1.3b	2.0b
30-Jun	279b	-	5.5b	-	1.2b	-
Linear contrast p value	0.002	0.005	<.001	<.001	<.001	0.001
LSD (0.05)	32	25	0.9	0.6	0.6	0.6
CV%	6.2	4.2	8.0	6.3	16	12.4
Seed rate (SR)						
20 kg ha-1	325a	351a	6.9a	5.3a	2.4a	3.3a
40 kg ha-1	317a	335a	6.8a	5.5a	2.2b	3.2a
60 kg ha-1	317a	346a	7.1a	5.5a	2.2b	2.9a
80 kg ha-1	313a	342a	6.7a	5.3a	2.0b	2.2b
Linear contrast p value	0.669	0.484	0.797	0.818	0.001	<.001
LSD (0.05)	32	17	0.76	0.4	0.2	0.6
CV%	14	6	15.6	8.6	14.0	23.4
Cut management (CM)						
No cut	341a	373a	8.1a	5.5a	2.5a	3.5a
One cut	313b	332b	7.0b	5.6a	2.1b	2.6b
Two cut	300b	325b	5.4c	5.1b	2.0b	2.5b
Linear contrast p value	<.001	<.001	<.001	0.03	<.001	<.001
LSD (0.05)	22	16	0.5	0.3	0.2	0.4
CV%	20	12	21.1	15.2	25.3	30.8
Significance of interactions of linear contrasts (p value)						
SD(lin) x SR(lin)	0.900	0.237	0.400	0.892	0.018	<.001
SD(lin) x CM(lin)	<.001	0.073	0.633	0.735	0.143	0.842
SR(lin) x CM(lin)	0.772	0.037	0.431	0.611	0.045	0.092
SD(lin) x SR(lin) x CM(lin)	0.681	0.387	0.587	0.543	0.175	0.911

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

4.4.2 Main effect means of sowing date, seed rate and cutting management on seed yield components of teosinte

4.4.2.1 Number of cobs per plant

Sowing date

A significant effect of sowing date was recorded for total cobs per plant in both 2017 ($P < 0.001$) and 2018 ($P < 0.01$) (Table 4.2). Cob numbers did not differ between the March and April sowing in either year, or between the April and May sowings. In 2017 the June sowing had significantly less cobs per plant than the earlier sowings. There was also a significant negative correlation ($R^2 = 0.98$) ($P > 0.01$) between sowing date and cobs per plant because cobs number decreased with a delay in sowing date (Figure 4.12).

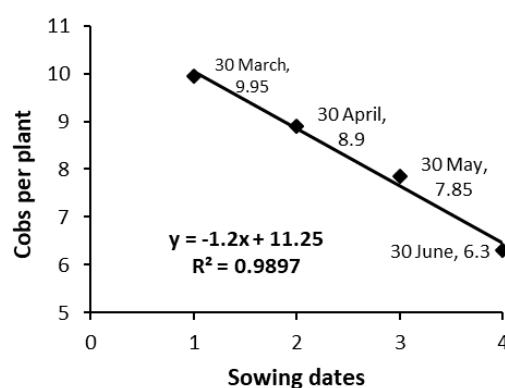


Figure 4.12 Correlation between sowing date and cobs per plant; data for March, April and May sowings are means for 2017 and 2018

Seed rate

Seed rates did not affect cobs per plant in 2017 but cobs per plant varied significantly with the seed rate in 2018 ($P < 0.001$) (Table 4.2). The highest number of cobs per plant was recorded from the lowest seed rate, those for 40 and 60 kg ha⁻¹ seed rate did not differ and the highest seed rate had the lowest number of cobs per plant (Table 4.2).

Cutting management

Cutting management was an important source of variation for total number of cobs per plant in both years ($P < 0.001$). In 2017, cob numbers did not differ between the no and one cut but the two cuts significantly reduced cob numbers (Table 4.2). However in 2018, uncut plants produced the highest number of cobs per plant with each cutting reducing cob number significantly (Table 4.2). There was also a significant negative correlation ($R^2 = 0.92$) ($P < 0.05$) between cutting management with cobs per plant decreasing as sowing date was delayed (Figure 4.13).

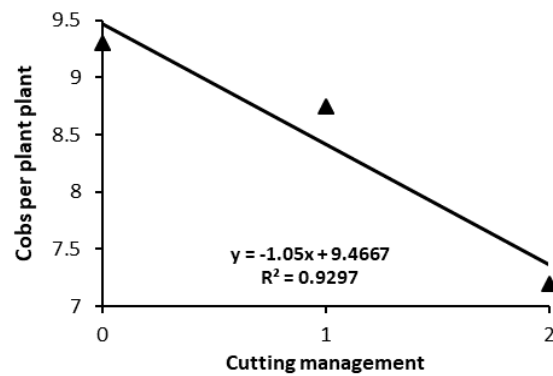


Figure 4.13 Correlation between number of herbage cuts and cobs per plant; data are means for 2017 and 2018

Interactions

In 2017, a significant interaction was recorded between sowing date and cutting management because the June sowing produced the least number of cobs from the twice cut plants ($P < 0.05$) (Table 4.2). A significant interaction also occurred between sowing date and seed rate in 2018, because the March sowing produced the highest cobs per plant at the lowest seed rate, but the March sowing did not differ from the April and May sowings ($P < 0.05$) (Table 4.2). The significant interaction between seed rate and cutting management occurred because uncut plants produced the highest number of cobs per plant from the lowest seed rate ($P < 0.01$) (Table 4.2). Similarly, the interaction among the sowing date, seed rate and cutting management occurred because uncut plants from the March sowing produced the highest number of cobs per plant at the lowest seed rate ($P < 0.05$) (Table 4.2). The interaction tables are attached in Appendix C.

4.4.2.2 Number of ears per cob

Sowing date

There was a significant effect of sowing date on ears per cob in both years. Ears per cob in the first and second sowing did not differ but the third sowing had significantly fewer ears per cob in 2017 ($P < 0.001$) and 2018 ($P < 0.05$). In 2017, the June sowing had the lowest number of ears per cob (Table 4.2).

Seed rate

Seed rate had no significant effect on ears per cob in 2017 (Table 4.2). In 2018, ears per cob did not vary among the three lowest seed rates but the highest seed rate had fewer ears per cob than the lowest sowing rate ($P < 0.005$) (Table 4.2).

Cutting management

Cutting had a significant effect on ears per cob in both years ($P < 0.001$) (Table 4.2). In 2017, ears per cob did not differ between uncut and once cut plants, but the twice cut plants had significantly fewer ears per cob. In 2018, ears per cob decreased significantly with each cutting ($P < 0.001$) (Table 4.2).

Interactions

A significant interaction between sowing date and cutting management occurred in both 2017 ($P < 0.001$) and 2018 ($P < 0.05$) (Table 4.2). The interaction in 2017 was because the first two sowings produced the highest ears per cob in the uncut and one cut plants (Table 4.2). In 2018, the significant interaction between sowing date and cutting management occurred because the uncut plants produced significantly more ears per cob from the March and April sowings (Table 4.2). The interaction tables are attached in Appendix C.

4.4.2.3 Number of seeds per ear

Sowing date

Sowing date was an important source of variation with respect to seeds per ear in 2017 and 2018 ($P < 0.001$). In 2017, seeds per ear for the March and April sowing, and the April and May sowing did not differ significantly from each other, but seeds per ear for the March sowing were significantly greater than for the May and June sowing ($P < 0.001$) (Table 4.2). In 2018, seeds per ear for the March and April sowing and for the April and May sowing also did not differ significantly from each other, but seeds per ear from the May sowing were lower than from the March sowing ($P < 0.001$) (Table 4.2). There was a negative correlation ($R^2 = 0.73$) ($P < 0.05$) between sowing dates and seeds per ear because seeds per ear reduced with the delay in sowing (Figure 4.14).

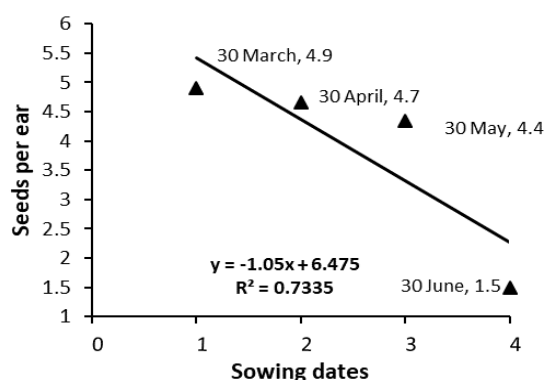


Figure 4.14 Correlation between sowing date and seeds per ear; data for March, April and May sowings are means for 2017 and 2018

Seed rate

In 2017, seeds per ear for the three lowest seed rates did not differ, but the 40 kg ha⁻¹ seed rate had more seeds per ear than the 80 kg ha⁻¹ seed rate (Table 4.2). In 2018, seeds per ear was significantly affected by seed rates ($P < 0.005$) (Table 4.2). Seeds per ear did not differ significantly between the two lower seed rates and also between the higher three seed rates, but the lowest seed rate had significantly more seeds per ear than the two highest seed rates (Table 4.2).

Cutting management

A significant effect of cutting on seeds per ear was recorded in 2017 and 2018. In 2017, seeds per ear did not vary significantly between uncut and single cut plants, but twice cut plants had significantly fewer seeds per ear ($P < 0.001$) (Table 4.2). In 2018, seeds per ear in uncut plants were significantly higher than that of single cut and twice cut plants ($P < 0.001$) (Table 4.2). There was a negative correlation ($R^2 = 0.87$) ($P < 0.05$) between cutting management and seeds per ear because seeds per ear reduced with increasing cutting frequency (Figure 4.15).

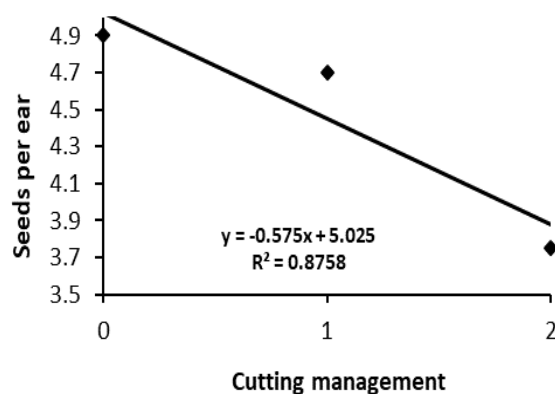


Figure 4.15 Correlation between number of herbage cuts and seeds per ear; data are means for 2017 and 2018

Interactions

There was a significant interaction between sowing date and cutting management on seeds per ear in 2017 because more seeds per ear were obtained from plants which were uncut ($P < 0.001$) (Table 4.2). Similarly, in 2018, seeds per ear were significantly affected by sowing date and seed rate, because more seeds per ear were produced from the March sowing at the lowest seed rate ($P < 0.01$) (Table 4.2). The interaction tables are attached in Appendix C.

4.4.2.4 Seeds per plant

Sowing date

Sowing date had a significant effect on seeds per plant in both years. March sowing had the highest number of seeds per plant and this reduced with each delay in sowing in both years ($P < 0.001$) (Table 4.2). There was a significant negative correlation ($R^2 = 0.99$) ($P < 0.05$) between sowing date and seeds per plant because seeds per plant were higher at the earlier sowing dates than the later sowing dates (Figure 4.16).

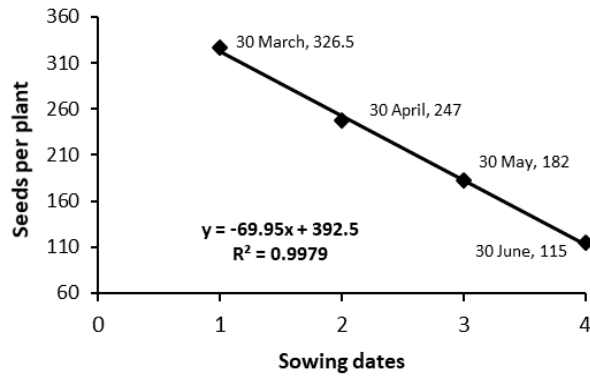


Figure 4.16 Correlation between sowing date and seeds per plant; data for March, April and May sowings are means for 2017 and 2018

Seed rate

In 2017, the seeds per plant did not differ among the seed rates. But in 2018, seeds per plant were significantly higher at the 20 kg ha^{-1} seed rate than the other three seed rates ($P < 0.001$) (Table 4.2).

Cutting management

Cutting was an important source of variation in both years. Cutting significantly reduced the number of seeds per plant in both years ($P < 0.001$) (Table 4.2). However, number of seeds per plant for uncut and once cut plants did not vary in 2017. There was a significant negative correlation ($R^2 = 0.95$) ($P < 0.05$) between cutting management and seeds per plant because seed numbers were reduced by cutting (Figure 4.17).

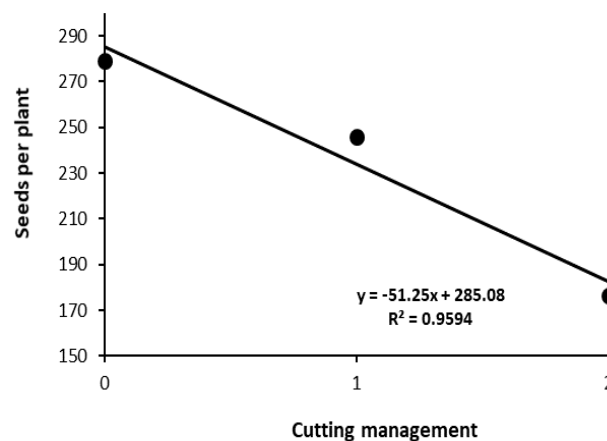


Figure 4.17 Correlation between number of herbage cuts and seeds per plant; data are means for 2017 and 2018

Interactions

In 2017, there was a significant interaction between sowing date and cutting management because the March sowing produced the highest number of seeds per plant ($P < 0.05$) (Table 4.2). No significant interactions occurred in 2018 (Table 4.2). The interaction tables are attached in Appendix C.

4.4.2.5 Seed yield (kg ha⁻¹)

Sowing date

Sowing date had a significant effect on seed yield in both years ($P < 0.001$) (Table 4.2). March sowing seed yield did not vary significantly with that of the April sowing, but May sowing reduced yield in both years. However, seed yield from the May sowing did not differ significantly from the June sowing in 2017 (Table 4.2). There was a significant correlation ($R^2 = 0.97$) ($P < 0.05$) between sowing dates and seed yield because seed yield reduced with each delay in sowing (Figure 4.18).

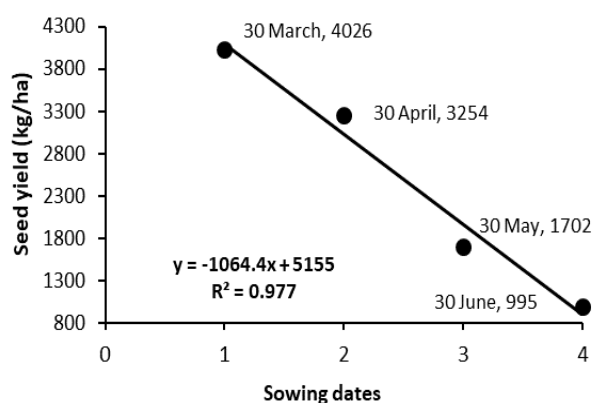


Figure 4.18 Correlation between sowing date and seed yield; data for March, April and May sowings are means for 2017 and 2018

Seed rate

In 2017, the seed yield did not differ among the three lowest seed rates, however seed yield from the 60 kg ha⁻¹ was greater than that for the 80 kg ha⁻¹ seed rate (Table 4.2). In contrast, in 2018 seed yield was significantly higher for the two lowest seed rates than the two higher seed rates ($P < 0.001$) (Table 4.2).

Cutting management

Cutting was an important source of variation in both years. In 2017, seed yield did not differ significantly between uncut and once cut plants, but it was significantly reduced by two cuts ($P < 0.001$) (Table 4.2). In 2018, cutting significantly reduced seed yield ($P < 0.001$) (Table 4.2). There was a significant negative correlation ($R^2 = 0.98$) ($P < 0.05$) between cutting management and seed yield because cutting reduced the seed yield (Figure 4.19).

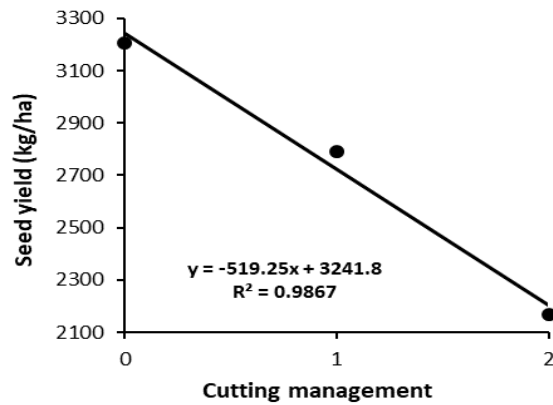


Figure 4.19 Correlation between number of herbage cuts and seed yield; data are means for 2017 and 2018

Interactions

In 2017, there was a significant interaction between sowing date and cutting management because the March sowing produced the highest seed yield in uncut plants, although the March sowing did not differ significantly with the April sowing and seed production from uncut plants did not differ from once cut plants ($P < 0.05$) (Table 4.2). Similarly, in 2018 there was a significant interaction between sowing date and cutting management because the March sowing produced the highest seed yield in uncut plants but the March and April sowings did not differ significantly with each other ($P < 0.001$) (Table 4.2). The interaction tables are attached in Appendix C.

Table 4.2 Main effect means of sowing date, seed rate and cutting management for teosinte seed yield and yield components in 2017 and 2018 at NCRP, Chitwan, Nepal

Main effect means of:	Cobs per plant		Ears per cob		Seeds per ear		Seeds per plant		Total seed yield	
	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Sowing date (SD)										
30-Mar	11.5a	8.4a	6.8a	6.3a	5.0a	4.8a	394a	259a	4233a	3791a
30-Apr	10.1ab	7.8ab	6.0a	5.8a	4.6ab	4.7ab	287b	207b	3440a	3054a
30-May	8.7b	7.0b	5.1b	4.4b	4.4b	4.3b	212c	152c	1778b	1618b
30-Jun	6.3c	-	4.0c	-	3.0c	-	115d	-	998b	-
Linear contrast p value	<.001	0.007	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	1.6	0.9	0.8	0.6	0.4	0.5	72	28	924	859
CV%	11.2	6.5	9.3	6.9	6.3	5.8	18	8	23	19
Seed rate (SR)										
20 kg ha ⁻¹	9.2a	8.6a	5.3a	6.0a	4.2ab	4.9a	248a	253a	2605ab	3475a
40 kg ha ⁻¹	9.4a	8.0b	5.7a	5.5ab	4.4a	4.6ab	272a	212b	2628ab	3287a
60 kg ha ⁻¹	9.3a	7.5b	5.4a	5.5ab	4.3ab	4.4b	256a	193bc	2850a	2772b
80 kg ha ⁻¹	8.6a	6.8c	5.5a	5.0b	4.1b	4.5b	232a	167c	2366b	1751c
Linear contrast p value	0.153	<.001	0.793	0.001	0.191	<.005	0.325	<.001	0.439	<.001
LSD (0.05)	0.9	0.5	0.4	0.5	0.2	0.3	41	16	406	430
CV%	14	7.2	10.7	10.9	7.7	7.7	23	15	22	18
Cut management (CM)										
No cut	10.3a	8.3a	5.8a	6.2a	4.7a	5.1a	296a	262a	3092a	3311a
One cut	9.8a	7.7b	5.7a	5.7b	4.6a	4.8b	276a	216b	2725a	2850b
Two cut	7.3b	7.1c	5.0b	4.6c	3.5b	4.0c	184b	169c	2019b	2303c
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	0.7	0.4	0.3	0.4	0.2	0.2	30	21	406	227
CV%	20.7	11.8	17	18.6	11.1	13	34	25	44	20
Significance of interactions of linear contrasts (p value)										
SD(lin)x SR(lin)	0.575	0.022	0.722	0.508	0.261	0.016	0.772	0.782	0.054	0.82
SD(lin)x CM(lin)	0.016	0.639	<.001	0.007	<.001	0.393	0.001	0.738	0.046	<.001
SR(lin)x CM(lin)	0.66	0.01	0.844	0.087	0.088	0.954	0.655	0.42	0.781	0.632
SD(lin)x SR(lin) x CM(lin)	0.851	0.021	0.53	0.279	0.306	0.613	0.848	0.641	0.446	0.14

Note: Kg/ha = Kilograms ha⁻¹; LSD = Least Significant Difference; CV = Coefficient of Variation. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cut management.

4.4.3 Main effect means of seed yield based on the position of cobs

Sowing date

The seed yield from each inflorescence position (top (T), middle (M) and bottom (B)) was significantly affected by sowing date. T yield from the March and the April sowing did not differ but was significantly higher than the later sowings in both years ($P < 0.001$) (Table 4.3). For M cobs in 2017 seed yield for the March sowing was significantly higher than the other sowings and yield reduced subsequently with delayed sowing ($P < 0.001$). In 2018, seed yield for the March and the April sowing did not differ and that for the April and the May sowing did not differ with each other for the M cobs ($P < 0.05$) (Table 4.3). For B cobs in 2017, seed yield for the March sowing was greater than that for the April and the May sowings in both years ($P < 0.001$) (Table 4.3). There was a significant negative correlation between sowing dates and seed yield from the top positioned ($R^2 = 0.92$) ($P < 0.05$), the middle positioned ($R^2 = 0.97$) ($P < 0.05$) and the bottom positioned cobs ($R^2 = 0.94$) ($P < 0.05$) because seed yield from the top, middle and bottom positioned cobs reduced with each delay in sowing (Figure 4.20, 4.21, 4.22).

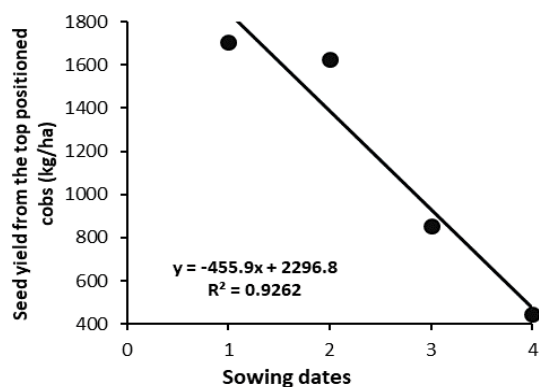


Figure 4.20 Correlation between sowing date and seed yield from top positioned cobs; data for March, April and May sowings are means for 2017 and 2018

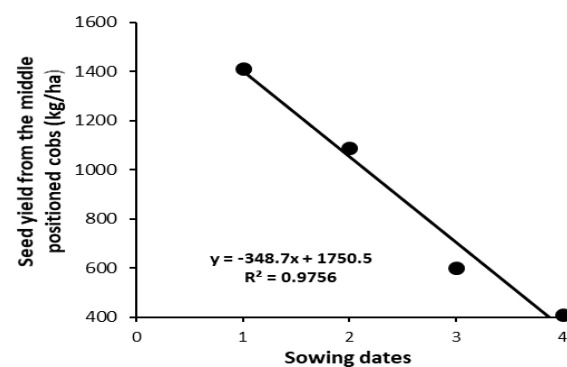


Figure 4.22 Correlation between sowing date and seed yield from middle positioned cobs; data for March, April and May sowings are means for 2017 and 2018

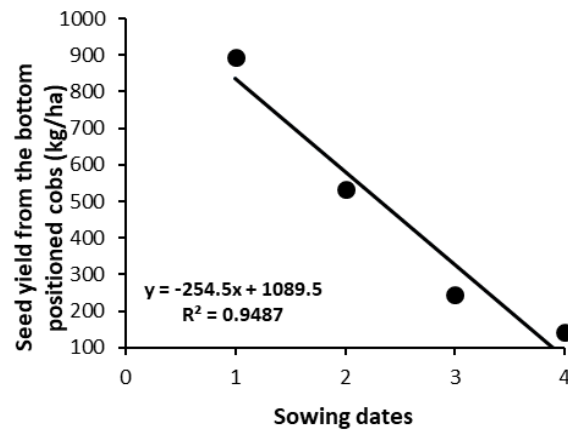


Figure 4.22 Correlation between sowing date and seed yield from bottom positioned cobs; data for March, April and May sowings are means for 2017 and 2018

Seed Rate

In 2017, there were no significant differences among the seed rates for T seed yield. But in 2018, the seed yield was highest for the 20 kg ha⁻¹ and the 40 kg ha⁻¹ seed rates but they did not differ with each other ($P < 0.001$) (Table 4.3). For the M cobs in 2017, seed yield did not differ among the three lowest seed rates while in 2018, seed yield did not differ between the 20 kg ha⁻¹ seed rate and the 40 kg ha⁻¹ seed rate while the 40 kg ha⁻¹ seed rate did not differ from that of 60 kg ha⁻¹ seed rate ($P < 0.001$) (Table 4.3). For B cobs in 2017 and 2018 seed yield for the three lowest seed rates did not differ and in 2018 yield from the three lowest seed rates was significantly higher than for the highest seed rate ($P = 0.001$) (Table 4.3). There was a negative correlation between seed rates and seed yield from the top positioned ($R^2 = 0.85$) ($P > 0.05$), middle positioned ($R^2 = 0.82$) ($P > 0.05$) and bottom positioned cobs ($R^2 = 0.42$) ($P > 0.05$) because seed yield from the top, middle and bottom positioned cobs reduced with an increase in seed rate (Figure 4.23, 4.24, 4.25).

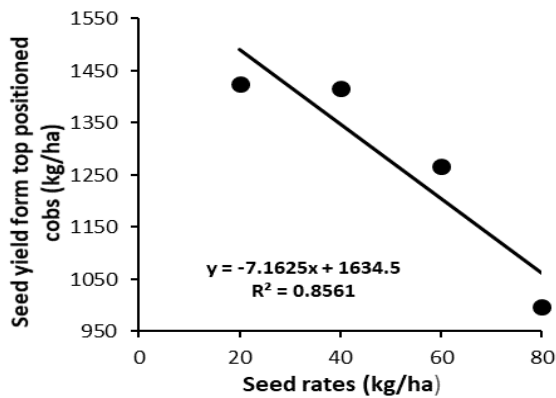


Figure 4.3 Correlation between seed rate and seed yield from top positioned cobs; data are means for 2017 and 2018

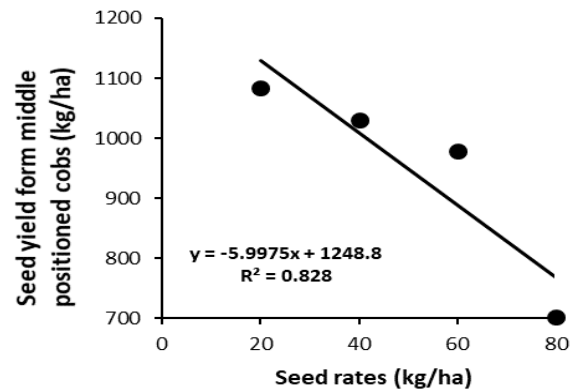


Figure 4.4 Correlation between seed rate and seed yield from middle positioned cobs; data are means for 2017 and 2018

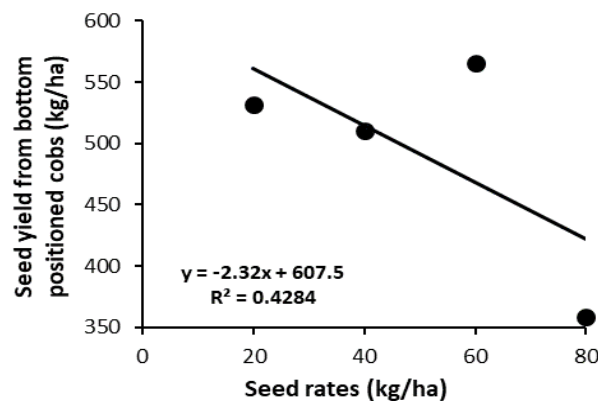


Figure 4.5 Correlation between seed rate and seed yield from bottom positioned cobs; data are means for 2017 and 2018

Cutting Management

Cutting management was an important source of variation with respect to seed yield from all cob position in both years ($P < 0.01$) (Table 4.3). The seed yield from the T and M cobs did not vary between uncut and once cut plants but was significantly higher than the twice cut plants in 2017 ($P < 0.001$) (Table 4.3). In 2018 cutting reduced the seed yield for the T cobs ($P < 0.001$) (Table 4.3). For the M cobs uncut and once cut plants produced higher seed yield than the twice cut plants in both years ($P < 0.05$) (Table 4.3). For the B cobs, seed yield did not vary for uncut and once cut plants and one cut did not differ from the twice cut plants ($P = 0.001$) in 2017 but cutting reduced the seed yield significantly ($P < 0.001$) (Table 4.3) in 2018. There was a significant negative correlation between cutting management and seed yield from the top positioned cobs ($R^2 = 0.97$) ($P < 0.05$) because seed yield from the top positioned cobs reduced with increase in cutting

frequency. For the middle ($R^2=0.99$) ($P<0.05$) and the bottom positioned cobs ($R^2=0.99$) ($P<0.05$), the correlation was highly significant and negative (Figure 4.26, 4.27, 4.28).

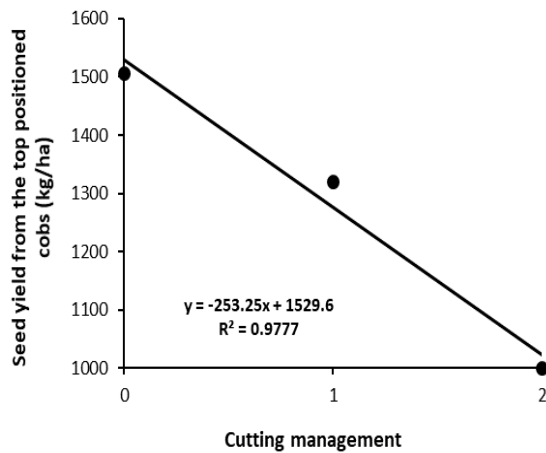


Figure 4.26 Correlation between number of herbage cuts and seed yield from top positioned cobs; data are means for 2017 and 2018

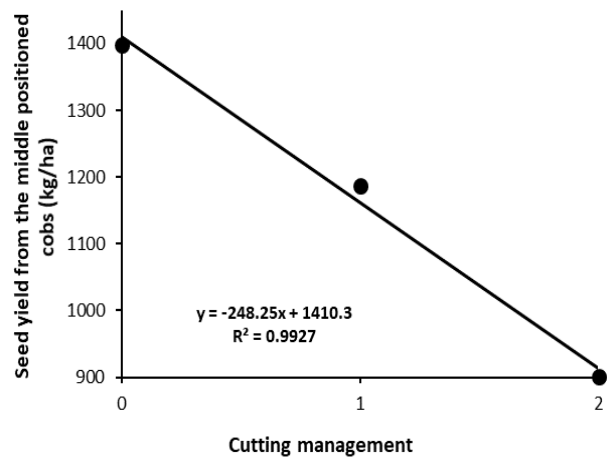


Figure 4.27 Correlation between number of herbage cuts and seed yield from middle positioned cobs; data are means for 2017 and 2018

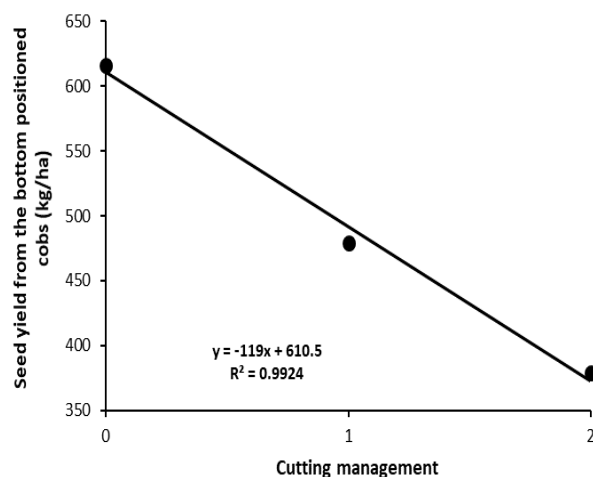


Figure 4.6 Correlation between number of herbage cuts and seed yield from bottom positioned cobs; data are means for 2017 and 2018

Interactions

A significant interaction between sowing date and cutting management was recorded for seed yield from the top positioned cobs in 2017 because the March and the April sown plants produced higher seed yield from uncut and once cut plants ($P<0.05$)(Table 4.3). A significant interaction was recorded between sowing date and cutting management for seed yield from the middle position cobs in both years because uncut and once cut plants produced higher seed yield in the March sowing. A significant interaction between sowing date and seed rate occurred for seed yield from the middle positioned cobs in 2018 because the highest seed rate produced the lowest seed yield

due to the reduced growth period of the May sowing ($P < 0.01$) (Table 4.3). Likewise a significant relationship between sowing date and cutting management for the bottom position cobs occurred because cutting reduced seed yield for the March sowing in 2018 ($P = 0.012$) (Table 4.3). The interaction tables are attached in Appendix C.

Table 4.3 Main effect means of sowing date, seed rate and cutting management on teosinte seed yield from different cob positions in 2017 and 2018 at NCRP, Chitwan, Nepal

Main effect means of:	Seed yield based on the cob position (Kg/ha)					
	Top		Middle		Bottom	
	2017	2018	2017	2018	2017	2018
Sowing date (SD)						
30-Mar	1695a	1717a	1487a	1336a	1051a	738a
30-Apr	1639a	1612a	1111b	1069ab	690b	374b
30-May	833b	872b	688c	515b	258c	231b
30-Jun	444b	-	412c	-	142c	-
Linear contrast p value	<.001	<.001	<.001	0.015	<.001	0.001
LSD (0.05)	389.7	267	339	616	276	246
CV%	22	12	24	40	34	34
Seed rate (SR)						
20 kg ha-1	1133a	1716a	928ab	1241a	544ab	519a
40 kg ha-1	1158a	1676a	957ab	1104ab	514ab	507a
60 kg ha-1	1161a	1373b	1022a	935b	667a	464a
80 kg ha-1	1158a	836c	791b	613c	416b	301b
Linear contrast p value	0.791	<.001	0.255	<.001	0.372	0.002
LSD (0.05)	190.8	260.9	192	190	162	129
CV%	23	22	29	23	42	34
Cut management (CM)						
No cut	1316a	1699a	1097a	1058a	679a	554a
One cut	1231a	1410b	963a	1010a	530ab	429b
Two cut	911b	1091c	712b	8514	396b	361b
Linear contrast p value	<.001	<.001	<.001	0.007	0.001	<0.001
LSD (0.05)	166.7	137	167	147	170	80
CV%	41	24	51	37	90	44
Significance of interactions of linear contrasts (p value)						
SD(lin)x SR(lin)	0.679	0.117	0.346	0.050	0.082	0.293
SD(lin)x CM(lin)	0.045	0.240	0.034	0.048	0.579	0.012
SR(lin)x CM(lin)	0.412	0.660	0.476	0.362	0.440	0.284
SD(lin)x SR(lin) x CM(lin)	0.602	0.445	0.284	0.412	0.201	0.163

Note: Kg/ha = Kilograms ha⁻¹; LSD = Least Significant Difference; CV = Coefficient of Variation. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cut management

4.5 Prediction of teosinte seed yield on the basis of growing degree days (GDD)

Table 4.4 gives the GDD accumulation until seed harvest and the seed yield for each sowing in 2017 and 2018. The earliest sowing accumulated the greatest number of GDD and the highest seed yield; both GDD and seed yield declined as sowing was delayed. This information was used to calculate the teosinte seed loss which would occur with each delayed sowing.

In Figure 4.29, the mean seed yield is plotted against mean GDD accumulation, where the mean is over the two seasons, 2017 and 2018. Then a regression line was fitted to the four points. There was a significant correlation ($R^2 = 0.97$) between GDD and seed yield. The estimated slope of the line was 2.2139, which meant that 2.21 kg/ha of seed yield was lost for each GDD unit lost. In other words, the prediction shows that for each gain in 100 GDD, seed yield will be increased by 222kg/ha.

Table 4.4 Growing degree day's accumulation for teosinte seed harvest for each sowing date

Main effect means of:	Growing degree days (GDD)		Seed yield(kg/ha)	
	2017	2018	2017	2018
Sowing date (SD)				
30-Mar	3799	3646	4260	3791
30-Apr	3364	3230	3454	3054
30-May	2857	2761	1785	1618
30-Jun	2330	-	995	-

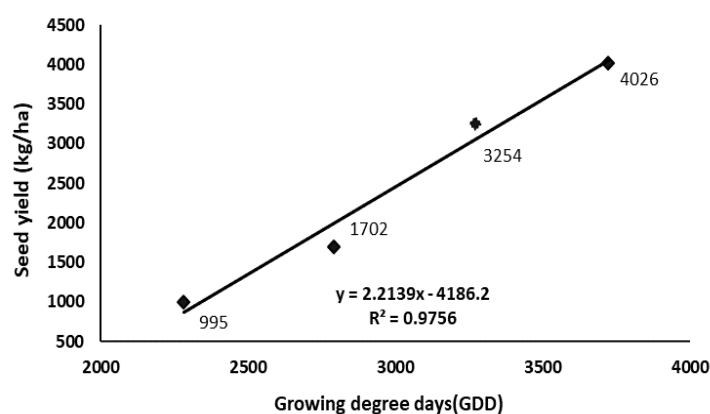


Figure 4.29 Relationship between sazGDD and seed yield of teosinte

4.6 Association of vegetative and reproductive components with seed yield in teosinte

For the different sowing dates, plant height, number of tillers, LAI and cobs per plant were the important contributors to determining the total seed yield of teosinte. Seed yield was positively correlated with plant height in both seasons. The correlation was not statistically significant in 2017, but was significant in 2018 ($P < 0.05$) (Figure 4.30). Similarly, tiller number was significantly correlated with seed yield in 2017 ($P < 0.05$) (Figure 4.31) and 2018 ($P < 0.01$) (Figure 4.31). In 2017, LAI was positively correlated with seed yield in 2017 ($P < 0.05$) and in 2018 ($P < 0.01$) (Figure 4.32). Cobs per plant were positively correlated with the seed yield ($P < 0.05$) (Figure 4.33). Ears per cob were significantly correlated with seed yield in 2017 ($P < 0.01$) and 2018 ($P < 0.05$) (Figure 4.34). Seed yield was positively correlated with seeds per ear in both seasons. The correlation was not statistically significant in 2017, but was significant ($P < 0.01$) in 2018 (Figure 4.35). No statistically significant correlations between the yield attributes and seed rates were recorded. For cutting management, seed yield was positively correlated ($P < 0.05$) with tiller number in 2018 but this was not significant in 2017 (Figure 4.36). Similarly, average cobs per plant were correlated with seed yield ($P < 0.05$) (Figure 4.37). Ears per cob were significantly correlated with seed yield in both years ($P < 0.05$) (Figure 4.38). Similarly, seeds per ear were positively correlated with the seed yield in both seasons ($P < 0.05$) (Figure 4.39).

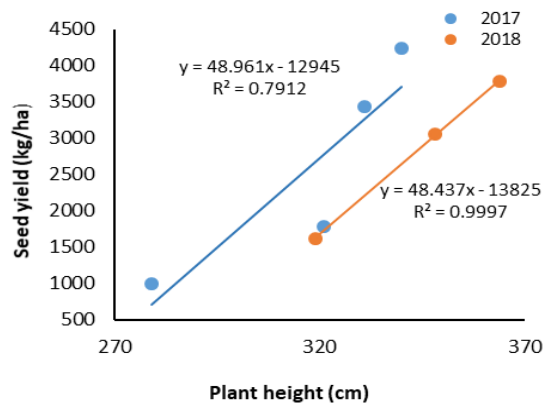


Figure 4.30 Correlation between plant height and seed yield using main effect means of different sowing dates

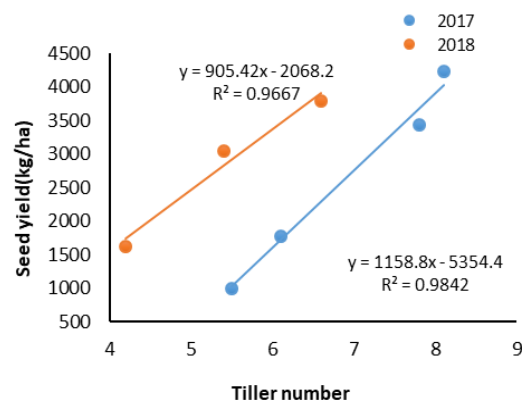


Figure 4.31 Correlation between tiller number and seed yield using main effect means of different sowing dates

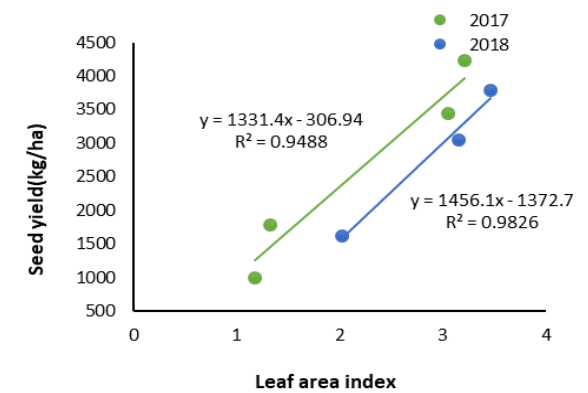


Figure 4.32 Correlation between leaf area index and seed yield using main effect means of different sowing dates

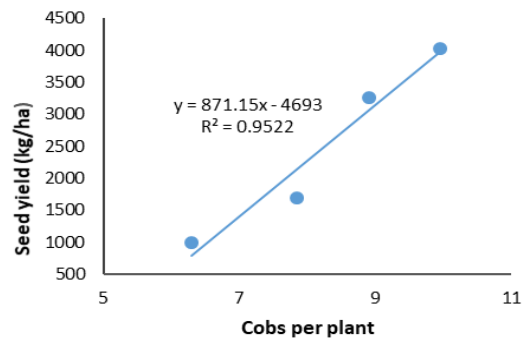


Figure 4.33 Correlation between cobs per plant and seed yield using main effect means of different sowing dates (Average mean of both years)

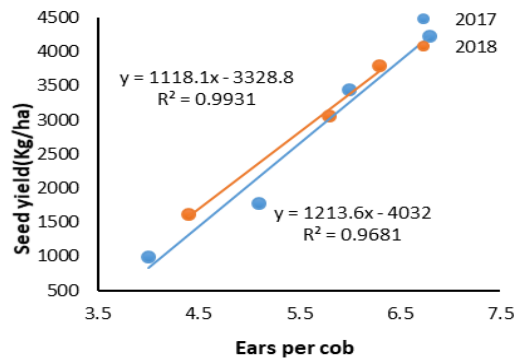


Figure 4.74 Correlation between ears/cob and seed yield using main effect means of different sowing dates

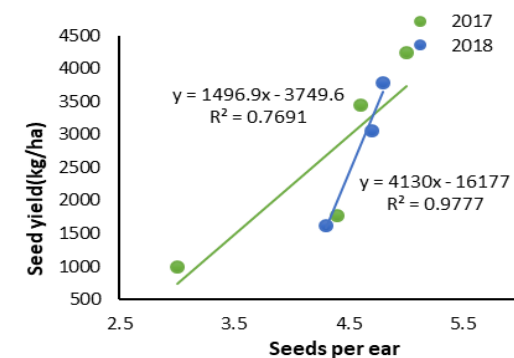


Figure 4.35 Correlation between seeds/ears and seed yield using main effect means of different sowing dates

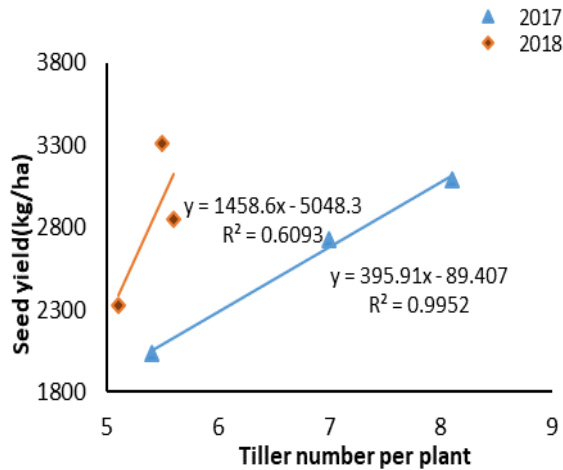


Figure 4.36 Correlation between tiller number and seed yield using main effect means of different cutting management

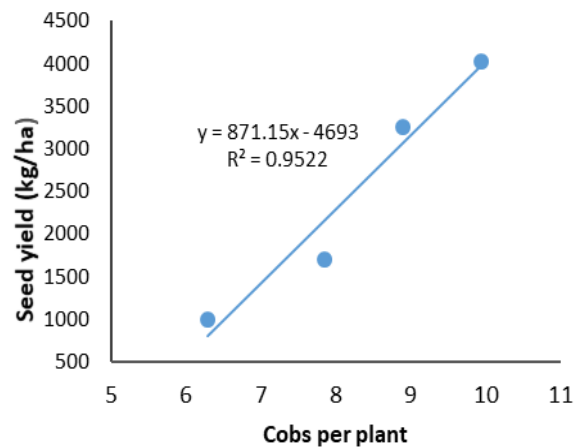


Figure 4.37 Correlation between cobs/plant and seed yield using main effect means of different cutting management (Average mean of both years)

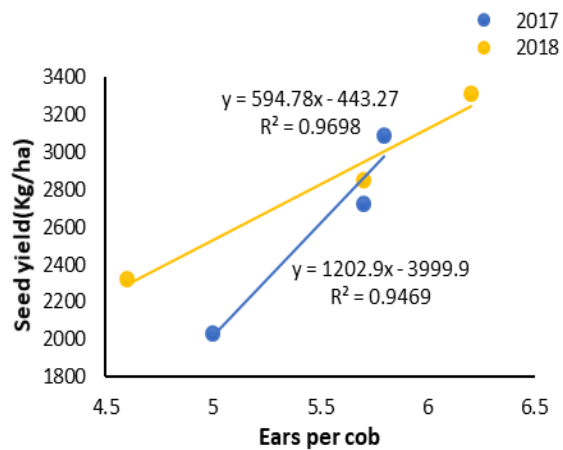


Figure 4.38 Correlation between ears/cob and seed yield using main effect means of different cutting management

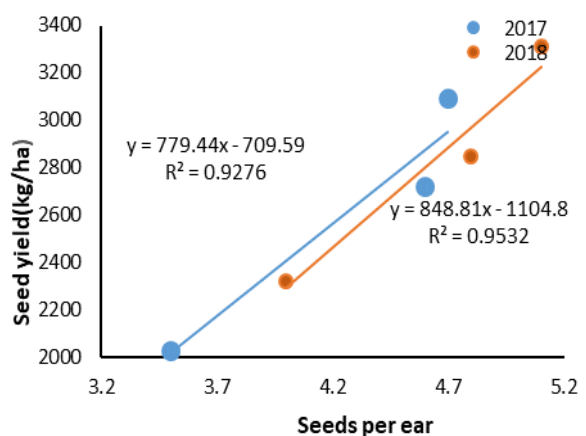


Figure 4.39 Correlation between seeds/ear and seed yield using main effect means of different cutting management

4.7 Discussion

4.7.1 Main effect means of sowing date, seed rate and cutting management on plant properties of teosinte

4.7.1.1 Final plant height

Sowing date

In 2017, plant height did not differ among the first three sowing dates but was reduced by the June sowing. This was because the shorter growing period in June was accompanied by lower temperature. The temperature during vegetative growth in June 2017, was lower by 1.5, 1.2 and 0.9°C than for the March, April, and May sowings respectively. This lower temperature and

shortened growth period decreased the accumulation of GDD. Lower GDD (Badu-Apraku et al., 1983) as a result of lower temperature following delayed sowing hinders plant growth (Bonaparte, 1975; Uprety & Reddy, 2016; Van Dobben, 1962). A shorter growing period due to later sowing limits the accumulation of assimilates because of a reduced opportunity for photosynthesis and plant height is reduced (Koireng et al., 2018). Similar findings have been reported by Al-Darby and Lowery (1987). High rainfall (11.9, 15.8, 10.0 mm rain in July, August and September) during vegetative growth for the June sowing may also have affected plant growth (Figure 2.2 in Chapter 2). Excess water during early vegetative growth hinders plant growth through poor root development due to aeration and nutrition problems (Sprague & Dudley, 1988). This result is in line with the findings of Reiad, Hamada, Elmaaboud and Khalil (2014) in pearl millet. Lower incident PAR for the delayed sowing might be another reason for shorter plant height. This result was in line with the findings of Cirilo and Andrade (1994) in maize. In 2018, the response differed because plants from the May sowing were shorter than the first two sowings. Reduced plant height for the May sowing in 2018 might have been because of lower GDD accumulation, otherwise it is not clear why the May sowing had a low plant height. Within sowing dates, the difference in plant height between the two years can be explained by the temperature and rainfall difference as in 2017 temperature was higher by 0.5°C and rainfall by 10.1mm than in 2018.

Seed rate

Seed rate had no major effect on the height of teosinte plants at maturity in either year. This result was similar to several plant density studies in maize (Carpici, Celik & Bayram, 2010; Yilmaz, Gozubenli, Knuskan & Atis, 2007). This might have been because the plant population at the highest seed rate was not sufficient to produce the interplant competition for light and resources which can result in reduced plant height (Abuzar et al., 2011).

Cutting management

At maturity, cutting had significantly reduced plant height in both years ($P < 0.001$). Reduction of food reserves and a shorter growing period accumulated lower GDD for the second cutting. Cutting immediately removes photosynthetic capability and the resources required to support regrowth (Arif et al., 2012; Noy-Meir & Briske, 2002). Similar results were reported by Abu-Shakra et al. (1977).

Interactions

In 2017, at maturity, cutting reduced the plant height for all sowing dates while in 2018, cutting reduced the plant height for different seed rates. The former result was in line with Asal (2019) who reported a significant interaction between sowing date and cutting time in sorghum because

cutting allowed the plant to carry on growing and elongating under a favourable environment as a result of timely sowing. Reduction in plant height due to cutting might be a result of decreased photosynthetic activity. Jones, Alpuerto, Tracy and Fukao (2017) reported similar findings where cutting reduced plant growth which they ascribed to reduced assimilates supply.

4. 7.1.2 Tiller numbers per plant

Sowing date

Tillers are a crucial component of plants directly related to biomass accumulation and seed yield in the majority of seed crops (Diawara, 2012; Kuraparthi, Sood & Gill, 2008). In the present study, tiller numbers did not differ among the first two sowing dates in 2017. In 2018 the March sowing had the highest tiller number and this reduced with each delay in sowing. In 2017, a longer growing period, high temperature, adequate soil moisture and better incident PAR allowed increased tiller numbers in early sown teosinte as a result of higher GDD accumulation (refer to Table 3.1-3.4 in Chapter 3). This result is also in line with the findings of Praeger (1977) and Stickler and Pauli (1961) in sorghum who reported that high temperature increased tiller number by accelerating the production of leaves. Similar findings of Yoshida (1973) in rice and Jan, Hamid and Muhammad (2000) in wheat showed that higher temperatures resulted in increased tiller numbers. Acharya, Marahatta and Amgain (2017) reported a shortened vegetative growth period, because of changes in photoperiod that lead to the start of stem extension for flowering in delayed sowings. Aspinall and Paleg (1964) and Friend (1965) reported an increase of tiller number with increase in light intensity as a result of increased photosynthesis which was similar to the findings of the present study where early sowings had higher incident PAR. Similar tiller numbers for the March and April sowings in 2017 was likely because of the similar climatic conditions (temperature lower by 0.3 °C and incident PAR lower by 0.3 MJ/m²/day) under higher moisture conditions. The difference in tillers number between 2017 and 2018 is likely again to be a result of temperature and rainfall difference in both years. As already noted 2017 had higher temperature and rainfall than 2018. Higher temperature allows the accumulation of more GDD, increasing tiller numbers in teosinte (Alam et al., 2014). Lower temperatures in 2018 reduced the GDD accumulation, resulting in less vegetative growth. Langer (1963) reported that tiller production by grasses is affected by environmental factors like photoperiod, solar radiation intensity, temperature, and soil moisture.

Seed rate

Seed rate had no major effect on tiller numbers of teosinte in either year in the present study. Breakage and lodging of tillers due to heavy rainfall and wind during the earlier stage of teosinte

(Figure 4.40-4.43) growth lead to a natural thinning of plants which might have reduced competition among the plants for light and nutrients. Similar findings were reported by Jan, Hamid and Muhammad (2000) and Nazir, Ahmad, Siddauqe and Riaz (1987) in wheat. Lodging was prominent in the first sowing and it reduced as the sowing was delayed in both years. Plants from the June 2017 sowings had the lowest damage due to rain and wind. Lodging was maximum in the plots with highest density of 80 kg/ha (73 % plants lodged) and decreased as the density decreased to 20kg/ha (55% plants lodged) for all the sowings. Similar results of stalk lodging of up to 60 % in maize at higher planting density have been reported by Gou et al. (2008). At higher planting density, mutual shading reduces the light intensity on the lower leaves which may affect stalk strength (Maddonni & Otegui, 2004; Tollenaar & Aguilera, 1992). Lodging resistance of plants at lower plant density can be explained by increase in thickness of stem due to better availability of resources to individual plants (Poehlmann & Sleper, 1995). Dahiya, Kumar, Chaudhary and Chaudhary (2018) also reported that plants at narrow spacing elongate very quickly resulting in thinner and lighter plants, which are weak and they tend to lodge. Less lodging in the June sowing can also be explained by the reduced plant height, which is in line with the findings of Cooper (1981) and Mancuso and Caviness (1991).



Figure 4.40 Breakage from the regrowth



Figure 4.41 Lodging of teosinte plants



Figure 4.8 Breakage of main stem



Figure 4.43 Breakage of stem from the base

Cutting management

In 2017, both cuts reduced tillers numbers, while in 2018 the number of tillers in uncut and single cut plants did not differ but was reduced in twice cut plants. Cutting reduced leaf area through the removal of green leaves, reducing the light interception which then interrupted photosynthesis, hindering the supply of assimilates to the root system for regrowth of tillers. This result is in line with the findings of Parsons (1988) and Jewis (1972) who reported that removal of axillary buds at the base of the plants by frequent cutting reduced tiller number in perennial grasses and sweet sorghum hybrids (Muldoon, 1985). Similar tiller numbers in uncut and once cut plants in 2018 are likely to be the impact of ambient temperature and solar radiation penetration to the tiller buds that were exposed after one cut. Exposure to favourable environmental conditions increases the tiller growth as a result of accumulation of more GDD and increased photosynthesis. Long days favour tiller initiation and development (Hakala, Kontturi, & Pahkala, 2009). The results in 2017 were similar to the findings of Naveed et al. (2014) in wheat and Schneider, Caron, Elli, Schwerz and Engroff (2019) in sorghum where uncut plants produced more tillers. del Pozo Ibanez (1963) and Devkota, Pokharel, Paudel, Upreti and Joshi (2015) also reported that tiller numbers were temporarily reduced by cutting. For 2017, differences in tiller numbers between uncut and cut plants may have been because of the lower radiation interception by the twice cut plants, which resulted in a reduced supply of photosynthates. A similar finding was reported by Hammer et al. (2010) in other field crops.

Interactions

There were no significant interactions among sowing date, seed rate and cutting management on tiller numbers in both years because cutting reduced the tiller number for all sowing date irrespective of seed rate.

4.7.1.3 Leaf Area Index

Sowing date

Distribution of leaves regulates the interception of solar radiation, gaseous exchange and the maintenance of temperature around the plant canopy. The highest LAI was recorded for the first and second sowing and LAI reduced in the subsequent sowings in both years. The extended growth periods at higher temperature which resulted in greater leaf areas and leaf area indices allowed better radiation interception for the first two sowings (March and April) resulting in higher LAI. This result is similar to the findings of Mukumbuta (1993) in maize. Tollenaar, Daynard and Hunter (1979) and Warrington and Kanemasu (1983) reported greater leaf canopy expansion under high temperature through increased cell expansion (Newton, 1958). Further, high temperatures activate enzymes and increase the rate of leaves unfolding (Milthorpe, 1959). Tsimba et al. (2013) reported that higher LAI in earlier sown plants might be a result of temperature variations between earlier and delayed sowings in maize. Similar findings were reported by El-Khoby (2004) in rice and Hay (1990) in cereals. Variation in LAI between the two years is mainly because of temperature and moisture variation as 2018 had lower temperature and precipitation than 2017. This result is in line with the findings of Kiziloglu, Sahin, Kuslu and Tunc (2009) in silage maize. There was also a reduced photoperiod due to the lower temperature during vegetative growth in 2018, as also reported by Allison and Daynard (1979) in maize.

Seed rate

LAI varied significantly with seed rate in both years where the lowest LAI occurred in the highest seed rate. However in 2017, LAI did not differ for the three highest seed rates and in 2018 LAI was similar for the lowest three seed rates. Factors affecting LAI include competition between the plants for water, sunlight, nutrients, and space, and other agronomic factors like sowing date, tillage, weed and pest control (Begna et al., 2001). Similar seed rate response were reported by Crozier, Gehl, Hardy and Heiniger (2014) and Van Roekel and Coulter (2012) in maize. The highest LAI at the lowest seed rate in both years is probably because there was less competition for space for each plant, which allowed more effective light interception and better conversion of photosynthates (Tollenaar, Aguilera & Nissanka, 1997). In the present study, at the highest seed rate, leaf shading reduced light interception and caused drying and decay of lower leaves. Similar

results have been reported by Bavec and Bavec (2002) in maize where up to 27 % of the plant leaves decayed at higher densities.

Cutting management

LAI in both years was higher in uncut plants than the cut plants. Cutting removed leaves, and reduced the plant's ability to regenerate the leaves and tillers. This reduced LAI because of the reduced photosynthetic tissue area which then lowered the assimilate supply for the regrowth of leaves and tillers. Similar results have been reported by Donaghy, Turner, and Adamczewski (2008) and Morvan-Bertrand, Boucaud, and Prudhomme (1999) in tall fescue and perennial ryegrass respectively.

Interactions

LAI was significantly affected by the interaction between sowing date and seed rate in both years. This was because better weather conditions for the early sowing extended the vegetative period and allowed greater light interception (Table 3.1-3.4 in Chapter 3). The interaction between seed rate and cutting management in 2018 might have resulted from the higher light interception at low seed rates and reduced apical dominance in cut plants.

4.7.2 Main effect means of effect of sowing dates, seed rates and cutting management on yield components

4.7.2.1 Cobs per plant

Sowing date

The higher the cob numbers, the higher will be the seed yield. Significant variation for total cobs per plant was recorded among the different sowing dates in 2017 and 2018, where the two earlier sowings produced the highest number of cobs per plant. Earlier sown plants have a better source sink relationship, as a result of prolonged growth period that allowed accumulation of higher GDD. Better solar radiation interception by a larger leaf area resulting in increased assimilate supply to the seed in an extended growing season has been reported to result in more cobs in early sown crops by Banotra et al. (2017) and Shaheenuzzamn, Saha, Ahmed, Rahman and Salim (2015) in sweet corn and by Earley (1974) and Sangoi (1996) in maize. A high number of cobs per plant is an important strategy to increase maize yield (Bos, Tijani-Eniola & Struik, 2000). Fewer cobs in the later sowings may be an effect of lower temperature, which consequently affected reproductive growth. The variations in cobs number per plant between the two years reflected the lower temperature and rainfall during the vegetative and reproductive stage in 2018, which reduced the amount of accumulated growing degree days that slowed down teosinte growth.

Seed rate

Seed rate had no major effect on the number of cobs per plant in 2017 which is possibly due to breakage of main stem and tillers which occurred during the vegetative growth stage in 2017 (Figures 4.31 - 4.34). Average rainfall during the vegetative stage in 2017 was higher than in 2018. This is in agreement with Duncan (1984) and Tollenaar, Deen, Echarte and Liu (2006) who reported that the impact of crowding stress is also influenced by climatic conditions (precipitation, temperature and solar radiation). Similar findings have been reported by Banotra et al. (2017), Norwood (2001) and Pavlista, Lyon, Baltensperger, and Hergert (2010) in maize. However in 2018, the highest seed rate produced the lowest number of cobs per plant and the lowest seed rate had the highest cobs per plant. Lemcoff and Loomis (1994), Shranabasappa and Basavanneppa (2019) and Raja (2001) all reported an increase in maize cobs per plant due to better physiological and photosynthetic activity of individual plants at lower plant densities. Assefa et al. (2016) reported that maize yield increases sharply at lower density and decreases at the higher density in the regions with lower latitudes. Higher number of cobs at lower density could be the result of great reserve accumulation per plant because that leads to better ventilation and light penetration to the base of the plant. This result is in line with the findings of Carmi et al. (2006) and Godsey, Linneman, Bellmer, and Huhnke (2012) who reported larger sorghum spikes at low plant density. Another possible reason for less cobs at higher density could be the shading effect which result in carbon starvation due to competition for resources between plants (Parsons, 1988).

Cutting management

Cutting was a source of variation for cobs per plant in both years. In 2017 uncut plants and once cut plants produced the same number of cobs per plant but this decreased in twice cut plants. But in 2018, uncut plants had more cobs than the cut plants, possibly because of the inability of the latter to compensate for the reduced photosynthesis after cutting (Barimavandi, Sedaghatoor, & Ansari, 2010). As reported by Koptur, Smith and Lawton (1996), absence of leaves due to cutting reduces the resources for reproductive growth. The higher temperature and precipitation in 2017 provided a better environment for regrowth of the once cut plants.

Interactions

There was a significant interaction of sowing date and cutting management in both years, because of the synergetic influence of early sowing on uncut plants which developed robust crowns producing better seed yield attributing parameters (Kumar et al., 2017). In 2018, there was also an interaction between the seed rate and cutting management; less competition among the plants allowed better utilization of solar radiation at lower plant density. The interaction among

sowing date, seed rate and cutting management arose because more leaves in uncut plants were better able to convert source into sink in a less competitive environment at a longer growth period.

4. 7.2.2 Ears per cob

Sowing date

While numbers of ears per cob are the primary determinant of yield, the number of seeds produced by each ear is also important. The number of ears per cob was significantly higher for the first two sowing dates in both years because of the longer growth period due to early sowing which allowed the accumulation of higher GDD. This result is similar to the findings of Farsiani, Ghobadi, & Jalali-Honarm (2011) who reported ears per cob being affected by environmental conditions and cultivation. Lower temperature at the seed filling stage at the late sowings slowed the reproductive process and also reduced the supply of assimilates (Tollenaar & Daynard 1978). This result is similar to the findings of Ugur and Maden (2015) in sweet corn and Idikut, Cesur and Tosun (2005), Oktem, Oktem and Coskun (2004), Turgut and Balci (2002) and Williams (2008) in maize. The difference between the numbers of ears per cob in the two years can be ascribed by the temperature difference that allowed accumulation of more GDD and faster growth in 2017. This result is in line with the findings of Sangoi (1996) in maize.

Seed rate

Seed rates had no effect on ears per cob in 2017 which has been explained in 4.7.2.1. However there was a difference in 2018 between the lowest and highest sowing rates. In 2018, the lower ears per cob for the highest seed rate is likely to be because of interplant competition for water, nutrients and light among the plants. This result is in line with the findings of Bhatt (2012) in sweet corn and Sobhana, Ashok and Ishwar (2013) in baby corn who reported that higher resources availability and better photosynthetic and physiological activity of the individual plants at lower plant densities increase the ears per cob.

Cutting management

Cutting had a significant effect on ears per cob in both years. In 2017 once cut and uncut plants produced the same number of ears per cob but in 2018 once cut plants had fewer ears per cob. In the first year the once cut plants, because of better growing conditions, were able to quickly recover from the effects of cutting, but growing condition in 2018 did not allow that to occur. Double cutting in both years did not allow plants to produce the same number of ears per cob because of resource constraints. Similar findings were reported by Musa, Gondal, Riaz, Hayat and Haider (2021) in berseem clover.

Interactions

There was a significant interaction of sowing date and cutting management in 2017 and 2018 because the longer growth period for the early sowing increased the capacity of plants to synthesize more photosynthates for growth and storage in other parts of the plant (Wardlaw, 1990).

4. 7.2.3 Seeds per ear

Sowing date

Seed number per ear is determined during the tassel emergence phase, and therefore temperature at this stage is very important to determine the seed yield (Tollenaar & Brulsema, 1988) in maize. Seed number reduction in the ear is the main cause of seed yield decreases with delayed sowings (Cirilo & Andrade, 1994). In the present study, seeds per ear in the earliest two sowings were higher than the later sowings in both years because of the higher accumulation of GDD which allowed more time to provide photosynthates for seed filling and retention. An increase in seed number per ear at higher temperature has been reported by Cooper and Law (1977) in maize. Lower solar radiation intensity from delayed sowings during early seed development and seed filling also results in fewer seeds (Lindquist et al., 2005) and lower seed weight as a result of limited source and sink capacities (Tsimba et al., 2013). Lower seeds per ear in 2018 were associated with the lower temperature and soil moisture during seed filling. Stansluos et al. (2020) reported less seeds per ear due to variations in the environmental conditions and cultivation techniques. Thus the result shows that teosinte responds better to higher temperature during seed development.

Seed rate

Seed rate had no significant effect on seeds per ear in 2017 which has been explained in 4.7.2.1. But seeds per ear were significantly affected by seed rate in 2018. In 2018, competition for resources at higher plant density is likely to have increased stress during pollination and seed filling that might have resulted in increased seed abortion and decreased seed filling rate. This is similar with the findings of Andrade et al. (1999); Bernhard and Below (2020); Greveniotis, Zotis, Sioki and Ipsilandis (2019); Kiniry and Richie (1985) and Hashemi, Herbert and Putnam (2005) in maize. Jacobs and Pearson (1991) and Lemcoff and Loomis (1994) also reported negative affect of seed rate on seeds per ear in maize because higher seed rate resulted in greater than the optimum plant density.

Cutting management

Uncut plants produced the most seeds per ear in 2018 which is because in the cut plants, the growth renewal of floral buds was adversely affected by the cutting. Similar results have been reported by Sardana and Narwal (2000) in Egyptian clover where cutting resulted in low seed setting as a result of poor regeneration. Delayed flowering leading to delayed seed maturation has also been reported in cut plants (Amato, Giambalvo and Ruisi, 2013). In 2017 the seeds per ear in uncut and once cut plants did not differ, which might be due to the high temperature effect on the crop phenology for the single cutting that led to a longer vegetative period and higher transmission of photosynthates from source to sink. A similar finding was reported by Tarandeep (2019) and Musa et al. (2021) in berseem clover.

Interactions

A significant interaction occurred between sowing date and cutting management in 2017 and sowing date and seed rate in 2018. For the first year there was a higher accumulated GDD and longer growth duration during the entire vegetative and reproductive growth period for the early sowing and better transmission from source to sink by uncut plants. Interaction between sowing date and seed rate in 2018 occurred because of a longer growth period and less competition among the plants for resources at the lower plant density.

4.7.2.4 Seeds per plant

Sowing date

Seed yield is a result of physiological conditions during the critical growth period of silking in maize (Andrade, 1999) and is closely related to the seeds per plant (Cirilo & Andrade 1994). Temperature and solar radiation during the flowering period are the major weather components affecting seed number in maize (Fischer & Palmer, 1984). In the present study, delayed sowing significantly reduced the total seed number per plant in both years. This is because of a longer growth duration in early sown plants which allowed a better accumulation of heat units. Higher temperature and incident PAR during the reproductive phase enhanced the dry matter partitioning from vegetative to reproductive growth resulting in an increased number of seeds. The temperature during the seed filling period for March sowing 2017 was higher by 0.6°C, 1.1°C and 2.2°C than the April, May and June sowings and the temperature during the seed filling duration for March sowing in 2018 was lower by 0.2°C than the April and higher by 0.6°C than the May sowing. Incident PAR for March sowing in 2017 was higher by (0.6, 0.7 and 1.1) MJ/m²/day for the April, May and June sowings and it was higher by (0.8 and 1.0) MJ/m²/day in the April and May sowings in 2018. Increased seed growth rate at higher temperature was reported by

Badu-Apraku et al. (1983) and Thompson (1986) in maize. This result is in line with the findings of Fischer and Palmer (1984) who reported that seed number is determined by the temperature and incident radiation from the stage of ear initiation to silking.

Seed rate

Seeds per plant was not affected by seed rate in 2017 which has been explained in 4.7.2.1. In 2018, the lowest seed rate produced more seeds per plant which might be because of more resources available to individual plants at the lower seed rate, due to less competition. Higher shading at greater density might have also resulted in reduced seed number. Similar findings on reduced seed number at higher plant density were reported by Lemcoff and Loomis (1994) and Paponov, Sambo, Presterl, Geiger, and Engels (2004) in maize.

Cutting management

In the present study, cutting reduced the seeds per plant in both years. The longer growth duration for uncut plants allowed higher GDD accumulation and better photosynthesis to produce carbohydrate reserves to support more seeds.

Interactions

There were variations in seeds per plant due to an interaction between sowing dates and cutting management in 2017. This occurred because of the longer duration of vegetative growth which allowed better source and sink relationships in uncut plants.

4. 7.2.5 Seed yield

Sowing date

The total seed harvested varied significantly among the sowing dates and the highest seed yield was recorded from the earliest two sowings in both years. Earlier sowing allowed a prolonged growth period at higher temperature for both vegetative and reproductive components accumulating higher GDD. A similar report of increased seed yield as a result of increased seed yield components from higher GDD accumulation was reported by Kakhki (2019) which was due to a better source-sink relationship (Kumar & Patel, 2017). This higher seed yield from the March and April sowings resulted from an increase in plant height, number of tillers, cobs per plant, ear per cobs and seeds per ear. This is consistent with the findings of Burgess, Retkute, Herman and Murchie (2017) who reported that tall plants offer greater sinks for photosynthates which can decrease the limits based on source-sink processes. This has also been supported by Kucharik (2006) for a maize growing season where moisture was not a limiting factor and other agronomical practices like plant population and fertilizer were optimum (Sheperd, Hicks & Schmidth, 1991). Lower seed yield from delayed sowing is most likely due to the seed filling and

maturing period coinciding with decreasing temperature and solar radiation which inhibited the period of seed filling. This result is similar to the findings of several authors (Ruget, 1993; Tsimba et al., 2013; Varma, Durga & Neelima, 2014 and Wilson, Muchow & Murgatroyd, 1995) in maize. Lower seed production is a result of limited assimilate mobilization to the sinks (Lizaso et al., 2018). Further, frost incidence with the delayed harvest could be another reason for lower seed yield and quality in delayed sowings which is similar to the findings of (Baum, Archontoulis, & Licht, 2019; DeVries, 2006; Wilson, Johnstone, & Salinger, 1994) in maize. Variation in the seed yield in the two years reflect the difference in temperature and ultimately GDD accumulation as a result of these temperature differences. As noted, temperature in 2017 was higher than in 2018. Low temperatures result in greater vegetative development than reproductive development that limits the seed set and growth as a result of reduced photosynthesis (Cirilo & Andrade, 1994; Hicks & Stucker, 1972; Shrestha et al., 2018). During the seed filling process, low temperature results in photoinhibition (van Hasselt & van Berlo, 1980) that reduces the photosynthetic activities of plants resulting in low seed yield (Duncan & Hesketh, 1968; Ying et al., 2000). This result is in line with the findings of Shrestha, Kandel and Chaudhary (2018) who reported the effect of high temperature and high thermal period on final seed yield of maize. The higher seed yield per unit area in 2017 was due to higher number of seeds per ear as a result of a longer growing period and heavier seeds as also reported by Sangoi (2001) in maize.

Seed rate

There was no major effect of seed rate on seed yield in 2017 which has been explained in 4.7.2.1. But plant density had a significant effect on the seed yield in 2018 as the lowest two seed rates produced the highest seed yield. At the lower density in the second year there was an increase in resource allocation at the individual plant scale and decreased plant-to-plant competition, and consequently increased per-plant yield potential. Similar results were reported by various authors (Andrade et al., 1999; Echarte, Andrade, Vega & Tollenaar, 2004; Kiniry & Richie, 1985 and Sarlangue, Andrade, Calvino, & Purcell, 2007) who found lower seed yield at the highest plant population as a result of greater stress during pollination and seed filling leading to increased seed abortion and a decreased seed filling period in maize. Further, crowding stress resulting from a higher seed rate was reported by Boyat, Kaan and Panouille (1990) which lead to leaf senescence as a result of shading (Hashemi-Dezfouli & Herbert, 1992) and decreased the net assimilation of individual plants. However, a contrasting result was reported by Jares, Balas, Krnjic, Vranic and Svecnjak (2019) who found seed yield significantly increased at higher density. However, planting density effects also depend on several factors like sowing date, genotype selection, planting pattern, soil fertility and also time of harvest (Oslan & Sanders, 1988).

Cutting management

In the present study, cutting was a source of variation in both years where uncut plants had a better seed yield than the cut plants in 2018. In 2017, there was no difference in the seed yield of uncut and once cut plants. Lower seed from cut plants in 2018 is likely due to lower level of carbohydrates reserve in the plants after cutting because of reduced photosynthesis due to reduced leaf and root growth and major removal of herbage. This result is in line with the findings of Patil and Merwade (2016) in sorghum, Everson (1966) in wheat grass and Davidson and Milthorpe (1966) in cocksfoot. Delayed phenology of plant due to cutting might be another reason for the lower seed yield (Harrison, Evans, Dove & Moore, 2012; Ramos, Moral, Marinetto & Moral, 1993). But in 2017, seed yield did not differ between uncut and cut plants because after the first herbage harvest at 45 DAS, there was plenty of regrowth time with favourable climatic parameters (higher temperature, and higher precipitation, better solar radiation) during the vegetative growth which resulted in quick growth of the plant without any penalty on seed yield. Similarly, higher seed yield was reported by Kumar et al. (2017) where a first cutting of alfalfa at 60 DAS produced more seeds than the uncut plants and plants cut at longer intervals.

Interactions

There were marked variations in seed yield due to interactions between sowing dates and cutting management in 2017 and 2018. The significant interaction between sowing dates and cutting management occurred because of higher GDD accumulation during the longer duration of vegetative growth in early sown plants which could trap more solar radiation from the greater herbage in uncut plants.

4.7.3 Main effect means of seed yield based on the position of cobs

Sowing date

Variation in the time of an individual cob growth, development, ripening and ageing until harvest is affected by the position of the inflorescence on the plant (Keigley & Mullen, 1986). This may result in variation in physical and physiological quality attributes of seed (Illipronti Jr, Lommen, Langerak, & Struik, 2000).

Teosinte is an indeterminate plant. It's flowering and thus seed development begins from the top towards the bottom position of the plant. In the present study, the seed yield of teosinte was higher from the top positioned cobs which for the March sowing produced 40% (2017) and 45 % (2018) of the total seed yield. The middle cobs produced 35 % of the total yield in both years and the bottom cobs produced 25 % (2017) and 20 % (2018) of the total seed yield. Higher yield from the top positioned cobs can be explained by competition for resources where flowers that open

early sequester higher resources by better utilization of the solar radiation. Emms (1996) and Espadaler and Gomez (2001) reported that more resources are utilized by the flowers that open early and there is a greatest possibility that the first flower is likely to produce more seeds as compared to the one opening later. Similar findings has been reported by Dohleman and Long (2009) in maize and Jacobsohn and Globerson (1980) in carrot. Similar inflorescence position effect on seed quantity and quality has been reported by Thomas, Biddington, and Otoole (1979) in celery and Smith, Welch, and Little (1973) in lettuce.

Being thermophilic in nature, teosinte can perform well at higher temperature of 35-40°C (Niazi, Rauf, Silva, & Munir, 2015). Ehleringer and Pearcy (1983) and Pearcy and Ehleringer (1984) have reported that C₄ plants have a very high productivity in an environment with higher temperatures and high light intensities, because of higher photosynthetic rates, but C₄ metabolism is highly sensitive under limiting light intensities (Ubierna, Sun, & Cousins, 2011). Therefore, inter and intraplant shading could be another reason for lower seed yield from the middle and the bottom positioned cobs because under shade conditions, the canopy light is poor, which means it inhibits photosynthesis (Lee, & Lopez-Molina, 2012; Shinomura, Nagatani, Chory, & Furuy, 1994). Chen et al. (2005), Cui et al. (2012) and Early, McIlrath, Seif, and Hageman (1967) reported that yield reduction in maize due to shading depends on the stage of plant growth but the highest yield reduction occurs during silking, where endosperm number is determined (Earley et al., 1967; Gerakis, Papkosta-Tasopoulou, 1980). Thus reduced seed yield due to shading is mainly associated with a reduction in seed number, as occurred in the present study. Zhao and Chen (1990) reported that maize yield reduction due to shading was due to reduced seeds per cob rather than reduced seed weight. Another possible reason for higher yield from the top positioned cobs can be that the tillers borne parallel to the first three cobs on the main stem were harvested together with the top three cobs in each plant. Therefore, lower seed yield in the middle and bottom positioned cobs can be well explained by the resource limitation (Wesselingh, & Arnold, 2003). Higher seed yield in the March sowing for all cob positions can also be the explained by the longer growth duration and higher temperature (Section 4.7.2.4 for temperature and incident PAR at seed filling) that accumulated higher GDD for each phenological growth stage allowing a higher transfer from source into sink (Fischer & Palmer, 1984). The difference in the seed yield between years at each cob position can be explained by the temperature differences already reported.

Seed rate

In 2017, there was no significant effect of seed rate on seed yield of teosinte for each cob position (Section 4.7.2.1). However, in 2018 seed rate did affect the seed yield for each cob position,

whereby seed yield was highest from the top positioned cobs from the lowest two seed rates. This result is similar to the findings of Sarlangue, Andrade, Calvino, and Purcell (2007) who reported reduced seed yield at higher planting density due to crowding and increased seed abortion. The middle and bottom positioned cobs produced a higher seed yield at the lowest three seed rates for similar reasons.

Cutting management

Cutting reduced seed yield at each cob position in both years. In 2017, for the top, middle and the bottom positioned cobs, uncut and once cut plants produced the same seed yield because there was plenty of time for vegetative regrowth after first harvest and the higher temperature, precipitation, and better solar radiation during this vegetative period allowed for quick re-growth of the plant without negative impacts for seed production. This result is similar to the findings of Kumar et al. (2017). In 2018, seed yield from the top and bottom positioned cobs was reduced by with cutting which is because of reduced carbohydrate resources on the cut plants (Ramos, Moral, Marinetto & Moral, 1993). However, the reason why seed yield for the middle position cobs did not vary for uncut and once cut plants is not known.

Interactions

A significant interaction between sowing dates and cutting management occurred for seed yield from the top positioned cobs because the longer duration for the March and the April sowings allowed the accumulation of higher GDD during vegetative growth and more leaves were available to intercept solar radiation in uncut and once cut plants. The interaction for sowing date and cutting management for seed yield from the middle positioned cobs in both years is because of the same reason. The interaction between sowing date and seed rate for seed yield from the middle positioned cobs in 2018 occurred because seed yield was reduced due to competition between plants for resources at higher plant density in a shorter growth period. Likewise the interaction between sowing date and cutting management for bottom positioned cobs occurred in 2018 because of higher solar radiation interception by more leaves from uncut plants.

4.8 Association of seed yield with vegetative and reproductive components in teosinte

Seed yield was positively correlated with plant height in both seasons although it significantly affected the seed yield in 2018 only. A strong association of maize plant height and seed yield has also been reported by Free, Winkelblech, Wilson, and Bay (1966), Pavan, Lohithaswa, Wali, Gangashetty, and Shekara (2011) and Reddy, Jabeen, Sudarshan and Seshagiri (2013). Plant height has been specified as an important factor affecting yield of maize because taller plants produce

more seeds as they are positively correlated with a greater number of ears (Ahmad, Khan, Ghaffar & Ahmad, 2011). The non-significant correlation between plant height and seed yield in 2017 is the result of the previous discussed environmental factors during the vegetative growth in 2017, although the relationship was positive. Arunah, Chiezey, Aliyu and Ahmed (2015) and Wilson, Clowes and Allison (1973) reported that the LAI contribution to seed yield is higher than any other parameters, because the photosynthetic ability of a plant is an index of assimilates production for yield. The positive correlation between LAI and seed yield of teosinte in the present study is similar to the findings of Bavec and Bavec (2002) in maize and Amare, Zeleke and Bultosa (2015) in sorghum. A significant interaction between the tiller number and seed yield occurred in the current research, as a greater leaf area in more tillers allowed more interception of radiation. Rotili et al. (2021) reported that plants with more tillers produce more seeds, which is true in the current research. Baum et al. (2003) reported that seed yield was largely contributed by the number of productive tillers in barley. In 2017, a significant positive correlation between seed yield and tiller number was because of higher tiller number in uncut plants which lead to more cobs per plant resulting in more seeds. Similar results were reported by Lannucci and Martiniello (1998) in clover. The significant correlation between cobs per plant and seed yield under cutting management in both years occurred because of the higher number of cobs in uncut plants. More cobs lead to higher number of ears and ultimately more seed. Similar results were reported by Verma, Yadav, Kumar and Gathiye (2020) in maize.

4.9 Prediction of teosinte seed yield on the basis of growing degree days

Temperature is an important climatic variable that affects the performance of plants, and its influence is obvious when plant phenological growth is closely related to the accumulation of degree days (García-Parra et al. 2020). This concept is regarded as the function of plant development rate (Montagnes, Kimmance, & Atkinson, 2003) and is used to predict crop development, which is the function of temperature at each critical growth stage of the plant on a daily basis (Baker & Reddy, 2001; McMaster & Wilhelm, 1997; Yang, Logan, & Coffey, 1995).

The relationship between temperature, phenological growth stage and maturity time in maize is calculated by GDD (Neild & Seeley 1977). The correlation between seed yield and GDD accumulation in the present study showed that increased GDD increased the seed yield and vice versa. The increase in seed yield as GDD increased was because of the extended growth period, longer day length and higher incident PAR during vegetative growth which allowed the accumulation of more carbohydrate reserves to support reproductive growth. Sangoi (1996) reported that the longer good weather conditions prevail during the reproductive period, more

GDD is accumulated and higher will be the potential seed yield. Similar results were reported by Hundal (1997) in wheat and Kumar, Ramesh, Singh, & Prasad (2010) in marigold.

4.10 Conclusions

In these field trials, the effects of different sowing dates, seed rates and cutting management on seed yield of teosinte in Nepal were investigated. The results showed that sowing date, seed rate, cutting management and their interactions significantly influenced plant growth and therefore seed yield of teosinte. Higher seed yield in the March sowing came from increased plant height, tiller number, LAI, number of cobs per plant, ears per cob and seeds per ear. The study showed that at this site:

- Sowing on March 30 and April 30 produced the greatest seed yield, compared to May and June sowing dates.
- Plant height (2018), tiller numbers per plant and LAI in both years were highest for the first and second sowing.
- Seed yield components (cobs per plant, ears per cob and seeds per ear) were higher for the first and second sowing which explained the higher seed yield.
- There were no significant effects of seed rate on plant height, tiller number or leaf number among the different sowing dates, but LAI was significantly affected by seed rates.
- Total cobs per plant, ears per cob, seeds per ear and seed yield did not differ with seed rate in 2017, but were reduced as seed rate increased from 20 to 80 kg ha^{-1} in 2018.
- Cutting had a significant negative effect on plant height, tiller number, LAI, total cobs per plant, ears per cob, seeds per ear, seeds per plant and seed yield, and the highest seed yield was produced by uncut plants.
- Top positioned cobs produced higher seed yield as compared to the middle and the bottom positioned cobs.
- Delay in sowing date reduced the GDD accumulation in teosinte and thus the seed yield.

Chapter 5

The effect of sowing date, seed rate and cutting management on seed quality of teosinte harvested on the basis of cob positions

5.1 Introduction

Teosinte, an ancestor of maize, is an important summer herbage crop around the world (Sallam, & Ibrahim, 2014) including Nepal, where it was first introduced from India. Since then, it has been under cultivation as a popular summer herbage in different geographic, climatic and soil types of Nepal. Its easy propagation and prolific nature has spread the crop widely (Khatefov & Appaev, 2019) over the years, and its profuse tillering ability and tolerance to multiple cuts makes the crop more profitable than maize fodder (Singh & Dutta, 2021). Teosinte is one of the popular summer herbages among the livestock farmers (Imtiaz, Saeed, & Hassan, 2015; Wang et al., 2020) because its ability to remain green for an extended period for herbage availability has lured farmers towards its cultivation (ICAR, 2011). In addition, it is equally good for sole feeding in the form of green herbage or in a mixture with legumes (Khanal, Devkota, Tiwari, & Gorkhali, 2020), while any surplus is suitable for making good silage and hay. Teosinte's uniqueness lies in the positions of flowers, whereby both male and female flowers are placed on separate places on the same plant (CIMMYT, 1988).

Being a self pollinated crop, teosinte produces a large number of seeds which disperse naturally by shattering after attaining physiological maturity. The shattering nature, in a way, has helped contribute to its persistence and spread over the years. Seed is one of the most important inputs to enhance crop production. Quality seeds must have high germination and seed vigour, along with high genetic, and physical purity, and freedom from pathogens (Hampton, 2002; Varma, Durga & Neelima, 2014). Seed quality alone can increase the crop yield up to 15-20% (BADCO, 2012). Quality seeds help to achieve a good crop stand resulting from a high and rapid emergence, and the production of vigorous plants able to withstand widespread environmental conditions (Ghassemi-Golezani, Tajbakhsh, & Raey, 2011).

5.1.1 Effect of sowing dates on seed quality

Seed quality is highly influenced by sowing date because the difference in temperature, solar radiation, humidity, and precipitation affects the seed development and maturation (Gutterman, 2000). Teosinte, being a short-day plant (Emerson, 1924; Rogers, 1950) requires uninterrupted

long nights to begin flowering (Minow, 2018) and the variation in flowering time with different sowing dates is pronounced. However, Melhus and Ahrens (1959) reported that a 10 hour dark period at least 5-20 days is required to induce flowering in different teosinte varieties. Timely sowing of quality seeds has the greatest impact of all other inputs (Giridhar, Reddy, Kumari, Kumari, Sivasankar, 2017; Kumar, Channakeshav & Siddaraju, 2017). However, both early and late sowing can result in lower yield and inferior seeds because of the uncertainty of climatic conditions which can occur after planting or during the growing period. Therefore, one or more than one planting window should be suggested to the farmers to assure against unpredicted seasons (Norwood, 2001) for quality seed production. The optimum sowing date allows enough opportunity for better use of time, precipitation, light, temperature, and other factors (Zaremohazabieh, Kazemeini, Ghadiri, & Edalat, 2017). Sowing date can also impact the phenological development of the crop as it depends on the temperature and heat units accumulated at the critical stages of crop growth (Greven, 2000). Untimely sowing results in variation in the temperature which might interrupt normal seed development during the seed filling stage if the temperature goes beyond the optimum (Hasan, Ahmed, Hossain, Mian & Haque, 2013; Spears, TeKrony, & Egli, 1997).

High temperature and moisture stress during flowering results in a rapid decrease in the number of ovules fertilized and able to set seeds (Claassen & Shaw, 1970). Additionally, high-temperature stress during seed development and maturation reduces the seeds ability to produce the storage compounds that will be required later in the germination process (Dornbos & McDonald, 1986) and causes physiological damage (Coolbear, 1995; McDonald & Nelson, 1986; Powell, 2006). Such seeds may lose the ability to germinate. Early sowing results in higher thousand seed weight because of exposure to a longer growing season (Koca, Canavar, & Kaptan, 2014) as seed weight is more dependent on reserve mobilization to post silking crop growth, while decreased radiation decreases the final seed weight due to the reduction in biomass production per seed. Further, higher temperature accompanied by delayed sowing reduces the length of the seed filling period because at low temperature the ability of seeds to store the available photo-assimilates is reduced (Muchow, 1990). Thus seeds produced from delayed sowing are of lower weight because of the limitation of the photoassimilates producing source (Didonet, Rodrigues, Mario, Ide, & Tissot, 2001). Seed quality degrades with delaying sowing dates, and therefore changing of sowing date could be a strategy to improve the quality of seed (Hampton, Boelt, Rolston, & Chastain, 2013).

5.1.2 Effect of seed rates on seed quality

Seed quality is further affected by variation in seed rates because of inter-plant competition for assimilates between vegetative development and seed production, in different environmental conditions (Severino, Auld, Vale & Marques, 2017). High plant density is accompanied with reduced assimilate availability due to increased inter-plant competition which impairs the cell membrane integrity (Flinn & Pate, 1968; Powell, 1988). While high planting density may be appropriate for higher seed yield, it is not necessarily better for seed quality (Rahman, Hossain, Anwar, & Juraimi, 2011). Planting density is one of the important aspects to synchronize the conflict between crop groups and individuals. Normally, a higher number of plants per unit area makes the most of the crop seed yield up to a certain limit, but after that limit, further increases in plant density reduce yield and quality (Fang et al., 2018; Wang et al., 2009; Watanabe, Nakano & Okano, 2003) as the supply of plant nutrients, water and other resources becomes limiting (Mandic et al., 2016). Thus yield can be limited under higher density because of the restrictions of endosperm growth (Salvador & Prerace, 1995). Higher yield cannot be expected without enough plants per unit area but interspecific competition for sunlight interception and nutrient uptake increases with increasing planting density which reduces the source to sink capacity (Sangakkara, Bandaranayake, Gajanayake, & Stamp, 2004). A linear decrease in thousand seed weight as a result of reduced resources caused by inter and intra plant competition for water and nutrients with increased crop density was recorded by Mandic et al.(2016) in maize, Rahman, Mwakangwale, Hampton and Hill (2005) in soybeans and Sahu, Tomar, and Nandeha (2018) in sweet sorghum. This could be due to smaller initial size of the spikelet primordia or a delay in development (Borras, Maddonni, & Otegui, 2003). The ultimate seed weight depends strongly on the numbers of cells and starch granules formed in the endosperm tissue, representing about 85% of the weight of mature maize seeds. Competition between the position of seeds and number for the substrates to grow might be another reason for lower seed weight at higher plant density (Jones & Simmons, 1983).

5.1.3 Effect of cutting management on seed quality

Seed quality is also affected by cutting of the herbage because cutting limits the time period for regrowth of tillers and development of leaves which might not be long enough to intercept sufficient radiation and convert it to assimilate through photosynthesis. A significant reduction in the germination of sudan grass after cutting herbage has been reported by Ibrahim and Sakr (2016). Thousand seed weight reduces with the number of cuttings due to the inability of the

remaining foliage to produce sufficient photosynthates to mobilize towards the developing seeds for seed production (Singh, Mor, Khan, & Kumar, 2017).

5.1.4 Seed development, maturity and harvest time of teosinte

Seed development is a process that begins after fertilization and finishes when a maximum dry weight is attained which is termed Physiological Maturity (PM). Seed maturation then occurs (Roberts, 1981) commencing when seed development terminates at PM and continuing as seeds reduce in moisture content until seed is harvested (Mehta, Hazra, & Mascarenhas, 1993). This whole process can be divided into four different stages, where the first two phases involve division of cells and their expansion, the third phase consists of accumulation of food reserves which is accompanied by an increase in dry weight of seed, and the final phase results in intensified seed moisture loss (Bareke, 2018). The third phase ends at PM (also known as relative maturity (Aldrich, 1943) and morphological maturity (Anderson, 1955). PM is a state of seed development when there is no further increase in seed dry weight as a result of restriction of the translocation of food materials to the seed. At PM seed quality is theoretically at its highest (Dayal, Rangare, Kumar, & Kumari, 2014).

The first three stages of seed development are collectively known as seed filling, which is a complex process that includes cell enlargement and accumulation of proteins, oils and carbohydrates. Seed filling begins with successful pollination and initiation of seed development and ends at PM (Yin et al., 2020). The seed weight continues to increase during the seed filling stage because of nutrient deposits and water uptake. At the start of seed filling, individual seed growth rate is determined on the basis of cotyledon cell number which is related to assimilate supply during cell division. Differences in photosynthetic activity after seed filling determine the seed growth rate which however depends on whether photosynthetic activity can produce the assimilates required for seed filling (Munier-Jolain, Munier-Jolain Roche, Ney, & Duthion, 1998). Sampling variation can make it difficult to decide the exact time of PM (TeKrony & Egli, 1997). Therefore some researchers prefer to call it a period rather than giving a precise time after which there is no increase in the seed dry weight (Bareke, 2018). The final stage of seed maturation is harvest maturity which depends on the environment and usually occurs 7-10 days after PM (Dayal, Rangare, Kumar, & Kumari, 2014). During this time seeds lose moisture until their moisture content comes into equilibrium with moisture in the air (Hampton, 2000).

Teosinte, with its indeterminate nature of growth, has seed maturity which is not uniform along the inflorescence. At a commercial level, teosinte seeds are commonly harvested before all of the seeds are fully mature to prevent shedding losses because of adverse weather conditions.

Harvesting the seeds at the right time is a key factor contributing to obtaining high quality seeds (Gu et al., 2017). Early harvesting of seeds may result in immature, poorly germinating seeds with less seed vigour (Deshpande, Kulkarni, & Kurdikeri, 1991; Fu, 2017). Delayed harvesting may result in deterioration due to the occurrence of adverse environmental conditions, and attack by insects and microorganisms which will hasten the seed deterioration process (Henning, Jacob Junior, Mertz, & Peske, 2011; Navazio & Colley, 2007) and the risk of losing seeds is high due to shattering (Navazio & Colley, 2007). Normal seed development is interrupted during the seed filling stage if the temperature goes beyond the optimum (Spears et al., 1997). A small variation in temperature during seed development and maturation can affect the quality of the seed (Gutterman, 2000). High temperature stress during seed development and maturation reduces the seeds ability to produce the storage compounds that will be required later in the germination process (Dornbos & McDonald, 1986). Heat stress causes physiological damage (McDonald & Nelson, 1986; Powell, 2006), which reduces seed vigour and may reduce the ability to germinate (Ellis, Summerfield, Edmeades, & Roberts, 1992; Shinohara, Hampton, & Hill, 2006; Spears et al., 1997).

Seed harvest time in teosinte depends on the time of sowing. Seed maturation time in teosinte differs from the top towards the bottom of the plant, and therefore it requires a couple of days to complete the seed harvest. Teosinte seeds after maturation shatter readily if not collected on time (Piperno, Holst, Winter, & McMillan, 2015). When the teosinte seeds are mature, the cobs opens naturally due to the influence of gene *tga1*, which is regulated by another gene, *tb1* (Piperno, Holst, Moreno, & Winter, 2019). During seed maturation, the uppermost cob opens up and the seeds are no longer attached to each other inside the row of each cob. They are arranged in a loose fashion and the colour of the uppermost seeds turns brownish, which is an indication of seed maturity for harvest. Seed moisture content at this stage is around 20-25%, so for safe storage, harvested seeds need further drying. The crop is manually harvested at around 25% SMC then dried before storage.

Teosinte is a good seed bearer but little is known about the effects of agronomic management on quality of teosinte seeds. Sowing date, seed rate and cutting management may have an impact on the quality of teosinte seeds, and indeed on seed development.

5.2 Objectives

The objective of this research was to study the seed development and maturity of teosinte, and to determine whether sowing date, seed rate and cutting management affected teosinte seed quality.

5.3 Methodology

The cropping history, weather, treatments and the experimental design are as explained in Chapter 2. Seed quality was determined on seeds from a subsample of 10 hand harvested plants per plot. Based on the position of cobs, seeds were harvested three times from each plant. The first harvest was taken from the top three cobs on the main stem and secondary tillers. The second harvest was taken from the middle three cobs, and the remaining cobs at the bottom were taken for the third harvest. Each time seeds were harvested at a moisture level of 25-27%, which was confirmed using a moisture meter. The cobs were manually harvested and threshed seeds were weighed, placed in a paper bag and each lot of seed harvested was ambient air dried before sending to the laboratory for quality testing.

5.3.1 Seed development study

A separate seed development study was conducted using the 30 March sowing in 2017. Sampling started immediately a week after anthesis and every five days after that as reported by Ghassemi-Golezani, Tajbakhsh and Raey (2011) in maize. Sampling consisted of hand picking three cobs from the middle portion of five selected plants other than the 10 tagged plants. Cobs were threshed by hand and 50 fresh seeds were measured for fresh weight, dry weight, seed moisture content, thousand seed weight and germination in the laboratory. The seed tests were done as described in the following sections. Seed sampling and quality testing was continued every five days until the seeds matured. Further, GDD accumulation for seed development was calculated.

5.3.2 Seed Quality testing

Seed testing was done to assess seed lot attributes to determine overall quality and value for seedling production and storage. Seed testing methods are based on scientific evidence and provide set procedures and facilities to conduct tests in a uniform manner and ensure comparable results for seed owners. As there are no published methods for testing tesosinte seed quality, the seed testing methods described below were closely aligned with the International Rules for Seed Testing (ISTA 2018) for maize, and followed these procedures for sampling, germination, moisture content and seed weight determination.

Fresh Weight

The fresh weight of seeds was taken on the day when seed was harvested from the plant. After the harvest, seeds were immediately placed into a paper envelope and weighed with the envelope. The weight of the envelope was then subtracted to get the final weight of fresh seeds.

Dry weight

The seeds after harvesting were dried in the shade to 14 % seed moisture content before sending to the laboratory. Once the seeds were taken to the laboratory, they were weighed again which is the dry weight of seed.

Dormancy Breaking

The inability of an intact viable seed to complete the germination process under a favourable environment is called seed dormancy (Bewley, 1997), which in some species is controlled by an inhibitor-promoter balance (Finch-Savage, Cadman, Toorop, Lynn, & Hilhorst, 2007). More than 90% of Mexican teosinte populations have seed dormancy, where higher dormancy is observed in plants grown in the regions of lower altitude, which generally have hotter and drier (lower relative humidity) environments (Avendano Lopez et al., 2011).

Dormancy in teosinte has been previously studied (Flannery, 1973; Mondrus, 1981; Wilkes, 1977). Different extent of dormancy is seen in various species of teosinte. Deep and very deep dormancy has been reported for *subsp. parviglumis* and *Zea luxurians*, which are well distributed in hot and very hot environments (26 to 30°C). Seeds grown in semi-hot to hot environments (18 to 26°C) were reported to have moderate dormancy which corresponds to subtropical and tropical conditions (Ruiz et al., 2003). Moderate dormancy was also found under temperate to semi hot conditions (12 to 22°C) (Garcia, 2004) and weak dormancy has been reported for *subsp. mexicana*, *Zea perennis*, and *Zea diploperennis* populations which are distributed in the highlands of Mexico (12 and 18°C) (Medina et al., 1998). Seeds without dormancy have been reported largely around semi cold areas (Avendano Lopez, et al., 2011). Contact with pure oxygen or an oxidizing agent like 20% hydrogen peroxide (H₂O₂) and gibberellic acid have been suggested as treatments to break dormancy in teosinte by Mondrus (1981), Taba, Ginkel, Hoisington, and Poland (2004) and Wilkes (1977).

In this study, gibberellic acid (GA3) was used to break the dormancy in the laboratory as suggested by Aboelgoud (2015). A 1000 ppm solution of GA, was prepared by dissolving 1000 mg of GA3 in 1000 ml of water (ISTA, 2018). Teosinte seeds were placed in this solution for 24 hours before transferring to the germination test (Figure 5.1).



Figure 5.1 Dormancy breaking by soaking seeds in a GA3 solution

Germination Test

The germination tests were conducted using the between paper (BP) method (ISTA 2018). For each plot, a total of 200 seeds were tested, divided into four replicates of 50 seeds. Each sample of 50 pure seeds was planted on a double sheet of the moistened paper leaving 2-3 cm along the lower edge and the sides and about 3 cm at the upper edge. It was then covered by another moist paper sheet, which was placed exactly over the first sheet or 2-3cm up from the lower edge depending upon the thickness of the bottom layer. The lower edge of the bottom layer was folded over the second sheet. It was then rolled loosely from left to right into a loosely packed cylinder-like roll. Identification number of the sample being tested was written on the end side of the outer layer. Then each roll was tied up at both ends with a rubber band and placed in a plastic bag. The rolls were placed in an upright position in the germinator at $25\pm 1^{\circ}\text{C}$ (Figure 5.2, 5.3). Seedlings were evaluated according to ISTA Rules (ISTA 2018). The first count was done at 4 days and the final count was done at 14 days. The result of the germination test (% normal seedlings) were calculated as the average of four replicates of 50 seeds. Only normal seedlings were removed and counted at the first count. All others were left until the end of the test, where the seedlings were categorized as normal and abnormal as per guidelines of ISTA (2018). Ungerminated seeds at the end of the test period were considered as either ungerminated fresh seeds or dead seeds by dissecting the seeds. The result was expressed as the percentage of normal and abnormal seedlings and fresh and dead seeds (ISTA, 2018). Normal seedlings are defined by ISTA as having all the essential structures (root system, shoot axis, cotyledons, terminal buds and coleoptile), well developed and healthy. Seedling with slight defects, and seedlings with secondary infection are considered as normal seedlings. Abnormal seedlings were considered those where the essential structures were damaged, seriously deformed, disordered or decayed as a result of seed infection (ISTA, 2018). Variations in results among the germination test replicates were assessed using the tolerance table for germination (ISTA, 2018). If the difference

between the highest and lowest germination percentage of replicates was not in the range of tolerance allowed, the germination test was repeated.



Figure 5.2 Teosinte seed germination test; single paper roll containing 50 seeds each



Figure 5.3 Germination test ready for evaluation

Seed Moisture Content (SMC)

SMC was measured for each seed lot using the ISTA high temperature oven method (ISTA 2018). Two replicates of 4 g seeds were ground and sieved through a wire sieve with meshes of 0.5 mm so that not more than 10% remained on a wire sieve with a mesh of 1.0 mm. The ground material was then transferred to an aluminium can which was placed in a hot air-oven for 4 hr at 130°C. After drying in the oven, the sample in the aluminium can was placed in a desiccator for 1 hour to cool and then weighed. Seed moisture content was determined using the following formula:

$$\% \text{ MC} = (M2 - M3) * 100 / (M2 - M1)$$

M1 is Weight of aluminium can with its cover

M2 is Weight of aluminium can with its cover and seed material before drying

M3 is Weight of aluminium can with its cover and seed materials after drying

1000 seed weight (TSW)

The working sample was taken from the pure seed. From the working sample eight replicates of 100 seeds were counted by hand using a counting board. Each replicate was weighed in grams to three decimal places. To obtain TSW, the mean value of the eight replicates of 100 seeds was multiplied by 10 (ISTA, 2018).

5.3.3 Statistical Analysis

Analyses of variance (ANOVA) to determine the effect of sowing date, sowing rate and cutting management on seed quality and their interactions among the factors were performed using

general linear model (GLM) of Genstat 19th Edition (VSN International, 2019). Accordingly, replication was considered as the block, whole plots as sowing plot, sowing rate as a sub-plot and cutting management as a sub-sub-plot factor, respectively. For presenting the significance of different factors on seed quality, $P < 0.05$, $P < 0.01$, and $P < 0.001$ were used for 5, 1, and 0.1 percent level of significance respectively. Significantly different means of each level of factors considered were compared using Fisher's unprotected test of least significant difference (LSD). The relationship between the sowing date, seed rate and cutting managements with the seed quality parameters was determined by regression analysis.

5.4 Results

5.4.1 Teosinte seed development and maturation

Seed development in teosinte was assessed to determine the time when physiological maturity (PM) occurred. The first sample was taken at 5 days after anthesis (DAA) which was around 80% seed moisture content (SMC). SMC steadily declined until reaching equilibrium SMC at 56 DAS (Figure 5.4). Some seeds germinated at 25 DAA and germination then steadily increased to reach a maximum at 56 DAA, after which there were no changes (Figure 5.4). Seed dry weight increased rapidly between 30 and 50 DAA, reaching a maximum at 56 DAA (Figure 5.5) indicating that the seeds had reached PM. The seed development period in 2017 was therefore 56 days for the March sowing. This seed development period accumulated a GDD of 617⁰C days for seed to reach PM (Table 5.1).

Table 5.1 The GDD requirements for seed development of Teosinte at the March sowing 2017

Days from sowing to PM	DAA to PM	Accumulated GDD from the end of flowering to PM
246	56	617

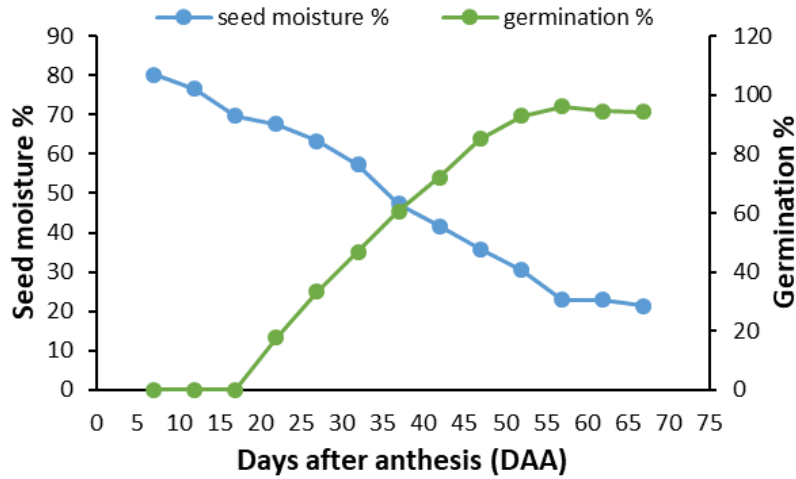


Figure 5.4 Teosinte seed moisture and germination during seed development

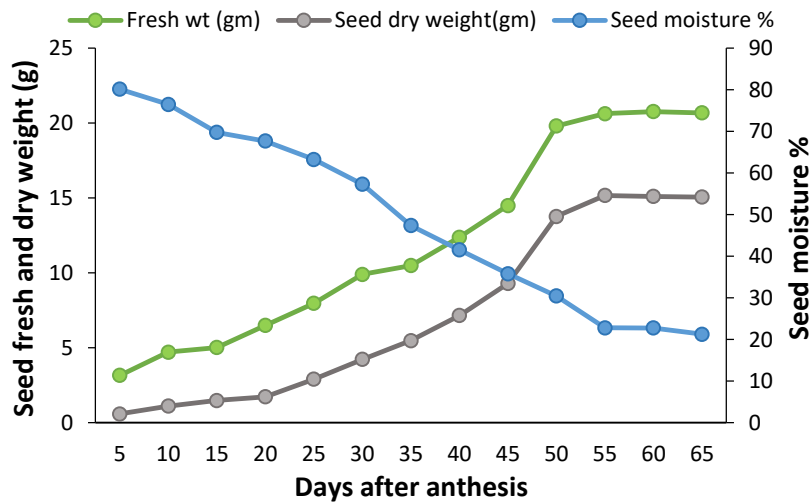


Figure 5.5 Teosinte seed moisture, seed fresh weight and seed dry weight during seed development

5.4.2 Germination

Sowing dates

For seed quality, data for the June sowing in 2017 were not included in the data analysis as June sown plants had very few seeds with low germination.

The overall germination (all cobs) differed significantly among the sowing dates in both years ($P < 0.001$) (Table 5.2). Further, the germination of seeds harvested from each cob position (top (T), middle (M) and bottom (B)) was also significantly influenced by sowing dates ($P < 0.001$). Seeds harvested from the March sowing had a greater germination for all cobs seeds and also for seeds harvested from each of the three positions in both years followed by the April and the May sowings in all positions except for the bottom position cobs where the April and the May sowing

in 2018 did not differ from each other ($P < 0.001$) (Table 5.2) Germination of seeds harvested from the top cob position varied significantly from the middle and the bottom positioned seeds at each sowing in both years ($P < 0.01$). The seeds harvested from the middle position had a higher germination than the seeds from the bottom position ($P < 0.01$) (Table 5.2). There was a highly significant negative correlation ($R^2 = 1$) ($P < 0.01$) between sowing dates and all cobs germination because germination was reduced with delay in sowing (Figure 5.6).

The data for abnormal seedlings could not be retrieved from the seed laboratory at the Agriculture and Forestry University, Chitwan, Nepal due to the Covid-19 situation. But from observation, most abnormal seedlings were because of the absence of roots. This number increased with the delay in sowing.

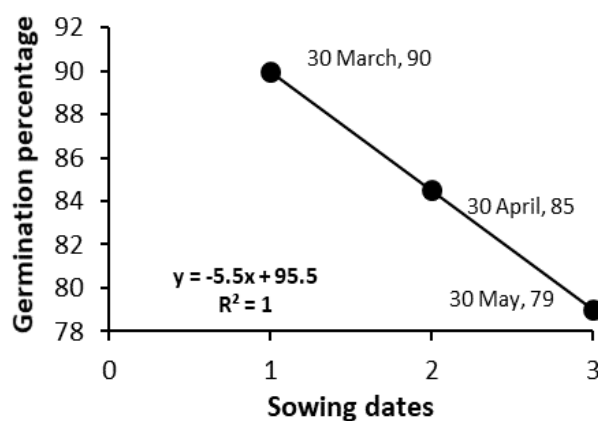


Figure 5.6 Correlation between sowing date and germination percentage; data are means for 2017 and 2018

Seed Rates

Seed rate was a significant source of variation for the germination of teosinte in 2017 and 2018 ($P < 0.05$) (Table 5.2). In 2017, the 20 kg ha^{-1} and 40 kg ha^{-1} seed rates produced seeds with the highest germination (all cobs) but they did not differ with each other. They also had germinations significantly higher from that of the 60 kg ha^{-1} and 80 kg ha^{-1} seed rates for the seeds from the top and middle positioned cobs ($P < 0.001$). In 2017 for bottom positioned cobs, 20 kg ha^{-1} , 40 kg ha^{-1} , and 60 kg ha^{-1} seed rates did not vary with each other while the 80 kg ha^{-1} seed rate had the lowest germination. In 2018, the 20 kg ha^{-1} seed rate had the highest germination. Germination for the 40 kg ha^{-1} and 60 kg ha^{-1} seed rate did not vary but was significantly higher than that of the 80 kg ha^{-1} seed rate ($P < 0.001$).

For the seeds harvested from the top and middle position cobs in 2017, germination for the two lower seed rates was significantly higher than the two higher seed rates but the 20 kg ha^{-1} and 40 kg ha^{-1} and 60 kg ha^{-1} and 80 kg ha^{-1} did not differ with each other ($P < 0.001$) (Table 5.2). In 2018,

germination for top and middle positioned seeds did not differ between the 20 kg ha^{-1} and 40 kg ha^{-1} seed rates and the lowest germination was recorded for the highest seed rate (80 kg ha^{-1}) ($P<0.001$) (Table 5.2).

In 2017, germination percentage of teosinte seeds from the bottom cob position differed significantly with each other ($P<0.001$). The 20 kg ha^{-1} seed did not vary from the 40 kg ha^{-1} and 60 kg ha^{-1} seed rates. The highest seed rate (80 kg ha^{-1}) had the lowest germination for the bottom positioned seeds. In 2018 germination also varied significantly with seed rates for the seeds from the bottom of the plant (Table 5.2). The germination was significantly higher for the 20 kg ha^{-1} and the 80kg ha^{-1} seed rate had the lowest germination (Table 5.2). There was a significant negative correlation ($R^2=0.98$) ($P<0.01$) between seed rates and germination because germination percentage decreased with an increase in planting density (Figure 5.7).

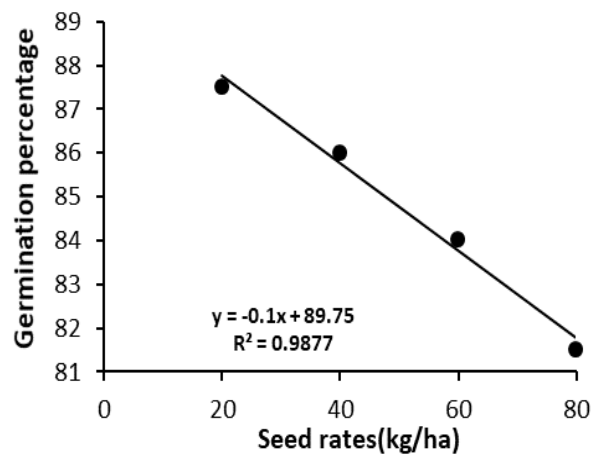


Figure 5.7 Correlation between seed rate and germination percentage; data are means for 2017 and 2018

Cutting Management

Cutting management was an important source of variation with respect to germination in both years ($P<0.01$) (Table 5.2). The overall germination (all cobs) of teosinte seeds from uncut plants was significantly higher ($P<0.001$) than once and twice cut plants in both 2017 and 2018 and it was significantly higher for top, middle and bottom positioned seeds than once or twice cut plants in both years ($P<0.001$) (Table 5.2). There was a highly significant negative correlation ($R^2=1$) ($P<0.01$) between cutting management and germination percentage because germination percentage decreased with an increase in cutting frequency (Figure 5.8).

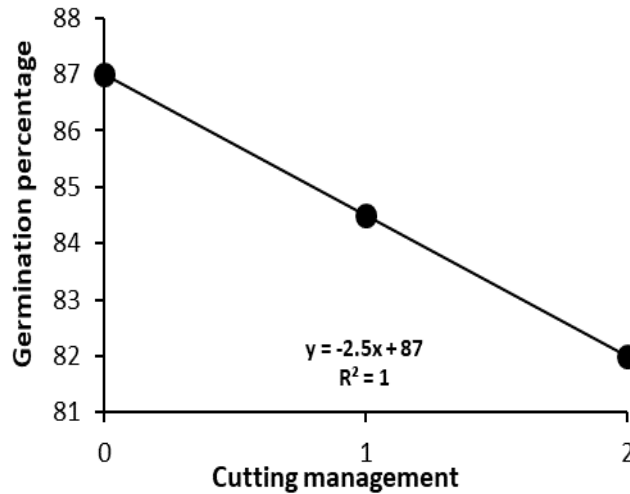


Figure 5.8 Correlation between cutting management and germination percentage; data are means for 2017 and 2018

Interactions

A significant interaction between seed rate and cutting management was recorded for overall germination and germination for seeds from the top and middle positions in 2017 because 20 kgha⁻¹ and 40 kgha⁻¹ seed rates produced seeds with higher germination in uncut plants (P<0.05) (Figure 5.9, 5.10 and 5.11). A significant interaction occurred between sowing date and cutting management for overall germination and germination for seeds at the bottom position in 2018 because the March sowing produced seeds with highest germination in uncut plants (P<0.001) (Figure 5.12, 5.13).

Table 5.2 Main effect means of sowing dates, seed rates and cutting management on the germination percentage of teosinte seeds harvested based on the position of cobs in 2017 and 2018 at NCRP, Chitwan Nepal

Main effect means of:	Germination percentage							
	All cobs		Top		Middle		Bottom	
Sowing date (SD)	2017	2018	2017	2018	2017	2018	2017	2018
30-Mar	91a	89a	93a	91a	91a	88a	88a	85a
30-Apr	86b	83b	87b	85b	84b	82b	83b	79b
30-May	81c	77c	82c	79c	80c	76c	79c	75b
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	3	3	3	4	3	4	3	4
CV%	2	2	2	2	2	3	2	3
Seed rate (SR)								
20 kg ha ⁻¹	88a	87a	89a	88a	87a	86a	85a	84a
40 kg ha ⁻¹	88a	84ab	89a	86ab	86a	83ab	85ab	80b
60 kg ha ⁻¹	85b	83b	86b	85b	84b	82b	82ab	79b
80 kg ha ⁻¹	84b	79c	85b	80c	83b	78c	82b	75c
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	2	2	2	2	2	3	2	2
CV%	3	4	3	4	2	4	3	4
Cutting Management (CM)								
No cut	89a	85a	90a	87a	88a	84a	86a	81a
One cut	86b	83b	88b	84b	85b	82b	84b	80b
Two cut	83c	81c	84c	83c	82c	80c	80c	78c
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	1	1	1	1	1	1	1	1
CV%	3	2	3	2	3	2	3	3
Significance of interactions of linear contrasts (p value)								
SD(lin) x SR(lin)	0.657	0.461	0.714	0.193	0.500	0.150	0.672	0.237
SD(lin) x CM(lin)	0.330	<.001	0.593	0.928	0.147	0.170	0.237	0.047
SR(lin) x CM(lin)	0.018	0.581	0.028	0.806	0.029	0.420	0.060	0.680
SD(lin) x SR(lin) x CM(lin)	0.548	0.155	0.590	0.548	0.545	0.240	0.742	0.501

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management

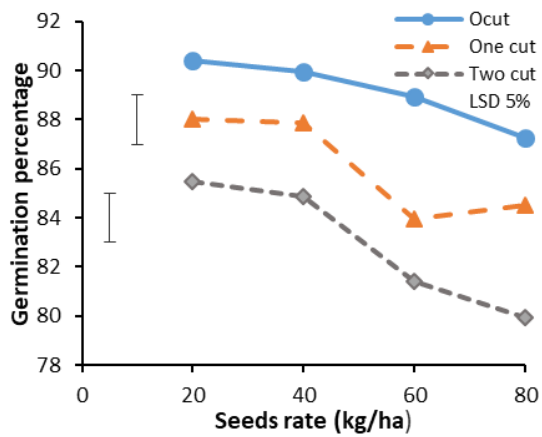


Figure 5.9 Interaction between seed rate and cutting management for overall germination percentage in 2017

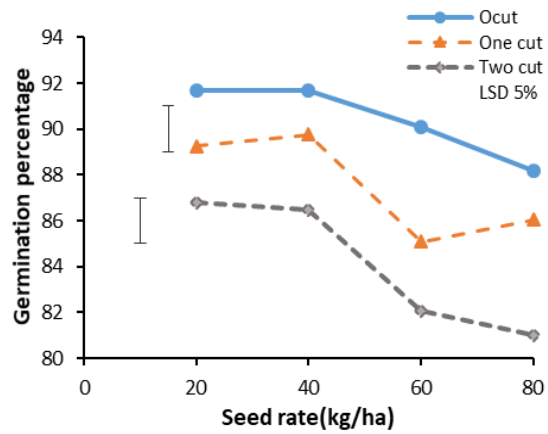


Figure 5.10 Interaction between seed rate and cutting management for germination percentage of seeds based at top position of plant in 2017

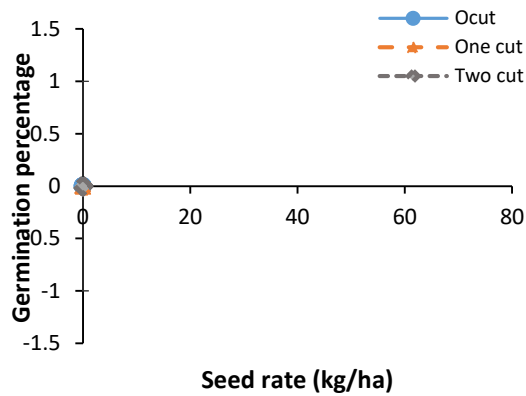


Figure 5.11 Interaction between seed rate and cutting management for germination percentage of seeds based at middle position of plant in 2017

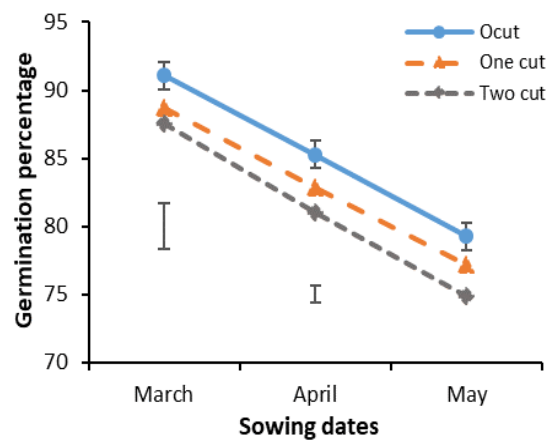


Figure 5.12 Interaction between sowing date and cutting management for overall germination percentage in 2018

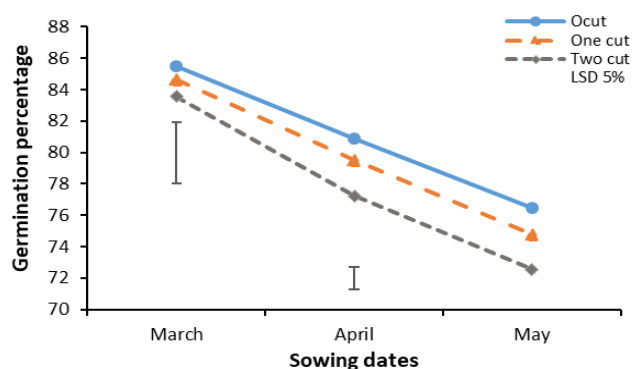


Figure 5.13 Interaction between sowing date and cutting management for germination percentage of seeds based at bottom position of plant in 2018

Note: In figures 5.9-5.13 the left-hand vertical bar is the LSD (5%) for comparisons within a sowing date, or seed rate and the right-hand vertical bar is the LSD (5%) for all other comparisons.

5.4.3 Difference between the germination among the cob positions

Germination percentage along the teosinte cobs decreased from the top to the bottom for the different sowing dates, seed rates and cutting management. To determine whether these variations were statistically different, analyses of variance for germination were done between seeds from the different cob positions, T & M, M & B and T & B. A significant difference in germination between top and bottom positioned cobs was recorded ($P < 0.001$) (Table 5.3). Delayed sowing increased the difference between the seed germination for top and bottom positioned cobs. A similar effect was observed for seed rate and cutting management. A significant interaction for germination occurred between sowing date and cutting management for T-B because May sowing and one cut plants produced the least difference in germination ($P < 0.05$) (Table 5.3).

Table 5.3 Differences in germination of seeds from different sowing dates, seed rates and cutting management based on different inflorescence positions

Main effect means of:	Germination percentage					
	2017			2018		
	T-M	M-B	T-B	T-M	M-B	T-B
Sowing date (SD)						
30-Mar	2.0	2.6	4.7	2.3	3.5	5.9
30-Apr	2.4	1.5	4.3	2.5	3.1	5.7
30-May	2.8	0.5	2.9	2.6	1.5	4.0
Linear contrast p value	0.625	0.008	<.001	0.553	0.005	0.018
LSD (1%)	6.8	10.6	5.4	0.9	1.7	2.1
LSE (1%)	4.8	7.5	3.8	0.6	1.2	1.5
LSD (5%)	1.6	1.3	0.3	0.6	1.1	1.4
LSE (5%)	1.1	0.9	0.2	0.4	0.8	1.0
Seed Rate (SR)						
20 kg ha-1	2.3	1.8	4.1	2.2	2.2	4.4
40 kg ha-1	3.2	1.8	4.7	2.8	3.1	6.0
60 kg ha-1	1.6	1.5	3.5	2.8	2.7	5.5
80 kg ha-1	2.4	1.0	3.4	2.1	2.8	5.0
Linear contrast p value	0.356	0.219	<.001	0.899	0.503	0.537
LSD (1%)	3.3	4.6	2.9	1.1	1.7	1.6
LSE (1%)	2.3	3.3	2.1	0.8	1.2	1.1
LSD (0.05)	1.0	0.9	0.4	0.8	1.2	1.2
LSE (5%)	0.7	0.6	0.3	0.6	0.9	0.9
Cut Management (CM)						
No cut	2.2	2.1	4.4	2.7	3.1	5.9
One cut	2.7	1.1	3.8	2.2	2.5	4.8
Two cut	2.2	1.4	3.6	2.5	2.4	5.0
Linear contrast p value	0.853	0.06	<.001	0.506	0.015	0.008
LSD (1%)	2.3	2.5	2.2	0.8	0.7	0.8
LSE (1%)	1.6	1.8	1.6	0.6	0.5	0.6
LSD (0.05)	0.8	0.8	0.3	0.6	0.5	0.7
LSE (5%)	0.7	0.6	0.3	0.7	0.9	0.9
Significance of interactions of linear contrasts (p value)						
SD(lin) x SR(lin)	0.638	0.688	0.820	0.617	0.482	0.694
SD(lin) x CM(lin)	0.305	0.676	0.124	0.109	0.299	0.020
SR(lin) x CM(lin)	0.961	0.604	0.214	0.186	0.086	0.826
SD(lin) x SR(lin) x CM(lin)	0.947	0.717	0.527	0.474	0.517	0.904

Note: LSD = Least Significant Difference; LSE = Least Square Estimator; Difference CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

5.4.4 Thousand seed weight

Sowing dates

There was a significant effect of sowing date on the TSW of seeds from all cobs in both years ($P < 0.001$) (Table 5.4). TSW of teosinte for the March sowing did not differ from the April sowing, but the May sowing had lower TSW in both years. June sowing in 2017 had the lowest TSW. A similar trend followed for the seeds harvested from the top, middle and bottom of the plant ($P < 0.001$) (Table 5.4). There was a negative correlation ($R^2 = 0.79$) ($P > 0.05$) between sowing dates and TSW because seed weight decreased with the delayed sowing (Figure 5.14).

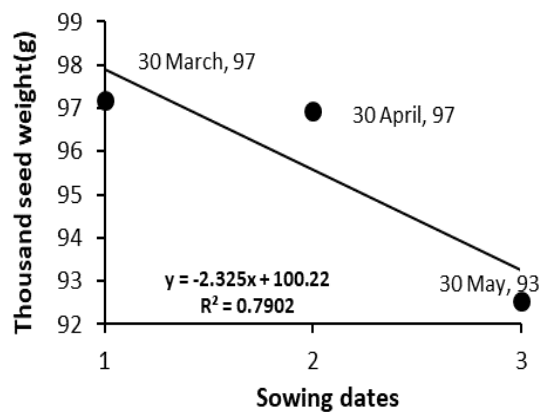


Figure 5.14 Correlation between sowing dates and TSW; data are means for 2017 and 2018

Seed Rate

Seed rate had a significant effect on the overall TSW in both years ($P < 0.001$) (Table 5.4). In 2017 and 2018 TSW for all cobs seeds, and the seeds harvested from the top, middle and bottom positions did not differ between the 20 kg ha^{-1} and 40 kg ha^{-1} seed rate, but their TSW was significantly higher than the 60 kg ha^{-1} and 80 kg ha^{-1} seed rate ($P < 0.001$) (Table 5.4). There was a significant negative correlation ($R^2 = 0.93$) ($P < 0.05$) between seed rate and TSW because seed weight decreased as the plant density increased (Figure 5.15).

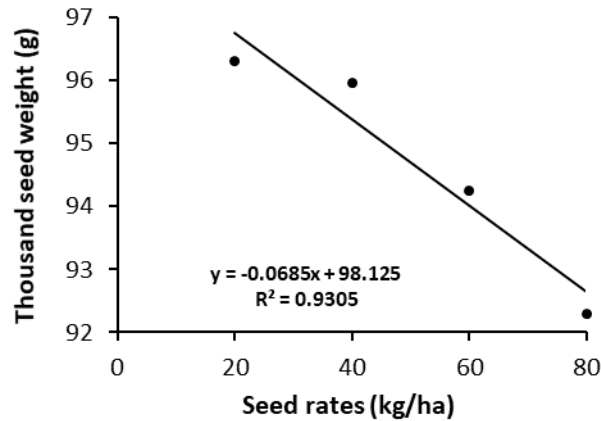


Figure 5.15 Correlation between seed rates and TSW; data are means for 2017 and 2018

Cutting Management

Cutting significantly reduced the TSW for all cobs and for all cob positions in both years. Uncut plants had higher TSW than once and twice cut plants in both years ($P < 0.001$) (Table 5.4). There was a significant negative correlation ($R^2 = 0.97$) ($P < 0.05$) between cutting management and thousand seed weight because seed weight decreased with increase in the cutting frequency (Figure 5.16).

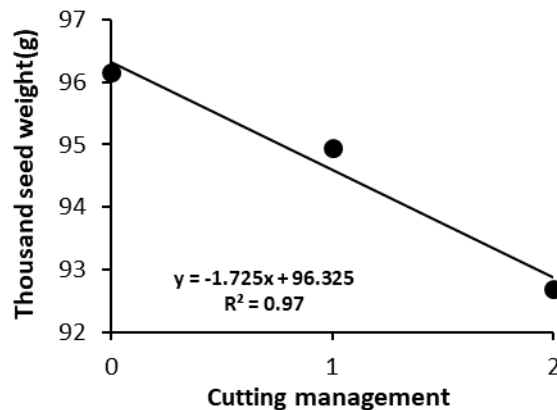


Figure 5.16 Correlation between cutting management and TSW; data are means for 2017 and 2018

Interactions

In 2017, there was a significant interaction between sowing date and cutting management for TSW of seeds from all cobs and top, middle and bottom positions, because the earliest two sowing dates produced seeds with the highest TSW (Figure 5.17, 5.18, 5.19, 5.20) ($P < 0.001$) in uncut plants. A significant interaction also occurred between sowing date and seed rate for bottom positioned cobs in 2017 because the June sowing produced seeds with the lowest TSW at the highest seed rate (Figure 5.21) ($P < 0.05$). Likewise, there was a significant interaction between sowing date, seed rate and cutting management for TSW for all cobs and seed from top

positioned cobs in 2017($P<0.05$) (Table 5.5,5.6) because the June sowing had the lowest TSW at the highest seed rate from twice cut plants ($P<0.001$). A similar interaction occurred in overall cobs and the seeds harvested from the top and middle positions ($P<0.05$) in 2018 because the May sowing produced the lowest TSW from twice cut plants (Table 5.7, 5.8, 5.9).

Table 5.4 Main effect means of sowing dates, seed rates and cutting management on the TSW of teosinte seeds based on the position of cobs in 2017 and 2018 at NCRP, Chitwan, Nepal

Main effect means of :	Thousand seed weight (g)							
	All cobs		Top position		Middle position		Bottom position	
Sowing date (SD)	2017	2018	2017	2018	2017	2018	2017	2018
30-Mar	97.8a	96.6a	98.2a	97.2a	97.6a	95.9a	96.5a	95.2a
30-Apr	97.6a	96.3a	98.1a	97.0a	97.2a	95.9a	96.3a	95.2a
30-May	92.7b	92.4b	93.2b	93.2b	93.6b	91.8b	91.3b	91.2b
30-Jun	88.2c	-	88.8c	-	87.8c	-	86.7c	-
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	2.7	0.6	2.7	0.7	2.8	0.6	2.9	0.6
CV%	1.9	0.4	1.8	0.4	1.9	0.3	2	0.4
Seed Rate (SR)								
20 kg ha-1	95.6a	97.0a	96.0a	97.2a	95.2a	96.1a	94.2a	95.2a
40 kg ha-1	95.2ab	96.8a	95.6ab	97.1a	94.7ab	95.8a	93.7ab	95.0a
60 kg ha-1	93.9b	94.6b	94.5b	95.8b	93.6b	94.0b	92.5b	93.5b
80 kg ha-1	91.7c	92.9c	92.1c	93.6c	91.3c	92.2c	90.3c	91.5c
Linear contrast p value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001
LSD (0.05)	1.3	1.3	1.3	1.2	1.2	1	1.3	1
CV%	1.9	1.6	1.9	1.5	1.9	1.3	1.9	1.3
Cut Management (CM)								
No cut	95.7a	96.6a	96.2a	97.3a	95.4a	95.9a	94.4a	95.2a
One cut	94.5b	95.4b	94.9b	96.1b	94.1b	95.0b	93.1b	94.1b
Two cut	92.1c	93.3c	92.6c	94.0c	91.7c	92.8c	90.7c	92.1c
Linear contrast p value	<.001	<.001	<.001	0.044	<.001	<.001	<.001	<.001
LSD (0.05)	0.5	0.5	0.6	0.5	0.9	0.5	0.6	0.5
CV%	1.7	1.2	1.7	1.2	1.8	1.3	1.9	1.4
Significance of interactions of linear contrasts (p value)								
SD(lin) x SR(lin)	0.08	0.986	0.102	0.948	0.06	0.907	0.033	0.53
SD(lin) x CM(lin)	<.001	0.567	<.001	0.312	<.001	0.741	<.001	0.437
SR(lin) x CM(lin)	0.649	0.234	0.805	0.194	0.561	0.362	0.579	0.519
SD(lin) x SR(lin) x CM(lin)	0.055	0.007	0.049	0.007	0.105	0.024	0.085	0.071

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

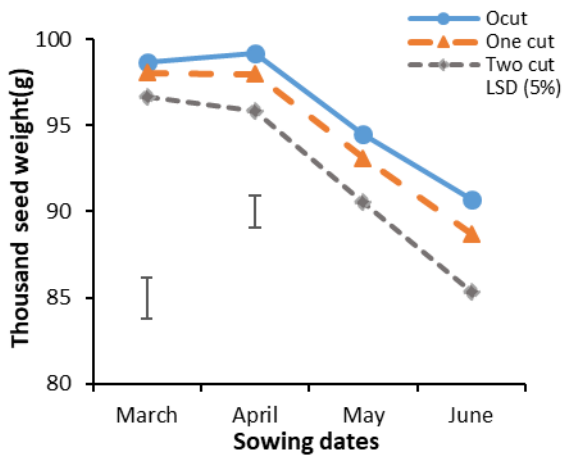


Figure 5.17 Interaction between sowing date and cut management for overall TSW in 2017

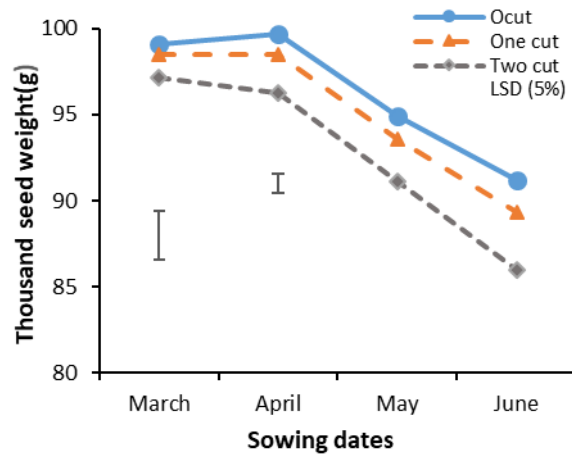


Figure 5.18 Interaction between sowing date and cut management for TSW at top positioned cobs in 2017

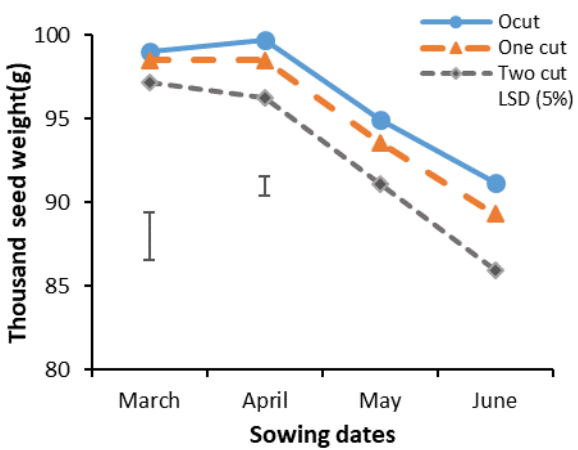


Figure 5.19 Interaction between sowing date and cut management for TSW at middle positioned cobs in 2017

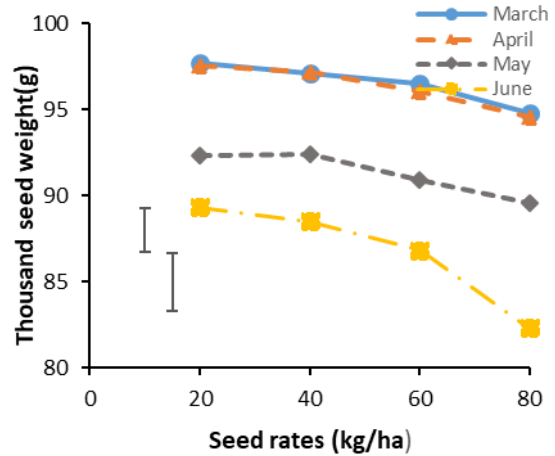


Figure 5.20 Interaction between sowing date and seed rate for TSW at bottom positioned cobs in 2017

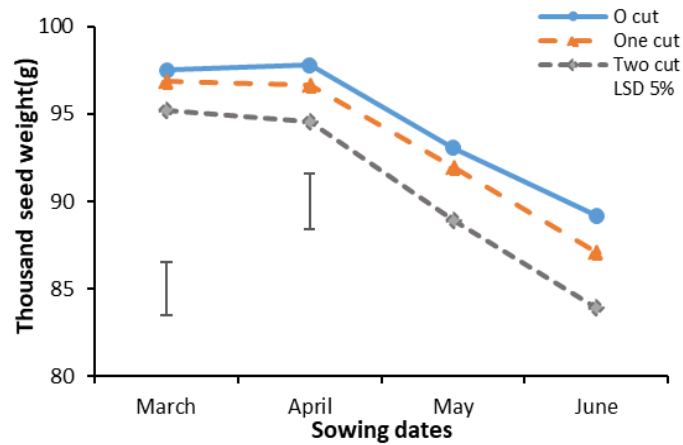


Figure 5.21 Interaction between sowing date and cut management for TSW at bottom positioned cobs in 2017

Note: In figures 5.17-5.21 the left-hand vertical bar is the LSD (5%) for comparisons within a sowing date or seed rate respectively and the right-hand vertical bar is the LSD (5%) for all other comparisons.

Table 5.5 Interaction among sowing date, seed rate and cutting management for TSW (g) of all cobs in 2017

Sowing date	Seed rate(kg/ha)	Uncut	One cut	Two cut
30 March	20	100.0	100.0	98.5
	40	100.0	99.0	98.0
	60	98.5	98.3	96.8
	80	96.5	96.8	95.5
30 April	20	100.0	99.5	97.7
	40	100.0	99.3	96.0
	60	99.3	98.0	96.5
	80	98.0	97.3	95.0
30 May	20	96.3	94.2	92.0
	40	95.8	94.8	92.3
	60	94.8	93.8	91.0
	80	93.0	91.6	89.0
30 June	20	93.0	92.0	88.5
	40	92.3	92.1	87.3
	60	90.4	89.5	87.3
	80	89.1	83.6	80.7
LSD(5%)(within a sowing date)		3.2		
LSD(5%)(all other comparisons)		3.8		

Table 5.6 Interaction among sowing date, seed rate and cutting management for TSW (g) of top positioned cobs in 2017

Sowing date	Seed rate(kg/ha)	Uncut	One cut	Two cut
30 March	20	100.0	99.4	98.0
	40	99.7	98.4	97.4
	60	98.4	97.9	96.5
	80	96.1	96.6	94.9
30 April	20	100.1	99.1	97.4
	40	100.5	98.9	95.5
	60	98.6	97.3	96.1
	80	97.5	96.7	94.5
30 May	20	95.8	93.6	91.4
	40	95.3	94.1	91.8
	60	94.2	93.4	90.4
	80	92.8	91.4	88.5
30 June	20	92.4	91.6	87.9
	40	91.9	91.5	86.7
	60	89.8	88.8	86.7
	80	88.7	83.0	80.1
LSD(5%)(within a sowing date)		3.1		
LSD(5%)(all other comparisons)		3.8		

Table 5.7 Interaction among sowing date, seed rate and cutting management for all cobs TSW (g) in 2018

Sowing date	Seed rate(kg/ha)	Uncut	One cut	Two cut
30 March	20	99.6	97.9	96.3
	40	98.7	97.8	96.3
	60	97.7	96.4	94.5
	80	96.0	95.0	93.4
30 April	20	100.0	98.5	96.3
	40	99.9	97.6	95.6
	60	98.0	96.3	94.4
	80	94.3	93.3	91.7
30 May	20	93.4	95.2	92.2
	40	95.0	94.3	92.1
	60	93.2	91.9	89.7
	80	93.4	90.8	88.0
LSD(5%)(within a sowing date)		2.1		
LSD(5%)(all other comparisons)		2.0		

Table 5.8 Interaction among sowing date, seed rate and cutting management for TSW (g) of the top positioned cobs in 2018

Sowing date	Seed rate(kg/ha)	Uncut	One cut	Two cut
30 March	20	100.0	98.3	97.0
	40	99.4	98.5	96.8
	60	98.0	96.8	95.8
	80	97.0	95.8	94.1
30 April	20	100.0	99.3	97.0
	40	100.8	98.3	96.3
	60	98.8	97.3	95.0
	80	95.0	94.0	92.5
30 May	20	94.1	96.0	92.8
	40	96.0	95.2	93.0
	60	94.3	92.5	90.8
	80	94.3	92.0	88.6
LSD(5%)(within a sowing date)		2.0		
LSD(5%)(all other comparisons)		2.1		

Table 5.9 Interaction among sowing date, seed rate and cutting management for TSW (g) of the middle positioned cobs in 2018

Sowing date	Seed rate(kg/ha)	Uncut	One cut	Two cut
30 March	20	99.0	97.8	95.5
	40	98.0	97.0	95.8
	60	97.3	96.3	93.5
	80	95.0	94.5	92.3
30 April	20	99.8	98.0	96.0
	40	99.3	97.5	95.8
	60	97.3	96.0	94.0
	80	93.8	93.0	91.5
30 May	20	92.8	94.8	92.0
	40	94.3	93.8	91.5
	60	92.3	91.3	89.1
	80	92.5	90.3	87.8
LSD(5%)(within a sowing date)		2.1		
LSD(5%)(all other comparisons)		2.2		

5.4.5 Difference between the TSW among the cob positions

To determine whether the variations in TSW were statistically different among the cobs positions, analyses of variance for TSW were done between seeds from different cob positions, T & M, M & B and T & B. A significant difference between all cob positions was recorded ($P < 0.001$) (Table 5.10). The LSE value at 1% and 5 % showed that the TSW of top positioned cobs was significantly higher than the middle and bottom positioned cobs at each sowing date. The maximum difference in TSW was recorded between the top positioned and the bottom positioned cobs (Table 5.10). Delayed sowing increased the difference for TSW between the seed positions. A similar effect was observed for T & M, M & B and T & B for sowing date, seed rate and cutting management.

Table 5.10 Differences in TSW of seeds from different sowing dates, seed rates and cutting management based on different inflorescence positions

Main effect means of:	Thousand seed weight (g)					
	2017			2018		
	M-T	M-B	T-B	M-T	M-B	T-B
Sowing date (SD)						
30-Mar	0.6	1.1	1.7	1.3	0.6	1.9
30-Apr	0.9	0.8	1.8	1.5	0.5	2.1
30-May	0.8	1.0	1.9	1.5	0.6	2.0
30-Jun	1.0	0.8	2.1	-	-	-
Linear contrast p value	0.057	0.974	0.203	0.085	0.811	0.351
LSD (1 %)	0.5	0.7	0.8	0.2	0.3	0.4
LSE (1%)	0.4	0.5	0.6	0.2	0.2	0.3
LSD (5%)	0.4	0.5	0.6	0.2	0.2	0.3
LSE (5%)	0.9	1.9	2.3	0.4	0.3	0.4
CV%	26.9	28.4	19.8	6.8	22.1	8.5
Seed Rate (SR)						
20 kg ha-1	0.8	1.0	1.8	1.3	0.8	2.1
40 kg ha-1	0.9	1.0	1.9	1.6	0.5	2.1
60 kg ha-1	0.9	1.0	1.9	1.4	0.4	1.8
80 kg ha-1	0.8	1.03	1.8	1.4	0.6	2.0
Linear contrast p value	0.922	0.914	0.984	0.891	0.071	0.246
LSD (1%)	0.4	0.4	0.4	0.5	0.3	0.5
LSE (1%)	0.2	0.3	0.3	0.4	0.2	0.3
LSD (5%)	0.3	0.3	0.2	0.4	0.2	0.4
LSE (5%)	0.6	1	1.2	0.4	0.3	0.6
CV%	45	41.2	21.6	31.2	54.4	21.5
Cut Management (CM)						
No cut	0.8	1.0	1.8	1.6	0.4	2.0
One cut	0.8	1.0	1.8	1.4	0.6	2.1
Two cut	0.9	1.0	2.0	1.4	0.6	1.9
Linear contrast p value	0.323	0.664	0.204	0.124	0.112	0.592
LSD (1%)	0.2	0.2	0.3	0.4	0.3	0.3
LSE (1%)	0.2	0.2	0.2	0.3	0.2	0.2
LSD (5%)	0.2	0.2	0.2	0.3	0.2	0.2
LSE (5%)	0.5	0.5	0.6	0.3	0.2	0.4
CV%	61.6	53.1	31.6	53.5	99.9	28.0
Significance of interactions of linear contrasts (p value)						
SD(lin) x SR(lin)	0.355	0.185	0.027	0.383	0.073	0.696
SD(lin) x CM(lin)	0.761	0.246	0.427	0.165	0.693	0.129
SR(lin) x CM(lin)	0.262	0.977	0.332	0.523	0.209	0.703
SD(lin) x SR(lin) x CM(lin)	0.393	0.612	0.768	0.232	0.739	0.050

Note: LSD = Least Significant Difference; LSE = Least Square Estimator; CV = Coefficient of Variation; - = Not Applicable. Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level (P<0.05). SD=sowing date; SR=seed rate; CM=cutting management.

5.4.6 Correlation between thousand seed weight (TSW) (g) and germination percentage for all the cobs in 2017 and 2018

A positive correlation was observed between TSW and germination percentage for different sowing dates for all cobs in 2017 ($R^2 = 0.77$) ($P > 0.05$) and 2018 ($R^2 = 0.80$) ($P > 0.05$) (Figure 5.22, 5.23). A similar positive correlation was observed between TSW and germination percentage for different seed rates for all the cobs in 2017 ($R^2 = 0.87$) ($P > 0.05$) and 2018 ($R^2 = 0.85$) ($P > 0.05$) (Figure 5.24, 5.25). For cutting management, a significant positive correlation between TSW and germination percentage was recorded in 2017 ($R^2 = 0.96$) ($P < 0.05$) and 2018 ($R^2 = 0.97$) ($P < 0.05$) (Figure 5.26, 5.27). All these correlations indicate higher germination at higher TSW.

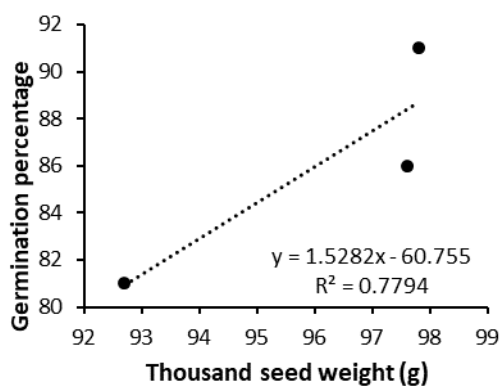


Figure 5.22 Correlation between TSW and germination percentage at different sowing dates for all cobs in 2017

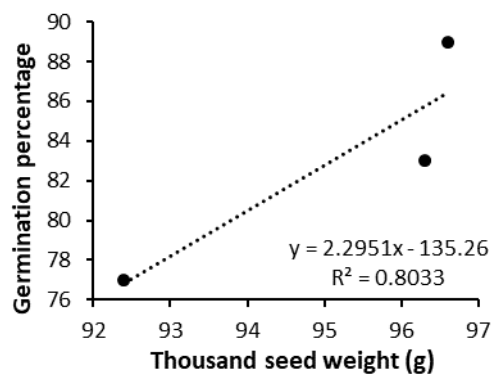


Figure 5.23 Correlation between TSW and germination percentage at different sowing dates for all cobs in 2018

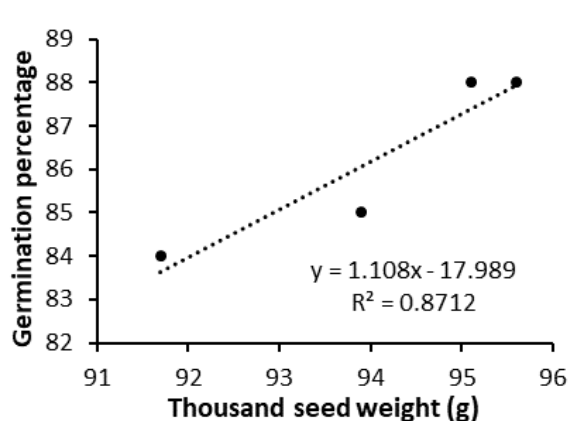


Figure 5.24 Correlation between TSW and germination percentage at different seed rate for all cobs in 2017

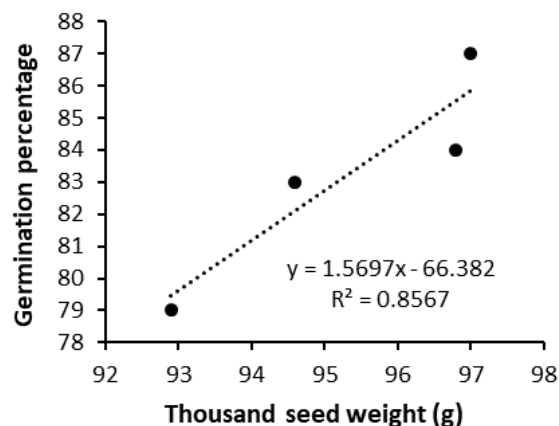


Figure 5.25 Correlation between TSW and germination percentage at different seed rate for all cobs in 2018

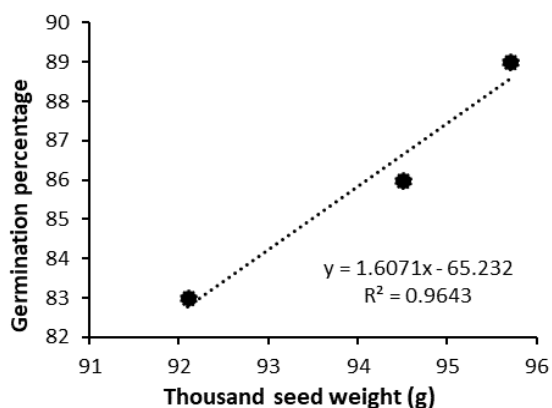


Figure 5.26 Correlation between TSW and germination percentage at different cutting management for all cobs in 2017

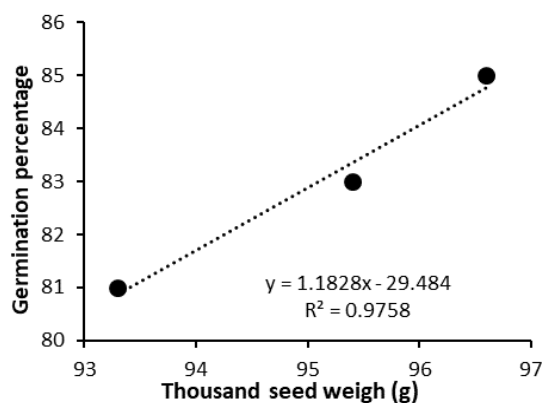


Figure 5.27 Correlation between TSW and germination percentage at different cutting management for all cobs in 2018

5.4.7 Correlation between thousand seed weight (TSW) (g) and germination percentage based on the position of cobs in 2017 and 2018

There was a positive correlation ($R^2=0.69$) ($P>0.05$) between TSW and germination at different sowing dates in 2017 and 2018, significant positive correlations occurred for seed rates ($R^2=0.92$) ($P<0.05$) and a highly significant positive correlation occurred for cutting management ($R^2=0.99$) ($P<0.01$) for top positioned cobs in 2017 and 2018 (Figure 5.28, 5.29, 5.30). For middle positioned cobs, a positive correlation ($R^2=0.70$) ($P>0.05$) occurred between TSW and germination percentage at different sowing dates in 2017 and 2018 (Figure 5.31) and a significant positive correlation occurred between TSW and germination percentage at different seed rates ($R^2=0.94$) ($P<0.05$) and cutting management ($R^2=0.96$) ($P<0.01$) in 2017 and 2018 (Figure 5.32, 5.33). For

bottom positioned cobs, a positive correlation ($R^2=0.68$) ($P>0.05$) between TSW and germination percentage occurred at different sowing dates in 2017 and 2018 (Figure 5.34). But for seed rates, the correlation between TSW and germination percentage was significantly positive ($R^2=0.92$) ($P<0.05$) and for cutting management the correlation between TSW and germination percentage was highly significant ($R^2=0.99$) ($P<0.01$) (Figure 5.35, 5.36) for bottom positioned cobs in 2017 and 2018.

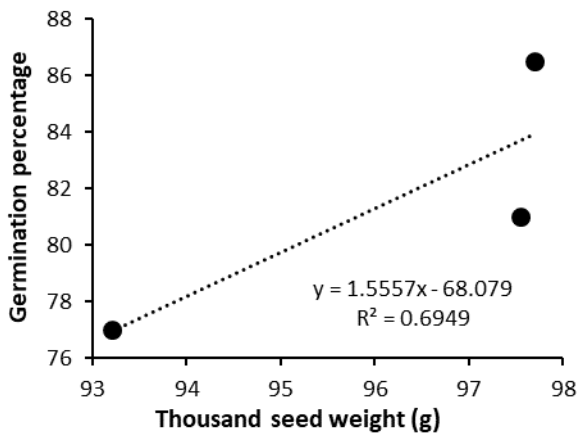


Figure 5.28 Correlation between TSW and germination percentage for top positioned cobs at different sowing dates; data for March, April and May are means for 2017 and 2018

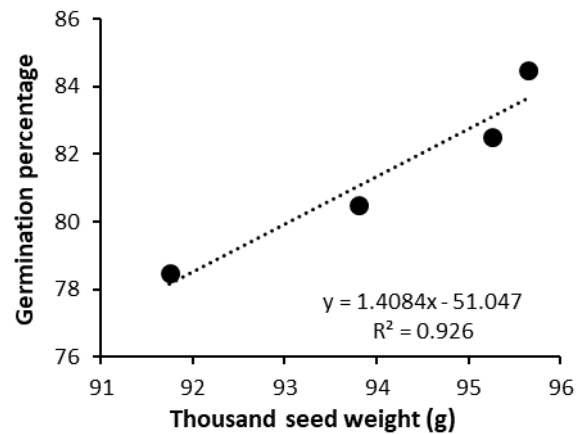


Figure 5.29 Correlation between TSW and germination percentage for top positioned cobs at different seed rates; data are means for 2017 and 2018

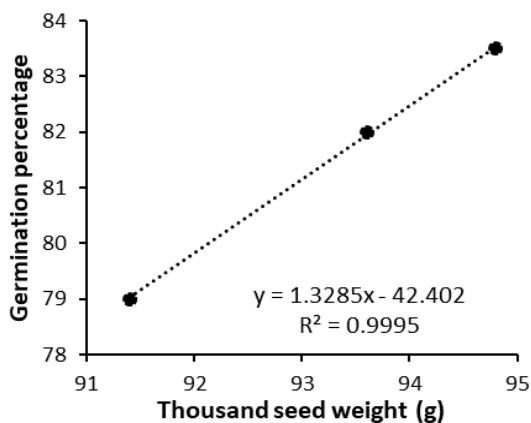


Figure 5.30 Correlation between TSW and germination percentage for top positioned cobs at different cutting management; data are means for 2017 and 2018

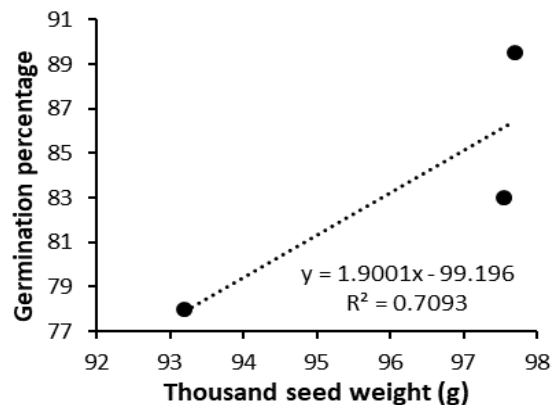


Figure 5.31 Correlation between TSW and germination percentage for middle positioned cobs at different sowing dates; data for March, April and May are means for 2017 and 2018

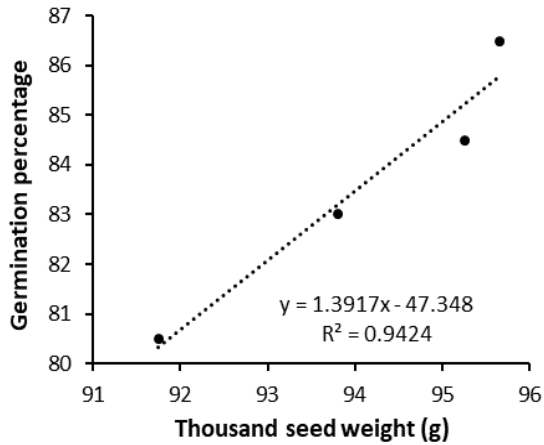


Figure 5.32 Correlation between TSW and germination percentage for middle positioned cobs at different seed rates; data are means for 2017 and 2018

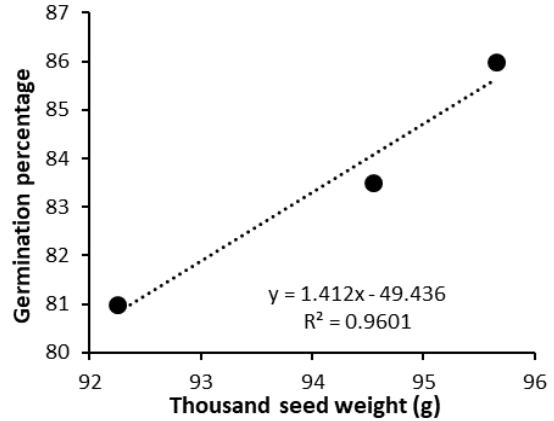


Figure 5.33 Correlation between TSW and germination percentage for middle positioned cobs at different cutting management; data are means for 2017 and 2018

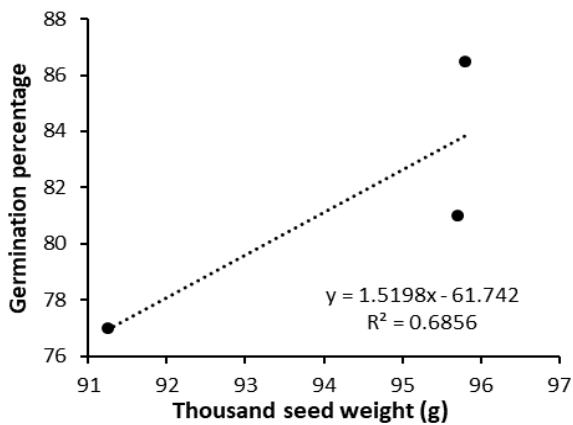


Figure 5.34 Correlation between TSW and germination percentage for bottom positioned cobs at different sowing dates; data for March, April and May are means for 2017 and 2018

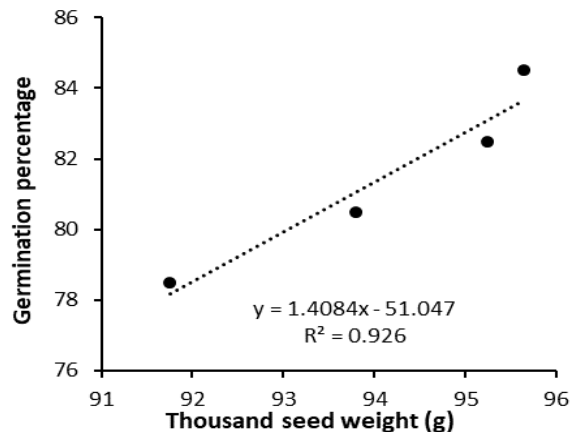


Figure 5.35 Correlation between TSW and germination percentage for bottom positioned cobs at different seed rates; data are means for 2017 and 2018

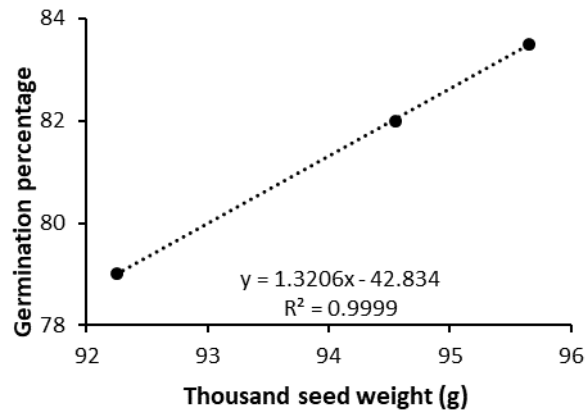


Figure 5.36 Correlation between TSW and germination percentage for bottom positioned cobs at different cutting management; data are means for 2017 and 2018

5.5 Discussion

5.5.1 Teosinte seed development and maturation

Seed development is determined by the environment prevailing at the time of seed growth and maturation (Hampton et al., 2013) as it is affected by various environmental components like water, light, temperature, soil nutrition and also by seed position on the plant (Bareke, 2018). The changes in seed morphology and physiology during maturation help to identify the ideal harvesting time for teosinte seeds, because early harvest may result in immature and undeveloped seeds, while late maturation may result in seed shattering and loss of seeds (Elias & Copeland, 2001 ; Wang et al., 2008). Understanding seed development allows the identification of PM, which is a significant indicator for harvest choices and also used in plant breeding and crop modelling (Li et al., 2021). PM for seed is defined as the time of attainment of maximum seed dry-weight (Aldrich, 1943; Ansari, Afridi, & Khan, 1990) and for maize PM is defined as the maximum kernel dry weight, the black layer formation, and disappearance of the milk line (Afuakwa & Crookston, 1984; Daynard & Duncan, 1969; Shaw & Loomis, 1950).

PM of teosinte in 2017 was attained at 23% SMC. The time from silking to PM in this study was 56 days, and after this seeds began to separate inside the row. Similar results have been reported for 15 different maize crosses, which took between 53-61 days from silking to PM (Ghassemi-Golezani, Tajbakhsh, & Raey, 2011; Hillson, & Penny, 1965) and for which the SMC at PM was 35% (Sala, Andrade, & Westgate, 2007). However, other authors have reported that SMC at black layer maturity (Carter & Poneleit, 1973; Daynard & Duncan, 1969; Rench & Shaw, 1971) can vary from 15.4-37.3% dependent on disappearance of the milk line (Afuakwa & Crookston, 1984). Because

SMC is associated with the unevenness in black layer formation among seeds and between plants, a longer time is required for complete black layer formation in the lower temperatures encountered with a late season (Sala, Andrade, & Westgate, 2007). Similar results have been reported by Li et al. (2017) in an experiment conducted on 11 varieties of summer maize which showed 100 seed dry weight ranging from 23.3 g to 37.4 g at PM and the average SMC at PM ranged from 21.5% to 33.1% which is consistent with the findings of Elias and Copeland (2001). The variation in the SMC at PM for maize and teosinte can be explained by differences in the temperature and the relative humidity in the area where crops were grown, because moisture loss from maturing seeds ceases when equilibrium seed moisture content is attained (TeKrony & Phillips, 1980).

The cumulative GDD for teosinte from completion of flowering to PM was 617°C for the March sown crops in 2017. Likewise, for maize, the mean GDD from anthesis to PM ranged from 512-821°C days (Carter & Poneleit, 1973). Similarly, Baum, Archontoulis, and Licht (2019) also reported a GDD of 648°C days for seed filling of maize. The variation between the number of GDD required for maize during the filling period depends on phenotypic correlations with SMC at PM, dry weight at PM and GDD from planting to pollination (Carter & Poneleit, 1973). The variation in the black layer formation is most likely affected by slower accumulation of GDD during the latter part of the season (Carter & Poneleit, 1973; Danyard, 1972). Further, the difference in the GDD requirements of teosinte and maize can be explained by the growth environment and duration of each crop. Since teosinte has an extended growth period it accumulates higher GDD.

5.5.2 Germination

Sowing date

Different sowing dates can result in variation in seed quality due to changes in environmental conditions (Rahman et al, 2005; TeKrony & Egli, 1997). The differences in environmental conditions the mother plant experience during the seed development and maturation phase may affect the germination (Fenner, 1991; Gutterman, 1980-81; Kaushal, Bhandari, Siddique and Nayyar, 2016; Rahman, Rahman & Hossain, 2013).

The present study showed that the March sowing produced seeds with higher germination for all the cob positions in the plant, and the germination was reduced with delayed sowings. Seeds from cobs from the top position had the highest germination for each sowing date, and germination was reduced for seeds from the middle and bottom cobs respectively. Higher germination in seeds from the early sown plants might be a result of the longer growth period which accumulated higher GDD for each phenological growth stage that allowed a better capacity

to store photoassimilates and produced seeds with a higher TSW. This result is in line with the findings of Ebadi and Hisoriev (2011) who reported that germination is dependent on the reserved nutrient quantity in seeds. Baskin and Baskin (1998) also suggested that larger seeds have a higher germination than smaller seeds and they produce more vigorous seedlings, but this however depends on the species and individual seed lots, as larger seeds do not always have better germination than smaller seeds (Hampton et al., 2013). Similar results have been reported by Msuya and Stefano (2010) in maize where the larger basal position seeds on the cob had higher germination than the smaller distal position seeds because of larger seed weight.

Higher germination from seeds at the top cobs is likely due to the competition among the developing seeds for assimilates because the sink strength of seed increases with size and age, and the earlier formed seeds will have a greater sink capacity. This result is in line with the findings of Diggle (1995) who reported that the stronger sink at the top positioned cobs is the result of resources allocation within the inflorescence, with preferential access to photosynthates, water and nutrients. Datta, Evenari, and Gutterman (1970) reported that early formed seed might be the preferential 'sink' for assimilates because of the competition between the florets inside the spikelet at an early stage is possibly before the occurrence of meiosis in the embryo. Similar findings have been reported by Dohleman and Long (2009) in maize, Emms (1996) in sand corn, Espadaler, and Gomez (2001) in Euphorbia, Hardesty and Elliott (1956) in wheat and Reddy, Ganiga, Devendra, Shankerogowda, and Mahadevappa (1976) in rice. Jacobsohn and Globerson (1980) in their study of seed positional effect in carrot found that seeds from primary umbels had better germination than those of the same size from secondary and tertiary umbels.

In this study delayed sowing reduced germination (i.e. the production of normal seedlings). Although some of the seeds germinated physiologically (the radicle emerged through the seed coat). They produced abnormal seedlings which had either missing primary roots or stunted root or absence of shoots in some cases. Similar findings to this result have been reported by ISTA (2016) and Rashid, Hampton, Rolston, Trethewey, and Saville (2018), where reduced germination was associated with higher abnormal seedlings because of root abnormalities, than with dead seeds. Germination capacity declines as seeds age physiologically, but complete death is usually preceded by the production of abnormal seedlings whose development is weak or unbalanced because the loss of vital functions does not occur simultaneously in the different tissues (MacKay, 1972; Rashid, Hampton, Rolston, Khan, & Saville, 2018). The production of abnormal seedlings can have many causes including chemical and mechanical damage, growing environment, harvesting methods, seed moisture, diseases and pests (Egli, TeKrony, Heitholt, & Rupe, 2005; Gillen, Smith,

Mengistu, & Bellaloui, 2012) and higher temperature during the plant growth period could also be one of the most prominent causes (Rashid, Hampton, Trethewey, & Rolston, 2017; Sanchez, Rasmussen, & Porter, 2014; Shinohara et al., 2006). In the present study, the seeds were hand harvested and sorting and drying was done manually so there was little chance of mechanical injury, and no disease or pest damage during the seed harvest was recorded. This indicates that the lower germination must be because of physiological deterioration of seed that had begun before seed harvest. A short period of heat stress is enough to initiate physiological deterioration in seeds (although it doesn't kill the seeds), which results in the production of abnormal seedlings (Rashid et al., 2018). High temperature disrupts the cell membrane stability and functions, enhancing the cell membrane permeability to ions (Bailly, Bogatek -Leszczynska, Come, & Corbineau, 2002; Ren, Bilyeu, & Beuselinck, 2009) leading to seed deterioration (McDonald, 1999) because cell membranes are sensitive to high temperature. Further seed deterioration occurs by changes in the phospholipid composition in the seed that results in loss of membrane integrity due to the changes in membrane configuration or changes the properties of membrane bound enzymes leading to leakage of ions in incubation water (Ren, Bilyeu, & Beuselinck, 2009; Taiz & Zeiger, 1998). Many physiological process are involved to allow root production, including the production of sufficient energy for germination and also the controlled production of reactive oxygen species (ROS), especially H_2O_2 (Bewley & Black, 2012; Rashid, 2016). This is because newly synthesised compounds and food reserves in the seed are mobilized for root development (Bailly, Bogatek -Leszczynska, Come, & Corbineau, 2002; Kupidlowska et al., 2006). High temperature interrupts the equilibrium between ROS production and ROS scavenging enzymes, which is likely to reduce the metabolic activity of seed that disturbs normal root growth (Rashid, 2016). Abnormal seedlings resulting from physiological aging are because of unrepaired cellular damage to mitochondrial DNA and RNA as a result of unrestrained deposit of H_2O_2 and reduced energy supply (Bailly, El-Maarouf-Bouteau, & Corbineau, 2008). At this stage the deterioration has not progressed sufficiently to stop radicle emergence but the seedlings produced are abnormal (Khajeh-Hosseini, Nasehzadeh, & Matthews, 2010). This result is in line with the findings of Rashid, Hampton, Rolston, Trethewey, and Saville (2018) who demonstrated that heat stress during seed development resulted in the production of abnormal seedlings. The difference in seed germination for the two years at each cob position is likely due to difference in the weather components.

Seed rate

The influence of the light a mother plant receives during seed production affects seed germination in several weed species (Bello, Owen, & Hatterman-Valentp, 1995; Brainard,

Bellinder, & DiTommaso, 2005; Kigel, Ofir & Koller, 1977). Seed quality is affected by plant densities depending on the seed position on the mother plant (Foroughi, Gherekhloo, & Ghaderi-Far, 2014). Being a C₄ plant, and a progenitor of maize, the light equilibrium of teosinte is believed to be very similar to that of maize, as reported by Idikut (2013). Therefore in partial light, crop growth is hindered.

In the present study the two lower seed rates (20kg ha^{-1} and 40kg ha^{-1}) produced the seeds with the highest germination percentage for the top and middle cobs, but for bottom cobs, the 20kg ha^{-1} seed rate produced seeds with highest germination in 2017. In 2018, the highest germination was recorded for the lowest two seeds rates and lowest germination was recorded for the highest seed rate for the overall cobs and for top and middle positioned cobs. For bottom positioned cobs, highest germination was recorded for the lowest seed rate and vice versa. Higher germination at lower density in both years for all cobs is likely due to higher light penetration at lower plant density. Lower competition between plants for resources allowed higher assimilates transfer from source to sink which is clearly evident in Table 5.4. This result is similar to the findings of Tuarira and Moses (2014) in green bean. Further, inter and intra plant shading might be another reason for lower germination at higher plant density. The difference in the germination percentage between the top, middle and bottom positioned cobs might be due to shading effect of the mother plant, which is similar to the findings of Baskin and Baskin (1998) and Gutterman (2000) who reported that position of seeds influenced seed germination. A similar result was reported by Brainard, Bellinder and DiTommaso (2005) in Powell amaranth where a 40 to 50% decrease in germination was observed in seed developed under shade in comparison to seeds developed under full sunlight. Weiner (1988), reported 67 to 78% greater germination from the seeds based on the top to middle third than those at the bottom third of the mother plant.

Cutting management

In the current study, seed germination was reduced with increase in cutting frequency in both years. A similar result was reported by Ibrahim and Sakr (2016) in Sudan grass under cutting management. Germination was also higher for the seeds at the top position and it reduced for the middle and bottom position which is likely due to competition for the assimilates between the developing seeds.

Interactions

In 2017, uncut plants produced seeds with higher germination at the lower seed rate for all cobs, and for top, middle and bottom positioned seeds due to reduced competition for assimilates among and between the plants where higher herbage in uncut plants allowed higher leaf photosynthetic rates due to better light penetration at lower density through larger leaf canopy

and greater LAI. Similar result has been reported by Dohleman, Heaton, Leakey, and Long, (2009) and Heaton, Dohleman, and Long (2008) who reported higher leaf photosynthesis at a higher LAI in miscanthus. In 2018 higher germination was recorded for the March sowing and uncut plants in overall and bottom position cobs which might be because of longer growth period of teosinte that allowed the accumulation of more assimilates in uncut plants in the early sowing.

5.5.3 Thousand seed weight

Sowing date

Seed weight is influenced by water availability (Brooks, Jenner, & Aspinall, 1982), assimilate availability (Blum, 1998; Egharevba, Horrocks & Zuber, 1976) and temperature (Wardlaw & Wrigley, 1994) as reported by Castro, Hodar, and Gomez (2006). Other reasons for variation in seed weight are a paternal genetic effect (Cavers & Harper, 1966; Datta, Evenari & Gutterman, 1970), progeny size, sibling rivalry, timing of flowering and prolificacy and position of seeds within a plant, or in the inflorescence, and the position of a seed inside the fruit (Obeso, 2012). In maize, genetics have been reported to be the main determinant of final weight achieved by seeds (Reddy & Daynard, 1983) but both high temperature (Hampton et al., 2013) and low temperature (Shim, Lee, Koo, Shin & Yoon, 2019) can reduce the seed weight by altering the seed filling period.

In the present study, the first two sowing dates produced the heavier seeds. The similarity between the two sowing dates for TSW can be explained by the adaptive capacity of teosinte to compensate for a longer growing season. The mean temperature during seed filling for the four sowing dates in 2017 was 24.9°C, 20.9°C, 20.5°C and 19.4°C in 2017 and 24.2°C, 20.7°C and 19.8°C for the three sowing dates in 2018. Despite the higher temperature and shorter seed filling period, TSW was not reduced in the first two sowings because teosinte was grown under a higher soil moisture and nutrient supply. In addition, the longer vegetative growth duration (Andrade, 1995) allowed a higher volume of assimilates to accumulate for transfer to the sink which is similar to the findings of the current study where phenologically the days required for vegetative growth for the March sowing was 157 and 160 days for 2017 and 2018, for the April sowing it was 136 days in both years and for the May sowing it was 107 days for both years. For the June sowing in 2017, the phenological days required for vegetative growth was 80 days (Table 3.1-3.4, Chapter 3). This result is similar to the findings of Koca, Canavar and Kaptan (2014) in maize. Nicolas, Gleadow and Dallin (1984) and Motzo, Giunta, and Deidda (1996) reported that higher seed weight in a shorter seed filling duration is associated with a greater seed filling rate in wheat which is similar to the findings of the current study where the phenological days for seed filling was 58 and 59 days for the March and the April sowings in 2017 and 2018 respectively; and 59

and 60 days for the May sowings in 2017 and 2018. It was 60 days for the June sowing in 2017. Lower TSW in the later sowings is likely because of smaller volume of seed filling at a lower temperature. This result is similar to the findings of Deivasigamani and Swaminathan (2018) in major field crops. Low temperature extends the cell cycle by reducing the cell division process (Francis & Barlow, 1988; Rymen et al., 2007) thus extending the seed maturation duration. It also affects the biomass partitioning to seeds through seed filling as seed weight is more dependent on reserve mobilization during post silking crop growth (Andrade, Uhart, & Cirilo, 1993; Ali, Ahsan, Mustafa, & Ejaz-ul-Hasan, 2013; Cirilo & Andrade, 1994; Kgasago, 2007). A shorter photoperiod due to delayed sowing reduces the daily net carbon gain in plants (Wulff, 1986). Taylor and Blackette (1982) reported a seed weight reduction of 10 percent in spring barley with delayed sowing. Further, delayed sowing reduced the GDD accumulation during the seed filling period of teosinte resulting in delayed seed maturation under lower temperature. Low daily incident solar radiation might be another reason which reduced the effective seed filling rate and lengthened the duration of seed filling. This is in line with Bonhomme, Derieux, and Edmeades (1994); Cirilo and Andrade (1996); Kiniry, Richie, and Musser (1983) and who reported that maize seed development is affected by photoperiod.

The variation in seed weight between seeds at the top and the bottom of the teosinte plant in both years is a result of competition for resources which is unfavourable for seed in the bottom positioned cobs that develop later in the plants. This result is in line with the findings of Hendrix (1984) in parsnip, Kolodziejek (2017) in fennel, Muchoq (1990) in sorghum, Thomas (1996) in parsley and Thomas, Biddington, and O'toole (1979) in celery where the seeds from the primary umbel had higher weight than the seeds from the lower order, secondary and tertiary umbels. Further, self-shading and interplant shading might be another reason for lower seed weight of the middle and the bottom positioned seeds as a result of insufficient water and nutrient levels for seed filling. This is consistent with the findings of Gao et al. (2018) and Wulff (1986) who reported that shading in a dense canopy produced smaller seeds because seed weight intensely depends on the photosynthetic ability of plants. Shi et al. (2015) also reported that shading limits the seed dry matter accumulation at later seed filling period leading to reduced seed weight. No such information on the difference in seed quality based on the cob positions of teosinte was found during this study. However, a contrasting result of TSW differences at different seed positions on the cob of maize was reported by Batistella, Moro, and De Carvalho (2002), Tollenaar and Daynard (1978), and Xu et al. (2015) where seeds at the top position on the cob were smaller than those at the middle and the distal positions, which is the consequences of inadequate food availability for the seed at the apex of the cob during seed development (Srinivas et al., 1991). In

the present study, the effect of seed position within a teosinte cobs was not assessed. The difference between two years can be explained by the difference in the environmental factors the mother plant received at each sowing in both planting seasons.

Seed rate

Maddonni, Otegui, and Bonhomme, (1998) reported that seed weight depends on the mobilization of reserves after silking. Teosinte, being a C₄ plant, has a metabolism which is highly affected by light intensities (Ubierna, Sun & Cousins, 2013) for canopy production (Kromdijk et al. 2008). Decreased incident solar radiation reduces the biomass production, which impairs seed filling (Ali, Ahsan, Tahir, & Basra, 2014). High yield reduction due to reduced seed weight has been reported as a result of shading during the reproductive growth stage of maize (Earley, McIlrath, Seif & Hageman, 1967; Li et al., 2005; Zhang, Dong, Wang, Hu, & Liu, 2006).

In the present study the two lower seed rates produced seeds with the highest TSW for overall and all cob positions in both years. This result is in line with the findings of Wulff (1986) who reported that seed weight is dependent on the light penetration through the canopy for accumulating photosynthates. Another reason for lower seed weight could be that at higher plant density seed filling duration is reduced as a result of higher plant to plant competition as reported by Poneleit and Egli (1979). Higher plant density increases respiration and decreases photosynthesis leading to reduced transmission of assimilates to seeds and therefore a reduction in the seed yield and weight as reported by Giridhar, Reddy, Kumari, Kumari and Sivasankar (2017) in black cumin, Novacek, Mason, Galusha and Yaseen (2013) and Mandic et al. (2016) in maize, Rahman, Mwakangwale, Hampton, and Hill (2005) in soybeans and Sahu, Tomar, and Nandeha (2018) in sorghum. The differences in thousand seed weight at different cob positions might be due to inter and intra plant shading. Shading reduces the ratio between red and far-red (R/FR) light lowering the PAR interception which limits the canopy productivity, reducing the assimilate synthesis under high density (Cui, Jin, & Zhang, 2015; Yang & Li, 2017; Yang et al., 2018). Low light intensity under shading can result in barren stalk (Zhong et al., 2013), reduced seeds per ear, higher shrivelled seed and reduced seed weight. Cui, Camberato, Jin, and Zhang (2015) reported that under shading silks, fertilized florets per ear, and floret fertilization rate were reduced, ear differentiation was delayed, and anthesis silking interval was lengthened resulting in lower seed weight. Jia, Li, Dong, and Zhang (2011) and Ubierna, Sun and Cousins (2013) found that one of the major causes of decreased seed weight under shading is the reduction in starch content, endosperm cell number, and volume which delayed the enlargement of the starch granules so that endosperm filling was abnormally hindered and the capacity of storage was limited. Reduced assimilate supply to the developing ear as a result of shading during

reproductive growth stage reduced the seed number per ear and seed weight (Cui, Camberato, Jin, & Zhang, 2015).

Cutting management

In the present study, cutting reduced the TSW in all cobs and all cob position in both years. Cutting by removing leaves and lowering LAI reduced the availability of assimilates able to be translocated to the seeds. This finding is consistent with the findings of Baswana and Pandita (1989) in fenugreek. Further, shorter growth period accompanied by cutting shortened the seed filling period which is similar to the findings of Frey (1981) in maize, where defoliation reduced the seed filling period and thus reduced the seed weight.

Interactions

A significant interaction between sowing date and cutting management for TSW for seeds harvested from all cobs and from top, middle and bottom position cobs occurred because the earliest two sowings produced seeds with the lowest TSW in twice cut plants due to lower assimilates production by the plant and intraplant competition for the assimilates. Similarly, a significant interaction also occurred between sowing date and seed rate for bottom positioned cobs in 2017 which is because June sown plants at 80 kg ha^{-1} produced seeds with lower TSW because of a shorter growing period and strong competition of plants for limited resources at the highest planting density. Likewise, a significant interaction between sowing date, seed rate and cutting management for TSW in all seed and seed from top and middle positioned cobs in 2017 occurred because lower temperature reduced assimilate supply to the seeds from twice cut plants, and there was more competition for reduced assimilate supply at higher density.

5.5.4 Seed position and seed quality

Teosinte has an indeterminate growth habit and flowering begins at the top and moves towards the bottom of the stem. Being thermophilic in nature, teosinte performs better at higher temperature. As a result, the foliage at the upper position from early flowering can intercept better solar radiation and produce more assimilates than the bottom, which faces the shading effect due to vigorous and profuse growth of the plant.

Seed position on the plant is one of the important components of variability for within plant difference and might be responsible for the physical (weight and shape) or physiological (viability and vigour) difference of seed attributes (Illipronti, Lommen, Linger, & Struik, 2000). Seed quality components such as seed size, embryo size, germination and vigour are influenced by seed position on the plant as it depends on the plant morphology, photosynthetic productivity and seed maturity (Hampton, 2000). At different phases of plant growth, the environment in which

the mother plant and the individual seed is grown will differentiate assimilate supply for growth between seeds produced on different positions on the plant (Gray, & Thomas, 1982). Maternal factors, either the location of the inflorescence on the mother plants or the location of the seeds in the inflorescence or even location of seed inside the fruit has been reported to significantly affect the germinability of seeds (Datta, Evenari, & Gutterman, 1970; Evenari, Kadouri, & Gutterman, 1977; Grey & Thomas, 1982).

Seed position within the ear has also been reported to influence seed size in maize (Bell, 1954). Anslow (1964) reported that early emerged spikes produced larger seeds in perennial ryegrass. In wheat, a severe dearth of assimilates results in lower weight in seeds that develop later in the plant (Bremner, 1972). The difference in seed weight on the same plants varies with the amount of carbohydrate being received by the seeds, at each position in a plant (Deleuran, Olesen, & Boelt, 2013).

5.5.5 Correlation between TSW and germination percentage

There was a positive correlation between the TSW and germination percentage for sowing dates and seed rates for all cobs and a significant positive correlation occurred for the cutting management for all the cob positions. Significant positive correlation between TSW and germination was observed for top, middle and bottom positioned cobs in this study which is because germination increased as the TSW increased. A positive correlation between TSW and germination might have occurred because the heavier seeds have a higher energy reserve which allows the growth of larger and healthier seedlings and faster seedling growth. Similar results have been reported by Moshatati and Gharineh (2012) in wheat. Until they are able to produce their own photosynthates, seedlings are dependent on the seed reserves and therefore the size of the embryo and the seed reserves are highly important for germination and emergence. Thus higher TSW will allow better seed germination (Baker, 1972; Cordazzo, 2002; Gross, 1984; Kolodziejek, 2017; Newbery & Newman, 1978; Noor-mohammadi, Siadat & Kashani, 2000). Similar results have been reported by Aharizad (2010) in chick pea, Ghassemi-Golezani (1992) in cereal, Ghassemi-Golezani, Mousabeygi, Yaeghoob, and Singh, Tripathi and Negi (1972) in soybean because large seeds have greater stored energy to support early seedling growth.

5.6 Conclusions

In this field trial, effects of different sowing dates, seed rates and cutting management on seed development and seed quality of teosinte in Nepal were investigated. Teosinte seed development is influenced by sowing dates. The results of this study also revealed that sowing date, seed rate,

cutting management and their interactions could significantly influence the seed quality of teosinte.

- Teosinte seeds reached physiological maturity (PM) at 56 DAA and the seeds required 617°C days (GDD) from anthesis to reach PM in March sown plants.
- There was a significant difference in overall germination among different sowing dates, where March sowing produced seeds with the highest germination (91% in 2017 and 89% in 2018). Sowing teosinte early improved the seed germination.
- Seeds sown at 20 kg/ha and 40 kg/ha had a statistically ($P < 0.05$) higher germination, (88% in 2017 and 87% in 2018), than the two higher seed rates; lower seed rates enhanced the teosinte seed germination.
- Cutting significantly reduced the seed germination.
- Thousand seed weight for the first and second sowing dates (March 30 and April 30) did not differ significantly but the thousand seed weight was reduced significantly for the last sowing (June in 2017 and May in 2018). Sowing early improved the thousand seed weight.
- Seeds sown at the lowest two seed rates had seeds with the highest thousand seed weight but these two seed rates did not differ statistically. Thousand seed weight was influenced by changing sowing dates.
- Cutting reduced the thousand seed weight in all the sowing dates and seed rates applied.
- Position of seed on the mother plant affected the germination percentage and thousand seed weight in teosinte, where seeds from the top cob position had higher germination and thousand seed weight than seeds from the middle and bottom positioned cobs.
- There was a significant positive correlation between thousand seed weight and germination for seeds from all the cob positions.

Chapter 6

Evaluation of different genotypes of *Euchlaena mexicana* in Nepal for herbage and seed production

6.1 Introduction

Human intervention through traditional practices for economic, cultural and nutritionally important crops has resulted in the domestication of wild crop relatives (Hernandez-Teran, Wegier, Benitez, Lira, & Escalante, 2017). One example of such domestication over thousands of years is development of maize from *Euchlaena mexicana*, as Balsas Teosinte (*Zea mays* subsp. *parviglumis*), is now agreed to be the biological ancestor of maize (Sahoo, Adhikari, Joshi & Singh, 2021). Abundant large and fairly undisturbed natural populations of teosinte species across the slopes of steep environments (Hufford et al., 2013; Sanchez, & Corral, 1997) makes teosinte suited for studies on local adaptation and natural selection. Besides, these populations also represent a predominantly beautiful learning system for natural and evolutionary genomics (Hufford, Bilinski, Pyhajarvi, & Ross-Ibarra, 2012).

The variation of hereditary characteristics in genetic makeup among related population of the same species is called genetic diversity (Hughes, Inouye, Johnson, Underwood, & Vellend, 2008). It is a multidimensional concept that can be applied to a population or group of populations, plant species and traits of specific interest for selecting individual genotypes from the closely related population for new breeding activities (Boakyewaa Adu, 2019; Hallauer & Miranda, 1988). It also helps to enable and sustain an effective long term plant breeding programme based on an understanding of the degree of genetic variability that is already existing in the germplasm (Mumtaz, Hussain, Saeed, Arshad & Yousaf, 2018). Natural mutation, geographic separation and genetic barriers are the major causes of genetic diversity (Appa, Rao, Mengesha & Reddy, 1996; Zongo, Gouyon & Sandmeier, 1993). Knowledge of genetic diversity gives researchers useful evidence for germplasm identification, conservation and utilisation (Ahmad, Khan, Ghaffar, & Ahmad, 2011). Quantifying the genetic variability among the genotypes growing in a particular area is necessary before starting any breeding program (Jotshi, Bhat & Bhan, 1988).

Crop diversity can be predicted using various criteria including phenotype, the pedigree and molecular markers (Matus & Hayes, 2002). However, morphological description is the first step in the classification of plant genetic resources (Smith, Hartnett, & Wilson, 1991) as the level of

diversity is visual. Visual observation of the characteristics makes the morphological study easier for the scientists and the farmers to select the characteristics of their interest (Bellon & Berthaud, 2006). Although morphological traits are affected by the environment, they indicate the diversity visually and impact the productivity of genotypes by acclimatisation to the environment (Geethanjali, & Jegadeeswaran, 2016). Many genetic variations can be identified through differences in morphology because they are ecologically adaptive (Agrama & Tuinstra, 2004; Torkpo, Danquah, Offei, & Blay, 2006) and the most obvious indicator of genetic diversity (Grenier, Hamon, & Bramel-Coxrenieret, 2001). While both natural and anthropogenic drivers are involved in the complex process of crop genetic diversity and its dynamics, they can be susceptible to variations at diverse levels of agrosystems between species, among varieties and within varieties (Deu et al., 2010). However, much of the variation among the crops can only be identified once the crop is cultivated and propagated in the field. Many farmers are able to recognise variation among cultivated crops and recognize that environmental and cultural drivers are based on crop adaptation and development (Bellon et al., 2018).

Teosinte has been grown and cultivated in different environments, thereby evolving different phenotypes and functional diversity. It has several useful characteristics, including its capacity to produce more herbage and withstand multi cutting better than maize, and a good ability to endure biotic and abiotic stresses (Jia, Yang, Wang, Wu & Sheng, 2008; Wang, Wang, Yuan & Xu, 2004; Warburton et al., 2011).

The only cultivar of teosinte currently used in Nepal (Sirsa) takes 8-9 months from sowing to seed harvesting, which makes growing this crop difficult for farmers with small land holdings wanting to establish winter crops. To investigate whether other teosinte genotypes might be useful for Nepal, 17 accessions of teosinte were imported from the gene bank of CIMMYT, Mexico and were evaluated along with Sirsa for yield, maturation time and regeneration capacity. Sirsa teosinte was introduced to Nepal in 1967 from India with an aim to supply high yields of green herbage for livestock (PFD 2015). It was highly admired by the farmers because of its multicut nature, profuse tillering, generous production and good regeneration capacity. Besides Sirsa, no other cultivars of teosinte have been imported formally into the country. Because of its expanding area and production, and an open border with India, there is a question as to whether any other teosinte cultivar has been introduced to Nepal through informal channels, or in effect a new cultivar has developed due to introgression among the genotypes. Knowledge on teosinte germplasm diversity and relationships among breeding materials have an important impact on the selection and improvement of a crop (Hallauer & Iranda, 1988). Therefore to investigate the prospects of

diversity of teosinte genotypes already in the country, the morpho-physiological and yield traits of seven teosinte genotypes from seven district of Nepal were evaluated.

6.2 Objective

The objective of this study was to (i) evaluate performance of different accession of *Euchlaena mexicana* imported from CIMMYT, Mexico in Nepal and (ii) to identify if the teosinte grown in different parts of Nepal belonged to the same genotype.

6.3 Methodology

6.3.1 Site description

Two experiments were conducted in the experimental block of the Agriculture and Forest University, Rampur, Chitwan during the summer season of 2018. The farm is situated in the central region of Nepal at Rampur, Chitwan District at 27° 84'' N Latitude and 84° 57' E longitude with an altitude of 228 m above mean sea level.

6.3.2 Description of soil properties

A soil sample was tested at the laboratory of the Agriculture Technology Center, Kathmandu in 2018. The soil is a slightly acidic, light textured sandy loam. The details of N, P, K and OM available in the soil of the experimental field are presented in Table 6.1.

Table 6.1 Soil analysis report of the experimental field at horticulture farm, AFU, Rampur, Chitwan, 2018

Soil	pH	Total N %	Available P ₂ O ₅ kg/ha	Available K ₂ O kg/ha	OM%
Depth of soil					
Top soil (0-5 cm)	6.9	0.17	42	298	2.67
Subsoil (20cm-25cm)	6.8	0.18	27	265	3.52

6.3.3 Weather during the study period

The weather data are presented in Chapter 2.

6.3.4 Land preparation and experimental lay-out

The first experiment was conducted using eighteen different accession of teosinte, seventeen of which were imported from CIMMYT, Mexico for evaluation in Nepal in 2018 (Table 6.2). These 17 accessions are landraces collected from various parts of the world and conserved in the active collection vault at the seed bank of CIMMYT, Mexico. Since only one certified cultivar of teosinte

(Sirsa) exists in Nepal, the experiment also includes this local cultivar as a control. The 18 accessions were randomized over the plot in a completely randomized block design (CRBD) with each accession replicated five times. Five seeds of eighteen accessions (17 from CIMMYT and Sirsa) were sown manually in each line giving a total of twenty five seeds of each accession sown in the trial. The 17 accessions of teosinte from Mexico were represented as T1, T2, T3, T4, T5, T6, T7, T8, T9, T10, T11, T12, T13, T14, T15, T16, and T17 respectively. Local accession Sirsa was represented as T18. The area required for each plot was 10.8 square meters (7.2 *1.5). Two plants in each plot were tagged to record the data.

The second experiment evaluated the genetic diversity of teosinte currently grown in Nepal. Seven seed lots of teosinte were collected from the southern and the central part of the country where teosinte has been continuously grown by farmers and at research stations for many years. The seed lots were collected from T1-Chitwan, T2-Sarlahi, T3-Mohattari, T3-Bara, T5-Gaughat, T6-Tikapur, and T7-Makwanpur (Figure 6.1) in collaboration with Forage Development Association (FDA) of Nepal. The 7 seed lots were randomized over the plot in a CRBD and each was replicated four times. The size of each plot was 2.4*2.4 square meter. The seed rate used for each treatment was 40 kg seed per hectare. Five plants in the middle row of each plot were tagged to collect the phenotypic data. The phenotypic characters were used as a basis for investigating genetic variation.

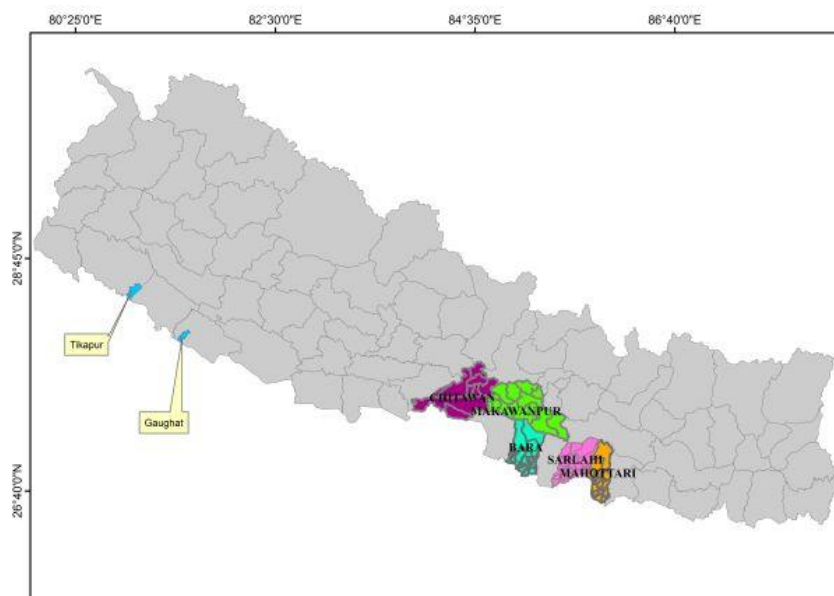


Figure 6.1 Location map for collection site of seven Nepalese seed lots

Both experiment were conducted at the experimental block of the horticulture farm, Agriculture and Forest University. The plant to plant distance for both experiments was 30 cm and the row to

row distance was 40cm apart. The spacing of 50 cm in between the plots and one meter at the upper and lower edge was maintained. Crop management was as described in Chapter 2.

Table 6.2 Teosinte accessions imported from CYMMIT, Mexico and used for the study (17 Mexican and one local)

Treatments	Accessions	Germplasm ID*	Population name	Origin	Number of seeds
1	CIMMYTMA 29740	265739	MGB-CI 51	MN-2008-U--	25 Seed
2	CIMMYTMA 29737	265736	MGB-CI 48	MN-2008-U--	25 Seed
3	CIMMYTMA 29736	265735	MGB-CI 47	GR-2008-U--	25 Seed
4	CIMMYTMA 29735	265734	MGB-CI 46	GR-2008-U--	25 Seed
5	CIMMYTMA 29733	265732	MGB-CI 44	MN-2008-U--	25 Seed
6	CIMMYTMA 29732	265731	MGB-CI 43	MN-2008-U--	25 Seed
7	CIMMYTMA 29730	265729	MGB-CI 41	MN-2008-U--	25 Seed
8	CIMMYTMA 29728	265727	MGB-CI 39	MN-2008-U--	25 Seed
9	CIMMYTMA 29726	265725	MGB-CI 37	MN-2008-U--	25 Seed
10	CIMMYTMA 29723	265722	MGB-CI 34	MN-2008-U--	25 Seed
11	CIMMYTMA 29722	265721	MGB-CI 33	MN-2008-U--	25 Seed
12	CIMMYTMA 29719	265718	MGB-CI 30	GR-2008-U--	25 Seed
13	CIMMYTMA 13576	252701	W.S.T. 92-6	BA-2009-U-2903-21	50 Seed
14	CIMMYTMA 13570	252695	E 86-3	TE-2008-B-6903-16	50 Seed
15	CIMMYTMA 13566	252691	W.S.T. 85-2	TL-2008-B-6903-18	50 Seed
16	CIMMYTMA 13562	252687	TEOSIN 62-?	BA-2006-U-2903-3	50 Seed
17	CIMMYTMA 13557	252682	TEOSIN 62-87	TL-2008-B-6903-17	25 Seed
18	Sirsa		Local cultivar	India	

*<http://mgb.cimmyt.org/gringlobal/search>

6.3.5 Recording of yield components

The eighteen accessions were assessed primarily for herbage production. The herbage production was evaluated after cutting at 30 days after sowing (DAS). Based on the survivability and performance of the 17 CYMMIT accessions at 30 DAS, the 7 accessions which had better survival and higher herbage yield plus Sirsa were harvested again for herbage at 60 DAS, and then left to produce seeds. The data collected for all accessions were plant height, tiller number, leaf number, Leaf Area Index (LAI), Herbage Yield (HY) and Dry Matter Yield (DMY) at 30 DAS, and additionally for the 7 better performing accessions flowering date, anthesis silking interval (ASI), cobs per

plant, number of ears per cob, number of seeds per ear, seed yield per plant and 100 seed weight. The difference between days to silking and days to anthesis of each entry was recorded as Anthesis-Silking Interval (ASI).

The yield components recorded for the second experiment were plant height, tiller number, leaf length, leaf width, days to silking, days to anthesis, seed yield per plant and 100 seed weight. Data for female flower length (cm), tassel length (cm), tassel width (cm), number of primary branches per tassel, number of secondary branches per tassel, number of cobs per plant, cob length (cm), cob width (cm), number of ears per cob, and number of seeds per ear were taken from the tagged plants from the middle of the row of each entry in each replication. The detailed methodology on recording yield components is as described in Chapter 2.

6.3.6 Statistical Analysis

Analyses of variance (ANOVA) was conducted to determine the effect of genotype on HY and DMY and associated yield components of the eighteen accessions in the first experiment and the seven Nepalese seed lots in the second experiment using General Linear Model (GLM) of Genstat 19th Edition (VSN International, 2019). Significantly different means of each level of factors considered were compared using Fisher's unprotected test of least significant difference (LSD). Multivariate analysis tool (cluster analysis) was used to identify the phenotypic diversity of the seven Nepalese seed lots using General Linear Model (GLM) of Genstat 19th Edition (VSN International, 2019). Twenty three traits were recorded which were standardised to mean 0 and standard deviation 1 for the cluster analysis using Microsoft Excel.

6.4 Results

6.4.1 Establishment of the 18 teosinte accessions

The emergence of the seedlings was recorded beginning at 5 DAS and continuing until 15 DAS. Days to first emergence ranged from 5.0 (Accession 1) to 7.4 (Accession 15). At 15 DAS, Sirsa had the highest emergence (80%), but this did not differ significantly from that of accessions 2,4-8, 10-13, 16 and 17 (Table 6.3). The lowest emergence (40%) was for accession 3 and 9, but this was only significantly lower than that for accession 11 and Sirsa. At 30 DAS, plant survival ranged from 32% (accessions 1 and 9) to 72 % (Sirsa), with survival for the other accessions ranging from 44-68 % (Table 6.3). Further plant losses occurred between 30 and 60 DAS. Accession 13 had the highest survival (64%), but this did not differ from all the other accessions except 1, 6, 9, and 17. Accession 9 had the lowest survival (18%), but this did not differ from that of accessions 1-3, 6-8, 12 and 14-17 (Table 6.3).

Table 6.3 Establishment and survival of 18 accessions of teosinte in Nepal

Accessions	Days to first emergence	Emergence % At 15 DAS	Survival % at 30 DAS	Survival % at 60 DAS
CIMMYTMA 29740 (1)	5.0c	48bc	32c	24cd
CIMMYTMA 29737(2)	6.2abc	64abc	44abc	32abcd
CIMMYTMA 29736(3)	7.0ab	40c	48abc	32abcd
CIMMYTMA 29735(4)	6.4ab	60abc	68ab	52abc
CIMMYTMA 29733(5)	6.6ab	52abc	52abc	52abc
CIMMYTMA 29732(6)	6.0bc	64abc	56abc	28bcd
CIMMYTMA 29730(7)	6.6ab	68abc	56abc	48abcd
CIMMYTMA 29728(8)	6.2abc	52abc	52abc	48abcd
CIMMYTMA 29726(9)	6.4aba	40c	32c	18d
CIMMYTMA 29723(10)	6.4b	52abc	64abc	52abc
CIMMYTMA 29722(11)	5.8bc	72ab	64abc	60ab
CIMMYTMA 29719(12)	6.4ab	68abc	64abc	40abcd
CIMMYTMA 13576(13)	6.0bc	64abc	68ab	64a
CIMMYTMA 13570(14)	6.4ab	44bc	52abc	40abcd
CIMMYTMA 13566(15)	7.4a	48bc	36bc	36abcd
CIMMYTMA 13562(16)	6.6ab	52abc	56abc	32abcd
CIMMYTMA 13557(17)	6.4ab	56abc	44abc	24cd
Sirsa(18)	7.0ab	80a	72a	56abc
Level of significance	0.258	0.356	0.422	0.201
LSD (5%)	1.3	30	34	33
CV (%)	16.5	42	50	64

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($P < 0.05$).

6.4.2 Vegetative performance and herbage yield of the 18 teosinte accessions at 30 DAS

The 18 accessions of teosinte were assessed for their vegetative production. In this study the HY was taken at 30 DAS, which is earlier than the traditional practice of taking the first harvest at 45 DAS. The idea was to have more time to observe the regeneration capacity of the accessions as well as early herbage availability (Table 6.4). At 30 DAS, the tallest plants were accessions 5(46.4 cm), 12(49.5cm) and 16 (48.3cm) but these did not differ significantly from accessions 1, 2, 4, 6-11, 13 and 18 (40.1cm to 44.8cm). The shortest plant was accession 17(27.6cm), but this did not differ from accessions 3-4, 11, 13-15 and 18. Highest tiller number per plant was recorded for accession 12(1.9) and the lowest was recorded from accessions 3 and 14 (1), but this was only significantly lower than that for accessions 5 and 8. Greatest LAI was recorded for accession 8 and the least was recorded for accessions 3 and 14, which was significantly lower than 2, 4, 6, 7, 10, 13 and 17. Accession 1 had the highest leaf number and the lowest was recorded for accessions 3

and 14. Sirsa produced the highest HY per plant but it did not differ from accessions 12, 13, 16 and 17. The lowest HY per plant was recorded from accession 3, but it did not differ from accessions 2, 4, 6, 7, 14 and 15. Similarly, Sirsa had the highest DMY per plant however it did not vary from accessions 8-13 and 16. Lowest DMY per plant was recorded from accession 3, but it did not differ from that of 1 and 14-15 (Table 6.4).

Table 6.4 Vegetative performance and herbage yield of 18 teosinte accessions at 30 DAS

Accessions	Plant height (cm)	Tillers per plant	LAI	Leaves per plant	HY/plant (g)	DM/plant (g)
CIMMYTMA 29740 (1)	44.1ab	1.6abc	0.6abc	6.1a	7.7cdef	0.3fg1
CIMMYTMA 29737(2)	44.3ab	1.4abc	0.5bc	5.5ab	7.4defg	0.6cdef
CIMMYTMA 29736(3)	32.2bc	1.0c	0.1c	5.0ab	3.2g	0.1g
CIMMYTMA 29735(4)	38.3abc	1.4abc	0.2bc	5.7ab	6.5defg	0.5cdef
CIMMYTMA 29733(5)	48.4a	1.8ab	0.4ab	5.4ab	8.3cdef	0.5cdef
CIMMYTMA 29732(6)	42.6ab	1.2bc	0.2bc	4.9b	7.4defg	0.7bcdef
CIMMYTMA 29730(7)	42.4ab	1.2bc	0.3bc	5.1ab	7.5defg	0.6cdef
CIMMYTMA 29728(8)	44.8ab	1.8ab	1.2a	3.8c	8.7bcde	0.7abcde
CIMMYTMA 29726(9)	42.4ab	1.2bc	0.6abc	5.2ab	8.1cdef	0.7abcd
CIMMYTMA 29723(10)	41.9ab	1.2bc	0.2bc	5.4ab	9.2bcde	0.8abc
CIMMYTMA 29722(11)	40.7abc	1.4abc	0.8ab	5.5ab	8.4cde	0.7abcd
CIMMYTMA 29719(12)	49.5a	1.9a	0.7abc	5.5ab	12.0abc	0.8abcd
CIMMYTMA 13576(13)	40.3abc	1.6abc	0.5bc	5.5ab	11.0abcd	0.9ab
CIMMYTMA 13570(14)	33.3bc	1.0c	0.1c	4.9bc	5.3efg	0.4defg
CIMMYTMA 13566(15)	33.2bc	1.4abc	0.2bc	5.6ab	4.0fg	0.4efg
CIMMYTMA 13562(16)	48.3a	1.6abc	0.8ab	5.9ab	12.8ab	1.0ab
CIMMYTMA 13557(17)	27.6c	1.2bc	0.1bc	5.3ab	9.9abcd	0.5cdef
Sirsa(18)	39.1abc	1.4abc	0.6abc	5.6ab	13.9a	1.0a
P value	0.321	0.726	0.726	0.377	<.001	<.001
LSD (5%)	7.7	0.7	0.4	1.1	4.4	0.189
CV%	28.4	60.3	108.6	26.8	41.3	43.7

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($P < 0.05$).

6.4.3 Vegetative performance and herbage yield of the 8 teosinte accessions at 60 DAS

The plants after the first harvest were allowed to regenerate for a further 30 days, after which the second harvest was taken. Based on the survival at 60 DAS (Table 6.3) and herbage yield (Table 6.4), the 7 best accessions plus Sirsa were selected for the 60 DAS data collection (Table 6.5). Plant height varied significantly among the genotypes ($P < 0.005$). Accession 12 had the greatest plant height, but this did not differ significantly from accession 5. The lowest plant

height was recorded for accession 17 while the other accessions were in between. Similarly, tillers per plant also varied significantly among the accessions ($P < 0.05$), where maximum tillers were recorded for accession 12 which did not significantly vary from accessions 5, 8, 11 and 13. Accession 17 produced the lowest tiller number but it did not vary significantly from accessions 7 and 18. Greatest LAI was observed for accession 5 and 8 but it did not differ from 7, 12, 13 and 18. Lowest LAI was from accession 11 and 17 but it did not differ from that of 7, 12, 13 and 18. No significant differences was recorded for leaves per plant among the accessions. HY per plant varied significantly among the accessions ($P < 0.005$). The highest HY was recorded from accession 12 but it did not vary significantly from accession 7. Accession 13 produced the lowest HY per plant but it did not differ significantly from that of 8, 17 and 18 and the other accessions were in between. Similarly significant variation ($P < 0.05$) occurred between the accessions for DMY per plant where accession 12 produced the highest DMY per plant but it did not differ from that of 7 and 11. The lowest DMY per plant was recorded from accessions 13 and 17.

Table 6.5 Vegetative performance and herbage yield of 8 teosinte accessions at 60 DAS

Accessions	Plant height (cm)	Tillers per plant	LAI	Leaves per plant	HY/plant (g)	DMY/plant (g)
T5	88.7ab	2.2ab	4.0a	9.9a	78.6bc	7.5bc
T7	81.0b	1.5bcd	3.0ab	9.5a	98.9ab	11.7ab
T8	72.8b	1.9abc	4.0a	10.5a	58.9bcd	7.6bc
T11	84.6b	2.0abc	2.3b	8.8a	83.7b	9.2ab
T12	110.1a	3.0a	3.5ab	8.3a	146.7a	14.7a
T13	75.6b	2.3ab	2.8ab	9.9a	17.3d	2.6c
T17	45.5c	0.8d	2.1b	7.2a	23.5cd	2.3c
T18 (Sirsa)	85.9b	1.0cd	3.3ab	9.9a	55.9bcd	6.1bc
P value	0.003	0.009	0.067	0.551	0.012	0.014
LSD (5%)	24.13	0.842	31.5	3.43	58.3	6
CV%	22.6	42.2	32.6	27.9	57.9	56

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($P < 0.05$)

6.4.4 Seed yield and yield components of 8 teosinte accessions

Plant growth parameters were again recorded at the seed harvest time for all 8 accessions (Table 6.6). There was no significant effect of accessions on plant height. Accessions 12 and 13 had the greatest numbers of tillers ($P < 0.05$), but did not differ significantly from accessions 5 and 8. The lowest tiller number was recorded for accessions 17 and 18 but this did not vary significantly from that of 7, 8 and 11. The greatest LAI was recorded for accession 5 and 8 but this did not vary from 7, 12, and 13. The lowest LAI was recorded from accessions 11 and 17 but this did not differ from

that of 7, 12, 13 and 18. Accession 5 had the earliest flowering among all accessions, which was followed by accessions 12 and 8. Accessions 11, 13, 7 had intermediate flowering and accessions 17 and Sirsa were the late flowering types ($P < 0.001$). Accession 5 began flowering 35 days earlier than Sirsa. There was a similar trend for silking among the eight accessions ($P < 0.001$). The shortest ASI was recorded for accession 8 and the longest ASI was recorded for Sirsa; the others were in between. Accession 5 produced the greatest number of cobs per plant but this did not vary from accessions 7, 8, 11, 13 and 18. The number of ears per cob varied significantly ($P = 0.001$) among accessions, where accession 8 produced the highest number of ears per cob but this did not vary significantly from accessions 5, 7 and 13. Accessions 12 and 17 produced the lowest number of ears per cob but this did not differ significantly from accession 11. The greatest number of seeds per ear was recorded from accession 7 and the lowest number of seeds per ear was recorded from accession 17. Both accessions did not differ significantly from accessions 5, 8, 11, 12, 13 and 18. Seed yield per plant was highest for accession 5 and the lowest seed yield per plant was recorded from accessions 8, 12 and 17 but these did not differ from accessions 7, 11, 13 and 18. Likewise, the greatest 100 seed weight was recorded from accession 5 which was followed by accessions 7, 13 and 18 respectively. The lowest 100 seed weight was recorded for accessions 8, 11, 12 and 17 (Table 6.6). Figure 6.2 represents the seed harvested from 17 accessions from CIMMYT, Mexico and local Sirsa from the research plots.



Figure 6.2 Seeds harvested from the 17 accessions from CIMMYT Mexico, and one local genotype

Table 6.6 Seed yield and yield components of 8 teosinte accessions

Genotypes	Plant Height (cm)	Tillers per plant	LAI	Days to tasselling	Days to silking	ASI	Cobs per plant	Ears per cob	Seeds per ear	Seed yield / plant (g)	Seed yield /sq. m (g)	100 seed weight (g)
T5	226a	2.6ab	4.2a	84a	89a	5ab	10.5a	7.4ab	5.8ab	40.3a	18.7a	12.2a
T7	232a	1.5bc	2.9ab	110f	115f	5ab	8.9abc	6.1abc	6.9a	20.1ab	9.6ab	9.1c
T8	290a	2.0abc	4.0a	91c	95c	4a	8.4abc	8.1a	6.6ab	12.7b	5.9b	8.2d
T11	224a	2.0bc	2.3b	94d	98d	5ab	10.3ab	4.4cd	6.8ab	18.3ab	8.5ab	8.0d
T12	256a	3.0a	3.4ab	88b	92b	4ab	5.4bc	3.4d	5.9ab	13.4b	6.2b	8.4d
T13	261a	2.8a	2.8ab	97e	102e	5ab	8.1abc	7.4ab	6.3ab	23.7ab	11.0ab	9.7c
T17	268a	1.0c	2.0b	119g	125h	5ab	4.7c	3.4d	5.3b	4.7b	2.1b	7.7d
T18	274a	1.0c	3.3ab	120g	126g	6b	7.2abc	5.8bc	5.8ab	26.4ab	12.2ab	10.7b
P value	0.616	0.011	0.120	<0.001	<0.001	0.313	0.21	0.001	0.314	0.135	0.135	<0.001
LSD (5 %)	83	0.81	1.5	2	2	1	5.0	2.1	1.5	23.2	10.8	0.7
CV %	22	38.3	32.6	1	1	19	42.6	23.7	15.9	78.0	77.9	5.0

Note: LSD = Least Significant Difference; CV = Coefficient of Variation; Lettering has been assigned using the unrestricted LSD procedure; means with no letters in common (in the same column) are significantly different at the 5% level ($P < 0.05$); g = gram; cm = centimetre. ASI= Anthesis Silking Interval

6.4.5 Analysis of variance (ANOVA) for yield related components in seven teosinte genotypes

The ANOVA (Table 6.7) reflects the descriptive statistics for 7 genotypes of teosinte. There were significant differences among some genotypes for number of tillers, number of leaves, days to 50 % flag leaf emergence, days to 50% silking, days to 50% anthesis, ASI, tassel width, number of primary branches per tassel, number of secondary branches per tassel, number of cobs per plant, number of ears per cob, number of seeds per ear, seed yield and 100 seed weight. The coefficient of variation (CV) ranged from 7.5-22.1% for the studied traits. The largest CVs were for number of tillers (22.1%), cob width (19.7%), anthesis-silking interval (19.5%), tassel length (19.5%), number of secondary branches/tassel (18.0%), and seed yield (19.9%) while there was less variation in days to 50% anthesis (7.5%) and silking (7.5%). The remaining traits showed intermediate levels of variation (Table 6.7). Figure 6.4 represents the seed harvested from seven genotypes.

6.4.6 Cluster analysis of seven teosinte genotypes

The cluster analysis of vegetative and reproductive traits (22 traits) of the seven teosinte seed lots collected across the southern and central region of Nepal grouped the genotypes into two main clusters (Figure 6.3). The teosinte from Makwanpur belonged to cluster I and those from Chitwan, Bara, Sarlahi, Mohattari, Tikapur and Gaughet belonged to cluster II. Thus it can be said that two genotypes of teosinte exist in Nepal, cluster I from the lower hills of the country and the cluster II from the Terai (eastern and western) (Figure 6.3). Cluster II differed from cluster I in terms of earlier flag leaf emergence, shorter ASI, and higher number of primary and secondary tassel branches. The number of ears per cob, number of seeds per ear and seed yield per plant for cluster I was lower than that of the other genotypes.

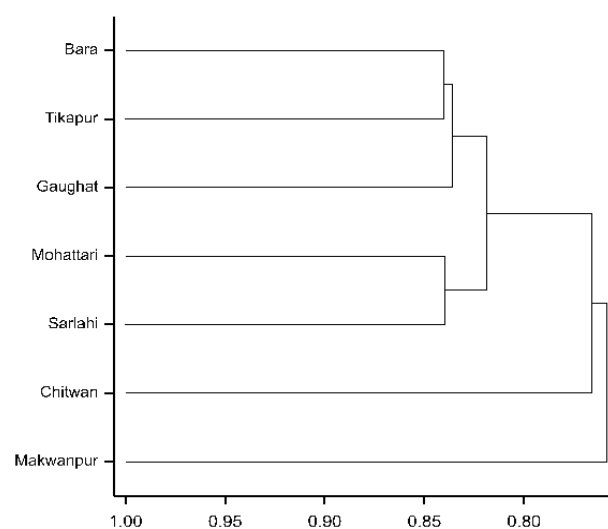


Figure 6.3 Grouping of 7 teosinte genotypes considering 22 agronomic traits

Table 6.7 Analysis of variance for yield components in seven teosinte genotypes

Yield attributes	Bara	Chitwan	Gaughat	Makwanpur	Mohattari	Sarlahi	Tikapur	P value	LSD (5%)	CV%
Plant height (cm)	389.9a	396.8a	374a	399.0a	385.0a	371.1a	355.2a	0.645	54.11	9.6
No. of tillers	4.1bc	4.5bc	3.6c	5.0ab	3.7bc	6.4a	3.7bc	0.007	1.45	22.1
No. of leaves	107.6de	142.8bc	86.5e	153.6bc	163.6ab	127.1cd	192.7a	<0.001	30.27	14.6
Leaf length (cm)	116.2a	122.5a	115.6a	122.1a	115.7a	110.6a	111.6a	0.780	14.3	10.9
Leaf width (cm)	6.6a	7.1a	6.6a	7.0a	7.0a	6.7a	6.8a	0.903	1.1	11.1
Days to 50% flag leaf emergence	94.8abc	117.2a	106.0ab	78.0c	100.2abc	110.0ab	87.9bc	0.064	25.4	17.3
Days to 50% silking	102.5bc	121.7a	120.6a	112.5ab	118.1a	118.7a	96.4c	0.003	12.6	7.5
Days to 50% anthesis	108.7bc	128.5a	128.1a	118.5ab	126.1a	126.1a	103.6c	0.003	13.3	7.5
Anthesis-silking interval (ASI)	6.3ab	6.8ab	7.4ab	6.0a	8.1b	7.4ab	7.2ab	0.387	2.0	19.5
Female flower length (cm)	5.7a	6.0a	6.0a	5.4a	5.0a	5.0a	4.7a	0.29	1.4	16.8
Tassel length (cm)	39.4a	36.7a	41.0a	36.9a	40.9a	31.9a	30.8a	0.308	8.1	19.5
Tassel width (cm)	20.0bc	36.0a	23.9b	17.8bc	23.1b	32.1a	16.5c	<0.001	6.3	4.2
Tassel internode length (cm)	19.0a	20.0a	21.6a	21.2a	20.5a	21.0a	18.7a	0.679	4.1	4.1
No. of primary branches/tassel	11.1b	13.0b	10.9b	16.2a	11.6b	13.7ab	11.1b	0.009	2.1	15.1
No. of secondary branches/tassel	11.5c	17.3b	16.0b	21.9a	18.0ab	19.0ab	9.1c	<0.001	4.3	18.0
No. of cobs/plant	9.7a	8.6ab	9.5a	9.0ab	8.5ab	8b	8.8ab	0.187	1.3	10.0
Cob length (cm)	14.5a	13.6a	14.7a	14.1a	15.0a	14.8a	15a	0.740	2.2	10.2
Cob width (cm)	2.1a	2.1a	2.0a	1.9a	1.9a	2.1a	2.1a	0.876	0.6	19.7
No. of ears/cob	11.5a	6.6d	9.8bc	5.4d	8.8c	9.8bc	10.8ab	<0.001	1.2	9.2
No. of seeds/ear	5.4ab	5.6ab	5.6ab	5.1b	5.9a	5.7ab	5.4ab	0.524	0.817	10.0
Seed yield (g/plant)	67.5a	46.0bc	66.0a	31.1c	43.2bc	49.7b	56.3ab	<0.001	15.2	19.9
100 seed weight (g)	11.3bc	14.8a	12.9ab	12.0bc	10.0c	11.3bc	11.9bc	0.024	2.6	14.7



Figure 6.4 Seed from different genotypes collected from different parts of Nepal

Discussion

6.4.7 Establishment of the 18 teosinte accessions

The time required for emergence and the uniformity of emergence are important factors for attaining highly productive potential in plants. Factors like soil moisture and depth of sowing also affect consistency in seed emergence (Finch-Savage, & Bassel, 2016). The time to emergence for the 18 teosinte accessions ranged from 5-7 days after sowing. Because only 25 or 50 seeds per accession were received from CYMMIT, it was not possible to test their quality (i.e. germination and vigour). This small difference in time to emergence and the difference in emergence percentage are more likely due to physiological differences between the seed lots as a result of diverse history of parental lines (Muchie & Fentie, 2016), which could have created differences in seed vigour for different seed lots which speeds up the germination for some accessions (Matthews, & Khajeh Hosseini, 2006). Higher emergence and survival for Sirsa may have been because it was a higher quality seed lot.

6.4.8 Vegetative performance and herbage yield of 18 teosinte accessions at 30 DAS

At 30 DAS, none of the 17 introduced accessions had produced more herbage per plant than Sirsa. However, herbage production by accession 12, 13, 16 and 17 did not differ from that of Sirsa, indicating perhaps adaptability to the environment at Chitwan. Herbage yield for the other accessions at 30 DAS was between 40-50 % lower than that of Sirsa, even though for many of them the lower population should have provided less interplant competition for resources. This result indicates these accessions were not well adapted to this environment because they produced smaller leaves. While leaf size was not directly measured, the fact that there were no

major differences in plant height, tillers per plant or leaves per plant compared to Sirsa, smaller leaf size is the only explanation for the lower herbage production. This result is in line with the findings of Sinclair (1994) who reported that plant growth is determined by individual organ development rate and its growth rate at different morphological stages. However, Amodu (2014) reported that variation in the yield components between the accessions depends on the variety, and environmental conditions.

6.4.9 Vegetative performance and herbage yield of 8 teosinte accessions at 60 DAS

By 60 DAS the herbage production comparison with Sirsa had changed. Accession T12 had triple the HY per plant of Sirsa while accession 17 had nearly double the yield. Similar increases were obtained for DMY per plant. Accession 12 was taller than Sirsa and had two more tillers per plant, but the leaves per plant were similar, once again indicating a large leaf size. Taller plants with more leaves enhance photosynthetic activity and increase dry matter production through greater sunlight capture (Duncan, 1971; Bingham & Lupton, 1987). Higher leaf size allows increased radiation interception capacity leading to increased HY (Elings, 2000; Mannetje, 1999; Sadras et al., 2016). Similar results have been reported by Khanal, Devkota, Tiwari, & Gorkhali (2020) in teosinte; Jing, Christensen, Sorensen, Christensen, and Rubaek (2019) in silage maize and Faji, (2021) in maize where plant height and tiller density influenced HY. Accession T7 did not differ from Sirsa for vegetative characters, and so larger leaves must have also explained the greater yield. This vegetative growth response from Accessions T12 and T7 indicates they were better able to recover from cutting at 30 DAS than all the other accessions and Sirsa. Conversely the latter group could not compensate for the loss of vegetative tissue at 30 DAS within the 30 days until the next harvest. Behaeghe (1986) and Hilbert, Swift, Detling, and Dyer (1981) noted that plant recovery is dependent on the length of time for regeneration, and that slow recovery after cutting may also trigger plant death. An inability to grow rapidly by these accessions after cutting might be because photosynthates were translocated to defoliated sites for tissue repair rather than new vegetative growth (McNaughton, 1979). Therefore a long recovery time is more beneficial during early defoliation and vice versa. However the variation among the yield attributes among the accessions might be because these seven accessions came from different origins and comprise a wide diversity of phenologies with different regeneration capacity in this environment.

6.4.10 Seed yield and yield components of eight teosinte accessions

Sirsa took 120 days to reach tasselling, and all accessions except T17 took less time with T5 took five weeks less. This accession also produced around 50 % more seeds than Sirsa because it had more cobs, ears and seeds as well as a higher 100 seed weight. Higher seed yield and higher 100 seed weight in accession 5 as compared to Sirsa might be because of higher biomass partitioning to seeds due to better ability to capture more photosynthates (Ali, Ahsan, Mustafa, & Ejaz-ul-Hasan, 2013; Kgasago, 2007). Similar results have been reported by Kakhki, (2019) and Kumar and Patel (2017) where higher yield components had a better source-sink relationship. Accessions T7, T11 and T13 produced the same seed yield as Sirsa and all three took significantly less time to reach tasselling than Sirsa. Thus the objective of finding one or more accessions which were earlier maturing than Sirsa was achieved. The long growing season for Sirsa in the lower altitude regions of Nepal restricts the ability of farmers to grow winter vegetables and this makes them reluctant to grow teosinte (Pariyar, personal communication, 2018). Having an early accession alternative would be of benefit to farmers.

While the focus was on seed production, vegetative production is most important for farmers who need to feed their livestock. A more rapid transfer to reproductive growth may limit vegetative production (Bodner, Nakhforoosh, & Kaul, 2015; Shavrukov et al., 2017). Among the introduced accessions, T5 was the best for seed (flowered early and had higher seed yield), and had a forage yield similar to that of Sirsa. T12 had the highest forage yield at 60 DAS but a low seed yield, while T7 was similar to Sirsa for both forage and seed yield, but only marginally earlier (10 days) to flower. These results, though only preliminary, and from a limited data set, suggest that at least three of these introduced accessions should be further evaluated for use in the Terai region of Nepal.

6.4.11 Phenotypic variations among the seven teosinte genotypes

High morphological (phenotypic) variability among genotypes is due to differences in capacity to utilize the resources in a particular area. Traits of domesticated plants deviate due to the influence of geography and climate and therefore agronomic factors can be phenotypically distinct (Iltis & Doebley, 1980; Chacon, Pickersgill, & Debouck, 2005; Brown, Myles & Kresovich, 2011). Phenotypic characterization allows the selection of useful traits for breeding purposes, but they are subject to environmental variations (Stanley, 2020).

In the present study, phenotypic analysis showed a difference among the seven genotypes with high variability for tillers, cob width, anthesis-silking interval, tassel length, number of secondary branches per tassel, silking, days to 50% anthesis, tassel width, number of primary branches per

tassel, number of secondary branches per tassel, number of ears per cob, seed yield 100 and seed weight. These variations in the morphological traits between the genotypes might have occurred because of differences in original introductions to Nepal, or to the influence of the environment where each genotype was grown over the years, along with natural and human selection. Ngugi and Maswili (2010) reported that morphological variation is an easy indicator to record genetic diversity in maize. This result is in line with the finding of Yucel, Hizli, Firincioglu, Cil, and Anlarsal (2009) in common vetch and Asfaw (1989) in barley. Ngugi and Maswili (2010) reported that a higher coefficient of variation for traits indicates large environmental influences on the phenotype which might result in weaker correlations between some of the traits. The present results are in line with the finding of Ndiso, Mugo, Kibe, and Pathaka (2013), Marker and Krupakar (2009), Rakszegi et al. (2010), Ranawat et al. (2013) and Sharma, Prasanna, and Ramesh (2010) who reported that phenotypic variation among maize genotypes indicated genetic variation. Morphological traits being used for identification and management of maize cultivars over the years has been reported by Camussi (1979) in Italy, Azar et al. (1997) in Canada and Louette and Smale (2000) in Mexico. The high variability for the flowering behaviour (ASI) of the seven teosinte genotypes could also be related to an adaptation to different environments (San Vicente, & Hallauer, 1993) because higher seed yield is related to early flowering and better plant and ear conformations (Oyekunle, & Badu-Apraku, 2017). Sharma et al. (2010) also reported that variations in maize genotypes can be differentiated by the differences in seed yield. The high coefficient of variation for tiller numbers indicates higher level of trait dispersion around the mean and the lower coefficient of variation for days to 50% anthesis relates to more precise estimate of the trait.

6.4.12 Cluster analysis of seven teosinte genotypes grown in Nepal

Genotypes having more than one desirable trait and belonging to different clusters represent genetic diversity (Bhusal et al., 2016). The clustering of genotypes into two groups according to their phenotypic representation clearly showed that there are two distinct lines of teosinte in Nepal. Areas at similar altitudes and with similar agro-ecology grouped together to form a teosinte cluster. The clustering pattern also showed some formal relationship between geographical diversity and genotypes because cluster I was from the lower hill region of central Nepal while cluster II was from the lower Terai region of eastern and western Nepal towards the south. Variability among the seed lots from mid-altitude and low altitude sites is not surprising, because the major entry of teosinte seed lots to Nepal was through the south where there is an open border with India. The close relationship among the genotypes cultivated across the southern region of the country might be because of the continuous cultivation of teosinte for a

very long time in those areas. These seeds are basically preserved by the farmers who either keep seeds from their previous harvest every year, obtain seed from the local community or buy seed from the local market, which increases the seed flow. Similar result was reported by Desmae, Jordan, and Godwin (2016) in sorghum. Further, farmer's preference for the Nepalese teosinte seed rather than the Indian seeds is possibly because the same Nepalese genotype has been cultivated for years. Therefore, seed exchange between the farmers might have attributed to regular gene flow across the southern belt resulting in weak differentiation, which is similar to the findings of Desmae, Jordan, and Godwin (2016) in sorghum. The presence of genotypes across the southern belt might also be explained by the warm and dry weather conditions that increase the frequency of specific phenotypes adapted to the prevailing climatic and edaphic conditions. Similar results have been reported by Jaradat, Shahid, and Maskri (2004) and Demisse and Bjornstad (1996) in barley. The latter authors reported that in Ethiopia, stress inclined towards homogenous genotypes rather than a higher degree of variation. This result is also in line with the findings of Grenier et al. (2001), who reported that ecological adaptiveness of morphological traits are good indicators of local differentiation, genetic differences or ecotypes that can be used to classify the phenotypic diversity. In contrast to the present result, Lahane et al. (2016) reported a non-formal relationship between geographical diversity and genetic diversity.

However, two theories can be generated for cluster I which is grown at higher elevation as compared to cluster II. Higher elevation and the cooler climate might have resulted in changes in the phenotypic behaviour of teosinte in cluster I as a result of geographical isolation. The importance of climatic conditions noticed in the structuring of crop diversity has been highlighted by several authors. A similar result was reported by Desmae, Jordan, & Godwin (2016) in sorghum where altitude and the climate influenced the phenotypic behaviour as a result of physical distances and climatic factors which executed physical and adaptive barriers to gene flow between agro-ecologically different and distant areas. This can also be explained by phenotypic plasticity as reported by Hausmann et al. (2012), where genotypes grown under different rainfall conditions acclimatize themselves towards the different environmental conditions by changing definite characteristics like the life cycle, HY, tillering, plant height etc. This result is in line with the findings of Manzelli, Benedettelli, and Vecchio (2005) who reported that the genotypic structures of landraces are affected by the local environment due to the close relationship between the agro-ecological conditions and morphological variation of the genetic material. A positive and significant association of diversity index with altitude and low temperature has been reported by Abay, Bjornstad, and Smale (2009) in barley. The second theory is that this is a different original genotype that was brought from somewhere else, probably India, and has been

cultivated in Makwanpur over many years. Because of a lack of regional seed exchange from this site, which is isolated from the southern corridor, this seed lot has continuously been grown in the same place. The south is a major teosinte growing area of the country and as it would have been easier to access teosinte seeds from the south, because of the road network, farmers in the eastern region prefer to get the seed source from the southern corridor rather than Makwanpur. This would also have contributed to the isolation of this genotype. Several other authors have also emphasized the significance of climatic conditions in shaping the crop. Asare, Tetteh, Twumasi, Adade, and Akromah, (2016) reported that within and between population variations could be a result of a large rate of gene flow between higher and lower altitude populations somehow restricted between geographically separate high land genotypes.

6.5 Conclusions

In this field trial, the performances of 17 introduced teosinte accessions and the local cultivar Sirsa were evaluated and the performance of seven seed lots of teosinte genotypes grown across Nepal was also evaluated in a separate trial.

- Out of 7 accessions, accessions 5, 7, and 12 showed either a better or comparable performances in terms of herbage yield, dry matter yield, earliness and seed yield than Sirsa. Therefore these accessions can be recommended for further evaluations in Nepal to develop high yielding teosinte cultivars. However more research is required as this result is based on the performance of 25 seeds and a single year trial at one site.
- There is considerable variability among the teosinte genotypes cultivated in Nepal. The hierarchical cluster analysis gave two distinct groups of teosinte. However, further study is necessary to confirm whether or not these two clusters have marked variations because of environmental influences on their morphological features.

Chapter 7

Outcomes, general discussion and future research

7.1 Overview of the study

Agriculture is a mainstay of the Nepalese economy. Around 60 % of the population are engaged in Agriculture (Nepal MoALD, 2020). Crops and livestock contribute 32.5% to the Gross Domestic Product (GDP); crops alone contributes 23 % to Agriculture Growth Domestic Product (AGDP) and livestock contributes 12%. Of the latter, milk contributes 33 % of the livestock GDP (MoALD, 2020). There is a feed balance (TDN) deficit of 20% (in 2016/17) at the national level which is basically due to the small area under herbage cropping, limited availability of high yielding herbage varieties, lack of quality seeds, poor herbage quality, and a changing cropping pattern in favour of cash crops (Singh & Singh, 2019). Herbage plays an important role in shaping the dairy industry in Nepal. Over recent years, demand for herbage has increased, because of the growing dairy industry and livestock commercialisation. The strength of animal health and their productivity lies in the availability of quality herbage (Somashekar, Shekara, Kalyanamurthy, & Lohithaswa, 2015). It is also vital to help in fulfilling the nutritional requirement of the livestock population, and therefore reduce supplement feed consumption, which ultimately will lower the cost of meat and milk production (SQCC, 2013). Herbage seed itself is an economic commodity that is linked with the financial development of farmers in Nepal and an important component for higher herbage yield. The area under teosinte (*Euchlaena mexicana*) cultivation in Nepal is 27,232 ha, which is the highest of all the herbage crops cultivated, and the seed production is 870 tonnes. A seed deficit of 220 tonnes per annum still prevails, which is 25% of the total teosinte seed requirement in the country (NPAFC, 2018). Along with other types of seeds in Nepal, the herbage seed system needs formalization as nearly 90% of the total seed requirements are traded through an informal system (SQCC, 2017). Therefore, it is crucial to develop a seed system that would provide improved seed and varieties to farmers at the required time throughout cropping seasons (Kansiime & Mastenbroek 2016).

Teosinte is an interesting cereal forage at the global level, both from socio-economic and environmental perspectives. However, this crop still does not occupy the place it merits in the world cropping systems. Being the progenitor of maize and having several genotypes with different maturity groups available, it is a possible source to develop new forage and grain maize

varieties, in the context of changing climate. This crop has been little studied and hence is poorly understood, hindering exploration of suitable niche areas for teosinte cultivation. The outcome of this study should help in boosting returns for Nepalese farmers. Increasing herbage yield through better management would allow replacement of concentrates and straw currently widely used as livestock feed and reduce the cost of milk production (Hampton et al., 2019). However, there is a shortage of quality teosinte seed in the country (Pariyar and Shrestha, 2016). Thus, farmers are forced to import poor quality seed from India. Therefore, the seed production technology developed in this study could be a very innovative approach that can be implemented at the provincial level through which the yield and quality of teosinte seeds in the country could be improved. Further niches for teosinte herbage and seed production could also be identified. Early maturing varieties with multicut nature identified from this study could be an asset for farmers. The economic study (see 7.2.6) clearly shows the profitability of growing teosinte for herbage and seeds (B: C ratio more than 5). There is no doubt, if teosinte is grown under good agronomic management and climatic conditions, it will be a model crop for economic stability of farmers in Nepal.

The main objective of this study was to examine seed yield and seed quality of teosinte in Nepal and to determine the impact of herbage cutting on seed production. This involved determining the major effects of sowing date, seed rate and cutting management, along with assessing harvesting time, studying seed development, seed quality assessment, and a diversity and economic study of teosinte in the Terai of Nepal. The lack of previous studies on this crop meant that there was very little information published on seed yield and quality. Moreover, the herbage yield, seed yield and seed quality cannot be compared directly with that of maize because of different physiology of these crops. As a complete discussion is included in each of the experimental chapters, this chapter only presents a brief summary of results, an overall discussion and future research directions.

7.2 Summary of outcomes and discussion

Teosinte, a wild ancestor of maize, is an annual cereal herbage crop with vigorous growth and generous tillering ability (Khanal, Devkota, Tiwari & Gorkhali, 2021; Sharma, 2018; Singh, & Dutta, 2021). It belongs to the Poaceae family and is a short day plant (Adhikari, Joshi, Kumar, & Singh, 2021; Wigge, & Jaeger, 2020) with indeterminate flowering. It is a tropical plant and produces high-quality herbage from multi-cuts in mid to late summer and can also produce a large quantity of seeds. Sowing date, seed rate and cutting management can all have an impact on herbage yield of teosinte. Teosinte seeds were sown in late spring/summer (30 March, 30 April, 30 May and 30

June in 2017 and 30 March, 30 April and 30 May in 2018) with an intention that the 30 days difference between sowings would provide different growing environments. Four seed rates (20, 40, 60 and 80) kg/ha were used to give different plant densities and two cutting managements, were used to study the regeneration potential and impacts on seed production. The first cutting was taken at 45 DAS and second cutting was taken at 75 DAS. Cutting was included as a treatment because Nepalese farmers traditionally take herbage cuts from their teosinte seed crops.

7.2.1 Effect of sowing dates, seed rates, and cutting management on herbage yield from a teosinte seed crop

The optimum sowing date makes the best use of climatic influences such as temperature, precipitation, and day length (Sawan, 2018). Plant density is also an important crop management tool to fully utilize the environmental factors, nutrients, and especially light, that has a positive effect on yield because photosynthetic efficiency and growth are significantly affected by the canopy structure and the vertical distribution of light (Rahmani et al., 2016). Cutting influences the carbohydrate reserves in plants and the utilization of these reserves for regeneration (Imoro, Kingsley, & Abukari, 2021).

In this study, the highest total HY and DMY was obtained from the April 30 sowing at the 60 kg ha⁻¹ seed rate and twice cut plants. At this site the earlier sowing experienced drought which caused a delay in seedling emergence, while delayed sowing resulted in poor early vegetative growth due to excess rainfall. Higher temperature for the April sowing allowed the accumulation of a higher GDD over a longer growing period. This favoured better plant growth (height, tillers and number of leaves), and permitted greater carbohydrate synthesis to produce more HY. A significant correlation also existed between plant height, tiller numbers, leaf number and LAI with the DMY. Seed rate is another factor which may affect the HY and DMY of teosinte. Higher HY and DMY were obtained from the 60 kg ha⁻¹ seed rate which can be explained by the higher vegetative yield components (number of tillers and leaves per square meter). This result was similar to that reported by Springer, Dewald, Sims, and Gillen (2003) in gamagrass and by Koireng et al. (2018) in maize. Khan, Shah, Khalil, and Karim, (2004) reported that higher DMY at higher plant density is due to higher solar radiation absorption allowing more photosynthesis due to the increased leaf area. Cutting frequency determines the quantity of herbage harvested and this depends on the regeneration potential of the crop under a cut and carry system. In this study the HY was higher for twice cut plants because HY was accumulation of the first and second cutting. However, this is a by-product from a crop grown for seed. The HY could be higher for a crop that is grown only for herbage production.

7.2.2 Assessing the influence of the environment on growth and yield of teosinte (*Euchlaena mexicana*)

As teosinte is a short day plant, it is sensitive to day length during its reproductive period (Stephenson et al., 2019; Troyer, 2001). Sowing date creates a different environment (photoperiod, temperature) that modifies the growth and expansion of the crop (Bhuiyan, Mondol, Bahaman, Alam, & Faisal, 2008). Both early and late sowing hampers plant growth because of a variation in temperature and precipitation (Wann, 1986), and differences in the thermal and radiative environments (Cirilo, & Andrade, 1994) during growth. Crop productivity is directly dependent on the environmental components; solar radiation, air temperature, growing degree days (GDD), precipitation and relative humidity (Dornbos, 2020; Hoogenboom, 2000). In Chapter 3, the effect of environmental components and GDD requirements (using a 10°C base temperature) for teosinte growth was studied. The temperature and GDD impacts were clearly evident in teosinte herbage yield where the April sown crops had higher HY due to higher GDD accumulation. Five critical growth stages for teosinte were defined during this study; emergence stage (GS1), vegetative stage (GS2), flowering stage (GS3), seed development stage (GS4) and seed maturity stage (GS5). The mean temperatures for the five critical growth stages were 26.2°C (GS1), 26.2°C (GS2), 24.1°C (GS3), 20.6°C (GS4) and 17.2°C (GS5). The mean GDD were 113°C, 134°C, 1632°C, 2218°C and 2330°C for GS1, GS2, GS3, GS4 and GS5 respectively. Based on these temperatures and phenology, the GDD accumulation and days from sowing for seed maturity were 3723°C (257d), 3297°C (234d), 2809°C (208d) and 2330°C (183d) for the March, April, May and June sowings respectively. The phenological days required from anthesis to physiological maturity of teosinte seed was 59-60 days.

7.2.3 Identifying the appropriate sowing dates and seed rates for maximizing seed production of Teosinte under different cutting regimes in Chitwan, Nepal

Being an indeterminate plant, teosinte flowering and thus seed development commences from the top to the bottom position of the inflorescence. Delay in sowing reduces seed yield and quality and harvesting early captures immature seeds, therefore seed harvesting time is critical for teosinte.

The early sowings on 30 March and 30 April produced the greatest seed yield because of better vegetative growth (plant height, tillers) and reproductive yield components (cobs per plant, ears per cob and seeds per ear). Similar effect of sowing date on plant height, LAI and DMY was reported by Von, Gehren, and Gansberger (2017). Lower seed yield from delayed sowing could be

due to reduced vegetative growth and also low seed filling rates (Alam, Ahmed, Nahar, Akter, & Uddin, 2020), where seed filling coincided with cooler autumn temperatures. No significant effect of seed rates was recorded in 2017 because of lodging of plants due to heavy rainfall and winds. In 2018 the two lower seed rates (20 kg ha^{-1} and 40 kg ha^{-1}) produced the greatest seed yield which can be explained by reduced interplant competition for resources. Cutting reduced the seed yield due to reduced photosynthates as a result of foliage removal (Patil & Merwade, 2016). In this study, uncut plants produced more seed than that of once and twice cut plants in 2018 but in 2017, seed yield from uncut and once cut plant did not differ from each other because in that season, there was rapid vegetative regrowth after the first cut at 45 DAS.

Seed shattering is a natural phenomenon, especially in wild and weedy plant species (Maity et al, 2021). It is chiefly controlled by genetics, but also significantly influenced by environment, management practices and their interaction, in the agro-ecosystems. Shattering is an undesirable character in domesticated crops and may lead to significant seed loss in commercial agriculture (Maity et al., 2021). However, some levels of shattering is preferred in pasture as a specific adaptation that guarantees self-seeding and pasture renewal (Dong & Wang, 2015). Harvesting teosinte seeds is very tricky because seeds mature from the top cobs towards the bottom and by the time seeds at bottom positioned cobs mature, seeds on the top positioned cobs are lost due to shattering. Early seed harvesting can result in immature, poorly germinating seeds that have low seed vigour (Fu, 2017) and harvesting late might result in deterioration of seed quality due to adverse weather condition (Henning, Jacob Junior, Mertz, & Peske, 2011). In this study, teosinte seeds from each sowing were hand harvested three times and seeds were weighed separately. Top positioned cobs of the plant produced the highest proportion of the seed yield (42.5%), followed by the middle positioned cobs (35%) and the bottom positioned cobs produced 22.5%. Seed harvested from the top and middle positioned cobs accounted for 77.5 % of the total, but that from the middle and bottom positioned cobs make up only 57% of the total. Therefore, it is recommended to harvest seeds from the upper and middle position cobs rather than waiting for seeds from the bottom positioned cobs to mature. For farmers in Nepal, time to harvest seeds is when 75 % of the seeds are mature, accepting that 25 % of the seed will be lost. They harvest teosinte seeds when the seeds inside the rows separate and the seed color changes to brown. It is desirable to harvest the seeds when the moisture content is around 25% SMC.

7.2.4 The effect of sowing date, seed rate and cutting management on seed quality of teosinte harvested on the basis of cob positions

Seed development is influenced by various environmental factors like water, light, temperature, soil nutrition and also seed position on the plant (Bareke, 2018). Seed development ceases at Physiological Maturity (PM) (Li et al., 2021). In the current study, teosinte seeds reached PM at 56 DAS and it required 617°C days to reach PM from anthesis. It required 14 days and 103°C days to reach harvest maturity from PM. Early sowing provides plenty of time for plant growth and seed development, but delay in sowing results in a shorter seed filling period which negatively influences seed development and quality (Deivasigamani & Swaminathan, 2018).

Sowing date affected the seed germination. Highest germination (91% in 2017 and 89% in 2018) was recorded for seeds sown in 30 March, with the plants subjected to longer period of growth. This is further enhanced by higher GDD accumulation for all the phenological stages of 30 March sown plants. Similarly, TSW was also higher for the 30 March and the 30 April sowings and it reduced with delay in sowing for a similar reason. Lower seed rates (20 kg ha⁻¹ and 40 kg ha⁻¹) had higher germination, 88% in 2017 and 87% in 2018, and higher TSW. Low plant density reduced the competition between the plants for resources resulting in better performance of individual plants. Higher plant density is not favourable for seed quality (Sahu, Tomar, & Nandeha, 2018) because of interplant competition for a fixed supply of resources (Postma, 2021). Cutting reduced the germination and TSW in teosinte because loss of foliage reduces the carbohydrate reserve in plants.

Cob position on the plant also affected the seed quality. Higher germination and TSW were recorded from seeds from the top positioned cobs. This can also be explained by competition, because more assimilates are available for the early developing seeds (i.e top cobs) (Georgieva, 2020; Ninh, Hiroshi, & Toru, 2007).

7.2.5 Evaluation of different genotypes of *Euchlaena mexicana* in Nepal for herbage and seed production

Teosinte has gained ready acceptance as a herbage crop in Nepal since it was introduced in 1967. However, only one certified cultivar (Sirsa) is currently available. It is high yielding but for seed production has a very long growth period that hampers the cultivation of winter crops. Therefore, this study was conducted to examine teosinte diversity and the production possibility of 17 teosinte accessions from CYMMIT, Mexico in Nepal. The main purpose was to identify if any accessions would perform better than Sirsa in terms of herbage and seed yield. Out of the 17 accessions, accessions 5, 7 and 12 out yielded Sirsa for herbage yield, seed yield and also were

earlier maturing. Accessions 5 flowered 35 days earlier than Sirsa and produced 50% more seed. Accession 7 was slightly earlier than Sirsa and had an almost similar seed yield and accession 12 had the highest herbage yield and was earlier by 32 days. This investigation was limited by the small number of seeds of each accession available, and the results were from one site and one season. Obtaining a further supply of seeds of these three accessions from CYMMIT will provide an opportunity for a further study and the eventual possibility of new cultivars of teosinte for Nepalese agriculture.

Hierarchical cluster analysis conducted on seven seed lots of teosinte collected from different regions of Nepal produced two clusters among these genotypes. Cluster I was from the lower hill region (Makwanpur) and cluster II (Sarlahi, Mohattari, Bara, Chitwan, Gaughat and Tikapur) was from the Terai regions of Nepal. However, this need further verification to confirm whether or not these two clusters have prominent differences because of environmental influences, or whether the major morphological variations could simply be because they were different when first introduced.

7.2.6 Gross margin of teosinte grown for herbage and seed yield

Feed and herbage costs constitute about 60-70% of the cost of milk production (Grover & Kumar, 2012). Without ensuring an adequate supply of quality herbage, the achievement of the desired growth of the Nepalese livestock sub-sector in the coming years looks difficult. Green herbage is one of the most important parts of sustainable agriculture for animals and the demand has been increasing for quality herbage and feed to increase milk yield (Jacob & Asokhan, 2020). The present study, therefore, was undertaken to estimate profitability of herbage yield in terms of milk yield, and seed income on the basis of gross margin and cost of cultivation of teosinte under different sowing dates, seed rates and cutting management.

The gross margin from teosinte cultivation is directly dependent on its herbage and seed yield. The gross margin was significantly higher for the March and the April sowings than for the May and (June) sowings. Gross margin was 326,314 and 385,807 NRs. in 2017 and 319,126 and 371,342 NRs. in 2018 for the March and the April sowings (Table 7.1). Similarly, in 2017, the lowest three seed rates had the highest gross margin and they did not differ significantly from each other. In 2018, the gross margin from the lowest two seed rates was significantly higher than two higher seed rates because of significantly higher income from seed. Similarly, the gross margin for uncut and one cut plants did not vary from each other and had higher gross margin than the twice cut plants in both years (Table 7.1). Higher gross margin is because of significantly higher seed yield from uncut and once cut plants than twice cut plants. Total cost was also low for

uncut and once cut plants than twice cut plants. For gross margin, a significant interaction occurred between sowing date and cutting management in 2017 ($P=0.008$) and 2018 ($P<0.001$) (Table A7.1 and A7.2). In general, the highest gross margins were for the March and the April sowing dates, with seed rates 20-60 kg ha^{-1} and none or one cut (Table A.7.1). There was no difference in the gross income from uncut and one cut plants because the income from milk for the latter was compensated for the reduction in income from seed. The detailed methodology on how the data in Table 7.1 were obtained is given in Appendix D.

Table 7.1 Effect means of sowing dates, seed rates and cutting management on the cost and income of teosinte in 2017 and 2018 at NCRP, Chitwan, Nepal (NRs. = Nepalese rupees, 1 NZ \$ =84.3NRs.)

Main effect means of:	Income from milk (Rs/ha)		Income from seed (Rs/ha)		Gross Return (RS/ha)		Total cost (Rs/ha)		Gross Margin (Rs/ha)		B:C	
	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Sowing date (SD)												
30-Mar	41,059b	33,898b	424,566a	417,059a	465,624a	450,479a	79,817a	79,137b	385,807a	371,342a	5.8a	5.7a
30-Apr	62,800a	65,711a	343,951a	335,977a	406,751a	401,687a	80,437a	82,561a	326,314a	319,126a	4.9a	4.8a
30-May	23,267c	33,898b	177,847b	177,968b	201,115b	211,866b	68,733b	73,593c	132,381b	138,274b	2.9b	2.9b
30-Jun	17,526d	-	99,208b	-	117314b	-	65,766	-	51,548b	-	0.8	-
Linear contrast p value	<.001	0.953	<.001	<.001	<.001	<.001	<.001	0.003	<.001	<.001	<.001	<.001
LSD (0.05)	5,013	18,047	101,127	94,512	90431	90,958	2,205	3,161	88,329	89,115	1.12	1.02
CV%	9	25	25	19	20	16	2	3	26	20	19	14
Seed rate (SR)												
20 kg ha^{-1}	36,614ab	36,125b	265,539a	382,243a	298,362ab	418,368a	70,141d	75,135c	228,221ab	343,233a	4a	5.5a
40 kg ha^{-1}	37,613a	42,101b	259,036ab	361,585a	300,455ab	403,686a	72,751c	78,056b	227,705ab	325,630a	3.9a	5.1a
60 kg ha^{-1}	35,403ab	48,643a	280,750a	304,939b	320,396a	353,581b	75,379b	80,223a	245,017a	273,358b	4.0a	4.4b
80 kg ha^{-1}	35,021b	50,502a	240,247b	192,573c	271,591b	243,076c	76,483a	80,307a	189,050b	162,768c	3.3b	3.0c
Linear contrast p value	0.065	<.001	0.065	<.001	0.343	<.001	<.001	<.001	0.343	<.001	0.012	<.001
LSD (0.05)	2,353	5,990	42659	47329	40280	46,880	1,022	1,383	39,310	45,795	0.45	0.5
CV%	9	16	23	18	19	16	2	2	25	20	16	14
Cutting Management (CM)												
No cut	-	-	310,198a	364,156a	310,198a	364,156a	68,979c	72,376c	241,219a	291,780a	4.3a	5.0a
One cut	29,763b	36,033b	272,494a	313,470b	317,139a	367,520a	75,371b	80,107b	241,768a	287,413a	4.0a	4.6b
Two cut	42,563a	52,652a	201,486b	253,379c	265,765b	332,357b	76,715a	82,808a	189,050b	249,549b	3.2b	3.9c
Linear contrast p value	<.001	<.001	<.001	<.001	0.035	0.002	<.001	<.001	0.011	0.002	<.001	<.001
LSD (0.05)	3,036	4,104	42,634	24,958	41161	26,880	1,167	1,281	40,047	25,896	0.5	0.3
CV%	16	22	46	20	39	19	5	4	51	23	35	15
Significance of interactions of linear contrasts (p value)												
SD(lin) x SR(lin)	0.514	0.067	0.332	0.820	0.188	0.133	0.28	0.638	0.283	0.663	0.128	0.739
SD(lin) x CM(lin)	0.004	<.001	0.071	<.001	<.001	0.438	0.005	<.001	0.008	<.001	0.027	<.001
SR(lin) x CM(lin)	0.147	0.998	0.827	0.632	0.408	0.021	0.747	0.291	0.758	0.327	0.956	0.044
SD(lin) x SR(lin) x CM(lin)	0.168	0.487	0.528	0.140	0.313	0.090	0.460	0.096	0.465	0.100	0.548	0.085

Note : For income from milk, the main effect means for the SD and SR are averages over all three cutting management treatments for comparability with other variables, even though the “no cut” treatments were excluded from the statistical analysis (as in Table A4).

7.3 Recommendations for future research

From this study, a lot more is now known about crop management impacts on teosinte seed yield and seed quality. The earlier sowing dates produced higher yield and quality. However, there were other aspects which were not included in this study and need further investigations.

- This was a seed production study which included the impacts of cutting for herbage at 45 and 75 DAS on seed yield. A separate herbage study should be conducted to include different cutting management at different growth stages and in different locations within Nepal. Herbage quality should also be assessed.
- The seed production study was conducted in only one location, and more locations or regions for teosinte seed production should be included for further study. This would help to identify the most suitable locations/regions for seed production. Similarly the GDD requirements for teosinte seed production for each location/region could be calculated.
- In this study, seed harvest was done from three different cob positions on the plant. A study on source sink relationships and competition for assimilates at the different seed position could be conducted.
- In this study only germination and thousand seed weight were tested. In future work seed vigour should also be assessed to better understand management and environmental impacts on seed quality.
- The seed development study was conducted only in one year at one site. This needs further locationwise verification.
- The diversity study used seed lots from seven locations of Nepal. Seed lots from other altitude/regions could be assessed for more than one year to further investigate teosinte diversity.
- More seeds from at least three of the teosinte accessions from CYMMIT should be imported and further evaluated. This investigation should be done at different teosinte growing locations of Nepal.
- A heat stress study could be done to understand the highest temperature tolerance capacity of teosinte plant as well as heat stress impacts on seed quality. This would help in breeding high heat tolerant maize varieties by hybridisation with teosinte that could be useful under the context of climate change.

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Appendix A for Chapter 2

Appendix A2.1 Reference value of available nitrogen, phosphorus and potassium (from laboratory soil test)

	N %	P ₂ O ₅ (kg ha ⁻¹)	K ₂ O (kg ha ⁻¹)	OM %
Low (L)	< 0.1	< 30	< 110	< 2.5
Medium (M)	0.1-0.2	31-55	111-280	2.6-5.0
High (H)	> 0.21	> 55	> 280	> 5.1

Appendix A2.2 Effect of seed rate on plant population and HY and DMY (g/plant)

Seed rate (kg ha ⁻¹)	Plants per square meter	HY				DMY			
		45 DAS		75 DAS		45 DAS		75 DAS	
		20 17	20 18	20 17	20 18	20 17	20 18	20 17	20 18
20	22.2	76. 6	75. 2	0.3 6	0.4 1	13. 5	12. 6	7.2	8.1
40	44.4	39. 6	44. 3	0.1 8	0.2 0	6.9	8.5	3.8	4.2
60	66.7	24. 8	35. 7	0.1 1	0.1 2	4.8	6.5	2.4	2.5
80	88.9	18. 4	27. 5	0.0 8	0.1 0	3.1	5.0	1.7	2.2

Appendix A2.3 Effect of seed rate on plant population and leaf and tiller number per square meter

Seed rate (kg ha ⁻¹)	Plants per square meter	Leaves per square meter				Tillers per square meter			
		45 DAS		75 DAS		45 DAS		75 DAS	
		2017	2018	2017	2018	2017	2018	2017	2018
20	22.2	160	184	193	258	42	56	33	38
40	44.4	329	324	373	471	80	93	62	62
60	66.7	480	467	560	640	113	133	93	100
80	88.9	649	533	729	747	133	107	124	107

Appendix B for Chapter 3

Appendix B3.1 Soil temperature at 5 cm depth during the cropping season in 2017 and 2018

Months	2017		2018	
	Min	Max	Min	Max
March	19.6	27.4	21.1	29.8
April	20.3	28.4	25.7	33.7
May	N/A	N/A	28.3	34
June	28.8	31.4	-	-

Appendix C for Chapter 4

Appendix C4.1 Table of means for the interaction between sowing dates and cutting management on plant height in 2017

Sowing date	Cutting management		
	0	1	2
30-March	371	338	311
30-April	330	283	381
30-May	300	353	310
30-June	365	275	196

LSD (5%) (Comparisons within a sowing date) 46

LSD (5%) (Other comparisons) 47

Appendix C4.2 Table of means for the interaction between sowing dates and seed rates on LAI in 2017

Sowing date	Seed rates (kg/ha)			
	20	40	60	80
30-March	4.1	3.3	2.8	2.6
30-April	2.7	2.9	3.4	3.3
30-May	1.4	1.3	1.3	1.3
30-June	1.4	1.2	1.2	0.9

LSD (5%) (Comparisons within a sowing date) 0.4

LSD (5%) (Other comparisons) 0.6

Appendix C4.3 Table of means for the interaction between seed rates and cutting management on LAI in 2017

Seed rate (kg/ha)	Cutting management		
	0	1	2
20	2.9	2.3	2.1
40	2.4	2.0	2.2
60	2.6	2.0	1.9
80	2.1	1.9	2.0
LSD (5%) (Comparisons within a sowing date) 0.3			
LSD (5%) (Other comparisons) 0.4			

Appendix C4.4 Table of means for the interaction between sowing dates and cutting management on cobs per plant in 2017

Sowing date	Cutting management		
	0	1	2
30-March	12.6	12.2	10.0
30-April	10.7	10.7	8.8
30-May	9.9	8.9	7.1
30-June	8.1	7.3	3.4
LSD (5%) (Comparisons within a sowing date) 1.3			
LSD (5%) (Other comparisons) 1.9			

Appendix C4.5 Table of means for the interaction between sowing dates and cutting management on ears per cob in 2017

Sowing date	Cutting management		
	0	1	2
30-March	6.5	6.9	7.0
30-April	5.9	6.2	6.0
30-May	5.4	5.6	4.4
30-Jun	5.3	4.2	2.5
LSD (5%) (Comparisons within a sowing date) 0.6			
LSD (5%) (Other comparisons) 0.9			

Appendix C4.6 Table of means for the interaction between sowing dates and cutting management on seeds per ear in 2017

Sowing date	Cutting management		
	0	1	2
30-March	5.0	5.0	5.0
30-April	4.8	4.6	4.5
30-May	4.6	4.5	4.1
30-Jun	4.3	4.2	0.4

LSD (5%) (Comparisons within a sowing date) 0.3
LSD (5%) (Other comparisons) 0.5

Appendix C4.7 Table of means for the interaction between sowing dates and cutting management on plant height in 2018

Sowing date	Cutting management		
	0	1	2
30-March	381	351	359
30-April	377	353	313
30-May	361	293	303

LSD (5%) (Comparisons within a sowing date) 28
LSD (5%) (Other comparisons) 32

Appendix C4.8 Table of means for the interaction between sowing dates and seed rates on LAI in 2018

Sowing date	Seed rates (kg/ha)			
	20	40	60	80
30-March	5.0	4.1	2.7	2.1
30-April	3.1	3.2	3.6	2.8
30-May	1.9	2.4	2.3	1.6

LSD (5%) (Comparisons within a sowing date) 1.0
LSD (5%) (Other comparisons) 1.0

Appendix C4.9 Table of means for the interaction between sowing dates and seed rates on cobs per plant in 2018

Sowing date	Seed rates (kg/ha)			
	20	40	60	80
30-March	9.4	8.3	7.9	7.9
30-April	8.6	7.7	7.4	7.4
30-May	7.8	7.8	7.2	5.0

LSD (5%) (Comparisons within a sowing date) 0.8
LSD (5%) (Other comparisons) 1.0

Appendix C4.10 Table of means for the interaction between seed rates and cutting on cobs per plant in 2018

Seed rate (kg/ha)	Cutting management		
	0	1	2
20	8.7	8.5	8.6
40	8.8	7.8	7.1
60	7.7	8.1	6.6
80	7.8	6.5	6.0

LSD (5%) (Comparisons within a sowing date) 0.7
LSD (5%) (Other comparisons) 0.7

Appendix C4.11 Table of means for the interaction between sowing dates, seed rates and cutting management on cobs per plant in 2018

Sowing date	Seed rate(kg/ha)	0	1	2
30-March	20	9.6	9	9.5
	40	10	7.8	7.2
	60	8.2	8.3	7.1
	80	8.3	8.1	7.4
30-April	20	9	8.7	8.1
	40	8.3	8.2	6.6
	60	8	7.5	6.6
	80	8.2	7.2	6.7
30-May	20	7.7	7.7	8.1
	40	8.2	7.5	7.6
	60	6.9	8.4	6.2
	80	7	4.1	3.9

LSD (5%) (Comparisons within a sowing date) 1.2
LSD (5%) (Other comparisons) 1.4

Appendix C4.12 Table of means for the interaction between sowing dates and cutting management on ears per cob in 2018

Sowing date	Cutting management		
	0	1	2
30-March	6.4	6.5	6.1
30-April	7.1	5.6	4.7
30-May	5.0	5.1	3.2
LSD (5%) (Comparisons within a sowing date) 0.7			
LSD (5%) (Other comparisons) 0.8			

Appendix C4.13 Table of means for the interaction between sowing dates and seed rates on seeds per ear in 2018

Sowing date	Seed rates (kg/ha)			
	20	40	60	80
30-March	5.0	4.8	4.4	4.9
30-April	4.4	4.3	4.3	4.3
30-May	5.3	4.8	4.7	4.2
LSD (5%) (Comparisons within a sowing date) 0.8				
LSD (5%) (Other comparisons) 0.9				

Appendix C4.14 Table of means for the interaction between sowing dates and cutting management on seed yield in 2018

Sowing date	Cutting management		
	0	1	2
30-March	4678	3731	2965
30-April	3518	3015	2629
30-May	1735	1803	1316
LSD (5%) (Comparisons within a sowing date) 393			
LSD (5%) (Other comparisons) 895			

Appendix C4.15 Table of means for the interaction between sowing dates and cutting management on seed yield from top positioned cobs in 2017

Sowing date	Cutting management		
	0	1	2
30- March	1721	1861	1503
30-April	1805	1636	1475
30-May	984	885	630
30-June	754	544	35
LSD (5%) (Comparisons within a sowing date) 334			
LSD (5%) (Other comparisons) 461			

Appendix C4.16 Table of means for the interaction between sowing dates and cutting management on seed yield from middle positioned cobs in 2017

Sowing date	Cutting management		
	0	1	2
30- March	1545	1532	1383
30-April	1331	1056	946
30-May	764	778	520
30-June	748	487	0
LSD (5%) (Comparisons within a sowing date) 333			
LSD (5%) (Other comparisons) 421			

Appendix C4.17 Table of means for the interaction between sowing dates and seed rate on seed yield from middle positioned cobs in 2018

Sowing date	Seed rate (kg/ha)			
	20	40	60	80
30-Mar	1600	1470	1348	928
30-Apr	1543	1266	912	555
30-May	579	576	547	358
LSD (5%) (Comparisons within a sowing date) 586				
LSD (5%) (Other comparisons) 590				

Appendix C4.18 Table of means for the interaction between sowing dates and cutting management on seed yield from middle positioned cobs in 2018

Sowing date	Cutting management		
	0	1	2
30-Mar	1608	1321	1079
30-Apr	1040	1053	1114
30-May	526	658	361
LSD (5%) (Comparisons within a sowing date) 237			
LSD (5%) (Other comparisons) 316			

Appendix C4.19 Table of means for the interaction between sowing dates and cutting management on seed yield from bottom positioned cobs in 2018

Sowing date	Cutting management		
	0	1	2
30-March	953	658	604
30-April	427	399	296
30-May	281	230	183
LSD (5%) (Comparisons within a sowing date) 138			
LSD (5%) (Other comparisons) 262			

Appendix D for Chapter 7

Methodology for economic analysis

For each sub sub plot, herbage yield and seed yield was calculated. The economic analysis was done on the basis of two different components. The first calculation is based on the milk yield through the herbage production and the second was through the seed yield. For economic analysis from herbage yield, it was assumed that one kg of herbage will increase 0.67% milk yield (Shrestha, Timilsina, Sanjyal and Munanakarmi, 2011) which is equivalent to 0.04355 liter per animal per day. The average milk production used for economic analysis was 6.5 liter per animal per day in Nepal and farm gate price per liter of milk was Rs 60 (Hampton, Pariyar, Ghimire, Armstrong, Rolston, Stevens and Shrestha, 2019). The cost of green herbage used during the calculations is Rs 6 per kg. Further the seed yield from the top and middle (T+M); middle and bottom (M+B) was also calculated separately and the seed income from these two positions was calculated in Nepali rupees for both years

Cost of production

The gross return of a crop is highly reliant upon the cost of cultivation, yield and the marketing price. Cost of cultivation is the sum total of several cost components in the field. Therefore, component-wise costs were recorded to study cost of cultivation of teosinte. The costs that relate directly to the farm production are called variable costs. Variable cost can either be cash costs or non-cash costs. Cost of seed, fertiliser, plant protection measures, irrigation, human labour/machine labour for land preparation, fodder cuttings, seed harvest etc. are cash costs. The fixed costs are land rent, depreciation of implements, interest on fixed capital, land revenue etc. The item wise per hectare cost was recorded for each variables and the cost incurred during the cultivation was calculated and summed up for all the items. Net cost is the addition of all the costs incurred from sowing to seed harvesting. The net cost, gross return, gross margin and benefit cost ratio (BCR) ratio was calculated using the formula as follows:

Net cost (NRs/ha) = Cost of seed (kg ha^{-1}) + land rent (NRs) + land preparation (NRs) + labor cost (NRs) (sowing, irrigation, weeding, herbicide/insecticide spray, herbage harvest, seed harvest) + pesticide + (NRs) fertilizer (NRs).

Gross return (GR) from milk and seed

Gross return was calculated by adding the income from herbage (as milk) and seed yield. The following algebraic form of NR was used for estimation:

$$\text{GR} = \Sigma \text{Rs} + \Sigma \text{Rh}$$

where,

GR = Gross return (NRs/ha);

ΣRs = Total return from seed (NRs/ha); and

ΣRh = Total return from herbage (NRs/ha).

Gross margin (GM)

Gross margin was calculated by the difference between gross return and Total cost. The following equation was used to calculate GM:

$$\text{GM} = \text{GR} - \Sigma \text{C}$$

Where,

GR = Gross return (NRs/ha); and

ΣC = Total cost (NRs/ha).

Benefit cost ratio (BCR)

Benefit cost ratio (BCR) is a relative measure which is used to compare the return per unit of cost. BCR was estimated as a ratio of gross return to Total cost. The formula used for calculating BCR was as follows:

$$\text{BCR} = \text{GR} \div \text{GC}$$

Where,

GR = Gross return (NRs/ha); and

TC = Total cost NRs/ha).

Appendix A7.1 Table of means for the interaction between sowing dates and cutting management on net cost in 2017

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	73397	82010	84044
30-Apr	71883	82957	86471
30-May	65720	69683	70797
30-Jun	64917	66835	65546
LSD (5%) (Comparisons within a sowing date) 2334			
LSD (5%) (Other comparisons) 2821			

Appendix A7.2 Table of means for the interaction between sowing dates and cutting management on gross return in 2017

Sowing dates	Cutting management(NRs)		
	0	1	2
30-Mar	364727	411420	381274
30-Apr	342151	330931	305860
30-May	144128	153182	99833
30-Jun	113870	71540	-30766
LSD (5%) (Comparisons within a sowing date) 80094			
LSD (5%) (Other comparisons) 106490			

Appendix A7.3 Table of means for the interaction between sowing dates and cutting management on gross margin in 2017

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	291330	329409	297230
30-Apr	270268	247974	219389
30-May	78408	83500	29036
30-Jun	48954	4705	-96312
LSD (5%) (Comparisons within a sowing date) 77873			
LSD (5%) (Other comparisons) 103817			

Appendix A7.4 Table of means for the interaction between sowing dates and cutting management on B: C ratio in 2017

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	4.932	4.996	4.497
30-Apr	4.6	3.92	3.474
30-May	2.129	2.197	1.374
30-Jun	1.694	1.051	-0.469
LSD (5%) (Comparisons within a sowing date) 0.95			
LSD (5%) (Other comparisons) 1.32			

Appendix A7.5 Table of means for the interaction between seed rate and cutting management on net cost in 2018

Seed rates	Cutting management (NRS)		
	0	1	2
20	70390	75670	79344
40	72555	79912	81701
60	73995	82018	84657
80	72564	82827	85532
LSD (5%) (Comparisons within a sowing date) 2474			
LSD (5%) (Other comparisons) 2561			

Appendix A7.6 Table of means for the interaction between seed rate and cutting management on gross return in 2018

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	438661	360027	315337
30-Apr	314079	337415	305885
30-May	122599	164797	127424
LSD (5%) (Comparisons within a sowing date) 44852			
LSD (5%) (Other comparisons) 93716			

Appendix A7.7 Table of means for the interaction between seed rate and cutting management on gross margin in 2018

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	362747	282000	231866
30-Apr	241166	250513	218017
30-May	54298	89406	50338
LSD (5%) (Comparisons within a sowing date) 43194			
LSD (5%) (Other comparisons) 91682			

Appendix A7.8 Table of means for the interaction between seed rate and cutting management on seed income in 2018

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	514575	410414	326190
30-Apr	386992	331701	289238
30-May	190900	198294	144711
LSD (5%) (Comparisons within a sowing date) 43228			
LSD (5%) (Other comparisons) 98453			

Appendix A7.9 Table of means for the interaction between sowing dates and cutting management on B: C ratio in 2018

Sowing dates	Cutting management (NRs)		
	0	1	2
30-Mar	5.763	4.602	3.773
30-Apr	4.278	3.886	3.469
30-May	1.798	2.203	1.649
LSD (5%) (Comparisons within a sowing date) 0.48			
LSD (5%) (Other comparisons) 1.06			