Regenerating and Characterizing the Teosinte 'Wilkes 10', a *Zea mays* subsp. *parviglumis* Collection from Guerrero, Mexico

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Abstract

Tremendous variation was observed in the teosinte accession 'Wilkes 10', a population of *Zea mays* subsp*. parviglumis* in the leaf number and size, leaf sheath color, maturity, plant height, number of aerial roots, tassel size and structure, and anther and glume color. Under 10 to 11-hr natural daylength and 85/70ºC day/night temperature setting in a greenhouse in Lubbock, Texas, 200 plants produced over 54,000 seeds. This accession was susceptible to spider mites and common smut. Extending daylength with artificial lights induced the photoperiod response, which led the tassel spikelet to become a seedling. Ensuring short-day growing condition and preventing photoperiod response of teosinte plants is critical for normal plant growth, development, and seed production. Hybridization with inbred lines homozygous for *y1*, *su1*, and *sh2* that affect kernel chemical composition in maize did not detect the presence of the three recessive alleles in Wilkes 10.

Introduction

Wild relatives serve as an important reservoir of genes for crop improvement. The USDA National Plant Germplasm Systems holds 439 wild accessions from the genus Zea and only 74 accessions are available for distribution (Bernau et al., 2020). Very limited descriptive data is available for these wild accessions. Here we describe a procedure for successful seed regeneration and a trait description on the teosinte accession Wilkes 10 (PI 384071) during the short-day winter in a greenhouse in Lubbock, TX.

Objective

The objective was to regenerate the seeds of a teosinte accession 'Wilkes 10' and provide a morphological and developmental description.

Materials and Methods

A total of 240 seeds of the teosinte accession of Wilkes 10 (PI 384071), a population of *Zea mays* subsp. *parviglumis* collection from Guerrero, Mexico were planted in five-gallon pots in a greenhouse on January 8, 2020, five seeds per pot. The pots were filled with a mix (1:1 v/v) of field soil and Sungro Professional Growing Mix (Fig. 1). Greenhouse day/night temperatures were set at 29.4ºC/21.1ºC (85ºF 70ºF). Artificial light was not applied in the greenhouse. Average day length in Lubbock, TX is 10, 11, 13, and 14 hrs. on January 15, February 15, April 15, and May 15. Plants experienced less than 11-hr short-day during the first month. Plants were watered regularly and fertilized with Miracle-Gro all purpose plant food every two weeks.

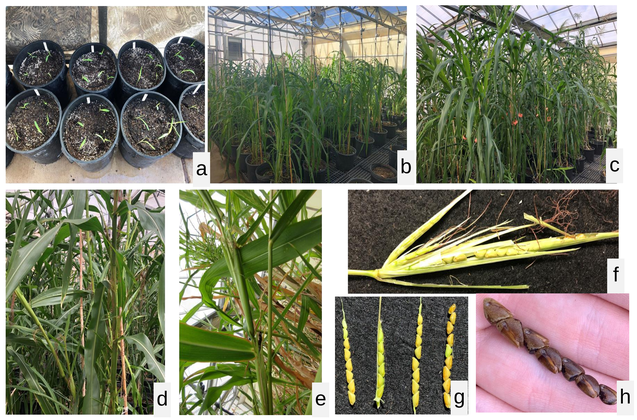

Fig. 1. Life cycle of the teosinte 'Wilkes 10' in the greenhouse: (a) emerging seedling; (b) growing plants; (c-e) flowering plants (f-g) developing and maturing seeds; (h) matured seeds .

Photoperiod response and reverse transition from reproductive to vegetative development:

Most plants showed normal tassel development, pollen shedding, and silking 100 days after planting. The neighboring room in the greenhouse complex had fluorescent lights. The plants closest to the fluorescent lights extended their vegetative stage (Fig. 1). When their tassels fully emerged, some or all spikelets did not produce anthers, instead turned into seedlings (Fig. 4). The daylength in Lubbock is 10 to 11 hr. from mid-October to mid-February. The reverse development indicates that a slight exposure to fluorescent light during the short-day winter could induce a strong photoperiod response. Photoperiod sensitivity is a critical factor that affects seed production and successful wide hybridization between maize and teosinte. Supplementing lights in the greenhouse before sunrise and after sunset may lead to abnormal teosinte plant development. When transplanted, these seedlings grew normally and become full plants that shed pollen, silked, and produced seeds. A teosinte plant can be cloned vegetatively with the seedlings formed by tassel spikelets.

Results

Tremendous variation observed within the teosinte 'Wilkes 10' collection:

A total of 229 seedling emerged, giving a 95.4% germination rate. The plants of this accession segregated in many traits such as coleoptile color (green vs. purple), sheath and stem color, plant height, number of leaves, leaf length, tassel size and branches, and anther color (Fig. 1). On average, it took 99 days from planting to pollen shedding. Plants that were exposed to longer daylength flowered and silked 20 to 40 days later. Most plants had 11 to 20 leaves. The average plant height was 256 cm. A small number of plants had no lateral branches. The first lateral branch appeared at the 7th node from bottom. The average number of nodes with aerial roots was 3. Some plants had 21+ nodes with aerial roots (Fig. 2 and 3). It has been reported that aerial roots exude large amounts of mucilage where unique diazotrophic microbials form community associations and fix atmospheric nitrogen (Van Deynze et al. 2018). Table 1 shows the correlation coefficients among the traits.

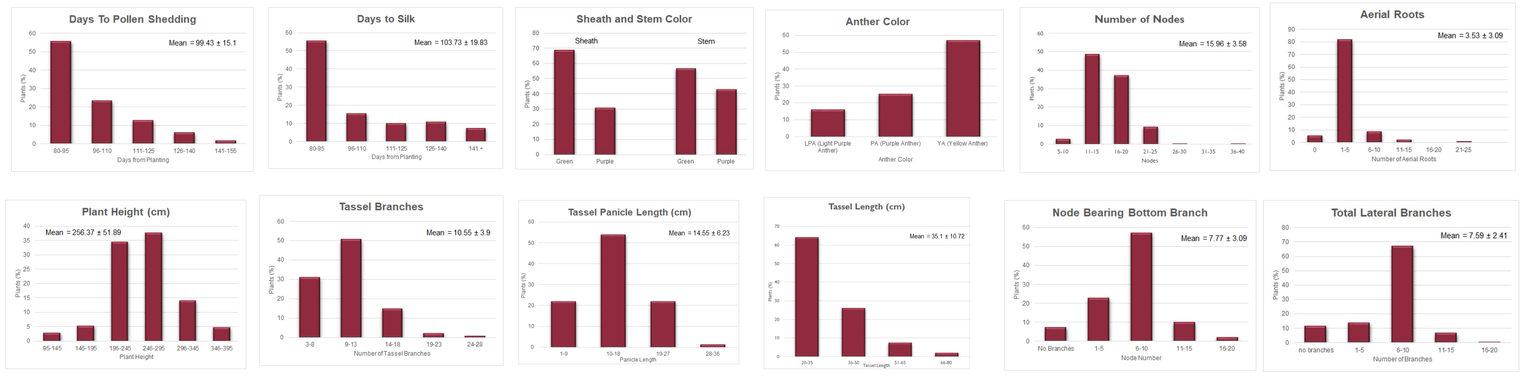

Fig. 2. Average, range, and distribution of morphological and developmental traits of the teosinte 'Wilkes 10'.

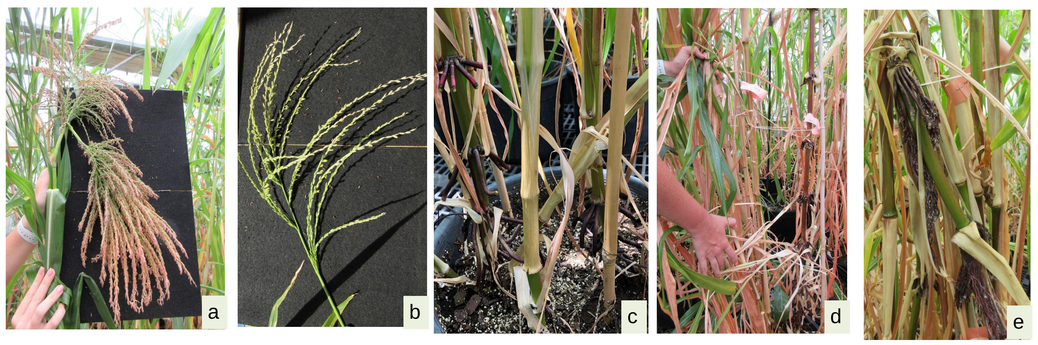

Fig. 3. Tassels with numerous branches and red anthers (a) or several branches and yellow anthers (b); stems with or without aerial roots (c-e).

Table 1. Pearson correlation coefficients among the traits. Values with * and ** represent the significance at 5% and 1% probability level.

	DTS	BBR	TBR	LBN	PTB	TSL	LFL	LFW	PHT	AER	NODE
DTP	0.91**	0.69**	0.66**	-0.04	0.16	0.32**	0.41**	0.60**	0.28**	0.50**	0.48**
DTS		0.49**	0.59**	0.16	0.36**	0.49**	0.40**	0.63**	0.29**	0.35**	0.35**
BBR			0.76**	-0.23**	0.25*	0.33**	0.34**	0.64**	0.43**	0.49**	0.64**
TBR				0.41**	0.43**	0.41**	0.31**	0.72**	0.65**	0.61**	0.80**
LBN					0.25*	0.11	-0.01	0.06	0.39**	0.22**	0.28**
PTB						0.48**	0.17*	0.35*	0.43**	0.26*	0.38**
TSL							0.33**	0.19	0.45**	0.20	0.36**
LFL								0.40**	0.38**	0.20*	0.25**
LFW									0.57**	0.60**	0.65**
PHT										0.42**	0.58**
AER											0.58**

DTP = days from planting to pollen shedding.
DTS = days from planting to silking.
BBR = the number of the node where first lateral branch appears.
TBR = the number of the node where top lateral branch appears.
LBN = the number of the lateral branches. (branch).
PTB = the number of primary tassel branches.
TSL = the tassel length from flag leaf collar to the top of tassel.
LFL = the leaf length where the first lateral branch appears
LFW = the leaf width where the first lateral branch appears
PHT = plant height from soil surface to the tassel tip.
AER = the number of nodes with aerial roots.
NODE = total number of nodes (i.e. the number of leaves on the central stem).

Detection of sugary 1 (*su1*), shrunken 2 (*sh2*), and yellow endosperm (*y1*) alleles:

Bulked pollen of the teosinte plants were pollinated to the inbred lines homozygous for *y1* (PHD03, *y1y1*), *su1* (P737M20, s*u1su1*), *sh2* (781 Ultra, *sh2sh2*), and AC 33892 (*su1su1* and *sh2sh2*). The resulting F_1 seeds from all the crosses (PHD03 x Wilkes 10, PHD03 x Wilkes 10, 781 Ultra x Wilkes 10, PHD03 x Wilkes 10, AC 33892 x Wilkes 10) were either yellow dent or yellow flint, indicating that 'Wilkes 10' does not have the three recessive alleles affecting the kernel chemical composition in maize.

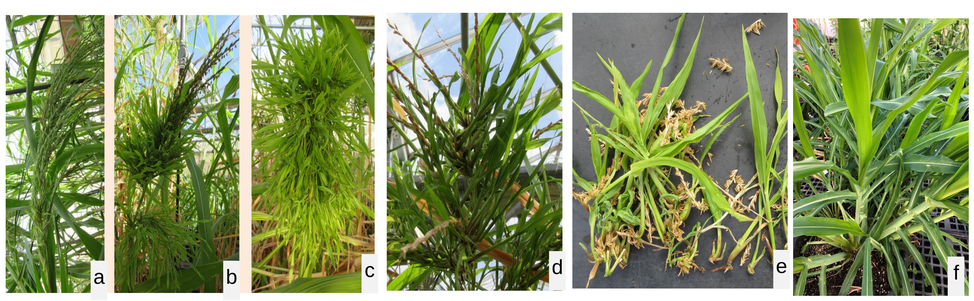

Fig. 4. Reverse change of tassel spikelets from reproductive to vegetative development. Parts or all tassel spikelets became seedlings (a to d). These seedlings formed by spikelets had leaves, stems and roots (e). When transplanted into soil, these seedlings grew normally and, in contrast to their mother plants, they produced numerous tillers (f). These plants were normal and eventually produced pollen and seeds.

This is an undergraduate internship research report by Morgan Molsbee. We appreciate the funding from Texas Corn Producers Board and Texas A&M AgriLife Research. For comments, contact: Morgan Molsbee (morgan.molsbee@ttu.edu) or Wenwei Xu (wxu@ag.tamu.edu).