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Plant density in maize (*Zea mays* L.): A review in perspective of Nepal

Jiban Shrestha¹ · Subash Subedi²

Abstract: Maize (*Zea mays* L.) is an important food and feed crop of the world. Maize yield is closely related to plant population. This article reviews on general overview of factors affecting optimum plant population, stress mechanism of plant population, plant architecture to high planting densities, relationship between plant population, growth, grain yield and yield attributing traits of maize. This review shows that at higher plant density, plant growth, development and production decreases and at low plant density also production decreases. The optimum density of plants varies on environmental factors and plant architecture. For higher production plant density of 66,000 plants/ha is better in subtropical region of Nepal. The promotion of desired plant density should be done to ensure increased maize production. This review serves as a useful tool to maize researchers and growers for making the right decision on management of plant density in maize.

Keywords: Growth · Maize · Plant density · Yield

Introduction

Plant population plays an important role in determining agronomic characteristic, growth and yield response of the crop. The optimum plant density utilizes the available resources and helps in increasing grain yield on particular environment. There is a strong correlation between optimum plant population and highest grain yield of any crop. The economic yield of the crop is low if the plant population is lower or higher than a optimum population. The abiotic and management factors like climate, soil and management practices determine the appropriate plant population for a particular location. The maize crop lack tillering ability that makes it more dependent for optimum plant density to maximize the grain yield or for forage. The proper plant population with recommended spacing is the most crucial factor to maximize the maize yield in agronomic practices. The thicker population promotes competition among the plants for light and nutrition while available nutrition are not optimally utilized if lower plant population is kept in the field and also lower plant population promotes weed population (Khan, 1972).

The improved agronomic practices including desired plant density and genetic advances determine the higher production and productivity of maize (Ciampitti and Vyn, 2012). The relationship between plant yield and density is highly variable due to some factors like rainfall, tillage system, fertilizer and soil type (Assefa *et al.*, 2016), although plant density has strong control on production and productivity of maize (Van Roekel and Coulter, 2011). The objective of this article is to present an overview of factors affecting optimum plant population, stress mechanism of plant population, plant architecture to high planting densities, relationship among plant population, growth, grain yield and yield traits of maize and also pinpoint optimum plant population for higher production and productivity of maize in Nepal.

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Factors affecting optimum plant density

The optimum density of plants varies on all environmental factors. The maximum number of plants/area depends on the variety, duration, productivity of the soil and water supply (Crickman, 1958). Maize being determinant crop, it is poor compensator to thinner plant population (Sharma *et al.*, 1991). Hybrid introduction caused high-density use in maize (Russel, 1991). Dawadi and Sah (2012) reported that in winter season at higher plant population of 66,666 plants/ha produced the higher grain yield (11.19 t/ha) compared to that of 55,555 plants/ha (9.52 t/ha) in Chitwan, Nepal.

The higher plant density is required for higher yield in subtropical and temperate climate (Anderson, 1995). Under rainfed condition, the amount of water and its availability both become important unmanageable factors for determining grain yield with optimum plant population (Loomis and Connors, 1992). The higher plant density resulted in high competition among plants for light, nutrients and other essential growth traits. The maize yield increases to reduce the width of the line to create a more balanced model of planting. The row width could be minimized for more equidistant planting pattern, which ultimately turn plant population to higher value in terms of maize grain yield. The maximum population of plants will be increased to use the distance between rows between 0.5 and 0.75 meters (Sangoi and Salvador, 1998a).

Stress mechanism of plant population

The grain yield per plant is linearly correlated with available space. The proportion between yield/plant and available space is a universal phenomenon observed in all soils, all countries, in fields, in pots or in culture solution (Reddy, 2000). Increasing the available space increased yield/plant. Under very dense plant stands, both inter and intra-plant competitions are sufficiently severe to reduce all components of yield. When there is closed spacing, the leaf surface per plant is reduced. At closer spacing, there is high competition between crop plants for light, nutrient, moisture, etc. If roots of another plant occupy the surrounding space, growth retarded (Papadakis, 1970). In general, high plant densities are conducive for lodging and built up of pests and diseases. The seedling mortality is common under higher plant densities.

Plant architecture to high planting densities

The reduced tassel size, rapid growth of first ear, short pollen-shed-to-silking interval and a more efficient

production of grain per unit of leaf were the major characters of plant density-tolerant genotypes (Smith *et al.*, 1982). The level of interference or competition of each individual maize plant over other is lower when there is a smaller and lesser leafy plant (Sangoi and Salvador, 1998a). The relative production and maintenance cost per plant (e.g. water, nutrients and assimilates) is lower with production of a smaller plant, with shorter stems, fewer and more erect leaves (Loomis and Connors, 1992). The leaf area index (LAI) is increased with a lower vegetative biomass per plant that allows the use of more individuals per area and the light interception is effective with more LAI (Tollenaar *et al.*, 1997).

The increase in dry matter yield is positively correlated with more intercepted solar radiation (Sinclair, 1998). The change in plant design in modern hybrid received higher rate of leaf photosynthesis than older hybrid even at higher plant population (Dwyer *et al.*, 1991). The problem of stalk and root lodging also minimized with improved maize stability at high plant population with more dense plant design (Sangoi *et al.*, 2000). The tassel is apical and differs in first place; priority is given to ears in maize. The production and dispersion of pollen through the development of ears and silk is ensured by higher plant population in maize (Sangoi and Salvador, 1998b). The balance between male and female inflorescences development is also compared with more number of ears per plants and fewer sterile plants gained from modern hybrids at higher plant population. Plant population density depends on both genotypic factors (Kumar *et al.*, 2013). Recently developed hybrids are more prone to withstand higher planting density than older hybrids (Sher *et al.*, 2017).

Effect of plant population on growth, grain yield and yield components of maize

The synchronization in flowering and grain yield influenced much with plant density. Aguila *et al.* (1971) reported that increment in plant population from 55,000 to 85,000 plants/ha, significantly delayed the silking time. Teasdale (1995) reported that maize grown in high plant population has weed control potential, thereby minimizing the herbicide use.

Lemkoff and Loomis (1986) reported the prolongation of the time interval between anthesis and silking due to the increase in the number of plants. The tassel requires a greater auxin concentration for its development than the ear. Therefore, greater growth hormone in the population can promote apical dominance over the ears, which contributes to infertility (Sangoi and Salvador, 1998b).

Cox (1996) noticed that with the increment in plant population, the forage dry matter is also increased in maize. Singh *et al.* (1997) reported higher dry matter accumulation under 1,11,111 plants/ha, followed by 83,333 plants/ha. Cox *et al.* (1998) found that the quality of silage was decreased at higher plant population so plant population under 86,500 plants/ha for milk yield compared with that under 97,500 plants/ha for dry matter yields is at best level in narrow rows. Athar (1979) noted that LAI and days to 50% silking increased and specific leaf weight decreased with increasing plant population.

Singh and Singh (2002) reported that during the regular season crop yields of 60-70 thousand plants/ha are important to increase yield. Corn grain yield decreases as plant density increases above optimal plant density, mainly due to a decrease in harvest index and an increase in stem lodging (Tollenaar *et al.*, 1997). Yao and Shaw (1964) observed that an increase in corn yield is associated with an increase in the plant population.

The lower yield is the outcome of low availability of light, moisture and other resources for individual plant in higher plant population. The loss of crop yield is mainly due to fewer ears, fewer kernels per ear (Baenziger and Glover, 1980), lower kernel weight (Poneleit and Egli, 1979), or addition of these components. In dense plant populations, many kernels may not develop. This occurs in some genotypes due to poor pollination as a result of the delayed period of silking compared with the appearance of tassels (Otegui, 1997) and/or due to the restriction of assimilation of nutrition, causing kernel and ear abortion (Karlen and Camp, 1985).

Interaction of densities with nutrition

Tajul *et al.* (2013) conducted a research in which three levels of plant populations (53000, 66000, and 800000 plants/ha) corresponding to spacing of 75×25 , 60×25 , and 50×25 cm) and 4 doses of N (100, 140, 180, and 220 kg/ha) were applied and they found that Crop Growth Rate (CGR) was the highest with the population of 80,000/ ha receiving 220 kg N/ha. Sharma and Gupta (1968) reported that under low plant population, smaller nitrogen rates produced maximum grain yield (100 kg N/ha for 40 and 50 thousands plants/ha) while under higher plant population only maximum dose of nitrogen (200 kg N/ha for 60 and 70 thousands plants/ha) produced maximum yield. They observed increased cumulative demand for nitrogen with increasing level of plant population. Shrestha *et al.* (2018) reported that the highest grain yield (6,925.79 kg/ha) of

maize was obtained with the application of 200 kg N/ha in population of 66,666 plants/ha in Chitwan, Nepal.

Ramirez (1965) reported that a positive interaction between N and plant population found the highest yields under plant densities of 60,000 and 75,000 plants/ha and N rates of 100-200 kg/ha. Adhikari *et al.* (2004) reported that the highest grain yield of 9,352 kg/ha was produced when the crop was fertilized with 120 kg N/ha on the crop planted under the plant density of 53,333 plants/ha and they noted the lowest yield (6,657 kg/ha) with the crop supplied with 60 kg N/ha under plant density of 44,444 plants/ha. Neupane and Basnet (2002) reported that non significant ($p > 0.05$) response on grain production of early maize varieties to 3 levels of N (60, 90 and 180 kg/ha) and 3 plant densities (53,000 71,000 and 95,000 plants/ha) were observed during summer season, however, the highest grain yield of 2.35 t/ha was produced by Arun-2 at 120 kg N/ha with 95 thousands of plant population.

Physiological changes with planting densities

The late ear differentiation and primordial growth resulted physiological changes that may be accompanied by infertility among high plant densities (Jacobs and Pearson, 1991). At higher rate of plant density, the axillary buds slow down the growth of the buds more than the point of the shoot. The initiated ear shoots may receive smaller amounts of such substances, thus making them less functional and less likely to form grains. For this reason, the growth observed for the ears among plants with dense populations may result in increased competition for assimilation between the ear and the rest of the plant.

A change in the plant population can affect the final kernel set by flowering, foliage and fertilization, as well as influencing the developing ear in the early stages of filling the grain. The number of spikelet fertilized determines the kernel set and grain yield of maize. There is low availability of nitrogen (Lemcoff and Loomis, 1994), photosynthates (Jacobs and Pearson, 1991), and water (Yao and Shaw, 1964) to growing ears due to high plant population. The lower spikelet number, poor silk formation, delay growth and development are due to restrictions in carbon or nitrogen metabolism and drought stress contributing to lesser spikelet that might be fertilized through concurrence of pollen shed with silking of individual spikelet (Jacobs and Pearson, 1991). Thus, infertility and the production of undeveloped or stub ears closely associated with higher plant population and delay in the growth of silk or ear.

Because of the low fertilized ovaries, the number of potential kernel/ears with a high plant population is also decreasing. The important substrate for grain is soluble sugar and organic nitrogen (Swank *et al.*, 1982). The lower supply of carbon and nitrogen to the ear resulted abortion after fertilization due to the consequences of higher plant population. The abortion of the young corn kernels contributes to a higher plant density. The number, size and activities of endosperm cells are reduced in higher plant densities as a result low endosperm capacity with lower grain yield.

Conclusion

The desired plant density is dependent on environmental factors, soil nutrition and plant architecture. In general, optimum plant density resulted in higher grain yield than other plant densities. High density planting, while important to increased yields, can also lead to greater competition for resources. The development of genotypes that withstand high plant densities should be necessary for enhancing higher production in maize.

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Turcicum leaf blight: A constraint to maize cultivation

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Abstract: Maize is cultivated extensively throughout the world and has the highest production among all cereals. However, Turcicum leaf blight caused by the pathogen *Exserohilum turcicum* is the most devastating disease of maize crop and causes immense yield losses. The disease has worldwide distribution and its development is favoured by cool to moderate temperature with high relative humidity. The prevalence of the disease has increased in recent years and new races of the pathogen have been reported worldwide. The fungus *E. turcicum* is highly variable in nature. Development of varieties with resistance to *E. turcicum* is the most efficient and cost-effective way to manage the disease particularly in high altitude mountain agro-ecologies.

Keywords: Disease · *Exserohilum turcicum* · turcicum leaf blight · symptomatology

Introduction

Turcicum leaf blight of maize also called Northern corn leaf blight is incited by the fungus *Exserohilum turcicum* (Pass.) teleomorph, *Setosphaeria turcica* (Luttrell) K. J. Leonard and E. G. Suggs. The disease is widely spread and economically most important disease of maize in the world (Chung *et al.*, 2010; Sibiya *et al.*, 2013). The disease causes enormous damage to crop and the loss in grain yield ranges from 24 to 91% (Pant *et al.*, 2000; Nwanosike *et al.*, 2015). The damage to maize crop can be manifold if the disease develops prior to silk emergence. The disease epidemics at an early stage cause premature death of blighted leaves thereby loss their fodder quality which is of great value under temperate agro climatic conditions as the same is being fed to the cattle during lean season (Payak and Renfro, 1968; Patil, 2000). Turcicum leaf blight has worldwide distribution particularly in areas where high humidity (75-90%) and moderate temperature (22-25°C) prevails during the cropping season (Khatri, 1993; Gregory, 2004). Deployment of resistant cultivars is most effective and cost-efficient way to control the Turcicum leaf blight. The genetic variability and pathogenicity of pathogen are the key factors for host-plant resistance and for the formulation of viable strategies for disease management. The fungus *E. turcicum* is known to be highly variable in cultural characteristics, pathogenicity and genetic traits. The lack/loss of substantial durable resistance in the maize genotypes may be attributed to the presence of variability and continuous change in racial spectrum of the pathogen (Pandurang Gowda *et al.*, 1993). The host plant resistance depends on the effectiveness of resistance against all the virulent races of the pathogen present in the region. Identification of variability among the isolates of a pathogen is an important step to devise a disease management programme for a particular region and for the development

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of disease resistant cultivars in many host pathogen system where major genes control resistance.

Occurrence and distribution

Turcicum leaf blight also known as Northern Leaf Blight is one of the most important diseases of maize (*Zea mays* L.) caused by a hemibiotrophic fungal pathogen *Exserohilum turcicum*. The disease has worldwide distribution particularly in areas where high humidity and moderate temperature prevails during the growing season (Shurtleff, 1980; Ceballos *et al.*, 1991; Juliana *et al.*, 2005). The disease was first reported by Passerine (1876) from Italy. Presently turcicum leaf blight is severe problem in the North-eastern United States, in sub-Saharan Africa, Europe, Australia and in areas of North Korea, China and India (Adipala *et al.*, 1993; Kim *et al.*, 2012; Wathaneeyawech *et al.*, 2015; Sartori *et al.* 2015). In India the disease was first reported by Butler (1918) on sorghum and later by Mitra (1923) on both sorghum and maize from the Punjab. Its prevalence in Punjab, Himachal Pradesh and Kashmir valley was also reported by Chenula and Hora (1962). Several other hot spots in the country such as, Dharward (Karnataka), Kolhapur (Maharashtra), Karim Nagar (Andhra Pradesh) and Dholi (Bihar) have also been identified (Laxminarayan and Shankerlingam 1983). This disease occurs sporadically in most temperate, humid maize grown areas (Lim *et al.*, 1974). Disease is most prevalent in all the major maize growing regions of India during rainy (*Kharif*) as well as winter (*Rabi*) season (Lal, 1991). This disease has also been observed to be the most important limiting factor for maize production in the hills, mainly in Jammu and Kashmir, Himachal Pradesh, Sikkim, West Bengal, Meghalaya, Tripura, Assam and Uttarakhand. Turcicum leaf blight is of particular concern in the tropical highlands, where conditions favour disease development, the disease was also found to be the major constraint of maize production under temperate agro-ecologies of Jammu and Kashmir (Ahangar *et al.*, 2016a).

Yield losses

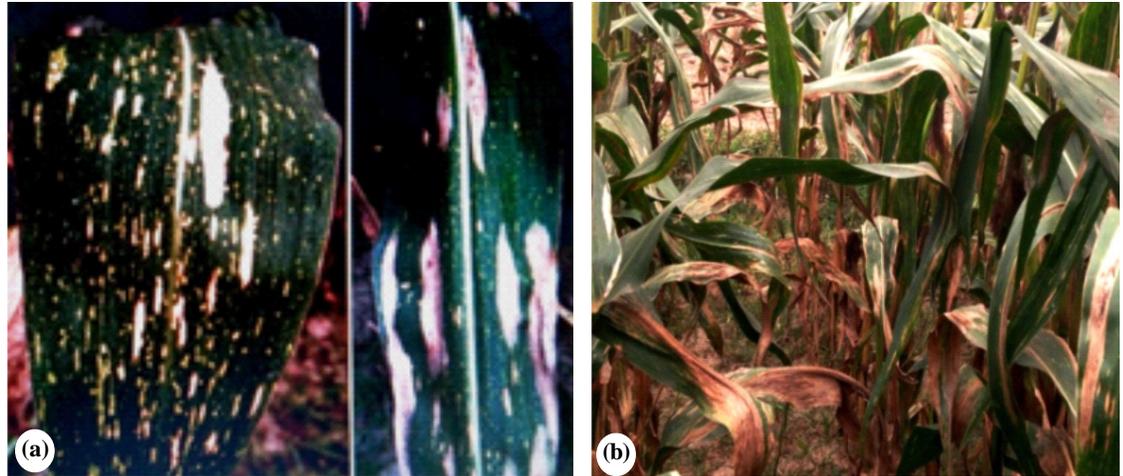
Northern corn leaf blight (NCLB) caused by *E. turcicum* is a constraint to global maize production. It is a ubiquitous foliar wilt disease of maize in many temperate and tropical environments which can cause yield losses up of to 70% (Yeshitila, 2003). Apart from yield loss, the disease causes qualitative changes in the seed resulting in decreased sugar content, germination capacity and severely infected plants

are predisposed to stalk rot (Cardwell *et al.*, 1997; Gowda *et al.*, 1992). The magnitude of yield losses caused by northern leaf blight depends on two factors; the stage of maize growth when the infection occurs and the severity of the disease. If the disease establishes before silking, yield reduction up to 40% may occur (Raymond and Hooker, 1981), but if infection delays until 6-8 weeks after silking, yield losses are minimal (Raymond, 1978). Yield losses approaches up to 50% when the disease occurs severely at 2-3 weeks after pollination (Shurtleff, 1980). The disease can substantially reduce the grain yield of maize over a wide range from 28 to 91% (Sharma and Aujla, 1968; Krasuz *et al.*, 1993; Pant *et al.*, 2000; Nwanosike *et al.*, 2015; Ribeiro *et al.*, 2016). Average losses of 60% have been reported in Kenya, Uganda, Ethiopia South Africa and Zambia (Nwanosike *et al.*, 2015). Maize crop in temperate belt of Kashmir is ravaged by this destructive disease to the losses in the range of 27.6-90.7% of total grain yield particularly if the disease develops prior to silk emergence (Chenulu and Hora, 1962). Moreover the disease cause immense damage to crop straw which is of great value under temperate agro climatic conditions of Kashmir as the same is being fed to the cattle during lean season.

Symptomatology

The disease starts at first as small, oval, greyish green and water soaked spots which grow into elongated, spindle-shaped necrotic lesions (Chenulu and Hora, 1962). They appear first on lower leaves and the number of spots increases with the development of plant, leads to complete blighting of the foliage (Figure 1a). Northern Leaf blight lesions are elongated elliptical greyish lesions, measuring up to 12 mm wide and 2.5-15 cm long which run parallel to leaf margin (Li and Wilson, 2013; De Rossi *et al.*, 2015a). On mature lesions distinct dark grey areas develop associated with fungal spores (Laxminarayan and Shankerlingam, 1983). Spore production causes the lesions to appear dark gray, olive or black (King and Mukuru, 1994). Symptoms can range from cigar-shaped lesions on the lower leaves to complete destruction of the foliage (Figure 1b), thereby reducing the amount of leaf surface area available for photosynthesis (Li and Wilson, 2013). Lesions of northern corn leaf blight can vary depending on the *E. turcicum* race present also lesion shape and size may vary with the genotype of the plant. This qualitative interaction between the resistance (R) gene of the host, and the Avirulence (Avr) gene of the pathogen directly affects conidial germination and ramification, and increases

Figure 1. Symptom of turcicum leaf blight on maize foliage



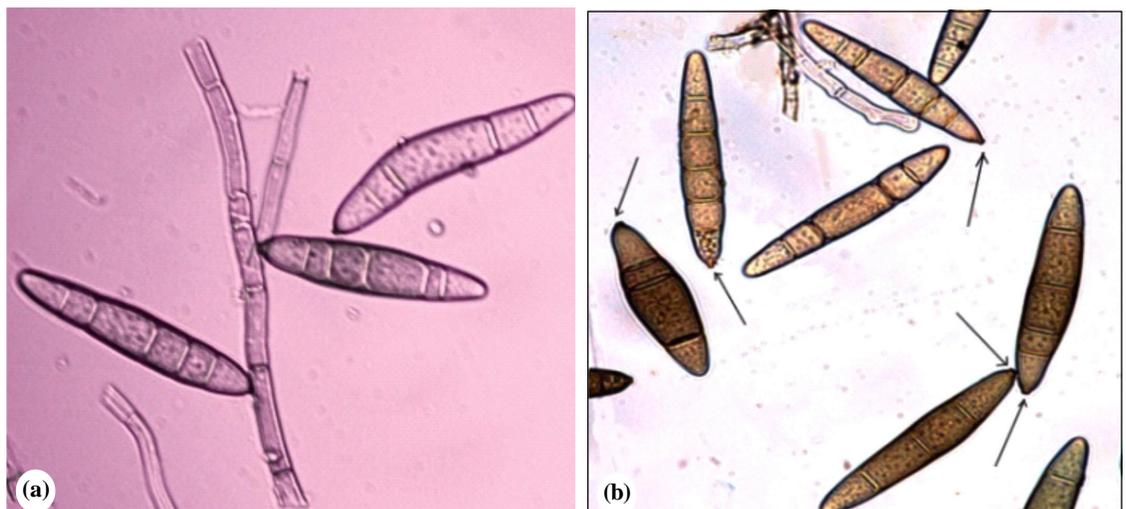
lesion size when the phytotoxin concentration is >250 ppm (Bashan *et al.*, 1996). Turcicum leaf blight is essentially a leaf disease. Symptoms usually appear at any stage of the crop as small grey spots on the lower leaves spreading upwards. Slowly the spots enlarge and appear as spindle shaped lesions with grey centre and dark brown margin (Sajeed and Chowdhury, 2014; Vieira *et al.*, 2014).

Taxonomy of the pathogen

Exserohilum turcicum is a polycyclic, heterothallic, facultative parasite of maize and was first reported by Passerine (1876) from Italy. Leonard and Suggs (1974) have proposed the nomenclature of the organism as *Exserohilum turcicum* (Pass.) K. J. Leonard and E. G. Suggs as imperfect stage and teleomorphic phase was identified in 1957 and named *Trichometasphaeria turcica* by Luttrell and later changed to *Setosphaeria turcica* (Luttrell) Leonard and Suggs (Leonard and Suggs, 1974; Alcorn, 1988). The causal agent of turcicum leaf blight of maize is normally

identified by its imperfect stage *Exserohilum turcicum* in which conidial hilum is strongly protuberant (Figure 2a). The sexual stage of the fungus, *Setosphaeria turcica* rarely occurs in nature (Luttrell, 1958). The teleomorphic phase of *E. turcicum* has been observed in the laboratory but typically not in nature. *E. turcicum* is a heterothallic fungus with three distinct mating types found in nature (Yongshan *et al.*, 2007). The study conducted by Bunkoed *et al.* (2014) is the first to investigate the sexual reproduction of *Setosphaeria turcica* in Thailand. Pseudothecia were found on heavily infected corn leaves from natural fields. The imperfect stage of *E. turcicum* is most often observed in nature. The conidia of the fungi are olivaceous-gray, elongated and Spindle shaped often less curved on one side (Figure 2b) compared with the conidia of *Helminthosporium maydis*, which are more curved. Average size is about 20-150 μ . Septation ranges from one to nine septa. A conspicuous spore feature of the conidia that distinguishes it from other of the more common species of *Drechslera* attacking corn is the protruding hilum

Figure 2. Taxonomy of *Exserohilum turcicum*



(Shurtleff, 1980; McGee, 1990). Conidiophores are simple, cylindrical, olivaceous brown which emerge individually or in groups of two to four from stomata in necrotic leaf lesions. Single conidium is formed terminally on the conidiophores (Figure 2) which then resumes growth to one side of the conidial attachment and eventually produced another conidium at the new tip (Leonard and Suggs, 1974; Alcorn, 1988; De Rossi *et al.*, 2015).

Etiology

The fungus *E. turcicum* has saprophytic survival. It overwinters as mycelia, conidia and chlamydospores on maize residues left on the soil surface. Local epidemics of turcicum leaf blight caused by *E. turcicum* usually originate from conidia on infected maize residue (Taken *et al.*, 1994). Mycelia and conidia of this fungus from infected crop residue, in or on the soil, act as the primary inoculum for the next crop. *E. turcicum* is also seed-borne and survives in the soil saprophytically, while on debris it survives as chlamydospores (Figure 3). Chlamydospores may become relatively ineffective if debris is ploughed deep in the soil in autumn. The secondary inoculum comes from lesions which produce conidia that are dispersed by wind and can be transported for long distances (Shenoi and Ramalingam, 1983; Ferguson and Carson, 2004). The fungus remains viable in seed for 28 months (Patil *et al.*, 2000). The turcicum leaf blight is favoured by mild temperature, high humidity, extended period of leaf wetness (rain or dew) and frequent light shower (Ullstrup, 1970; Shurtleff, 1980).

Germination of *E. turcicum* conidia is bipolar (Figure 3) and occurs 3-6 hours after inoculation. Germ tubes grow at an angle rather than parallel to the veins of the leaf. They produce simple or forked terminal appressoria from which

penetration pegs develop (Muiru, 2008). Penetration is usually direct and occurs only rarely through stomata. The pathogen allows penetration and colonization with the production of a range of secondary metabolites and toxins. The *E. turcicum* genome includes two genes encoding xylanase enzymes, which degrade arabinoxylan in the plant cell wall causing loss of integrity and aiding penetration (Degefu and Hanif, 2003). Infection pegs grow into or between epidermal cells of either the dorsal or the ventral side of the leaf (Hilu & Hooker, 1964; Chung *et al.*, 2010). Penetration usually occurs 12-18 hours after inoculation (Lilian *et al.*, 2002; Sajeed and Chowdhury, 2014). Following penetration, the fungus produces a vesicle-like structure inside or between the epidermal cells which gives rise to secondary hyphae that proceed intracellularly in the mesophyll tissue in various directions (Hilu & Hooker, 1964). The hyphae continue advancing inside the chlorenchyma tissue resulting in lesion formation. Cells eventually die in the vicinity of a lesion. These cells later appear devoid of all cytoplasm, separate and become disorganized (Tagne *et al.*, 2002). The hyphae continue to grow from the xylem into the surrounding healthy tissues resulting in the enlargement of the lesions. The hyphae penetrate the normal bundle sheath and grow rapidly in adjacent mesophyll cells. The cells become plasmolyzed and their protoplasm appears granular and the whole cell dies very rapidly (Jennings & Ullstrup, 1957; Muiru, 2008). Mycelial strands aggregate into pseudo parenchymatous masses in sub-stomatal chambers. Conidiophores produced from these dense masses emerge through the stomata and produce conidia abundantly (Hilu and Hooker, 1964, Lilian *et al.*, 2002).

Turcicum leaf blight of maize is more prevalent in humid weather with temperature between 20–28°C (Hooda

Figure 3. Etiology of *Exserohilum turcicum*



et al., 2017). The incidence and severity of Turcicum leaf blight varies from year to year and from one location to another depending largely on genetic makeup of the plants and prevailing environmental conditions (Gregory, 2004). The northern corn leaf blight is favoured by moderate temperature (18-27°C), high humidity (85-90%) besides low luminosity, the presence of large amount of inoculums, extended period of leaf wetness, frequent light showers and long dew periods favour turcicum leaf blight epiphytotic (Bentolila *et al.*, 1991; Gregory, 2004). Heavy dews, high humidity, and frequent rains are conducive environmental conditions for disease development (Jordan *et al.*, 1983). Northern leaf blight is generally known to be sporadic in occurrence, depending on the environmental conditions and the level of disease resistance of the plant (Perkins and Pedersen, 1987; Degefu, 1990). In general, the increase in the prevalence of the disease might be attributed to mono-cropping practice, high humidity, morning fogs and extended dew period, minimum tillage and the use of uniform and highly susceptible varieties (Babu *et al.*, 2004; Harlapur, 2005; Khedekar *et al.*, 2010; Reddy *et al.*, 2013).

Pathogenic variability

The genetic variability and pathogenicity of the pathogen are key factors for host-plant resistance and for the formulation of viable strategies for disease management. The fungus *E. turcicum* is known to be highly variable in cultural characteristics and pathogenicity. Greater genotypic diversity and gametic phase equilibrium was observed in *E. turcicum* populations from tropical regions than populations from temperate regions (Borchardt *et al.*, 1998). Higher frequency of sexual recombination was observed in tropical climates, while populations in temperate regions appear to be more clonal. *E. turcicum* populations are highly adaptable in both temperate and tropical climates, as an extensive migration was also found within agroecological zones (Borchardt *et al.*, 1998). Considerable variability among the isolates of *E. turcicum* was found under temperate agroclimatic conditions of Kashmir (Ahangar *et al.*, 2016a; Ahangar *et al.*, 2016b). Knox-Davies and Dickson (1960) reported sufficient evidence of heterokaryons and their perpetuation through the conidia and suggested that the high variability in the fungus population might be related to heterokaryosis. Assefa (1995) indicated significant difference among *E. turcicum* isolates in their virulence and the mean virulence rating was significantly correlated with spore length and rate of

germination. In nature a continuous build up of a new pathogen races/strains develops intermittently. Considerable variations in morphology (Harlapur *et al.*, 2007; Bunker *et al.*, 2011; Kutawa *et al.*, 2017) pathogenicity (Abebe and Singburadom, 2006; Bunker and Mathur, 2010; Muiru *et al.*, 2010) and genetic diversity (Eschholz *et al.*, 2010; Aci *et al.*, 2013) have been observed among isolates of *E. turcicum*.

Isolates from different locations differs in parasitic fitness with respect to infection efficiency, sporulation and lesion size besides differ in colour, type of mycelium, rate of growth and sporulation in culture (Mwangi, 1998; Levy, 1991; Abebe and Singburadom, 2006; Shree *et al.*, 2012). Highly virulent isolates exhibit higher infection types on different differentials. In a study by Muiru *et al.* (2010), the *E. turcicum* isolates from Kenya, Germany and Austria showed a varied response in the differentials indicating a high virulence complexity and variability of the pathogen. Aggressiveness of the various *E. turcicum* isolates differ in terms of lesion density, AUDPC, lesion size, length of incubation period and rate of lesion expansion (Reddy *et al.*, 2013; Yadav *et al.*, 2014). Bunkoed *et al.* (2014) first time investigated the sexual stage *Setosphaeria turcica*, of *E. turcicum* in Thailand and suggested that sexual reproduction of *S. turcica* has caused genetic variation in the fungal pathogen, supported by previous analysis with inter-simple sequence repeat markers. Furthermore, the virulence may be enhanced or new physiological races may be generated through sexual hybridization. Wathaneeyawech *et al.* (2015) found considerable variability among 478 isolates of *E. turcicum* collected from Thailand. Significant morphological variability was detected among ten *E. turcicum* isolates from Argentina and Brazil for all measured variables *viz.*, length, width and number of septa (De-Rossi *et al.*, 2015).

Genes for resistance and pathogenic races

Turcicum leaf blight is mainly controlled by resistant cultivars. The resistance is either qualitative or quantitative. Qualitative resistance is typically race specific and inherited by single gene (monogenic) whereas quantitative resistance is race nonspecific and oligogenic or polygenic (Geiger and Heun 1989; Pataky *et al.*, 1998). Monogenic or race specific resistance is controlled by *Ht1*, *Ht2*, *Ht3* and *Htm* genes. Gene *Ht1* (*Ht* for *Helminthosporium turcicum*) confers a chlorotic lesion type and was first single gene resistance, identified by Hooker in 1963 in the dent inbred line GE339 and in the popcorn cv 'Ladyfinger' (Hooker,

1963). In maize lines with *Htn* gene, lesion formation is delayed in such way that plants in the field remain free from lesion until shortly after pollination while as *Ht1*, *Ht2* and *Ht3* resistant gene occurs as chlorotic lesions with minimum sporulation (Leonard *et al.*, 1989). Six dominant genes (*Ht1*, *Ht2*, *Ht3*, *Htm1*, *Htn1* and *HtNN*) and two recessive genes *ht4* and *rt* were found to provide resistance to the various key races of *Setosphaeria turcica* (Gevers, 1975; Hooker, 1977; Hooker and Tsung, 1980; Welz and Geiger, 2000; Ogliari *et al.*, 2007). Qualitative resistant Ht genes have successfully been used in breeding programs. Several *Ht* genes have been mapped with molecular markers. *Ht1* and *HtP* are located on chromosome 2L and map 10 cM from each other (Bentolila *et al.*, 1991), *Ht2* and *Htm1* are on chromosome 8L (Zaitlin *et al.* 1992; Simcox and Bennetzen 1993), and *rt* is located on chromosome 3L (Ogliari *et al.* 2007). *Htm1* encodes a wall-associated receptor-like kinase that acts as an important component of the plant innate immune system by perceiving pathogen or host-derived elicitors (Hurni *et al.*, 2015). Co dominant SSR markers linked to the known *Ht* genes *Ht1*, *Ht2*, and *Htm1* have already been identified such as bnl1721 and umc 1042 being closely linked to the resistance gene *Ht1* ($R_2 = 0.2948$ and 0.2626 , respectively (Puttarach *et al.*, 2016).

E. turcicum races are defined based on their phenotypic reactions when inoculated onto a set of differential maize lines in this system (Leonard *et al.*, 1989). The disease has spread throughout the world with several different races such as Race 0, 1, 2, N, 2N, 3, 3N, 12, 13, 13N, 23 and 23N (Muiru *et al.*, 2010). An increasing number of *E. turcicum* races identified from China (0 and 1), Mexico (23N, 23 and 2N), Zambia (23, 23N and 0) and Uganda (0, 2, N, 23N) led to rapid resistance loss in many hybrids containing *Ht1*, *Ht2*, *Ht3*, or *HtN* gene (Welz *et al.*, 1993). Race designations are based on resistance genes that their virulence matches. For example *E. turcicum* race 0 is ineffective (avirulent) against all Ht genes while as race 1 is only effective (virulent) against *Ht1* gene; race 23N is virulent against *Ht2*, *Ht3* and *Htm1* genes; race 3 and 4 are virulent against the *Ht2*, *Ht3*, *Htm* genes and race 12 is virulent against *Ht1* and *Ht2* genes. Races 0 and 1 are most prevalent, whereas races 23, 2N, and 23N are rare (Fallah and Pataky, 1994). The race present in the Indian subcontinent has been determined to be race 2 (Payak and Sharma, 1985). Sexual recombination of *E. turcicum* likely occurs in nature (Borchardt *et al.*, 1998). Studies also have shown that *E. turcicum* migration over long distances is possible, which could transfer virulence to new regions

(Ferguson and Carson, 2004). Selection pressure, sexual recombination within the pathogen, and ability to migrate long distances could produce more virulent populations and lead to spatial and temporal race population shifts. The occurrence of novel pathogenic races therefore is a potent threat to maize with single-locus resistant genes. Sixteen races of the pathogen could be, theoretically, identified by four Ht genes. Among them, 13 races have already been detected in northern China (Hooda *et al.*, 2017) indicating a high race diversity of *S. turcica*.

Varietal resistance

The use of cultivars with genetic resistance is the most economical, effective and practical method of obviating loss in crop yield due to diseases (Ribeiro *et al.*, 2016). Inherent resistance or tolerance of crop plants to infection by the pathogen can most likely be a safe alternative and eco friendly disease management venture. Great efforts have been done worldwide to develop, identify and utilize germplasm with turcicum leaf blight resistance. Lot of maize germplasm has been evaluated to identify new resistance sources and establish durability of known resistance against Turcicum leaf blight (Harlapur, 2005; Shikari and Zafar, 2009; Kumar *et al.*, 2011; Chandrashekara *et al.*, 2014; Mitiku *et al.*, 2014; Singh *et al.*, 2004; Muiru *et al.*, 2015; Ribeiro *et al.*, 2016; Garoma *et al.*, 2016; Setyawan *et al.*, 2016). Resistance sources for turcicum leaf blight across the globe have been identified by screening germplasm against TLB, and high levels of resistance have been reported in a number of inbred lines. Resistant sources and their utilization in regular breeding program has been reviewed by Welz and Geiger (2000). Sustainable management of turcicum leaf blight with more resistance sources and their origins has well demonstrated by Hooda *et al.*, (2017).

Conclusion

Turcicum leaf blight caused by the pathogen *Exserohilum turcicum* is one of the most devastating disease of maize crop and causes immense grain and fodder yield losses. The disease has worldwide distribution and its development is favoured by cool to moderate temperature with high relative humidity. Epidemics of the disease usually originate from mycelia and conidia of this fungus from infected crop residues left in farm fields. The prevalence of the disease has increased in recent years and new races of the pathogen

have been reported worldwide. The fungus *E. turcicum* is highly variable in nature. Identification of races of the pathogen present in an area and the understanding of their geographical distribution are important steps for the development of disease resistant cultivars in many host pathogen system where major genes control resistance. Though chemical measures are available for the control of the turcicum leaf blight disease, it is difficult to sustain. Further it has not been adopted particularly in marginal farming system under high altitude rainfed conditions. Most efficient and cost-effective way to manage the disease is to develop varieties with resistance against *E. turcicum*. The resistant sources with varied levels of resistance do exist against the Turcicum leaf blight disease. The determination of genetic basis of these sources and incorporation of their resistant genes may help in the development of high yielding TLB resistant maize cultivars.

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LeasyScan-an efficient phenotyping platform for identification of pre-breeding genetic stocks in maize

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Abstract: The huge genetic resources among all the crop species is still underutilized in meeting the worldwide challenges of agriculture production systems. In maize only 5% of world's maize germplasm has been used. The utilization of the maize genetic resources, which hold the answers to most of the threats and challenges, would be enhanced by their precise characterization and evaluation. Also, the data needs to be generated at a faster rate to meet the onset challenges. In this study, we discussed and demonstrated the use of an imaging phenotyping platform-LeasyScan, coupled with lysimeters, to measure precise plant height and canopy traits *viz.*, leaf area and leaf area index (LAI) affecting water use in six experimental and two released maize hybrids *viz.*, 14746185, 8315622, 22525674, 18270413, 4695575, 783527, 900MG and 30V92. Of these, experimental hybrids 2 & 5 i.e 8315622 4695575 showed promising 3D-leaf area and LAI. We conclude that LeasyScan –phenotyping platform can be effectively used in the identification of genotypes/germplasm lines with high vigour (Plant height), efficient 3D-leaf area and LAI at early stage of around one month old seedlings. Identification of such genetic stocks/ germplasm lines can be an important step towards effective utilization of the genetic resources in pre-breeding programme.

Keywords: Phenotyping · LAI · Germplasm · Screening · LeasyScan · Maize

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Introduction

The challenges to the agricultural production systems across the globe are diverse and complex with the changing environmental conditions which impacts severely on the crop yields (Tester and Langridge, 2010). The agricultural approaches and practices that contribute to climate change adaptation and mitigation have been listed by FAO (2015). As the dual challenges of climate change and increased demand of food production for the rapidly growing population, are immense, therefore we need high throughput and efficient technologies in finding out ways and means for mitigating the challenges and meeting the basic food demands. A huge untapped potential is the underutilized genetic resources among all the crop species, for example, in maize only 5% of world's maize germplasm has been used (Tabata and Eberhart, 2004). The utilization of the genetic resources, which hold the answers to most of the threats and challenges, would be enhanced by their characterization and evaluation data (Drinic and Andjelkovic, 2012). The generation of data on huge genetic resources at the international level, for example over 7.4 lakh accessions conserved in 11 international gene banks; and total germplasm accessions conserved worldwide, *viz.*, 7.4 million (Commission on Genetic Resources for Food and Agriculture, 2010) is highly desired. The data needs to be recorded as precisely and at a faster rate, as it involves huge germplasm collections. Further, broadening the genetic base of the crop species and use of the phenomics have been settled as the technologies to look up to in the changing world (Araus *et al.*, 2012). The way to genetic gain is being increasingly seen through effective phenotyping and phenomics approaches, this necessitates the amalgamation of high throughput technologies such as phenotyping and the utilization of genetic resources. The use of imaging phenotyping platform- LeasyScan, coupled

with lysimeters, to measure canopy traits affecting water use, *viz.*, leaf area, leaf area index, transpiration has been demonstrated in peanut, cowpea and pearl millet (Vadez *et al.*, 2015).

In maize, most of the yield improvement (about 75%) has been attributed to genetic gain and remainder to the improvement in agronomic practices (Araus *et al.*, 2012) further, interestingly, the genetic gain was not attributed to heterosis but to more stress tolerance (Duvick, 1999) relating to increased leaf area index and higher harvest index (Lee and Tollenaar, 2007). Bolaños and Edmeades (1996) concluded that yield variation explained more by the reproductive traits like HI than traits like leaf extension rate, canopy temperature, leaf erectness, leaf rolling, and leaf senescence which contributed to plant water status, water use and WUE. Leaf area and Leaf Area Index plays an important role in ultimate plant yield in maize as it has considerable influence on yield in maize (Lukeba *et al.*, 2013). The use of LeasyScan-a phenotyping platform for measuring and validation of 3D leaf area and the destructive leaf area in pearl millet, cowpea and peanut have been demonstrated (Vadez *et al.*, 2015). In this experiment using maize hybrids, we have attempted the use of LeasyScan phenotyping platform and to see if LAI can be used as a tool to identify potential early experimental hybrids for yield.

Materials and methods

Eight hybrids of maize, which included two released and six experimental hybrids, were made part of the LeasyScan phenotyping platform. The platform is equipped with a set of scanners (PlantEye F300, Phenospex, Heerlem, The Netherlands) which are made to slide above the plants using well established moving device that enabled the generation of 3D point clouds of the crop canopy structure. The leaf area and other parameters are derived through a process known as segmentation. The hybrids were sown as per the standard spacing of 60 × 20 cm in the lysimetric tubes fitted in the platform in a Replicated Block Design (RBD) with three replications. The sowing was done on 13th January 2016. The LeasyScan is equipped to record the

plant height, total leaf area (which is called 3D-leaf area) and the projected leaf area which is the equivalent to Leaf Area Index using the eye camera. The details on working of LeasyScan were explained by Vadez *et al.* (2015). The data on the plant height, leaf area and projected 3D-leaf area were recorded at 3-day interval up to one month beginning from one week after sowing. The data was subjected to analysis using the Wasp 2 (Web Agri Stat Package 2.0) available online at <http://www.ccari.res.in/wasp2.0/index.php>.

Results and discussion

The platform was designed, initially, with an aim to assess the range of genetic variation in leaf area, transpiration and transpiration rate (i.e. canopy conductance) for mapping and screening purposes (Vadez *et al.*, 2015). The analysis of variance for the eight hybrids of maize has been presented in table 1. There was no significant difference ($p > 0.05$) in the plant height among the maize hybrids, whereas the 3D-Leaf area and Leaf area index values recorded significant variation ($p < 0.05$) among the hybrids.

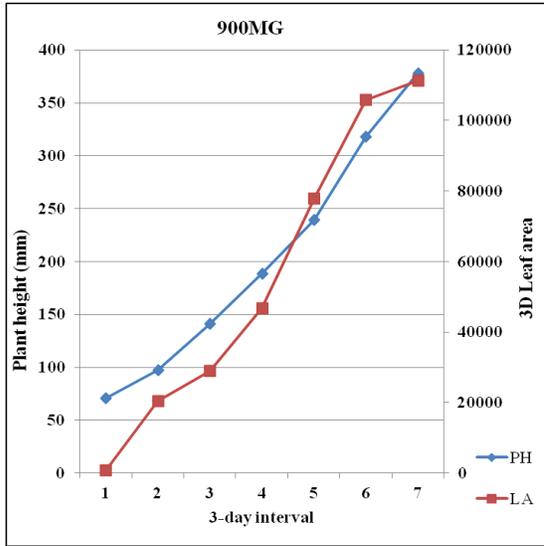
Taking the cue from the significant variation among the hybrids, we observed if there was any difference between the released hybrids-which are popular among the farming community for their high yield and unique leaf architecture. The leaf architecture of these hybrids also promotes high density planting resulting in high production.

3D-Leaf area

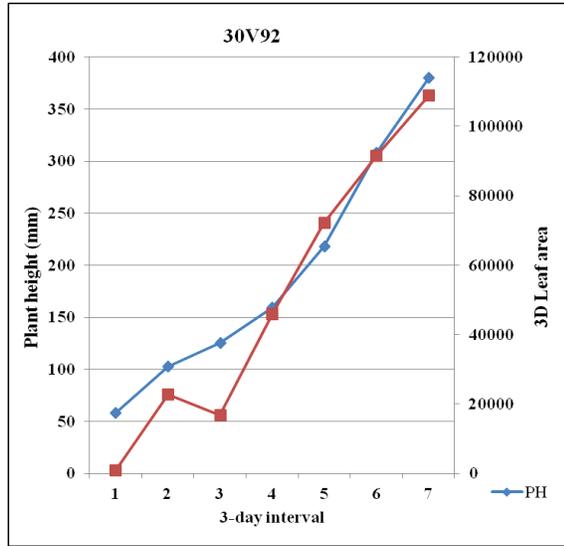
The two released hybrids, *viz.*, 900MG and 30V92 were characterized by high 3D-Leaf area values (Figure 1a to 1h). The 900 MG recorded value of 111230.4 at the end of 30 DAS, whereas 30V92 recorded a value of 108945. All the other experimental hybrids, except hybrids 2 (No.8315622) and 5 (4695575) which recorded a 3D leaf area value of 109296 and 103174, respectively, recorded values lower than 100000 mm². Hence, these two hybrids which recorded higher 3D-leaf area values may be selected as potential hybrids for promotion as they have fared on

Table 1. Analysis of variance for different traits recorded in maize hybrids

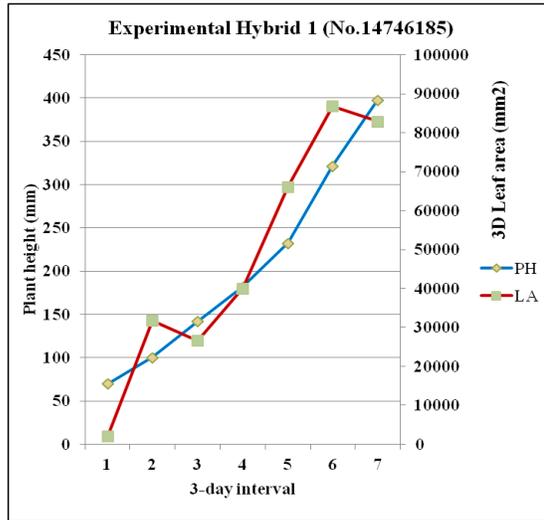
Source	df	Mean sum of squares		
		Plant height	3D-leaf area	Leaf area index
Replications	2	967.24	189042024.64	16163.60
Hybrids	7	895.22	838171802.45	45573.18
Error	14	718.78	102191266.61	4710.60



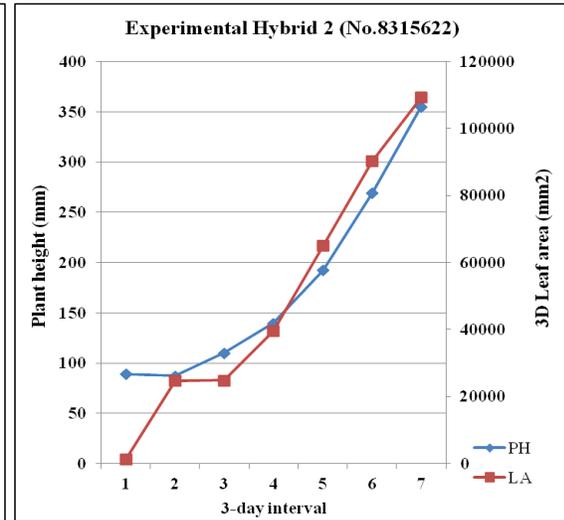
1a



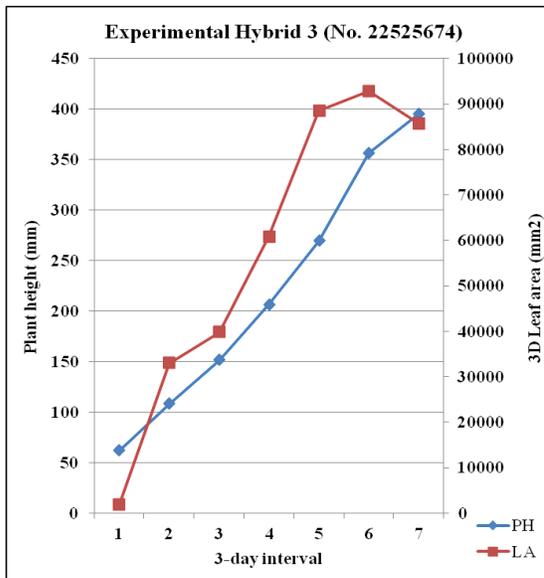
1b



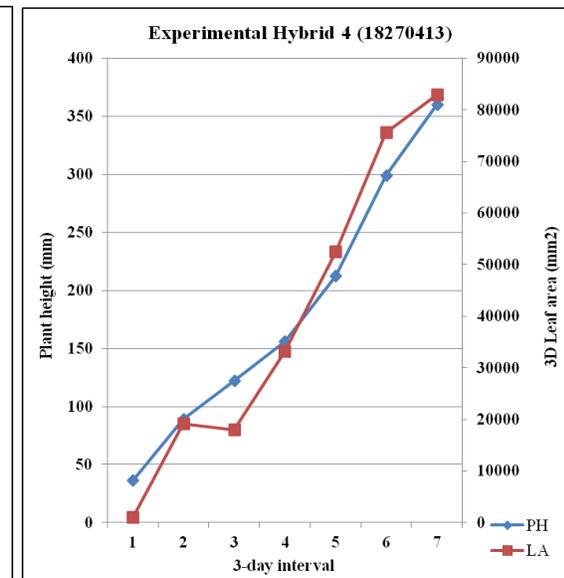
1c



1d



1e



1f

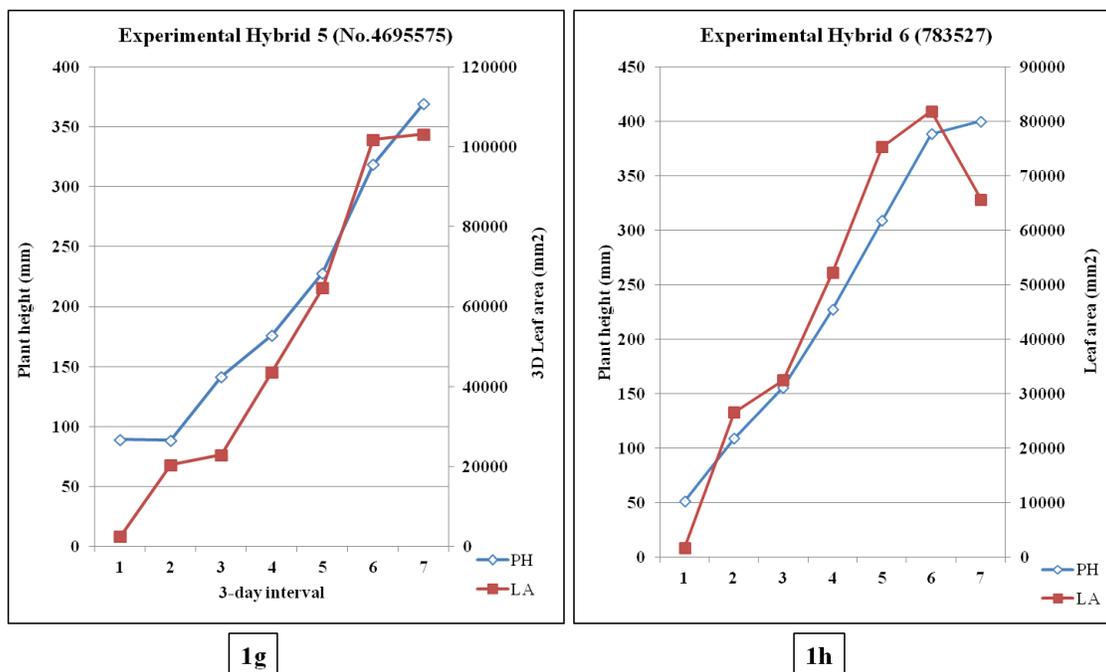


Figure 1a to 1h. Variation observed for plant height and 3D leaf area in maize hybrids

par with the highly popular released hybrids in case of 3D leaf area.

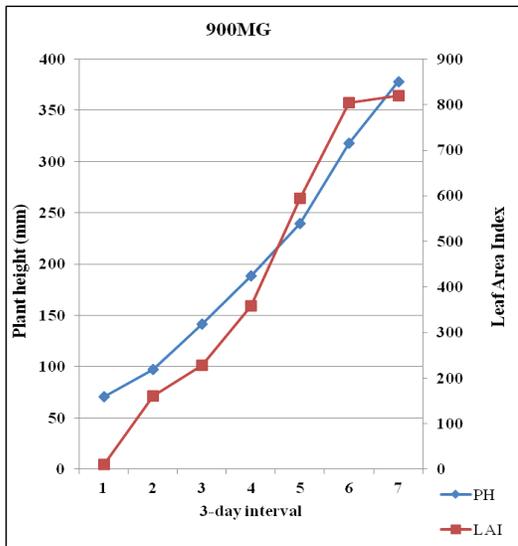
Leaf area index

Leaf area Index, is the Specific Leaf Area, which is considered as the key determinant for the growth rate of the species and system productivity (Norberg *et al.*, 2001) is one of the important agronomic traits that is used as an indirect measurement of yield. Asner *et al.* (2003) have termed LAI as the most important bio-physical factor in climatologically, meteorological, ecological, and agricultural modelling. It has also been used in yield forecasting using LAI based yield model (Baez-Gonzalez *et al.*, 2005). Although, destructive assessment of LAI, involving manual collection of leaves and direct measurement by Planimeters, has been used to with accurate results, but its main drawback is, it is time-consuming, as a result, expensive and often limited to small areas (Jonckheere *et al.*, 2004).

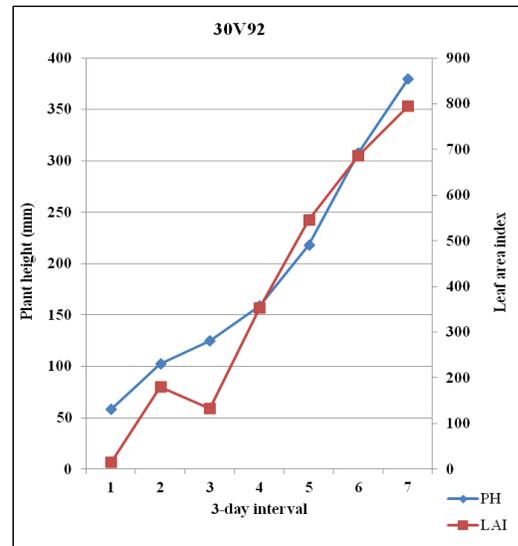
Consequently many indirect methods of measuring LAI, have been developed, grouped as *in-situ* and remote sensing approaches (Bauer *et al.*, 2016).

As in the 3D-leaf area, the LAI values in the released hybrids were superior to the experimental hybrids (Figure 2a to 2h). The LAI values recorded in 900MG was 820 and in 30V92 it was 794; Two of the experimental hybrids, viz., Experimental hybrid 2 (No. 8315622) and 5

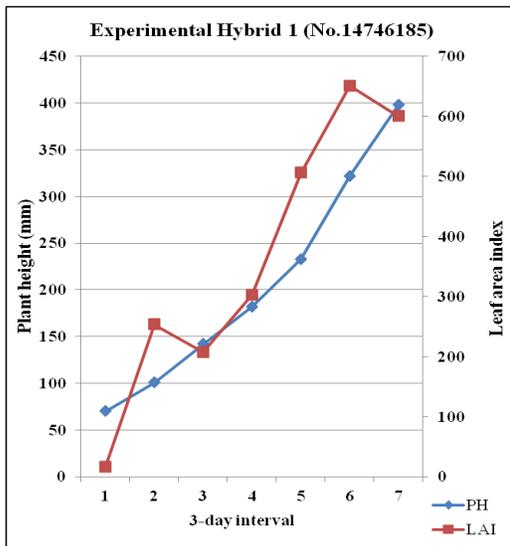
(No.4695575) recorded higher LAI. The LAI recorded in both these hybrids is more than 750. These hybrids may be selected as potential hybrids as they meet the higher LAI. A smaller leaf angle results in a more upright leaf orientation. This is beneficial for increasing the leaf area index, reducing maize shade syndrome and improving photosynthetic efficiency (Sakamoto *et al.*, 2006). High Density planting also promoted LAI, which resulted in higher light interception, higher Dry Matter Accumulation (DMA) and hence higher yields (Shi *et al.*, 2016). The QTLs (Li *et al.*, 2015) and genes (Zhang *et al.*, 2014) related to leaf angle in maize have been mapped and cloned. Identification of genotypes at an early stage, as with using the LeasyScan, can help us to identify genetic stock with good potential for high leaf area index through leaf angle. The benefits of high leaf area index post flowering, which is a result of delayed leaf senescence leading to improvements in maize grain yields have been reported in newer hybrids (Lee and Tollenaar, 2007). New methods like satellite based LAI yield forecasting have been attempted, although with error of 2-5%, but will prove useful in large areas avoiding laborious ground leaf area measurements (Baez-Gonzalez *et al.*, 2005). The importance of LAI as the main driving force of net primary production, water and nutrient use, and carbon balance and the impact of canopy LAI on the understory communities especially in the soil were dealt by Bréda



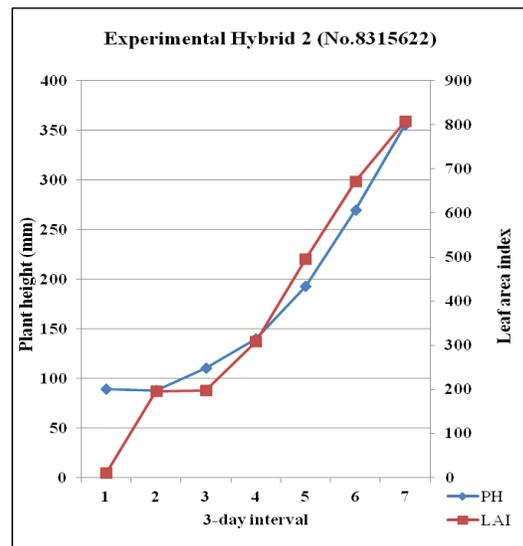
2a



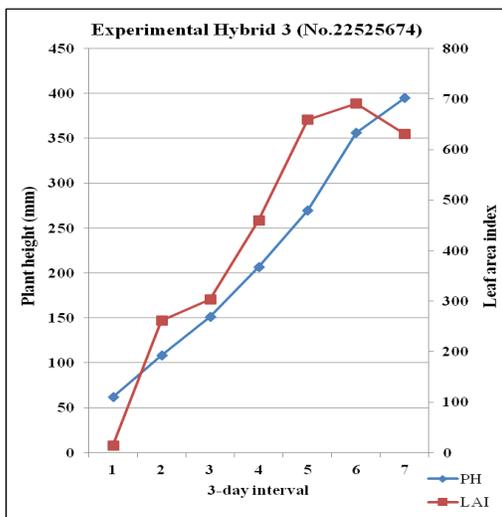
2b



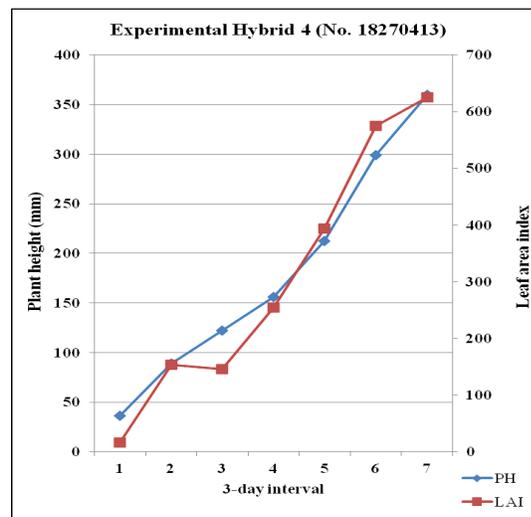
2c



2d



2e



2f

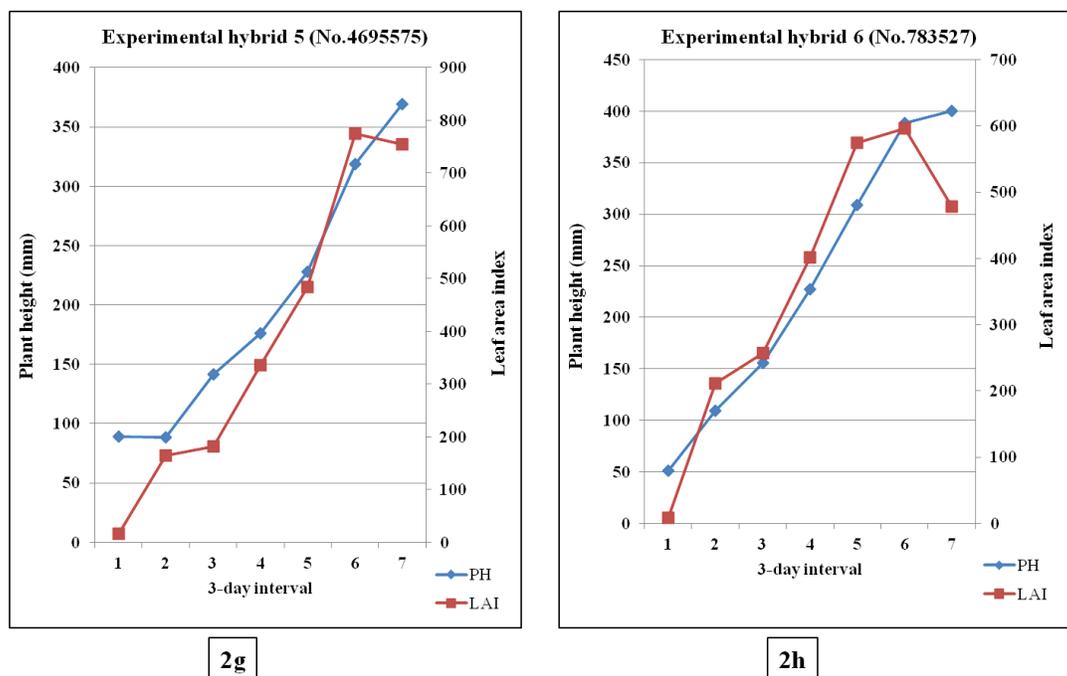


Figure 2a to 2h: Variation observed for plant height and leaf area index in maize hybrids

(2008). Ewert (2004) concluded that for estimating the future plant productivity under elevated CO₂ cannot be achieved without improved modeling of LAI.

Conclusion

The LeasyScan–phenotyping platform can be used in the identification of genotypes/germplasm lines with high vigour (Plant height), efficient 3D-leaf area and LAI. Identification of such genetic stocks/germplasm lines forms the important pre-breeding steps towards the effective utilization of germplasm lines.

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Studies on genetic variability, heritability and genetic advance in maize (*Zea mays* L.) for yield and its components

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Abstract: The present investigations were carried out during the *kharif* 2018 with the objective to estimate genetic variability for yield and yield contributing traits in 37 maize inbred lines. The analysis of variance revealed that all the treatments were highly significant for the yield and its contributing traits indicating sufficient variation among the treatment/materials under study. PCV values were higher than the GCV for all the characters indicating influence of environment. The traits grain yield per plant, ear height, cob length, plant height and test weight had higher GCV and PCV as well as high genetic and phenotypic variability. High heritability coupled with high genetic advance was observed for the traits grain yield per plant, ear height, cob length, plant height and test weight indicating the presence of additive gene action. Hence, emphasis should be given to select these quantitative traits to enhance the yield potential of maize.

Keywords: Genetic variability · Heritability · Genetic advance · Maize

Introduction

Maize (*Zea mays* L.; $2n=20$) is known as “queen of cereals” and it is a cross-pollinated crop with wider genetic variability and able to grow successfully all over India covering tropical, subtropical and temperate agro-climatic conditions. It is the primary staple food of many developing countries. India produced 26.26 million tons of maize during the year 2016-17 (Anonymous, 2017). Maize is an important food crop after rice and wheat in India. Maize grain serves as valuable nutrient feed for poultry and the green plant were used as forage or made into silage for dairy and beef industries. Grains are also used as basic raw material for number of industrial products from feed to fuel.

Maize and agriculture are still fundamental to the economic development of most of the Asian and African countries. In much of Asia, maize plays a central role in politics, society and culture, directly or indirectly employs more people than any other sector. A healthy maize industry, especially in Asia’s poorer countries, is crucial to the livelihoods of maize producers and consumers alike. Underpinning this, a strong maize research sector can help to reduce costs, improve production and ensure environmental sustainability. Indeed, maize research has been a key to productivity and livelihood.

Yield augmentation is the major breeding objective in maize breeding programme and knowledge on the nature and amount of the genetic variation governing the inheritance of quantitative traits like yield and its components is important for effective genetic improvement. The knowledge on the genetic variability parameters, such as phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability and genetic advance for different characters of economic importance is a main pre-requisite for any plant breeder to work with crop breeding programs. Hence, the present study was

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undertaken in this circumstance to elucidate information on variability, heritability and genetic advance of the promising maize genotypes. A good understanding of genetic resources might also assist in identifying desirable genotypes for upcoming hybridization program.

Materials and method

Utilization of natural genetic variability assist to meet short-term objective as often breeders have to meet instant requirement of the farmers, consumers and end-users (Gayatonde *et al.*, 2018). The present investigation was conducted during the *kharif* season 2018 at the Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi using thirty seven maize inbred lines, *viz.*, HUZM-185, HUZM-36, HUZM-53, HKI-536, HKI-162, HUZM-80-1, HKI-287, HKI-1105, HKI-193, HUZM-211-1, HUZM-97-1-2, HUZM-509, HUZM-121, HKI-323, HKI-164-4-(1-3)-2, HUZM-88, HKI-PC-8, HUZM-47, HUZM-60, V-25, CM-126, V-348, V-386, V-388, V-351, CM-212, HUZM-356, V-335, CM-145, V-336, M-341, CML-395, HKI-209, HKI-586, HKI-1352-5-8-9, CML-140 and CML-152. The experiment was laid out in randomized block design (RBD) with three replications and crop was maintained as per the standard agronomic practices. The field observations were recorded for 9 traits which includes days to tasseling (DT), days to silking (DS), anthesis-silking interval (ASI), Plant height (PH), ear height (EH), cob length (CL), days to maturity (DM), test weight (TW), grain yield per plant (GP).

The analysis of variance was completed as recommended by Panse and Sukhatme (1967). Phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and heritability broad sense (h^2) were calculated (Burton, 1952). Genetic advance (GA) was estimated by the method recommended by Johnson *et al.*,

(1955). Analysis of variance for all the traits was carried out by INDOSTAT software.

Results and discussion

The analysis of variance revealed significant variations over the characters under study exhibiting a wide range of phenotypic as well as genotypic coefficient of variation (Table 1). The relative magnitudes of the genotypic as well as phenotypic variances between the characters were compared based on the genotypic and phenotypic coefficient of variation (Table 2). PCV was recorded highest for grain yield per plant (41) followed by ear height (40.6), cob length (24.8), plant height (23.8) and test weight (20.7). Moderate PCV was exhibited by anthesis silking interval (14) and Low magnitude of PCV was recorded for days to 75% dry husk (4.0) followed by days to 50% silking (6.4) and days to 50% tasseling (7.0). Similarly, GCV was also high for grain yield per plant (40.5) followed by ear height (40.2), cob length (24.4), plant height (23.4) and test weight (20.1). Moderate GCV was exhibited by anthesis silking interval (10.8). Whereas, low magnitude of GCV was exhibited for days to 75% brown husk (3.1) followed by days to 50% silking (5.3) and days to 50% tasseling (5.8) (Table 2).

Heritability (broad sense) in the present study ranged from 59.3% to 97.9%. The highest heritability was registered for ear height (97.9) followed by grain yield per plant (97.7), plant height (96.5), cob length (96.3), test weight (93.9), days to 50% tasseling (67.4) and days to 50% silking (67.7). Whereas moderate heritability was observed for anthesis silking interval (59.3) followed by days to 75% dry husk (58.8) (Table 2).

High genetic advance as percent of mean was observed for grain yield per plant (82.5) followed by ear height (81.9), cob length (49.2), plant height (47.3) and test weight (40.1).

Table 1. Analysis of variance for yield and yield component traits in maize

Source	d.f	Days to 50% tasseling	Days to 50% silking	Anthesis and silking interval	Days to 75% brown husk	Plant height (cm)	Ear height (cm)	Cob length (cm)	Test weight (g)	Yield/plant (g)
Replication	2	8.2	11.2	3.4	155.6	161.2	21.9	1.4	9.7	6.4
Treatment	36	30.8	30.3	0.8	27.1	1859.4	1099.8	27.9	33.6	560.5
Error	72	4.3	4.2	0.2	5.1	22.3	7.6	0.4	0.7	4.4
SEm		1.2	1.2	0.2	1.3	2.7	1.6	0.3	0.5	1.2
C.V. (%)		4.0	3.6	9.0	2.6	4.5	5.8	4.8	5.1	6.2
C.D. 5 %		3.4	3.3	0.6	3.7	7.7	4.5	1.0	1.4	3.4
C.D. 1 %		4.5	4.4	0.8	4.9	10.2	6.0	1.3	1.8	4.5

Table 2. Estimation of genetic parameters for yield and yield components traits in maize

Traits	Mean	Range		PCV (%)	GCV (%)	h ² bs (%)	GAM
		Min.	Max.				
Days to 50% tasseling	51.7	42.8	55.7	7.0	5.8	67.4	9.7
Days to 50% silking	56.0	47.5	60.4	6.4	5.3	67.7	8.9
Anthesis and silking interval	4.3	3.0	5.3	14.0	10.8	59.3	17.2
Days to 75% brown husk	87.2	81.4	93.2	4.0	3.1	58.8	4.9
Plant height (cm)	105.9	75.7	161.1	23.8	23.4	96.5	47.3
Ear height (cm)	47.5	24.9	88.1	40.6	40.2	97.9	81.9
Cob length (cm)	12.4	6.4	17.1	24.8	24.4	96.3	49.2
Test weight (g)	16.5	8.8	20.9	20.7	20.1	93.9	40.1
Yield/plant (g)	33.6	15.8	60.0	41.0	40.5	97.7	82.5

PCV=Phenotypic coefficient of variation

GCV= Genotypic coefficient of variation

h² bs=Broad sense heritability

GAM= Genetic advance as per cent of mean at 5 per cent level

Moderate value was observed for the trait anthesis silking interval (17.2). Lowest value was registered by days to 75% dry husk (4.9) followed by days to 50% silking (8.9) and days to 50% tasseling (9.7) (Table 2). These findings are in agreement with that of Rajesh *et al.* (2013), Mahmood *et al.* (2004), Kumar *et al.* (2014).

The early flowering and maturity were seen in inbreds namely M-341, V-335, HKI-PC-8, CM-145, HKI-209 and V-348 whereas inbreds HKI-193, CML-152 and CML-140 exhibited late flowering and maturity. These inbreds can very well be utilized in developing early and late maturing hybrids based on the agro-ecological zones. Late maturity in maize is a healthier ideotype trait and in addition inbreds showing superior grain filling can be further used in various crop improvement programme. Kumar *et al.*, (2014) reported negative as well as positive value for earliness in maize inbreds. The negative significant value for plant height is preferable because dwarf plant stature is indispensable to utilize for directed biomass buildup in cobs, even thought to be lodging resistant.

The amount of genetic variability determines the effectiveness of selection. The variability should be heritable so that it can be used in the breeding programme. The heritable portion of the whole of observed variation can be determined by studying the components of variation like PCV, GCV, heritability and predicted genetic advance. In this study, the estimates of PCV were higher than their respective GCV for all the characters studied. These findings are in close agreement with the findings of researchers Bharathiveeramani *et al.*, (2012), Rajesh *et al.*, (2013), Mahmood *et al.*, (2004), Kumar *et al.*, (2014).

The PCV and GCV were high for grain yield per plant followed by ear height indicating that these characters were under the main influence of genetic control and less variable as a result of environmental factors. So, such characters are essential for further improvement. The similar results were reported by Ali *et al.*, (2007) and Nagaraju (2012). The GCV often provides some indication pertaining to validity of characters for selection. But it does not give clear picture of the extent of genetic gain to be achieved from selection of phenotypic characters, unless heritable portion of variation (heritability) is recognized (Burton, 1952). The difference between the estimates of PCV and GCV were small for almost all the characters except ASI, indicating less influence of environmental factors in expression of these characters suggesting phenotypic differences may be measured as genetic difference among genotypes for selection.

This also shows that *per se* performance of these characters should not be used directly on the basis of selection and variability for these traits however heritability may also be taken into account. In the present study, high heritability was observed for most of the traits except few characters. Heritability and genetic advance are indispensable selection parameters. Heritability values beside with genetic advance are generally more helpful in predicting the gain under selection than heritability values alone and shows presence of additive variance or Additive × Additive kind of interaction. It is not obligatory that a trait showing high heritability will also show high genetic advance. The breeder should be careful during selection based on heritability as it shows both non-additive and additive gene action.

Therefore, heritability estimates together with genetic advance would be more reliable and meaningful in making selection procedure as it shows that most probably the heritability is due to additive gene action. In the current set of materials, high heritability together with high genetic advance as percent of mean was observed for ear height, grain yield per plant, cob length, plant height and test weight, demonstrating effectiveness of selection for the development of these characters while high heritability along with low genetic advance exhibited by traits days to 50% silking and days to 50% tasseling are indicative of non-additive gene effects. High heritability along with high genetic advance may be recognized as additive gene effect (Addisu and Addisu and Shumet, 2015). The high heritability is being showed due to positive influence of environment rather than genotype and selection only for such characters may not be fruitful for further progress. Ali *et al.* (2007) and Krishna *et al.* (2010) also indicated such results in their work adding value to the present findings.

Conclusion

Analysis of variance for 37 maize inbreds revealed the existence of significant differences among them for all the characters. The majority of traits showed close PCV and GCV estimates except ASI. This indicates the meagre influence of environment on these characters which are useful to improve yield *per-se*. Five traits exhibited high heritability coupled with high genetic gain under selection indicating existence of additive variance and more scope for selection. Majority of the characters also showed high expected mean yield in the subsequent generation, demonstrating prediction is the better choice if at all the objective is to select the inbreds for double- or three-way crosses.

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Combining ability and heterosis in early maturity maize (*Zea mays* L.)

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Abstract: Combining ability studies among the newly developed early inbred lines identified DMEI-70 and DMEI-195 as early based on the significant and high General Combining Ability (GCA) effects (-19.03) and (-28.57) in negative direction for days to 50% tasseling. Similarly, the line DMEI-207 recorded significant and high GCA effects (23.11) for grain yield followed by DMEI-165. Among the crosses the contribution of lines × tester interaction for grain yield was high (54.91%) as compared to lines (44.51%) and tester (0.58%) indicating that importance of hybrids for exploitation of heterosis. The cross combination of DMEI-70 × CM-500 recorded significant Specific Combining Ability (SCA) effect for days to 50% tasseling (23.9) and was early by 5-6 days over PEMH-2 with highest mean grain yield of 75.55 q/ha and standard heterosis of 46.0%. Further, all possible cross combination among the selected lines from the two heterotic groups resulted in identification of the cross DMEI-165 X DMEI-73 as very promising hybrid combination with 100 q/ha grain yield and was also early in flowering by 5 days over check hybrid PEMH-2.

Keywords: Maize · Line × tester combining ability · Grain yield · Non-additive

Introduction

Maize is an important cereal crop in Karnataka with an area of 1.35 m ha and *kharif* is the major cropping season for maize. Majority of the area is under rainfed situation and is entirely occupied by hybrids with full season maturity (115-120 days duration). Under rainfed situation with the changing climatic scenario, presently the cultivated hybrids show susceptibility to terminal moisture stress and the situation gets further aggravated with delayed onset of monsoon. Further, these late maturing hybrids are also not suitable for the double cropping system which is very important from the point of farmer's income. However, the late maturity hybrids have yield advantage. Since, the area under rainfed maize is increasing every year, this along with delayed onset of monsoon will hamper maize production, farmers cropping system and sustainability. Under such situation, maize hybrids with earliness and appreciable level of heterosis for yield and resistance to Turcicum leaf blight (TLB) would be a boon to the farmers. Therefore, in order to address the issue, early maturing hybrids developed under AICRP system were evaluated at Dharwad (Karnataka) but, majority of these promising hybrids were found to be susceptible for the major foliar disease TLB. Hence, with this background information a breeding programme was initiated on development of high yielding hybrids which can adapt to local environment with earliness and resistance to TLB.

Materials and method

The present study was initiated at Agriculture Research Station, Arabhavi during *kharif* 2010. To begin with advanced varietal trial-II- of early maturity maize was the source material and the promising genotypes with TLB resistance were selfed to derive inbred lines. The selfed

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ears were planted as ear to row for selfing and advanced up to S_4 . At S_4 generation 117 lines were selected based on earliness, *per se* and uniformity of the line. In order to identify the promising lines, they were crossed with two lines belonging to late and early maturity during Rabi/Summer season of 2014-15 and this top cross nursery was evaluated at Main Agriculture Research Station, Dharwad during *kharif*, 2015. Based on top cross nursery evaluation only top 10 potential lines with high yielding ability and earliness as the main criteria were selected.

The selected lines were again crossed with two known testers CM-111 and CM-500 in line \times tester method (Kempthorne, 1957) in order to know the combining ability and to group these lines into different heterotic groups. The crosses thus generated were evaluated during *kharif* 2016 in a replicated trial along with early maturity hybrid checks (PEMH-2) at Main Agriculture Research Station, UAS, Dharwad under rainfed situation with one protective irrigation. Based on the combining ability status of the lines, SCA and standard heterosis, the lines were classified into two heterotic groups (A & B) and those lines with desirable GCA effects were crossed in all possible cross combination and were evaluated at AICRP-Maize, Dharwad during *kharif* 2017.

Each test hybrid was raised in two rows of 4.0 m length with a spacing of 60 cm between rows. Planting was done using two seeds per hill with a spacing of 20.0 cm between hills. The recommended package was followed to raise the healthy crop. Thinning was done at three to five leaf stages to have a optimum plant stand. Observations were recorded on days to 50 per cent tasselling, cob length, cob diameter, number of kernel rows and number of kernels/row and grain yield. The compiled data was analyzed using TNAU STAT statistical package.

Results and discussion

From the Analysis of variance, it was possible to know the

existence of significant differences among the parents and hybrids for all the morphological traits (Table 1). This indicated that the experimental genotypes had sufficient genetic variability for all the characters under study. Analysis of variance for combining ability revealed that the lines exhibited differences in their combining ability for all characters. Whereas, the testers also consistently differed in combining ability for all the traits except yield. Similarly, the interaction of line \times testers was also significant for all the traits indicated that the parental lines were highly diverse (Table 1). Sumalini *et al.* (2014) also opined that the lines behaved differently for all the traits were reported greater diversity among the lines in their study on line \times tester in maize over the environments. The combining ability analysis facilitates partitioning of genotypic variation of the crosses into variation due to GCA (main effects) and SCA (interaction). Such information on grain yield and associated traits would greatly help in designing strategy for breeding better genotypes in maize.

Among the genetic parameters, for days to 50% tasselling which is the indicator for earliness most of the new crosses were early as compared to the check. Hence, these hybrids met the criteria of mean \pm 1.5 days over early check hybrid PEMH-2 used in AICRP trials for identifying early maturing hybrids.

The GCA and SCA effects are the main criteria used for selection and classification of parents in terms of their potential performance in various cross combinations. GCA effects for grain yield showed that DMEI-207 to be the best general combiner with consistent and significant GCA effects in positive directions for all the traits (Table 2) followed by DMEI-165, DMEI-196, and DMEI-73. The desirable GCA effects for grain yield have been also reported earlier by Srivastva and Singh (2002), Todkar and Navale, (2006) and Kumar and Kumar (2014). However, DMEI-195, DMEI-134, and DMEI-70 also recorded significant GCA but in negative direction for grain yield

Table 1. ANOVA for combining ability in newly developed early maturing maize lines

Source	df	Yield (q/ha)	Cob length (cm)	Cob girth (cm)	No of Kernel rows	No of Kernels /row	Days to 50% tasselling
Replication	2	134.7	3.97	0.01	0.86	8.6	10.16
Cross	19	2036.6 *	120.16*	7.95*	86.25*	493.9*	1225.9*
Lines	9	1913.6 *	120.0*	6.16*	80.32*	432.4*	1107.7*
Tester	1	223.9	41.0*	3.7*	21.6*	132.01*	355.2*
Line x Tester	9	2369 *	129.0*	10.2*	99.37*	595.6*	1440.89*
Error	38	74.9	0.76	0.03	1.84	7.4	3.4
Total	59						

*Significant at 0.05 %

Table 2. General combining ability (CGA) of the lines

Lines	Yield (q/ha)	Cob length (cm)	Cob girth (cm)	No of kernel rows	No of kernels / row	Days to 50% tasselling
DMEI-68	-5.23	4.19**	0.76**	1.53**	5.12**	11.67**
DMEI-70	-19.03**	-4.86**	-1.53**	-5.13**	-10.05**	-20.33**
DMEI-73	12.95**	0.91	0.66**	1.87**	2.62*	7.83**
DMEI-134	-22.22**	-6.27**	-1.39**	-5.13**	-13.05**	-18.17**
DMEI-135	6.88	1.09**	0.13	1.87**	2.12	10.0**
DMEI-165	17.17**	2.84**	0.73**	3.53**	5.28**	6.50**
DMEI-173	4.71	1.21**	0.97**	2.53**	5.45**	8.0**
DMEI-195	-28.57**	-7.27**	-1.38**	-5.47**	-12.72**	-20.17**
DMEI-196	10.23**	3.13**	0.39**	2.20**	6.12**	7.67**
DMEI-207	23.11**	5.03**	0.66**	2.20**	9.12**	7.0**
Tester						
CM-111	-1.93	-0.83**	-0.25**	-0.60*	-1.48**	-2.43**
CM-500	1.93	0.83**	0.25**	0.60*	1.48**	2.43**

*Significant at 0.05 %; **Significant at 0.01 %

and other traits which is undesirable. For earliness i.e., days to 50% tasselling DMEI-70 recorded significant GCA effect (-19.03) in negative direction which is desirable followed by DMEI-195 and DMEI-134 respectively.

For other traits, DMEI-207, DMEI-196 and DMEI-165 were observed as consistently good general combiners for cob length. DMEI-173 and DMEI-68 were good general combiners for cob diameter, however the values were very low. For kernel rows per ear, DMEI-207 and DMEI-196 had significant GCA values. Overall, DMEI-207, DMEI-196 and DMEI-165 were found to be good general combiners for all the traits. High GCA effects may be due to additive effects and additive \times additive gene effects. So these parents could be used in hybrid breeding programme aimed for increasing grain yield or to develop populations with favourable alleles. Based on their GCA effects for different characters, the parental lines can be classified as good / high, average / moderate and poor / low general combiners.

The variance of SCA was higher than the GCA for all the traits indicating preponderance of non-additive gene action in the inheritance of the traits (Table 3). This fact is also supported by low GCA to SCA variance ratio. This suggests that greater importance of non-additive gene action in expression of traits. The results were in accordance with previous results of Akbar *et al.* (2008) and Dinesh *et al.* (2016). The proportional contribution of lines, testers and their interaction to the total variance showed that lines played an important role towards the trait manifestation. The contribution of lines for earliness was to the extent of

42.8% as compared to the testers (1.53%) indicating that the derived lines were early in maturity. The contribution of lines \times tester interaction to the total variance was greater than the tester alone for all the characters indicated higher estimates of variance due to specific combining ability. Similar results were also reported by Hussain *et al.* (2006) and Dinesh *et al.* (2016). The identified best inbred lines can be used in development of early maturing hybrids with high yielding ability. These lines show high potential to transfer desirable traits to their progenies.

The SCA effects are associated with dominance and epistatic components of variation i.e. mainly non-fixable components of variation. Significant specific combining ability is the indication of relative importance of interactions in determining the performance of single crosses. Out of the twenty crosses evaluated only three cross combinations, *viz.*, DMEI-70 \times CM-500, DMEI-134 \times CM-111 and DMEI-173 \times CM-111 were observed to possess desirable specific combining ability for days to 50% tasselling and grain yield along with significant standard heterosis of 46.07, 36.53 and 10.45 per cent over early check PEMH-2 (Table 5). However, the cross combination of DMEI-70 \times CM-500 also recorded significant SCA for cob length, number of kernel rows, number of kernels/row and days to 50% tasselling apart from grain yield. Presence of desirable SCA effects and high performance for most of the traits was also confirmed from the studies of Dar *et al.* (2015). The cross between DMEI-134 \times CM-111 showed significant SCA for No. of kernel rows, number of kernels/row and days to 50% tasselling. Likewise, the cross

Table 3. Genetic components of variance and contribution of lines, testers and line × tester to total variance

Lines	Yield (q/ha)	Cob length (cm)	Cob girth (cm)	No of kernel rows	No of kernels / row	Days to 50% tasselling
GCA	-7.33	-0.2	-0.05	-0.2	-2.3	-4.8
SCA	762.0	42.77	3.4	32.5	196.0	479.15
GCA/ SCA	0.0096	0.046	0.0147	0.0061	0.0117	0.01
Lines	44.51	47.22	36.6	44.1	41.47	42.8
Testers	0.58	1.8	2.45	1.32	1.47	1.53
Lines x Testers	54.91	50.89	60.89	54.57	57.12	55.67

Table 4. Mean grain yield of the diallele crosses of the selected lines

Pedigree	Mean grain yield (q/ha)	Plant height (cm)	Shelling %	Days to 50 % tasselling
DMEI 165 × DMEI 73	100.1	154.7	87.0	55
DMEI-73 × DMEI- 196	97.8	159.7	84.9	57
DMEI 73 × DMEI 207	92.9	154.7	85.9	57
DMEI 196 × DMEI 207	90.2	160.7	85.0	56
DMEI 165 × DMEI 196	88.4	158.0	84.0	55
DMEI 165 × DMEI 207	86.5	163.7	84.9	57
DMEI 73 × DMEI 173	85.6	168.7	85.4	57
DMEI 196 × DMEI 73	83.9	148.0	83.3	56
DMEI 73 × DMEI 165	83.8	174.7	86.1	55
DMEI 207 × DMEI 196	77.5	170.7	84.7	58
DMEI 173 × DMEI 196	73.9	163.0	83.7	57
DMEI 173 × DMEI 73	70.3	154.7	85.0	58
DMEI 173 × DMEI 165	68.9	186.0	84.9	55
DMEI 165 × DMEI 173	65.8	163.3	85.2	55
Checks				
PEMH-2	71.8	165.3	81.9	60
PEMH-5	51.7	168.7	83.7	60
S.Em±	9.20			1.06
CD (@0.05)	26.16			3.0
CV (%)	18.19			3.2

combination of DMEI-173 × CM-111 recorded significant SCA for days to 50% tasselling. The crosses with high positive and significant SCA effect could be selected to use in maize population improvement program as opined by Abrha *et al.* (2013).

However, among the three crosses the cross combination of DMEI-70 × CM-500 was early in maturity by 7.0 to 8.0 days over PEMH-2 and also recorded higher grain yield of 75.55 q/ha with significant SCA and standard heterosis for grain yield. This study also revealed that GCA effects of the parents were not reflected in the SCA effects of the crosses as the promising hybrids identified in this study were with parents of low x moderate and low x low GCA type. Thus, in most cases, crossing the two good

general combiners may not necessarily result into a good specific combination and the same was true for certain poor combinations which involved one good combiner. These findings contradict the reports of Paul and Duara (1991) who have reported that parents with high GCA always produce with high estimates of SCA. However, the findings in the present study are supported by the results obtained by Kumar and Kumar (2014).

The experiment was further continued by grouping the lines into two Heterotic groups based on SCA and standard heterosis. The top 2-3 lines from each of the group with significant SCA effects and standard heterosis i.e., DMEI-165 and DMEI-196 (Het Grp A), DMEI-207, DMEI-73 along with one more promising line DMEI-173 (Het.grp

Table 5. Mean, SCA and standard heterosis of top three hybrids and their performance for yield contributing traits and earliness along with checks and testers

Treatments	Yield (q/ha)			Cob length (cm)			Cob diameter (cm)			No. of Kernel rows			No. of Kernels/rows			Days to 50% Tasselling		
	Mean	SCA	Std. Het (%)	Mean	SCA	Std. Het (%)	Mean	SCA	Std. Het (%)	Mean	SCA	Std. Het (%)	Mean	SCA	Std. Het (%)	Mean	SCA	Std. Het (%)
DMEI-70 × T2	75.55	35.84**	46.0**	18.57	8.4**	14.85	4.37	1.9**	-4.38	14.0	6.4**	-25.0	38.0	17.52**	20.0**	52.0	23.9**	-7.6**
DMEI -134 × T1	69.19	36.53**	33.7**	15.53	8.6**	-2.68	4.63	2.5**	1.46	14.0	7.6**	-25.0	32.0	17.48**	1.05	57.0	30.9**	0
DMEI-173 × T1	70.03	10.45**	35.4**	15.2	0.98	-4.12	4.53	0.1	-0.73	16.0	1.93*	-14.2	36.0	2.98	13.68	55.0	2.70*	-3.51
PEMH-2 (Early Check)	51.72			16.2			4.6			18.7			31.7			57.0		
T1 (CM-111)	21.32			13.0			3.3			12.7			26.3			58.0		
T2 (CM-500)	30.03			15.0			4.5			16.0			23.7			59.0		

*Significant at 0.05 %; **Significant at 0.01 %

B) were crossed in all possible combination to generate the heterotic hybrids and these were evaluated in a replicated trial along with other combinations during *kharif* 2017. From the data (Table 4) it was possible to identify the cross combination of DEMI-165 × DMEI-73 as heterotic hybrid with highest grain yield of 100.0 q/ha and was also early w.r.t. days to 50% tasselling by five days over PEMH-2. Overall, among the top five hybrids, the female parent was either DMEI-165 or DMEI-73.

Conclusion

The general and specific combining ability are the main criteria for rapid assaying of test genotypes under line x tester analysis and a comparison of the combining ability effects of the parents and their corresponding crosses indicated that the GCA effects of the parents were not reflected in the SCA effects of the crosses for most of the traits studied. Thus, in most cases, crossing the two good general combiners may not necessarily result into a good specific combination and the same was true that crossing two poor combinations will not yield always poor hybrids. Hence, based on the present results, it could be concluded that the production of hybrids based on the parental performance was not practically true. The cross combinations observed as good specific combiners can be utilized as such under heterosis breeding or in obtaining desirable recombinants/segregates as evident from the present study that DMEI-73 and DMEI-165 resulted in high yielding heterotic early maturity hybrids.

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Heterotic studies for grain yield and quality parameters in Pop corn

Goutam Rana · Preeti Sharma · Mehar Chand Kamboj · Narender Singh

Abstract: In the present study eight inbred lines, viz., HKI PC-1, HKI PC-3, HKI PC-4, HKI PC-7, HKI PC-4B, HKI PC BT-3, HKI PC-8B and HKI PC-1473-5, were used to develop twenty-eight single cross pop corn hybrids by using half diallel mating design during *Rabi* 2014-15. These twenty-eight single cross popcorn experimental hybrids along with their eight parental lines and two standard checks, viz., one of pop corn (Bajaura Popcorn) and another of normal maize (HM 4) were sown in a randomized block design with 3 replications during *kharif* 2015 at research farm of CCS Haryana Agricultural University, Regional Research Station, Karnal. Observations recorded for fifteen yield and quality characters and data recorded was analyzed to assess economic heterosis over two standard checks. Major findings of the experiment revealed significant difference due to mean sum of squares values for all the characters except final plant stand per plot, which indicated presence of enough genetic variability in the experimental material. Twenty one cross combinations showed positive heterosis for grain yield against check 1 (pop corn check) but none of the cross showed positive heterotic response against check 2 (normal maize check). Highest economic heterosis for grain yield per plot was exhibited by the cross HKI PC-8B × HKI PCBT-3 (66.74%) followed by HKI PC 7 × HKI PCBT-3 (52.02%), HKI PC 8B × HKI PC 1473-5 (49.38%), HKI PC-3 × HKI PC-7 (30.22%), HKI PC-7 × HKI PC-8B (30.17%), HKI PC-4 × HKI PC-8B (27.92%), HKI-1473-5 × PCBT-3 (26.57%) and HKI PC-7 × HKI PC-4B (24.44) against pop corn check (Bajaura pop corn) indicating their suitability to increase grain yield. The crosses HKI PC 1473-5 x HKI PCBT-3 and HKI PC-7 × HKI PCBT-

3 showed high heterosis for grain yield for popping quality parameters (popping per cent and popping volume) showing simultaneous improvement of grain yield and popping quality. These hybrids may be exploited commercially for maximizing productivity after multi location testing.

Keywords: Diallel mating design · Heterosis · Maize · Popcorn

Introduction

Globally, maize is referred as ‘Miracle crop’ or ‘Queen of the Cereals’ due to its high productivity potential compared to other Poaceae family members. In terms of area and production, it ranks next to wheat and rice while in yield, it surpasses all the cereal crops. As per fourth advance estimate, in India, area under maize was 9.50 million ha with total production of 26.30 million tonnes and productivity of 2630 kg/ha (Anonymous, 2018). India is also on threshold of maize revolution and states including Haryana has an ample scope to increase its acreage and productivity. Maize is one of the most diverse grain crop found in nature. The most common types of corn include flint, dent, floury, sugary, waxy and popcorn. Amongst these, the most popular is the “popcorn” type of corn (*Zea mays var. everta*) that puffs up when heated. Its consumption has greatly increased in recent years because of the advent of microwavable popcorn and proliferation of flavoured, ready- to- eat products. The popping process not only retains the actual nutritional profile of grains but also enhance its protein digestibility, bioavailability of iron and dietary fiber content. Popping also reduces some of the anti nutrients, viz., phytates, tannins, acid detergent fiber, lignin and cellulose (Reddy *et al.*, 1991). Producers and sellers of popcorn consider two major factors in evaluating the quality of popcorn, viz., popping percent and popping volume. The expansion or volume is more

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important for both the vendor and consumer. The popping volume is the ratio of pop volume (ml) per weight (g) of the popped kernel. For the consumer, large sized popcorn being tender is associated with higher quality while for producer, distributor and vendor expansion is closely related to profit. Vendors buy popcorn by weight and sell it by volume hence higher expanding popcorn fetches higher profit per unit weight.

Currently in India, popcorn is commercially grown on a fairly small scale. All the commercially grown varieties of popcorn in India are composite varieties, *viz.*, Amber popcorn, Jawahar popcorn and VL popcorn with low yield and less popping quality. Modern plant breeding aims to improve yield and quality together. The popping volume and yield are the traits which are controlled by hereditary factors. It is difficult to obtain superior genotypes for both traits but it is possible to develop genotypes with good popping volume and satisfactory yield (Pajic *et al.*, 2008). The genetic improvement work in popcorn is neglected in India and there is good scope for its improvement in near future. Looking at this fact, there is a need to develop single cross hybrids of popcorn for higher grain yield with better popping quality. For the development of hybrids, the foremost step is the development and evaluation of the inbreds and to know the nature and magnitude of heterosis for grain yield and its component traits (Sharma *et al.*, 2015; Sharma *et al.*, 2017). Therefore, for the development of pop corn hybrids with high yield and good popping quality, it is necessary that we screen our pop corn inbred lines for their genetic potential for yield, popping quality and economic heterosis. In popcorn, enhanced level of heterosis for grain yield and popping expansion has been reported by previous researchers. Therefore considering the above facts, the present investigation has been undertaken with the following objectives (a) to determine extent of heterosis in respect of yield and its component traits and quality traits and (b) to identify best experimental hybrids having balanced level of grain yield and better popping quality.

Materials and methods

The 28 experimental hybrids were generated by crossing, in half diallel mating design, eight inbred lines of popcorn, *viz.*, HKI PC-1, HKI PC-3, HKI PC-4, HKI PC-7, HKI PC-4B, HKI PC BT-3, HKI PC-8B and HKI PC-1473-5 during *rabi* 2014-15. During *kharif* 2015, the final experiment involving eight parents (inbred lines of popcorn), two

checks (HM 4 hybrid of normal maize and Bajauara popcorn cultivar of popcorn) and 28 F₁s were grown in Randomized Block Design with three replications in one environment, on July 10 (timely sown) 2015, at the experimental area of CCS Haryana Agricultural University, Regional Research Station, Uchani, Karnal. The entries were sown in a two row plot of 4 m with inter and intra-row spacing of 60 cm and 20 cm, respectively. Recommended agronomic practices were adopted to raise a good crop.

From every row, five competitive plants were randomly selected from each replication and observations on following 15 quantitative characters, *viz.*, days to 50% tasseling, days to 50% silking, days to maturity, plant height (cm), first cob placement (cm), final plant stand per plot (numbers), number of cobs per plot, cob weight per plot (kg/plot), shelling percent, grain yield per plot (kg/plot), 100 grain weight (g), moisture (%), popping volume (cm³/g), popping (%) and grain protein content were recorded on plants in each replication. The data on days to 50% flowering and maturity were taken on the plot basis and quality traits were determined by adopting following methods.

Popping volume

Twenty five gram of grain were collected from each replication. These were popped by traditional method on an iron tawa over an open flame. After popping, these popped corn were transferred to a flask and its volume was recorded in cm³. The popping volume was recorded by following formula in cm³/g.

$$\text{Popping volume (cm}^3\text{)} = \frac{\text{Volume of 25 g popped corn}}{25}$$

Popping

After recording the data on popping volume the data was recorded on total grains and no. of popped grains in 25g sample and the popping (%) was recorded by following formula

$$\text{Popping percent} = \frac{\text{No. of popped grains}}{\text{Total of popped grains}} \times 100$$

Shelling

After recording the data on cob weight per plot, five randomly selected cobs were taken and total weight of these five cobs was recorded. These five cobs were

threshed and grain weight was recorded by the following formula.

$$\text{Shelling (\%)} = \frac{\text{Grain weight of five cobs}}{\text{Total weight of five cobs}} \times 100$$

Statistical analysis

The Analysis of variance was carried out using mean values of observations on five randomly selected plants for each character. To know the precise genetic worth of crosses, the magnitude of heterosis for yield, its contributing characters and grain quality traits were estimated in relation to better parent and two standard check hybrid, viz., HM 4 hybrid of normal maize and Bajaura Popcorn cultivar by using formula:

$$\frac{\bar{F}_1 - \bar{CC}}{\bar{CC}} \times 100$$

Where CC = mean performance of commercial cultivar
 F₁ = mean performance of a cross

Results and discussion

The mean sum of squares for all the traits presented in Table 1 revealed that the mean sum of squares due to genotypes were highly significant for all the traits except final plant stand per plot which further indicated that the material selected for the present investigation was quite appropriate for further genetical analysis as considerable amount of variability existed in the experimental material under investigation. The values of mean sum of squares were found non significant for final plant stand per plot which indicated that the experiment was conducted properly and difference in the values of traits are due to genotypes, not due to differences in plant population. The *per se* performance of inbreds and hybrids along with their range for different characters presented in Table 2 (a) and 2 (b) revealed scope of improvement through hybridization. The pertinent results based on *per se* performance of hybrids and check varieties and percentage of heterosis measured as increase or decrease over checks (Bajaura popcorn, check-1 and HM-4, check-2 of normal maize) have been presented in Table 3. In our study several hybrids were found to exhibit substantial amount of heterosis over the standard checks for grain yield and quality parameters.

Among all the characters studied, only one character *i.e.* final plant stand per plot showed that population in all

Table 1. Analysis of variance in respect of 15 characters in popcorn genotypes

Source of variation	df	Days to tasseling	Days to 50% silking	Days to maturity	Plant height (cm)	First Cob placement (cm)	Final plant stand per plot (numbers)	No. of cobs per plot	Cob weight per plot (kg/plot)	Shelling (%)	Grain yield (kg/plot)	100 grain weight (g)	Moisture (%)	Popping (%)	Popping volume (cm ³ /g)	Grain protein content (%)
Replication	2	12.93	12.34	19.84	918.09	235.15	3.11	58.18	0.17	8.44	0.26	0.05	0.28	11.45	0.26	0.78
Treatment	35	8.29**	10.69**	19.74**	1836.19**	393.39**	1.01	87.55**	1.33**	15.33**	2.92**	27.46**	3.70**	21.18**	13.30**	1.48*
Error	70	0.16	0.21	0.24	13.35	4.81	0.77	3.18	0.02	1.63	0.01	0.05	0.10	2.49	0.60	0.91

*** and ** represent significance at 1% and 5% level, respectively.

Table 2 (a). Mean performance and range of parental inbred lines for the various characters in popcorn.

Source of variation	Days to 50% tasseling	Days to 50% silking	Days to maturity	Plant height (cm)	First cob placement (cm)	Final plant stand per plot (numbers)	No. of cobs per plot	Cob weight per plot (kg/plot)	Shelling (%)	Grain yield (kg/plot)	100 grain weight (g)	Moisture (%)	Popping (%)	Popping volume (cm ³ /g)	Grain protein content (%)
HKI PC 1	53.33	56.33	84.67	73.00	45.67	35.33	45.33	1.28	78.00	1.00	12.32	22.53	91.00	15.70	10.43
HKI PC 3	51.00	54.00	80.67	75.67	47.33	35.67	49.33	1.34	80.85	1.08	12.59	20.33	83.67	15.13	10.23
HKI PC 4	53.33	56.33	84.00	92.00	60.67	34.67	50.33	1.01	74.57	0.76	9.25	22.03	84.33	14.20	10.37
HKI PC 7	52.67	55.67	86.00	93.00	50.67	34.33	46.33	1.43	79.20	1.13	10.40	22.70	92.33	16.73	10.77
HKI PC 4B	56.33	59.33	87.00	83.33	50.33	34.67	44.00	1.37	79.12	1.09	10.44	23.20	83.33	13.47	10.87
HKI PC 8B	53.67	55.00	88.33	97.00	54.67	35.00	37.00	1.47	75.71	1.12	13.37	24.23	91.00	11.20	11.33
HKI PC 1473-5	53.67	56.67	87.33	88.67	50.00	35.67	49.67	1.44	80.78	1.16	9.27	24.00	91.33	16.67	11.00
HKI PCBT 3	55.67	59.67	90.00	65.33	30.67	34.33	44.67	1.51	82.91	1.26	12.78	22.50	90.67	16.70	9.38
Mean	53.71	56.63	86.00	83.50	48.75	34.96	45.83	1.35	78.89	1.07	11.30	22.69	88.46	14.98	10.55
Range	51.00-56.33	54.00-59.67	80.67-90.00	65.33-97.00	30.67-60.67	34.33-35.67	37.00-50.33	1.01-1.51	74.57-82.91	0.76-1.26	9.25-13.37	20.33-24.23	83.33-92.33	11.20-16.73	9.38-11.33
C.D.	1.62	1.76	2.80	5.96	3.58	1.43	2.91	0.20	2.09	0.18	0.36	0.51	3.58	1.26	0.56

plots was observed to be statistically same which further revealed that the experiment has been conducted properly. So far plant height is concerned, medium height is preferred because tall plants tends to lodge and dwarf types are slow growing and face more weed problem and also perform poor under moisture stress condition.

Moisture percent at harvest was calculated because at the time of harvest different genotypes have different moisture in grain but we have to dry it upto 15 percent for calculation of cob yield/plot, grain yield per plot and shelling per cent. Drying should be upto 14 percent for recording data on popping parameters. As there is no relevance in estimating the economic heterosis and combining ability effects for the parameters such as final plant stand at harvest, moisture % at harvest and plant height, so it has not been calculated for these parameters.

If first cob placement is high then plant tend to lodge. So the genotypes with low placement are preferred. All the hybrids had negative heterotic values against both the checks which shows that all the hybrids had low placement for first cob which is desirable. The best performing hybrids for this trait were HKI PC 1 × HKI PC 8B, (-57.19, -54.93), HKI PC 4 × HKI PC1473-5(-54.18, -51.76), HKI PC 3 × HKI PC 4(-50.50, -47.89) against the pop corn check and normal maize check, respectively.

For grain yield per plot, which is most important aspect of breeding, 14 hybrids exhibited heterotic value of more than 10 percent over the standard check Bajaura popcorn and none of the hybrids showed positive hetrotic response over the standard check HM 4 which indicated that the grain yield potential of pop corn hybrids was very low as compared to normal maize hybrids. Eight of the crosses have shown more than 20 per cent economic heterosis for grain yield per plot over the pop corn check (Bajaura pop corn) but none were found superior against the normal maize check (HM 4).

The highest heterosis for grain yield has been reported in cross HKI PC 8B × HKI PCBT 3 (66.74 %) followed by HKI PC 7 × HKI PCBT 3 (52.02 %), HKI PC 8B × HKI PC 1473-5 (49.38 %), HKI PC 3 × HKI PC 7 (30.22%), HKI PC 7 × HKI PC 8B (30.17 %), HKI PC 4 × HKI PC 8B (27.92%), HKI 1473-5 × PCBT 3 (26.57%) and HKI PC 7 × HKI PC 4B (24.44) against pop corn check (Bajaura pop corn) which indicated their suitability to increase grain yield. Heterosis for grain yield per plot and other related characters in popcorn has also been reported earlier by Vieira *et al.* (2009), Munhoz *et al.* (2009) and Soni *et al.* (2013). All the crosses which showed high heterosis for grain yield showed high positive heterosis for cob weight

Table 2 (b). Mean performance and range of experimental hybrids for the various characters in popcorn.

S. No	Cross	Days to 50% tasseling	Days to 50% silking	Days to maturity	Plant height (cm)	First Cob placement (cm)	Final stand per plot (numbers)	No. of cobs per plot (kg/plot)	Cob weight per plot	Shelling (%)	Grain yield (kg/plot)	100 grain weight	Moisture (%)	Popping volume (cm ³ /g)	Grain protein content (%)	
1	HKI PC 1 × HKI PC 3	49.33	51.33	82.33	106.33	50.67	34.67	52.00	2.29	81.83	1.92	18.03	20.67	88.33	19.07	11.50
2	HKI PC 1 × HKI PC 4	52.67	54.33	85.33	107.67	67.33	34.33	49.33	2.34	83.30	1.95	16.72	22.63	89.00	18.57	11.33
3	HKI PC 1 × HKI PC 7	52.33	55.33	85.67	134.67	59.67	35.00	54.67	2.40	81.03	1.94	18.52	21.13	91.33	20.40	10.43
4	HKI PC 1 × HKI PC 4B	53.33	56.67	88.00	122.67	65.33	34.67	51.33	2.53	84.63	2.12	18.60	23.60	87.33	16.87	10.70
5	HKI PC 1 × HKI PC 8B	52.33	55.33	90.00	91.33	42.67	33.33	40.67	2.45	79.90	1.98	19.56	23.50	88.67	16.87	10.47
6	HKI PC 1 × HKI PC 1473-5	52.00	55.00	85.33	117.67	59.33	34.33	50.33	2.42	83.43	2.02	16.39	21.43	91.33	20.00	10.73
7	HKI PC 1 × HKI PCBT 3	53.33	56.33	85.67	123.00	65.33	34.67	53.00	1.49	83.93	1.25	17.48	22.30	92.67	20.70	9.90
8	HKI PC 3 × HKI PC 4	51.33	53.33	82.00	103.00	49.33	34.00	48.33	1.82	82.77	1.51	16.23	20.67	88.00	17.07	11.20
9	HKI PC 3 × HKI PC 7	50.00	53.00	83.00	147.67	64.33	35.67	53.67	2.74	84.63	2.32	17.29	21.53	89.33	17.67	10.07
10	HKI PC 3 × HKI PC 4B	51.00	53.00	83.00	117.00	63.00	34.67	53.00	2.28	84.07	1.92	18.80	21.57	83.67	16.33	11.27
11	HKI PC 3 × HKI PC 8B	51.67	54.67	84.33	121.00	60.00	36.00	41.00	2.62	80.10	2.10	17.66	21.53	87.67	17.37	10.63
12	HKI PC 3 × HKI PC 1473-5	50.00	53.00	82.00	111.67	51.33	35.33	52.67	2.14	84.13	1.80	15.43	20.53	87.00	17.77	11.03
13	HKI PC 3 × HKI PCBT 3	50.33	53.33	82.67	121.67	62.33	32.67	49.67	1.85	83.03	1.54	16.01	21.20	90.67	19.70	10.60
14	HKI PC 4 × HKI PC 7	51.33	54.33	89.67	133.33	51.67	36.00	56.67	2.17	82.30	1.79	15.34	22.37	87.33	17.77	10.73
15	HKI PC 4 × HKI PC 4B	53.33	56.33	87.33	96.00	51.33	34.00	51.67	1.10	82.53	0.91	13.32	23.27	83.33	15.73	11.13
16	HKI PC 4 × HKI PC 8B	51.00	54.00	88.67	135.00	70.33	35.67	40.67	2.86	79.60	2.28	18.37	22.52	89.33	15.93	10.90
17	HKI PC 4 × HKI PC 1473-5	54.00	56.67	84.67	88.67	45.67	35.33	54.67	1.53	83.10	1.27	15.31	21.60	88.67	17.87	10.63
18	HKI PC 4 × HKI PCBT 3	54.33	57.33	90.00	126.67	59.67	34.67	49.67	2.44	83.17	2.03	18.40	23.80	86.67	18.03	10.47
19	HKI PC 7 × HKI PC 4B	51.00	54.00	88.00	140.67	60.33	35.33	40.00	2.73	81.27	2.22	18.41	23.09	87.67	16.50	11.23
20	HKI PC 7 × HKI PC 8B	50.33	51.67	87.67	148.67	77.67	34.67	38.33	2.84	79.53	2.32	18.81	23.73	86.67	15.93	10.53
21	HKI PC 7 × HKI PC 1473-5	51.00	53.00	87.67	152.00	85.67	35.67	42.33	2.61	80.30	2.10	19.31	24.47	90.33	18.67	10.70
22	HKI PC 7 × HKI PCBT 3	50.67	53.67	87.33	147.67	72.33	34.33	50.67	3.26	83.10	2.71	18.38	23.53	92.00	21.17	10.37
23	HKI PC 4B × HKI PC 8B	53.33	56.33	87.00	129.67	50.00	34.33	51.67	2.06	82.93	1.70	18.48	22.37	90.00	19.10	11.27
24	HKI PC 4B × HKI PC 1473-5	54.00	57.00	85.33	105.33	54.67	35.33	52.67	1.44	82.07	1.18	18.69	23.09	89.67	17.77	11.43
25	HKI PC 4B × HKI PCBT 3	51.33	54.33	88.00	139.67	67.00	33.67	52.00	2.25	82.03	1.84	17.67	22.60	89.33	18.30	10.60
26	HKI PC 8B × HKI PC 1473-5	50.33	53.67	87.33	105.33	51.67	35.00	53.00	3.21	84.80	2.66	17.60	23.40	88.00	17.87	11.47
27	HKI PC 8B × HKI PCBT 3	51.67	55.00	89.67	135.00	64.67	35.00	41.00	3.65	81.30	2.97	18.98	23.53	91.00	18.67	10.74
28	HKI PC 1473-5 × HKI PCBT 3	50.33	53.33	87.67	165.33	84.33	35.00	56.00	2.78	80.93	2.25	17.65	22.27	92.00	21.07	11.26
	Mean	51.70	54.48	86.26	124.08	60.99	34.76	49.31	2.37	82.42	1.95	17.55	22.43	88.82	18.17	10.83
	Range	49.33-54.33	51.33-57.33	82.00-90.00	88.67-165.33	42.67-85.67	32.67-36.00	38.33-56.67	1.10-3.65	79.60-84.63	0.91-2.97	13.32-19.56	20.53-24.47	83.33-92.67	15.73-21.17	9.90-11.50
	C.D.	1.62	1.76	2.80	5.96	3.58	1.43	2.91	0.20	2.09	0.18	0.36	0.51	2.58	1.26	0.56
	Mean (Standard checks)															
1	Bajaura Popcorn	50.00	52.67	86.33	191.33	96.67	34.00	54.67	2.15	82.93	1.78	18.75	22.57	92.00	26.17	11.32
2	HM 4	50.33	53.33	85.00	187.00	94.67	35.33	58.67	3.78	78.93	2.99	25.61	21.57	8.00	1.26	9.87

Table 3 (a). Nature and magnitude of economic heterosis (over standard checks) for yield and quality characters

S. No.	Cross	Days to 50% tasseling		Days to 50% silking		Days to maturity		First cob placement (cm)		No. of cobs per plot		Cob weight per plot (kg/plot)	
		Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4
1	HKI PC 1 × HKI PC 3	4.00	3.32	4.42	3.13	-4.63	-3.14	-49.17	-46.48	-4.88	-10.85	6.65	-39.34
2	HKI PC 1 × HKI PC 4	5.33	4.64	3.16	1.88	-1.15	0.39	-32.44	-28.88	-9.76	-15.42	8.93	-38.04
3	HKI PC 1 × HKI PC 7	4.67	3.98	5.06	3.76	-0.77	0.78	-40.14	-36.97	-0.01	-6.28	11.40	-36.64
4	HKI PC 1 × HKI PC 4B	6.67	5.97	7.59	6.26	1.93	3.53	-34.45	-30.99	-6.10	-12.00	17.86	-32.96
5	HKI PC 1 × HKI PC 8B	4.67	3.98	5.06	3.76	4.25	5.88	-57.19	-54.93	-25.61	-30.28	13.91	-35.21
6	HKI PC 1 × HKI PC 1473-5	-1.33	-1.98	-2.54	-3.74	-1.15	0.39	-40.47	-37.33	-7.93	-13.71	12.60	-35.95
7	HKI PC 1 × HKI PCBT 3	6.67	5.97	6.95	5.63	-0.77	0.78	-34.45	-30.99	-3.05	-9.14	-30.70	-60.58
8	HKI PC 3 × HKI PC 4	2.67	1.99	1.26	0.01	-5.02	-3.53	-50.50	-47.89	-11.59	-17.14	-15.26	-51.80
9	HKI PC 3 × HKI PC 7	0.00	-0.66	0.63	-0.62	-3.86	-2.35	-35.45	-32.04	-1.83	-7.99	27.40	-27.54
10	HKI PC 3 × HKI PC 4B	2.00	1.33	0.63	-0.62	-3.86	-2.35	-36.79	-33.45	-3.05	-9.14	6.00	-39.71
11	HKI PC 3 × HKI PC 8B	3.33	2.66	3.79	2.51	-2.31	-0.78	-39.80	-36.62	-25.00	-29.71	22.05	-30.58
12	HKI PC 3 × HKI PC 1473-5	0.00	-0.66	0.63	-0.62	-5.02	-3.53	-48.50	-45.78	-3.66	-9.71	-0.56	-43.44
13	HKI PC 3 × HKI PCBT 3	0.67	0.01	1.26	0.01	-4.24	-2.74	-37.46	-34.16	-9.15	-14.85	-14.05	-51.11
14	HKI PC 4 × HKI PC 7	2.67	1.99	3.16	1.88	3.87	5.49	-48.16	-45.42	3.65	-2.85	0.93	-42.59
15	HKI PC 4 × HKI PC 4B	6.67	5.97	6.95	5.63	1.16	2.74	-48.50	-45.78	-5.49	-11.42	-48.65	-70.79
16	HKI PC 4 × HKI PC 8B	2.00	1.33	2.53	1.26	2.71	4.31	-29.43	-25.71	-25.61	-30.28	32.98	-24.37
17	HKI PC 4 × HKI PC 1473-5	8.00	7.29	7.59	6.26	-1.93	-0.39	-54.18	-51.76	-0.01	-6.28	-28.93	-59.58
18	HKI PC 4 × HKI PCBT 3	8.67	7.95	8.85	7.51	4.25	5.88	-40.14	-36.97	-9.15	-14.85	13.67	-35.34
19	HKI PC 7 × HKI PC 4B	2.00	1.33	2.53	1.26	1.93	3.53	-39.47	-36.27	-26.83	-31.42	26.79	-27.88
20	HKI PC 7 × HKI PC 8B	0.67	0.01	-1.90	-3.12	1.55	3.14	-22.08	-17.96	-29.88	-34.28	32.14	-24.84
21	HKI PC 7 × HKI PC 1473-5	2.00	1.33	0.63	-0.62	1.55	3.14	-14.05	-9.51	-22.57	-27.42	21.53	-30.87
22	HKI PC 7 × HKI PCBT 3	1.33	0.67	1.89	0.63	1.16	2.74	-27.43	-23.59	-7.32	-13.14	51.44	-13.86
23	HKI PC 4B × HKI PC 8B	6.67	5.97	6.95	5.63	0.78	2.35	-49.83	-47.18	-5.49	-11.42	-4.42	-45.63
24	HKI PC 4B × HKI PC 1473-5	8.00	7.29	8.22	6.88	-1.15	0.39	-45.15	-42.26	-3.66	-9.71	-33.07	-61.93
25	HKI PC 4B × HKI PCBT 3	2.67	1.99	3.16	1.88	1.93	3.53	-32.78	-29.23	-4.88	-10.85	4.42	-40.61
26	HKI PC 8B × HKI PC 1473-5	0.67	0.01	1.89	0.63	1.16	2.74	-48.16	-45.42	-3.05	-9.14	49.35	-15.05
27	HKI PC 8B × HKI PCBT 3	3.33	2.66	4.42	3.13	3.87	5.49	-35.12	-31.69	-25.00	-29.71	69.77	-3.44
28	HKI PC 1473-5 × HKI PCBT 3	0.67	0.01	1.26	0.01	1.55	3.14	-15.39	-10.92	2.43	-3.99	29.40	-26.40

Table 3 (b). Nature and magnitude of economic heterosis (over standard checks) for yield and quality characters

S. No.	Cross	Grain yield per plot (kg/plot)		Shelling (%)		100 grain weight (g)		Popping (%)		Popping volume (cm ³ /g)		Grain protein content	
		Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	HM 4	Bajaura Popcorn	Bajaura Popcorn	HM 4	Bajaura Popcorn
1	HKI PC 1 × HKI PC 3	8.03	-35.69	1.09	6.16	-3.39	-29.61	-3.99	1004.16	-5.47	1413.25	1.59	16.99
2	HKI PC 1 × HKI PC 4	9.61	-34.75	0.45	5.48	-10.41	-34.72	-3.26	1012.50	-7.95	1373.57	0.11	15.29
3	HKI PC 1 × HKI PC 7	9.04	-35.08	-2.29	2.61	-0.75	-27.68	-0.73	1041.66	1.14	1519.05	-7.84	6.13
4	HKI PC 1 × HKI PC 4B	19.10	-29.10	0.85	5.90	-0.31	-27.36	-5.07	991.66	-16.38	1238.65	-5.48	8.85
5	HKI PC 1 × HKI PC 8B	11.35	-33.71	-2.45	2.44	3.50	-24.59	-3.62	1008.34	-16.38	1238.65	-7.54	6.48
6	HKI PC 1 × HKI PC 1473-5	13.48	-32.44	0.61	5.65	-12.15	-35.99	-0.73	1041.66	-0.84	1487.30	-5.19	9.19
7	HKI PC 1 × HKI PCBT 3	-29.72	-58.16	1.21	6.28	-6.32	-31.75	0.73	1058.34	2.63	1542.86	-12.54	0.71
8	HKI PC 3 × HKI PC 4	-15.28	-49.57	-0.20	4.81	-13.01	-36.61	-4.35	1000.00	-15.38	1254.52	-1.06	13.94
9	HKI PC 3 × HKI PC 7	30.22	-22.47	2.05	7.17	-7.34	-32.49	-2.90	1016.66	-12.41	1302.14	-11.07	2.41
10	HKI PC 3 × HKI PC 4B	7.64	-35.92	1.37	6.45	0.77	-26.58	-9.06	945.84	-19.02	1196.27	-0.47	14.62
11	HKI PC 3 × HKI PC 8B	18.09	-29.70	-3.41	1.43	-5.38	-31.05	-4.71	995.84	-13.90	1278.33	-6.07	8.17
12	HKI PC 3 × HKI PC 1473-5	1.07	-39.83	1.45	6.54	-17.29	-39.74	-5.43	987.50	-11.91	1310.08	-2.54	12.24
13	HKI PC 3 × HKI PCBT 3	-13.76	-48.66	0.12	5.14	-14.22	-37.50	-1.45	1033.34	-2.33	1463.49	-6.36	7.83
14	HKI PC 4 × HKI PC 7	0.34	-40.27	-0.76	4.22	-17.79	-40.10	-5.07	991.66	-11.91	1310.08	-5.19	9.19
15	HKI PC 4 × HKI PC 4B	-48.76	-69.50	-0.48	4.51	-28.63	-48.00	-9.42	941.66	-22.00	1148.65	-1.65	13.26
16	HKI PC 4 × HKI PC 8B	27.92	-23.85	-4.02	0.80	-1.55	-28.27	-2.90	1016.66	-21.01	1164.52	-3.71	10.89
17	HKI PC 4 × HKI PC 1473-5	-28.65	-57.53	0.20	5.23	-17.95	-40.22	-3.62	1008.34	-11.42	1318.02	-6.07	8.17
18	HKI PC 4 × HKI PCBT 3	14.21	-32.01	0.29	5.31	-1.41	-28.16	-5.80	983.34	-10.59	1331.19	-7.54	6.48
19	HKI PC 7 × HKI PC 4B	24.44	-25.92	-2.01	2.91	-1.34	-28.11	-4.71	995.84	-18.20	1209.52	-0.77	14.27
20	HKI PC 7 × HKI PC 8B	30.17	-22.51	-1.68	3.25	0.82	-26.54	-5.80	983.34	-21.01	1164.52	-6.95	7.15
21	HKI PC 7 × HKI PC 1473-5	17.87	-29.83	-3.17	1.68	4.81	-23.64	-1.81	1029.16	-7.45	1381.51	-5.48	8.85
22	HKI PC 7 × HKI PCBT 3	52.02	-9.50	0.20	5.23	-1.48	-28.22	0.00	1050.00	4.94	1579.92	-8.42	5.46
23	HKI PC 4B × HKI PC 8B	-4.27	-43.01	0.00	5.02	-0.98	-27.85	-2.17	1025.00	-5.30	1415.87	-0.47	14.62
24	HKI PC 4B × HKI PC 1473-5	-33.65	-60.50	-1.04	3.92	0.14	-27.03	-2.54	1020.84	-11.91	1310.08	1.00	16.31
25	HKI PC 4B × HKI PCBT 3	3.54	-38.36	-1.08	3.88	-5.29	-30.99	-2.90	1016.66	-9.27	1352.38	-6.36	7.83
26	HKI PC 8B × HKI PC 1473-5	49.38	-11.07	-0.16	4.85	-5.66	-31.27	-4.35	1000.00	-11.42	1318.02	1.30	16.65
27	HKI PC 8B × HKI PCBT 3	66.74	-0.74	-1.97	2.95	1.73	-25.88	-1.09	1037.50	-7.45	1381.51	-5.15	9.23
28	HKI PC 1473-5 × HKI PCBT 3	26.57	-24.65	-2.41	2.49	-5.43	-31.09	0.00	1050.00	4.45	1571.98	-0.53	14.55

per plot, also indicating the importance of this trait for the high heterotic value of grain yield. These findings are in line with the findings of Vieira *et al.* (2009), Munhoz *et al.* (2009) Soni *et al.* (2013) and Elmyhum *et al.* (2013) and Solalinde *et al.* (2014). Same crosses showed poor heterosis response over normal maize check in negative direction. Similar findings were reported by Krupakar *et al.* (2013), Elmyhum *et al.* (2013), Kumar *et al.* (2014) and Munhoz *et al.* (2009).

Among the superior crosses, the positive heterosis has been reported for shelling per cent in some of the crosses (HKI PC 1 × HKI PC 4B, HKI PC 1 × HKI PC 1473-5, HKI PC 3 × HKI PC 7, HKI PC 3 × HKI PC 8, HKI PC 4 × HKI PC 8 B, HKI PC 4 × HKI PC 3 and HKI PC 7 × HKI PCBT 3) although the magnitude was found low. Among the superior hybrids the positive heterosis has also been reported for 100 grain weight in some of the crosses (HKI PC 1 × HKI PC 8, HKI PC 7 × HKI PC 8B, HKI PC 7 HKI PC 1473-5 and HKI PC 8B × PCBT 3). Highest heterotic effect (3.65%) for number of cobs per plot was recorded in hybrids HKI PC 4 × HKI PC 7 against pop corn check followed by cross HKI PC 1473-5 × HKI PCBT 3. Thus, the number of cobs per plot was mainly responsible for increase in grain yield per plot. Similar results were reported by Goncalves *et al.* (2014), Soni *et al.* (2014), Elmyhum *et al.* (2013) and Vieira *et al.* (2009).

For days to flowering (days to 50 % tasselling and days to 50% silking) and days to maturity, the hybrids with less number of days are preferred because we need early maturing varieties for fitting in different cropping system. So, hybrids with negative heterotic values are desirable from breeding point of view. For earliness in tasseling, high negative heterotic response was observed in cross HKI PC 1 × HKI PC 1473-5 i.e. -1.33 and -1.98 against check 1 (bajaura popcorn) and check 2 (HM 4) respectively. This was followed by HKI PC 3 × HKI PC 1473-5 (0.00 and -0.66) and HKI PC 3 × HKI PC 7 (0.00 and -0.66), indicating vigour for earliness in tasseling. The cross HKI PC 1 × HKI PC 1473-5 also showed highest negative heterotic response over both standard checks for earliness in silking (Table 3a). Other high heterotic hybrids for earliness in silking were HKI PC 7 × HKI PC 8B, HKI PC 3 × HKI PC 1473-5, HKI PC 3 × HKI PC 4B and HKI PC 3 × HKI PC 7, over both standard checks.

For earliness in maturity better performing hybrids such as HKI PC 3 × HKI PC 4 and HKI PC 3 × HKI PC 1473-5 showed the maximum desirable negative heterotic effect followed by HKI PC 1 × HKI PC 3 and HKI PC 3 × HKI PCBT 3 against check 1 and check 2, respectively, showing them to be having vigour for earliness in maturity. These

results confirm the finding of Mohammed and Al-Falahy (2015) and Junaidu, *et al.* (2015).

The popping quality (popping per cent and popping volume) of normal maize check HM 4 was found very poor so it is not logical if we compare the heterosis of crosses against this check for popping parameters. So in popping parameters, for practical utility, the heterosis has been compared only against pop corn check. For popping per cent parameter, highest positive heterosis was reported in cross HKI PC 1 × HKI PCBT 3 (0.73%) against pop corn check (Bajaura pop corn) and second best positive performing hybrid was HKI PC 1473-5 × HKI PCBT 3 followed by HKI PC 7 × HKI PCBT 3. Four hybrids, viz., HKI PC 7 × HKI PCBT 3 (4.94%) and HKI PC 1473-5 × HKI PCBT 3 (4.46%), HKI PC 1 × HKI PCBT 3 (2.63%) and HKI PC 1 × HKI PC 7 (1.14%) also showed high heterotic response for popping volume. The crosses HKI PC 1 × HKI PC 3 (1.59% and 16.99%) and HKI PC 8B × HKI PC 1473-5 (1.30% and 16.65%) were best performing hybrids with positive heterotic response for grain protein content over check 1 and check 2, respectively. These results are in agreement with the findings of Khan *et al.* (2015), Pajic *et al.* (2008), Ahmet *et al.*, (2011), Phumelele *et al.* (2014), Moterle *et al.* (2012) and Sonia *et al.* (2014).

Conclusion

The results obtained in the present investigation were optimistic and major findings revealed that two better performing hybrids HKI PC 1473-5 × HKI PCBT 3 and HKI PC 7 × HKI PCBT 3 had high *per se* performance and also had high heterosis for popping quality parameters (popping per cent and popping volume) showing that simultaneous improvement of grain yield and popping quality is possible. Therefore, these hybrids which have given more than 10% grain yield improvement / enhancement warrant their further testing under different agro climatic conditions to confirm the results for exploitation of heterosis. Also the better performing hybrids and parents could be used to generate more number of desirable segregates for grain yield, popping expansion and other component traits.

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Studies on Southern corn leaf blight disease in West Bengal

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Abstract: Southern leaf blight disease caused by *Helminthosporium maydis* is one of the important biotic stresses of maize crop reported from most maize growing regions of the world including India. For the purpose of better understanding and finding out the suitable management practices a detailed study of the fungal pathogen including the different types of symptoms observed in the field, isolation and pathogenicity of the pathogen, host plants and suitable culture media to grow the fungus under laboratory condition was conducted in New Alluvial Zone of West Bengal. Different types of symptoms are observed in the experimental plots of District Seed Farm, Kalyani Simanta. Five different media were taken to identify the most suitable media for maximum growth of the fungal pathogen to study the fungus in details under laboratory condition and Potato Dextrose Agar medium was found best for growing the fungus under artificial condition. Four cereals crops viz., Paddy, Wheat, Pearl millet and Sorghum were taken for host range study of the fungus and it is found that all of them may be the host plant of this fungal pathogen.

Keywords: Biotic stress · Host plant · Culture media · Pathogenicity · Artificial condition · Southern corn leaf blight

Introduction

In West Bengal (North Eastern Plain Zone/ NEPZ) maize is getting importance day by day specially during *rabi* season. Maize crop may be affected by 112 biotic stresses. The important biotic stresses faced by maize in West Bengal are Southern corn leaf blight, Northern corn leaf blight, banded leaf & sheath blight, curvularia leaf spot, stalk rot, rust, etc. Southern corn leaf blight (SCLB) is considered as one of the most serious diseases and has attained the status of the economically important disease. This disease is widely distributed in India during *kharif* season. Southern corn leaf blight (SCLB) caused by the fungus *Helminthosporium maydis* is one of the serious diseases of maize in NEPZ and also a serious disease in warm & humid condition during crop growing season (White, 1999). The predominant form of *Helminthosporium maydis* is Race O which can cause yield loss upto 40% (Byrnes *et al.*, 1989).

Considering the economic importance of this disease, detailed study of the fungus in relation to symptomatology, isolation and pathogenicity, growth evaluation of the causal fungus in different solid media, host range is very essential for better understanding and finding out the suitable management strategies of the fungus as well as the disease. In present study a detail account of the fungus was taken in West Bengal condition.

Materials and methods

Symptomatology

Symptoms of different types found in the experimental plots of AICRP on Maize of Kalyani were critically observed and defined. Different types of symptoms found in hybrids and inbreds were observed carefully.

Isolation of the fungal pathogen

Maize leaves having characteristic symptoms of southern

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leaf blight were collected from the experimental plots of AICRP on maize and farmer's field. The small pieces of infected portions of leaves were sterilized with mercuric chloride (HgCl_2) solution (1 : 1000) for thirty seconds and thoroughly washed in sterilized water for three times and placed on a sterilized filter paper in a petri dish to remove excess water. Finally 3-4 pieces were carefully plated on water agar inside laminar air flow chamber under aseptic condition. Inoculated plates were incubated in a B.O.D. incubator at $27\pm 1^\circ\text{C}$ with 12 h light and 12 h darkness. The growth of fungus was conspicuous after 24 h of inoculation. The pure colonies developed from the leaf bits were transferred to PDA slants and incubated for 10 days. After proper observation of the fungal, spores under microscope the pathogen was confirmed as is *Helminthosporium maydis*.

Pathogenicity

Pathogenicity of the fungus was studied on maize variety; Kaveri 50 of in earthen pots (diameter 30 cm) containing autoclaved sterilized soil. Healthy seedlings of maize plants of 3-4 leaf stage were artificially inoculated with pure spore suspension from 15 days old fungal culture of *Helminthosporium maydis* (8×10^4 spore/ml). Inoculated plants were kept in glasshouse. The data of percent infection were taken after every seven days. The fungus was re-isolated from the diseased leaves and Koch's postulates were proved.

Confirmation of the pathogen

The characters of fungal growth was observed carefully and critically under microscope (40X) when the pure culture was 15 days old. Observed characters were compared with evidences given in different literatures. Thorough examination of the mycelial growth on Potato Dextrose Agar (PDA) plates, hyphal and conidial characters were used for confirmation of the fungus.

Study of the fungus on different culture media

To study the fungus under *in vitro* condition effectively, pure culture and proper growth of the fungus is a prerequisite much. Identification of suitable culture media for the specific fungus is the basic need for this purpose. For identifying the ideal culture media five different solid culture media (Potato Dextrose Agar medium, Carrot Dextrose Agar medium, Corn Meal Agar medium, V_8 media

and Corn Leaf Extract media) were used to find out the best media for the growth of *Helminthosporium maydis*. 20 ml of each sterilized media was poured into each sterilized Petri plate. The center of each medium of Petri plate was inoculated with *Helminthosporium maydis* using 5mm disc from 15 day old cultures of each isolate with a sterilized corks borer. The inoculated plates were incubated at $27\pm 2^\circ\text{C}$ for 12 h of alternate light and darkness. Observations on colony diameter, conidia germination and sporulation were recorded after 15 days of incubation while dry mycelial weight was taken after drying and harvesting the cultures.

The radial growth of fungal colony on different solid media at $27\pm 1^\circ\text{C}$ was measured every 24 hrs for a period of 8 days. The measurement of colony growth of three plates of each medium was taken.

For estimation of sporulation / conidia formation, the spores were collected carefully and gently with the help of a brush by adding 10ml distilled water in the plates. The spore solution passed through a funnel mated with absorbent cotton to stop the mycelia from passing through. The conidia of the suspension were counted with the help of Haemocytometer and spore concentration was expressed as number of conidia/ml.

Host range study

The experiment was conducted in the field of District Seed Farm (A,B Block) Kalyani Simanta as of four cereal crops – Paddy, Wheat, Pearl Millet and Sorghum. Plants were maintained by proper agronomical practices. Artificial inoculation with fungal spore solution was done in field as 25 days old plants.

Result and discussion

Symptomatology

Symptoms of Southern Corn Leaf Blight (SCLB) first appeared on lower leaves of 45 days old plants, gradually increased in number and vary in different hybrids and inbreds depending on the method of inoculation. Lesions elongate between the veins, tan coloured with buff to brown or dark reddish brown border - similar symptoms were also observed by Singh and Srivastava (2012). With maturity, the lesions become elongated. Lesion size is 0.40 to 3 cm length and 0.2 to 0.7 cm width that was similar to the observation of Hulagappa (2012). Lesions first appear on lower leaves, increase in number and size with time,

progresses upward the plant till maturity and destroys large area of leaf (Figure 1).

Isolation and pathogenicity

Infected maize leaves with characteristic symptoms of SCLB were collected for preparing from field and pure culture of the fungus. The isolated fungus was critically examined for characterization. Initially fungal colonies appeared dull whitish, brown, pale brown and afterwards slightly dark in colour. Septate hyphae produced loose or little dense brownish septate conidiophores bearing slightly curved, cylindrical, smooth walled conidia having 7-10 septa.

Kumar *et al.* (1979) and Bhavani and Gohilo (2016) also observed the similar characters of mycelium, conidiophore and conidia of *Helminthosporium maydis*. Pathogenicity test was successfully carried out by artificial inoculation with pure culture of *Helminthosporium maydis* spore suspension (5×10^4 spore /ml). Inoculated plants exhibited greyish tan, parallel straight sided or diamond shaped 1-3cm long, lesions with buff or brown borders with prominent colour banding or irregular zonation after 15 days of inoculation. Same experiment was also conducted with same type of observation by Zamani and Mehriyan (2006). The reisolated culture of the fungus was identified as *Helminthosporium maydis* and matched with the original pathogen. Muhammad and Amusha (2003); Bhavani and Gohilo (2016) proved the pathogenicity of

Helminthosporium maydis in maize in pot condition by inoculation of mycelial suspension @ 5g/ml sterilized deionized water (Figure 2).

Study of suitable culture media

The data on observations taken on the growth and sporulation of *Helminthosporium maydis* in different solid culture media is presented in Table 1. Significant results were obtained from this study. Among the five solid media maximum fungal growth (8.75 cm after 192 h) was observed in PDA media followed by growth (8.23 cm after 192 hrs) in Carrot Dextrose Agar media. Next best media was V_8 media (4.99 cm after 192 h) and Maize Leaf Extract media (3.23 cm after 192 h). Lowest growth (2.46 cm after 192 h) of *Helminthosporium maydis* was obtained in Corn Meal Agar media (Figure 3,4 & 5).

Similarly same type of media such as – Potato Dextrose Agar, V_8 Juice Agar, Garraway medium and Corn leaf media were found effective for growth of *Helminthosporium maydis* (Garraway, 1973).

Sanjeev and Rani (2009) studied the culture media and nutritional studies in relation to growth and sporulation of *Helminthosporium maydis* and reported highest fungal growth (78.6 mm) in PDA. Rabbani *et al.* (2011) also reported that PDA as the best media for growing the fungus *Dreschlera hawaiiensis*. We also found as best media.

So, PDA best among the five media used for growing the fungus *Helminthosporium maydis*. Variation of colour

Figure 1. Symptoms of Southern corn leaf blight. [(a) Healthy plants in the field and (b-f) Symptoms of infected leaf in the fields]

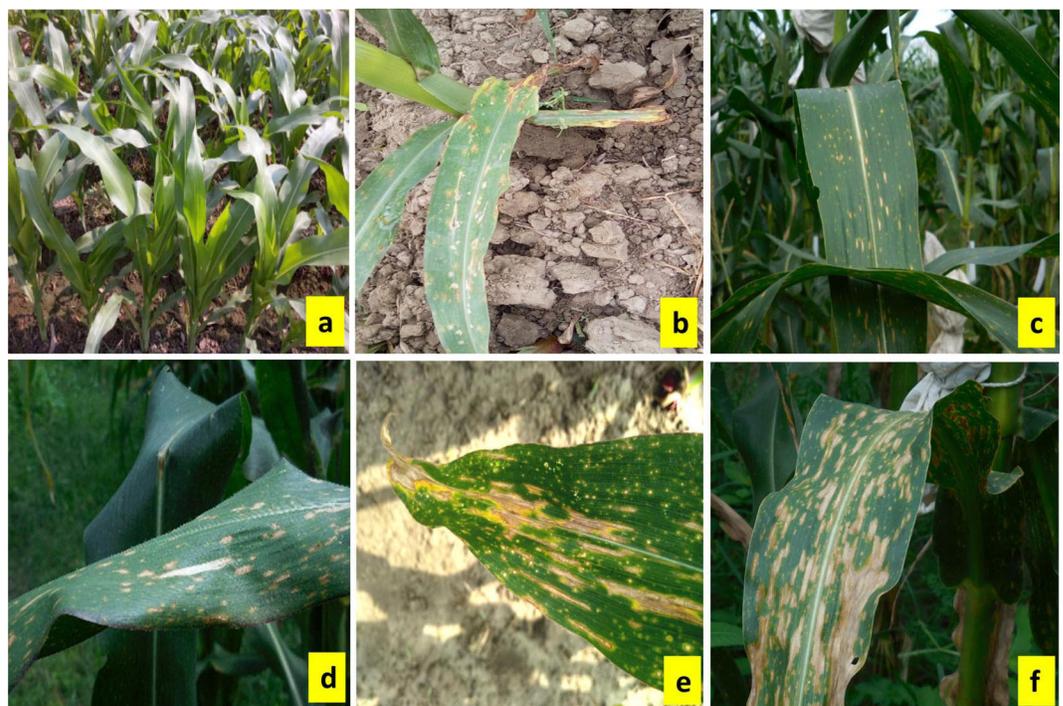


Figure 2. Morphology of *Helminthosporium maydis* [(a) Initial growth on PDA, (b&c) conidia on infected leaf sample, (d-e) conidia with mycelium and (f) formation of conidia]

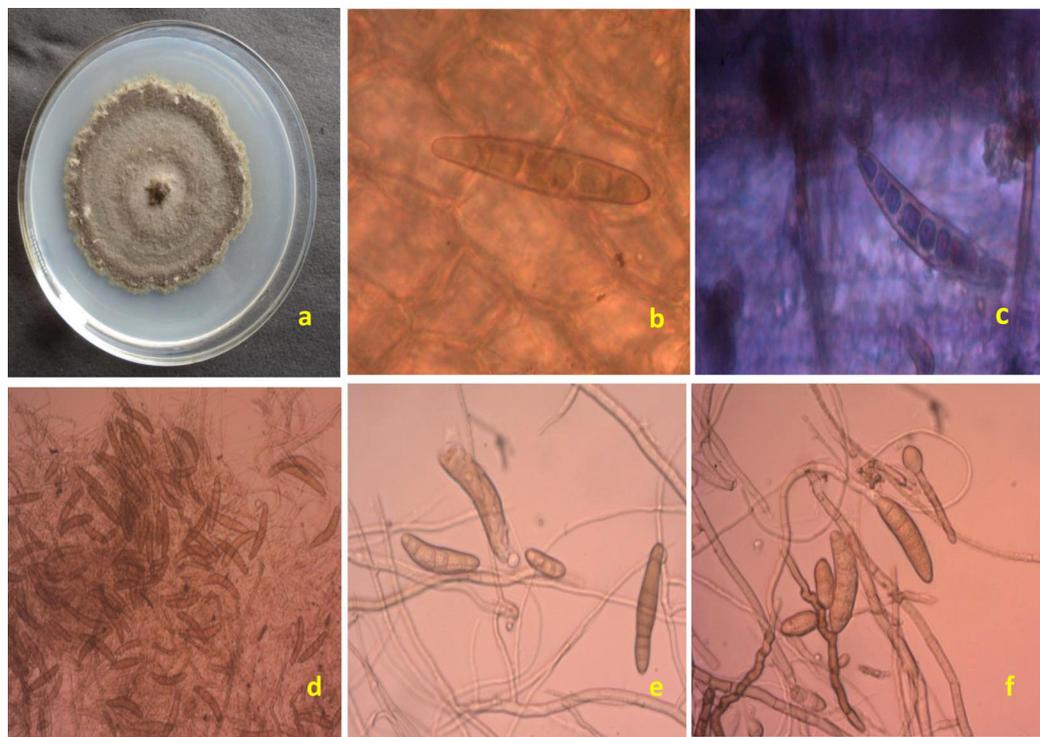


Figure 3. Growth of *Helminthosporium maydis* in different solid media [(a) PDA, (b) CDA, (c) v8 media, (d) CMA and (e) LEA]

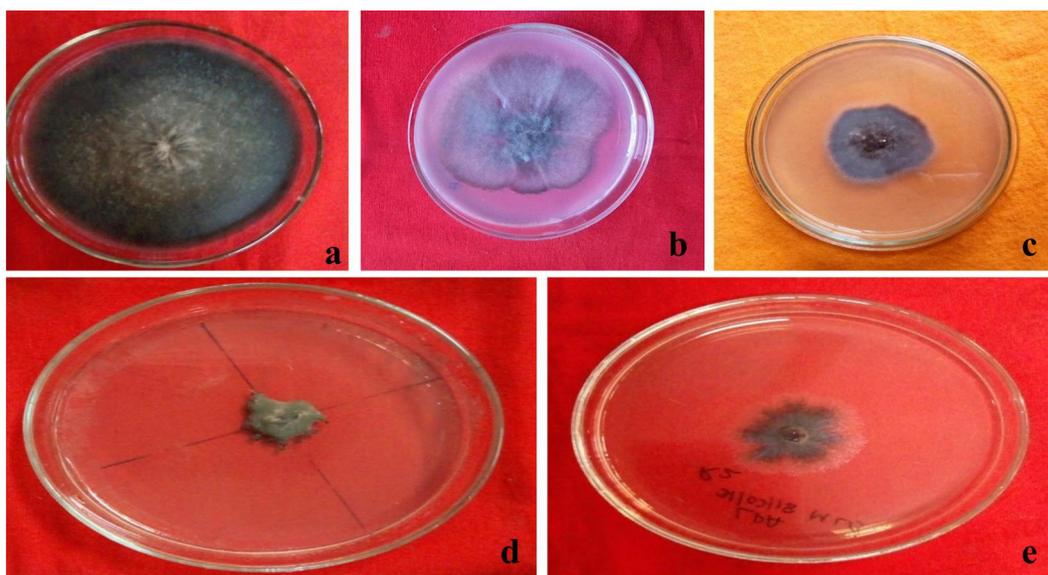


Table 1. Growth evaluation of *Helminthosporium maydis* in different solid media at different hours of incubation

Solid media used for fungal growth	Growth of fungus at different incubation periods (cm)							
	24hrs*	48hrs	72hrs	96hrs	120hrs	144hrs	168hrs	192hrs
Potato dextrose agar	1.03	2.01	2.82	4.18	5.48	6.83	7.95	8.75
Carrot dextrose agar	1.04	1.61	2.99	4.16	5.29	6.17	7.015	8.23
Corn meal agar	0.78	1.02	1.21	1.52	1.73	1.89	2.13	2.46
V ₈ media	0.90	1.50	2.17	2.67	3.54	4.08	4.71	4.99
Maize leaf extract	0.92	1.22	1.52	1.75	2.07	2.47	2.82	3.23

*Average of three replications

Figure 4. Growth of *Helminthosporium maydis* in PDA, CDA, V8 media, CMA and LEA at 24 hrs. interval up to 8 days

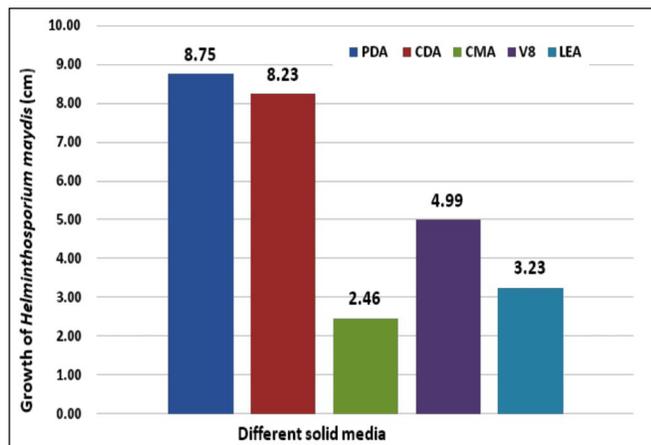
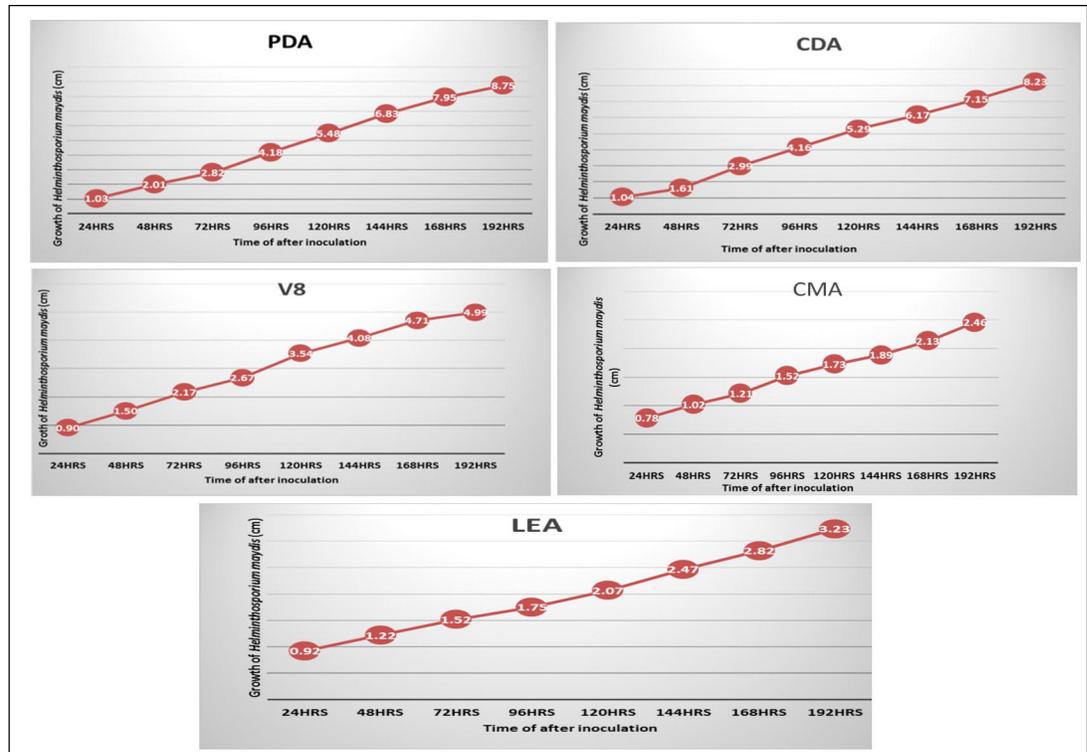


Figure 5. Average growth of *Helminthosporium maydis* on different solid media after 8 days of incubation

of fungal growth were observed in different media. Fungal growth was black in PDA & Corn Meal Agar media, light brown in Carrot Dextrose Agar media and Corn Leaf Extract media and whitish in colour in V₈ media. Growth of the fungus *Helminthosporium maydis* was found to be in increasing trend with time in all the solid media used.

Reaction of Helminthosporium maydis to some cereal crops

The causal organisms of plant diseases spreads through the alternate hosts and for better understanding of the ways of disease spread, study of the host range is of great importance. Among the four field crops (Paddy, Wheat,

Figure 6. Some cereals host of *Helminthosporium maydis*. [(a) Pearl millet, (b) Wheat, (c) Rice and (d) Maize]

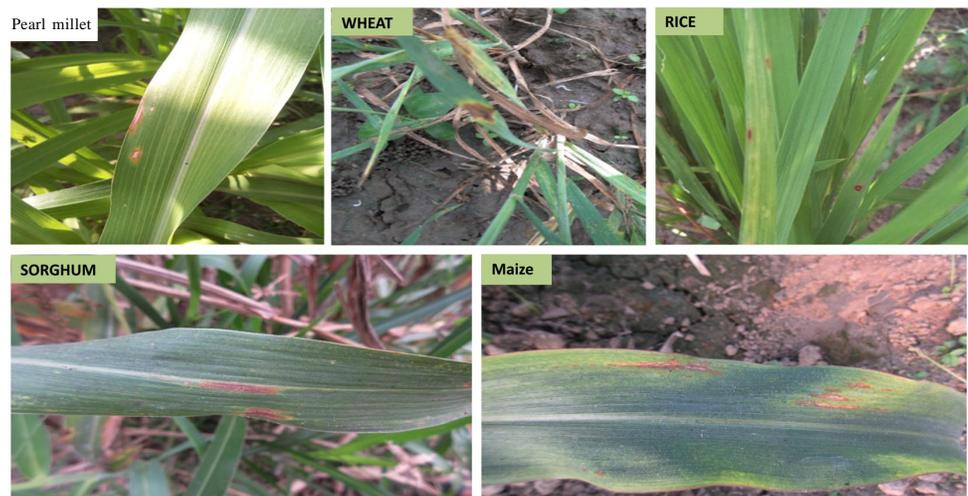


Table 2. Reaction of *Helminthosporium maydis* to some cereal crops

Common name of host tested	Scientific name	Reaction - Infection (+) or noninfection (-)
Pearl Millet	<i>Pennisetum glaucum</i>	+
Wheat	<i>Triticum aestivum</i>	+
Sorghum	<i>Sorghum bicolor</i>	+
Paddy	<i>Oryza sativa</i>	+

Pearl Millet and Sorghum) that were inoculated artificially with *Helminthosporium maydis*, showed disease symptoms identical to the symptoms produced by *Helminthosporium maydis* in maize plant (Figure 6 and Table 2). All these four cereal crops may act as the host plant of *Helminthosporium maydis*. This result supports the work done by Duan *et al.* (1992). They tested ten crops (Barley, Oat, Paddy, Rye, Sorghum, Sugarcane, *Zizania latifolia* and Wheat) against *Bipolaris maydis* and obtained the identical symptoms.

Conclusion

In the present study different types of symptoms of southern corn leaf blight were found in the fields of alluvial zone of West Bengal. All the four plants artificially inoculated with *Helminthosporium maydis* showed typical symptom of SCLB. Among five artificial culture media the PDA

showed best performance in growing the fungal pathogen of SCLB.

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