Can Germination Requirements Predict Tolerance to Extreme Weather? — An Assessment for Heirloom Maize from the P'urhépecha Plateau

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Abstract

The temperature and water potential requirements for seed germination were determined for five heirloom maizes from three different agroclimatological regions from the P'urhépecha community of San Francisco Pichátaro, Michoacán, Mexico. Germination experiments were conducted in environmental controlled chambers exposing seeds to day/night air temperatures (12 h photoperiod) of 15/5, 20/10, or 35/25°C, and to water potentials of 0, −0.01, −0.05, −0.1, or −0.5 MPa, which were created with aqueous solutions of polyethylene glycol (molecular weight 20,000). Germination was maximal and occurred at a faster rate for seeds incubated at 35/25°C. In turn, a high water potential of 0.0 or −0.01 MPa was necessary to achieve maximum germination, which decreased under more negative water potentials until germination was completely inhibited for seeds incubated under −0.5 MPa failed. Although an environmental specialization for germination was not observed among the heirloom maize evaluated, their ability to germinate maximally under high temperatures is indicative of their cultivation potential under the increasing air temperatures expected to occur during the present century. In contrast, germination sensitivity to relatively mild water potentials may be a conservative ecophysiological trait of drought-escape for this tropical annual plant.

Keywords: Cereal ecophysiology; Climate change; Domestication; Phyto genetic diversity; Traditional agriculture

Introduction

Global climate change poses an impending threat to food production, considering that the most productive areas where crops are currently cultivated are likely to become reduced or at least shifted in response to increasing air temperature and lower precipitation (Saucz-Romero et al., 2010; Monterroso-Rivas et al., 2011; de la Barrera, 2016). In addition, a higher frequency and severity of extreme weather events such as droughts, hail storms, hot spells, and unusually high precipitation, can become catastrophic for food production (Intergovernmental Panel on Climate Change, 2012). An adaptation strategy is the development of the so-called "climate ready" varieties capable of growing or even thriving under such anticipated environmental conditions (Lipper et al., 2014). In tropical regions, where a decrease in precipitation is estimated to be more severe than the sole increase in air temperature, the development of drought resistant crops is most desirable. Regardless of the breeding technique utilized for developing such varieties, the source of genetic materials are existing native varieties from the regions of original domestication where the genetic diversity of crops is usually highest. This is true for coffee in Kenya, for rice throughout Asia, and most certainly for maize in the Mesoamerican region (Boege, 2008; Aerts et al., 2013; Kumagai et al., 2016).

Screening for climate-ready traits in heirloom materials, such as resistance to drought or to extremely high temperatures, can be slow and costly. For instance, conventional breeding programs involve the experimental cultivation of various materials under environmental conditions that are similar to those projected to occur in the future, an evaluation that can span from several months to various growing seasons (Farshad and Sutka, 2005). Given that ample areas are required to establish proper agronomical trials, this procedure only permits the simultaneous evaluation of a few materials. Alternatively, physiological and molecular assays can be utilized to screen numerous materials in relatively short periods. For instance, molecular methods allow the thorough and rapid screening of numerous materials for tolerance to environmental stress, such as drought (Xu et al., 2009, 2011). Also, the buildup of osmolytes such as proline or gas exchange and photosynthesis parameters have been utilized to screen for ecophysiological specialization to drought and high temperature (Campos et al., 2004; Guerrero-Jiménez and de la Barrera, 2015). However, these methods require specialized equipment and training precluding their...
implementation by traditional producers. A rapid, inexpensive, and "low-tech" assay for screening desirable traits facing climate change is thus needed.

For many annual and ephemeral plants, the maximum and fastest germination occurs under environmental conditions similar to those that are prevalent during their growing season (Vázquez-Yanes et al., 2005; Baskin and Baskin, 2014). For instance, germination for the African bunch grass *Pennisetum ciliare* requires water potentials of at least –0.1 MPa and air temperatures above 30ºC, which effectively increases the chances of germinating during the summer and avoiding the winter, when a substantial precipitation can occur, but lethal low temperatures are prevalent (Villa-Reyes and de la Barrera, 2016). Also, germination for the neotropical weed *Lopezia racemosa* (Onagraceae) requires water potentials of at least –0.05 MPa but can withstand temperatures above 35ºC (Martínez and de la Barrera, 2017). For the case of maize, Mexico is a region of particular interest as the crop was originally domesticated here and numerous varieties have been bred in the adapting to an ample diversity of agroclimatic regions (MacNeish and Eubamks, 2000; Benz, 2006). For instance, seedlings of heirloom lines from the Purhépecha plateau in west-central Mexico have different temperature and water requirements according to the altitudinal gradient of cultivation despite a restricted geographical amplitude (Guerrero-Jiménez and de la Barrera, 2015). In fact, the Purhépecha, the most prevalent of the four indigenous peoples from the state of Michoacán, near the region of original domestication, preserve various traditional festivities that require the cultivation of various specific heirloom maizes (Barrera-Bassols and Zinck, 2003; Alarcón-Chaires, 2009).

Because the particular environmental requirements leading to germination of heirloom maizes from the Purhépecha plateau were not known, the present study aimed to elucidate whether the environmental optima for germination of materials planted in distinct various agroclimatic zones reflect the environmental conditions leading to adequate plant development and proper grain yield. We hypothesized that the seeds of maizes cultivated in a drier and warmer environment would germinate under higher temperatures and lower water potentials than those from more mesic environments. By means of laboratory experiments, the germination germination responses to incubation temperature and water potential were characterized for seeds of five heirloom maizes from different agroclimatic regions from the Purhépecha plateau, Mexico.

**Materials and Methods**

**Study Region and Plant Material**

Seeds of five heirloom maize genotypes were obtained during a field campaign in March 2010 directly from Purhépecha producers from the community of San Francisco Pichátaro, Tingambato, Michoacán, Mexico (19.58º N; 101.83º W). At this locality a steep altitudinal gradient has created three distinct agroclimatic zones (Barrera-Bassols et al., 2009). In particular, a "humidity agriculture" zone is at the highest elevation where lower air temperature and higher annual precipitation are prevalent. At mid-elevations, a region with warmer climate and lower annual precipitation allows for "residual humidity agriculture." Finally, at the lowest elevation occurs the valley where "rainfed agriculture" is practiced given its highest air temperature and lowest annual precipitation. In this lower valley, however, "homegarden agriculture" also occurs allowing the cultivation of several plant species, including some short-cycle maizes (Alarcón-Chaires, 2009; Barrera-Bassols et al., 2009). Approximately ten Mexican landraces of maize have been identified to be cultivated in Pichátaro, representing about nearly 15% of the Mexican landraces (Astier and Barrera-Bassols, 2006; Comisión Nacional para el Conocimiento y Uso de la Biodiversidad, 2009). However, only a few senior citizens maintain the tradition of cultivating the local heirloom varieties, thus only five maizes were found during the field campaign (Table 1), which were utilized in the present work, namely: *azul* (blue, in Spanish) is usually cultivated within the humidity agriculture zone, *blanco* (white) and *Amarillo* (yellow) are cultivated within the residual humidity agriculture zone, and *negro* (black) and *aurate* (blue, in Purhépecha) are cultivated in homegardens within the lowest valley. The seeds were kept in the laboratory (dark, 23ºC, 42% relative humidity) until utilized, usually within three months of collection.

**Temperature**

The effect of incubation temperature on germination for the five maize genotypes was evaluated in I-35LL environmental controlled chambers fitted with fluorescent light bulbs (Percival Scientific, Boone, Iowa, USA), where regimes of day/night air temperatures were set at 15/5, 20/10, and 35/25ºC with a 12 h photoperiod. Experimental units (n = 6 for each maize variety) were created by placing 50 seeds inside petri dishes (10 cm diameter) that contained two layers of sterile filter paper to which 50 mL of sterile, deionized, distilled water were added. The seeds were monitored every day until germination of new seeds ceased and germination was scored as the number of seeds with radicle protrusion.

**Water Potential**

The effect of water potential on seed germination was also evaluated in environmental chambers (30/20ºC, 12 photoperiod). These experimental units (n = 6 for each maize variety) were similarly created consisting of 50 seeds placed in petri dishes (10 cm diameter) that contained two layers of sterile filter paper to which 50 mL of a sterile aqueous solution of PEG 20000 were added.
Germination responses to temperature and water potential were evaluated for each maize by means of a repeated measures ANOVA on ranks followed by pairwise Tukey tests. All analyses were conducted with SigmaStat (SPSS, San Rafael, CA, USA). Data are means ± 1 S.E.

Results

Higher temperatures led to higher final germination for the five maizes considered, which also tended to germinate at faster rates ($p < 0.001$; Fig. 1). In most cases, no significant difference was found between seeds incubated under 35/25 and 20/10°C (Fig. 1A, C, D and E). The exception was blanco whose germination under 20/10°C was lower than under 35/25°C ($p < 0.05$; Fig. 1B). For all the maize varieties considered in the present study, the lowest germination was observed for seeds incubated at the lowest temperature of 15/5°C ($p < 0.05$).

Seed germination decreased with decreasing water potentials for all five maizes ($p < 0.001$; Fig. 2). Maximum germination at 14 days after imbibition was observed for seeds incubated under 0 MPa. Germination for all maizes was reduced by more than half for seeds incubated under −0.05 MPa and it was inhibited under −0.1 and −0.5 MPa ($p < 0.05$; Fig. 2).

Discussion

Germination by the five maizes responded to the incubation temperature, with slower rates and a reduced final germination under the lower temperatures. This is consistent with previous studies where higher incubation temperatures led to faster and higher germination up to ca. 30°C, above which it decreases until full inhibition at 40°C (Alessi and Power, 1971; Blacklow, 1972). This response to incubation temperature is common among other tropical species, especially annual grasses, whose optimum germination temperature is similar to the prevalent temperatures during the spring and summer (Vázquez-Yanes and Orozco-Segovia, 1996; de la Barrera et al., 2009; Baskin and Baskin, 2014).

Regardless of their provenance, the maizes considered here were quite sensitive to decreasing water potentials, as a mere −0.5 MPa, well above the permanent wilting point of −1.5 MPa (Nobel, 2009), inflicted sufficient stress to inhibit germination for all the materials that were tested. This is similar to what has been found for hybrid maizes, for which −0.6 MPa reduces germination (Heidari and Kahrizi, 2018). A possible reason for the lack of germination that we observed at the lowest water potential may be the duration of our experiments, as up to 76 days are required for maize germination under −0.5 MPa (Schneider and Gupta, 1985). These observations reflect the fact that, as an annual tropical grass, the main drought coping strategy of maize is completing its lifecycle while sufficient water is available in the soil (DaCosta and Huang, 2009). However, germination may be improved by either exposing the seeds to cycles of wetting-drying or to osmopriming with solutions of PEG (Dubrovsky, 1996; Arif et al., 2014; Mahboob et al., 2015; Farooq et al., 2018). Under a scenario where the distribution of annual precipitation may become less even, priming seeds before sowing may become a useful practice to overcome the effect of an increasingly late rainy season in Mexico, where rain-fed agriculture is most prevalent and where sowing is done early in the year with up to 4 months of low or nil precipitation.

An additional factor to consider regarding the temperature and water potential requirements for germination is the fact that environmental variation is greatly reduced in the soil (Nobel, 2009). For instance, given an air temperature fluctuation of 15°C over the course of a clear day, the surface of bare soil can change its temperature almost 30°C, given its thermal properties, but the temperature at a depth of 25 cm fluctuates less than 5°C (Nobel and de la Barrera, 2003).
The water content in the soil may also be relatively stable, even during the dry months of spring because the very low hydraulic conductivity of the dry soil surface greatly reduces evaporation, which allows water to be available for triggering seed germination and sustaining seedling growth (Nobel, 2009). In any case, various climate change scenarios anticipate both increased air temperatures and reduced precipitations in large portions of Mexico.
(Saenz-Romero et al., 2010). In this respect, the sensitivity of maize germination needs to be considered when selecting and developing planting materials in order to maintain the high production of maize required for feeding 120 million Mexicans. Given the high genetic diversity of maize that exists in Mexico and the fact that traditional producers participate in a very vigorous seed exchange and movement (Dyer and López-Feldman, 2013; de la Barrera and Orozco-Martínez, 2016), future research for identifying climate-ready materials should consider young seedlings, a developmental stage that is most vulnerable both to environmental and biological threats and where environmental specialization has already been detected for maize collected within a small geographical region (de la Barrera et al., 2009; Guerrero-Jiménez and de la Barrera, 2015); perhaps during this most sensitive stage the ecophysiological differences for maize varieties will become evident, as they occur for older plants, but still allow for a relatively rapid, inexpensive, and potentially massive screening by non-specialists.

Conclusion

An ability of the heirloom maize to germinate under high temperature exhibited it innate potential for cultivation under high air temperature. On the contrary, maize sensitivity to germinate under relatively mild water deficit may be a conservative ecophysiological property to drought-escape.

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