

Review Article

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Gametophytic Selection: A Simple Technique for Thermo Tolerance Genotypes Identification in Maize

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ABSTRACT

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Temperature of environment increase day by day so present time identification of heat tolerant genotypes is great challenge. Because tolerance to high temperature is a multi-genic character and also many screenable traits are available. But there no single techniques available which give holistic idea about plant level heat tolerance. Gametophytic selection could be one of the alternative strategies. There are many successful attempt have been made to improve the crop plants by gametophytic selection. In this review discuss the advantages of gametophytic selection over sporophytic selection, the effect of high temperature on maize and how gametophytic selection is a potential option for high temperature screening in maize.

Introduction

The main goal of any plant breeding program is the improvement of all characteristics that contribute to economic value. These characteristics may be increased yield, ability of tolerant to biotic and abiotic stresses (Fehr 1991). Traditional selection has mainly focused on the sporophytic phase of a plant's life cycle. But sporophytic selection has many limitations. With the improvement of breeding techniques and biotechnological advances, now many options available to plant scientists interested in improving an array of plant responses and characteristics.

In recent time information available from plant genome and many advance techniques

are developed a small but growing resurgence of research focused on male gamete selection has developed (McCormick, 2004). The life cycle of angiosperms are consist of two parts (Gametophytic and sporophytic) and it provide opportunity to introduce early selection into breeding program via gametophytic selection (Fig. 1). The most prominent phase of angiosperm life cycle is the sporophytic generation and it develops from zygote followed by the fertilization.

Constrains of sporophytic selection

Traditional selection has mainly focused on the sporophytic phase of a plant's life cycle.

But sporophytic selection has many limitations. It is extremely time consuming, labor intensive and can occupy vast quantities of space. Sporophytic selection also requires large population sizes in order to obtain the essential combination of attributes needed to improve a crop.

Many woody perennial species especially have lagged behind in new cultivar development due to larger physical size and long juvenile phases (Hormaza and Herrero, 1996). Sporophytic selection needs to repeat several seasons that increase the cost of selection. The environment changes day by day so it is very difficult to screen the crops at the field.

The life cycle of angiosperms offers the opportunity to introduce early selection into breeding programs via gametophytic selection. The alternation of generations that occurs in angiosperms is a conserved attribute of the plant kingdom. The most prominent phase of the angiosperm life cycle is the sporophytic generation. Sporophytes develop after fertilization of male and female gametes. But gametophyte generation develops after meiosis of spores (Hormaza and Herrero, 1996). These two different generations gives idea for early selection pressure and it can be applied to both male and female gametophytes. It is reported that male gametes are more sensitive for biotic and abiotic stresses so identification of pollen which are more resistance to these stress can be produce more resistant crop plants. And, competition between the large numbers of male gametophytes during development in the anther, pollen germination and tube growth in the style, as well the independent phase that pollen experiences between shedding and stylar deposition, create an opportunity for easier manipulation using the male gametophyte (Ottaviano and Sari Gorla, 1993).

Gamete selection

Gametophytic selection could be either male or female gametophytic selection. But the male gametes are more susceptible for stress so crop improvement mainly focuses on male gametophytic selection. Male gametophytic development is a very complex process that requires the coordinated participation of various cell and tissue types and their associated specific gene expression patterns. In this process both sporophytic (anther, tapetum) and gametophytic (pollen) tissues participate in this process. Gametophytic selection is highly reduced system as compared with the sporophyte and therefore it provides opportunities to study the developmental regulation of gene expression and functional interactions between different cell types (Honys *et al.*, 2004).

Male gametophytic selection can occur in two styles: selection for pollen competitive ability or selection for tolerance to environmental stresses. The concept of selecting for pollen competitive ability relies on the assumption that the most vigorous pollen grains will confer vigour to the sporophyte (Ottaviano *et al.*, 1991). The main components of pollen competitive ability are faster pollen germination and pollen tube growth rates which were shown to give rise to positive responses in kernel weight, because germinated pollen which have large pollen tube length can easily fertilize female gametes (Ottaviano *et al.*, 1983). Selective pressure can be most effectively applied at two times: during pollen development or during pollen germination and tube growth (Hormaza and Herrero, 1996). Selection for such traits as herbicide resistance is effective during the quiescent stage, although for many environmental stresses it is important to apply the selection pressure when genetic differences are fully expressed.

Temperature tolerance in maize

The global temperature increase day by day and different climate models predict that the global mean temperature will continue this trend, increasing by 1–4°C by the end of the twenty-first century, additionally climatological extremes such as heat waves leads to increase temperature more frequently (IPCC, 2013; Tebaldi *et al.*, 2006; Hansen *et al.*, 2015). Above data suggested that average global temperature increases and it also significant different between seasonal and also varies regional that leads to potential impact on the agriculture (IPCC, 2007). Most of the area under maize crop is in the warmer parts of temperate regions and in humid subtropical climate. Highest production is in areas having the temperature 21°C to 27°C. Maize production and productivity are prone to rapid and constant changes due to global warming related environmental changes (Porter, 2005; Wahid *et al.*, 2007). In india also 10-20% maize yield loss because of temperature fluctuation.

High temperature stresses affects all stages of crop. Among all stages reproductive stage of maize is most sensitive. As the majority of our food supply is a product of sexual reproduction in flowering plants, understanding how different plants cope with stress during their reproductive (gametophytic) phase is critical to managing the future of agricultural productivity. During the short time surrounding fertilization, even a single hot day cause fatal to reproductive success for many plant species. Thus, it is important to consider what aspects of plant sexual reproduction may become the weak links in agricultural productivity. High temperature affect at vegetative as well as reproductive stages of maize.

At temperatures above 38°C (reproductive stage), poor seed set in maize has been

attributed to both a direct effect of high temperature (Carberry *et al.*, 1989) and pollen desiccation (Schooper *et al.*, 1986; Lonquist and Jugenheimer, 1943). High temperature affects pollen fertility directly, since the crop showed no visible symptoms of water stress (Schooper *et al.*, 1987).

Pollination is critical process for corn seed set and yield. For kernels to develop, silks must emerge and be fertilized by viable pollen. Silks grow about 1 to 1.5 inches per day and will continue to elongate until fertilized. Temperatures greater than 35°C with low relative humidity will desiccate exposed silks, but not greater on impact silk elongation rates greatly. Pollen is no longer viable once temperatures reach the more than optimum, especially with low relative humidity. Fortunately, pollen shed usually occurs from early to mid-morning when temperatures are lower.

The temperature stress induces slow desiccation of silks and result in low seeds set. Historically, this has been the most important problem leading to yield reduction, particularly in stressful years. Once silks begin to desiccate, they lose their capacity for pollen tube growth and fertilization (Madhiyazhagan Ramados *et al.*, 2005). Heat also affects pollen production and viability. First, temperature more than 35°C depresses pollen production. Even continuous heat, over several days before and during pollen-shed, results in only a fraction of fertile pollen being formed, probably because of the reduced sugar available. In addition, high temperature reduces the period of pollen viability to a couple hours (or even less). While, there is normally a surplus of pollen, heat can reduce the fertility and amount available for fertilization of silks. It's been also showed that exposure to high temperature reduce the pollen shedding and fertility (Tom Hoegemeyer, 2011).

There are also plenty of report which showed that high temperature affects at seedling, photosynthesis and even all metabolic of crop plants.

Approaches to crop improvement for heat stress tolerance

As photosynthesis and reproductive developments are two very sensitive physiological processes for heat stress (Prasad *et al.*, 2008). So screening of genotypes which are resistance on these stages, they are also heat tolerance. There are many approaches available for improvement of heat stress. But there is lack of comprehensive screening methods for heat tolerance and screening at field level is very difficult because fluctuation of temperature in field condition. So need to develop methods by which we can screen the crops at control condition.

During the last few years, interest has been generated in the gametophytic selection/response and it act as an important alternate technique involving for crop improvement. Many researchers examined the possibility of increasing the efficiency of plant breeding through the selection of pollen (Ottaviano *et al.*, 1990; Hormaza and Herrero, 1992).

In vitro screening of pollen grains for temperature tolerance on of effective strategy for selection of plants because high temperature leads to decrease in pollen germination and pollen tube growth and it is also demonstrated in many crops.

Associations between gametophyte and sporophyte have been reported for tolerance to several environmental factors (Ravikumar and Patil, 2004; Clarke *et al.*, 2004). One important area in which pollen response may parallel to that of sporophyte is its response to biotic and abiotic stress factors. Indirect

evidence is obtained through an analysis of the behaviour of gametophytic and sporophytic phases in relation to different biotic and abiotic agents.

Variety of selection pressures such as temperature (Zamir *et al.*, 1982; Clark *et al.*, 2004; Dominguez *et al.*, 2005), pathotoxin (Laughnan and Gabay, 1973), herbicide (Sari Gorla, 1989), metal (Searcy and Mulcahy, 1985) and water stress (Ravikumar *et al.*, 2003) were enforced during gametophytic generation and were found very effective in increasing the frequency of resistant individual in the progeny. These studied support that gametophytic selection is potential method which could alternative approach of sporophytic selection.

Gametophytic selection for high temperature tolerance

Pollen screening is an effective technique of screening the genotypes of crop plants for various stresses. It has been shown that gametophytic (haploid) and sporophytic phase (diploid) do share a common gene expression profile (Mulcahy *et al.*, 1996; Hormaza and Herrero, 1986; Ravikumar *et al.*, 2003). Selection pressure enforced at gametophytic stage improves sporophytic performance.

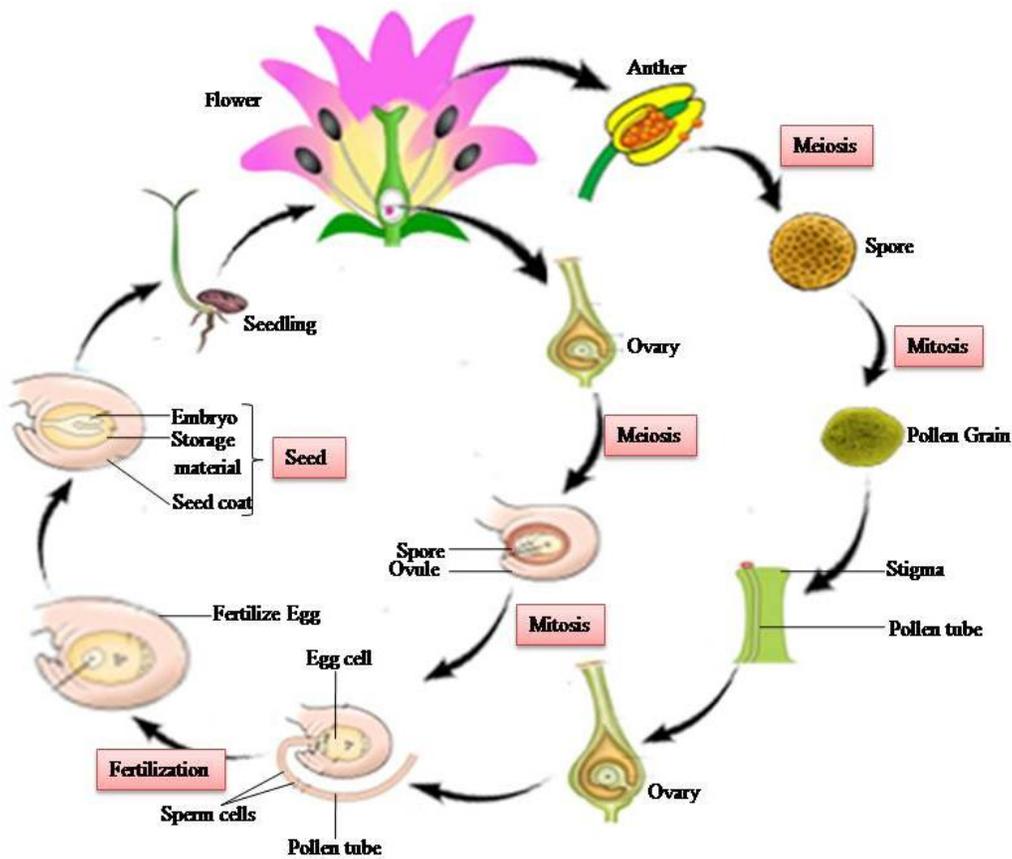
Maize is susceptible for high temperature and its susceptibility depends on stage of crops. Among all stages, tasseling stage is most susceptible stage for temperature. Temperature is more than ambient at tasseling stage leads to zero seed set at severe condition. There are several attempt have been done to identify heat tolerant genotype at pollen stage.

Pollen grains are simple structured plant cells. Pollen tube formation is a good and simple model of growth and development (Taylor and Hepler, 1997).

Table.1 Composition of different pollen germination media

Composition	Reference
5% sucrose, 0.0025 % boric acid, 0.056% calcium chloride, 0.025 % potassium dihydrogen phosphate, 3 % PEG 4000	Herrero and Johnson (1980)
Sucrose 15 %, 0.6 % bactoagar, 0.03% calcium nitrate, 0.01 % boric acid	Pfahler and Linskens (1981)
10% Sucrose, 0.6% Agar, 0.03 Boric acid, 0.05% CaCl ₂ , 0.01% MgSO ₄	Shivanna <i>et al.</i> , (1991)
Sucrose 150 g/L, boric acid 200 mg/L, potassium nitrate 200 mg/L, calcium nitrate 200 mg/L, magnesium sulphate 200 mg/L, PEG 223.6 g/L	Reddy <i>et al.</i> , (2001)
10% sucrose, 0.7% agar and 100 ppm IAA	Geetha <i>et al.</i> , (2004)
1% agarose, 25% sucrose, 0.52 mM KNO ₃ , 3.06 mM MnSO ₄ , 1.66 mM H ₃ BO ₃ , 0.42 mM MgSO ₄ 7 H ₂ O and 1 μM of gibberellic acid	Prabhu and shankar, (2005)
Sucrose 18 %, 0.72 % bactoagar, 0.036 % calcium nitrate, 0.012 % boric acid	Singh <i>et al.</i> , (2016)

Fig.1 Life cycle of angiosperm



Pollens generally germinate on stigmatic surface of plants (Ünal, 1986). In some plant groups they germinate in anther loculus (Yldrm and Ünal, 1996; Dane, 2000). During the past few years pollen tubes grown under *in vitro* conditions became a popular model system for cell biology studies in plant cells (Moutinho *et al.*, 2001). The media used for pollen germination vary according to the plant species (Vasil, 1960; Baker and Baker, 1979). Pollens of some species need more complicated media (Çetin *et al.*, 2000). The required environment for pollen germination *in vitro* is related to genetic composition and also the quality and quantity of nutrient reserves of pollen. There are several media standardized for different crops. There are several media summarized in table 1.

When high temperature treated pollen exposed in germination media different genotypes respond differently (Geetha *et al.*, 2004 and Singh *et al.*, 2016). These evidences showed that different pollen respond differently at high temperature. The exposure of maize pollen to high temperature (40°C) reduces the *in vitro* fertilization percentage, and after 6 hours of stress no fertilization occurs because pollens are susceptible to high temperature (Dupuis and Dumas, 1990). Those genotypes whose pollen germination percentage and large pollen tube length they are resistant for high temperature. The genotypes which showed gametophytic tolerance to heat stress under *in vitro* conditions produced more number of seeds per cob after heat treatment to pollen grains. A strong correlation exists between pollen and the sporophyte tolerance to various stresses (Herrero and Hormaza, 1996; Ravikumar and Patil, 2004).

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