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# Breeding Perennial Warm-season Grasses for the Subtropical Belt in South America

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**Key words:** Polyploidy; Apomixis; Forage breeding

## Abstract

Perennial warm-season grasses share several agronomic characteristics, such as marked seasonal growth, cold susceptibility and photoperiod sensitivity. Breeding efforts in South America have been focused on attempting to improved cool-season growth, cold tolerance and also adaptation to the alternation of flooding and drought periods. Warm-season grasses also have in common that most of them are polyploid and some have very low fertility. Apomixis is also a common trait among these species. For polyploid species with limited seed yield, which commonly have stolons or rhizomes, F<sub>1</sub> hybrids are created and released as cultivars. *Acroceras macrum* and *Hemarthria altissima* will be used as examples. For polyploid species without seed fertility issues, such as *Setaria sphacelata*, recurrent phenotypic selection (RPS) is used to generate improved populations adapted to these transition zones. For polyploid and apomictic species, such as *Paspalum* spp. and *Brachiaria* spp., several breeding approaches are now available. The generation of F<sub>1</sub> apomictic hybrids is currently used. It has been recently observed that the efficiency of this breeding method can be improved if the genetic distance among parents is considered. There is also new information indicating the great potential of using apomixis-linked molecular markers for the early identification of apomictic hybrids. Population-breeding approaches, such as RPS and selection based on combining ability, can also be used to assess the generation of superior apomictic hybrids. Finally, the challenge of breeding perennial warm-season grasses for the subtropics mainly relates to improving adaptation to extreme conditions (cold winters and warm summers and alternation of flooding and drought), developing specific breeding techniques for polyploid or apomictic species.

## Introduction

Subtropical South America (the land area located between 27 and 31° latitude) represents a transition zone between temperate and tropical regions where warm summers, cool winters, and high photoperiod variation area typical. The amount of precipitation is also highly variable across the region, and drought and flood conditions are also common. Soils usually have low fertility, and acidity or salinity issues are also commonly found. Forage breeders must consider all these characteristics to develop adapted cultivars, particularly perennial forages that are expected to produce across several years.

Perennial warm-season grasses are the most common type of cultivated forages in Subtropical South America, and the genera *Brachiaria*, *Panicum*, *Chloris*, *Cynodon*, and *Paspalum* predominate. Polyploidy, variable modes of reproduction, and reduced seed yield are common characteristics in this group of forages (Vogel and Burson 2004).

In some species, seed fertility issues limit the possibility of seed reproduction, and vegetative propagation is used. Several species of *Cynodon*, *Acroceras macrum*, *Hemarthria altissima*, and some species of *Paspalum* and *Brachiaria*, which usually have rhizomes or stolons are vegetatively propagated. Ecotypes or F<sub>1</sub> hybrids are evaluated in the target environment and superior genotypes are released as cultivars. This type of propagation has become popular in waterlogged soils or areas with periodic floodings in northern Argentina (Ferrari Usandizaga et al. 2015).

Some polyploid species with a more regular meiosis are cross-pollinated and set a moderate amount of seed, which make seed propagation at large scale possible (Vogel and Burson 2004). *Chloris gayana*, *Panicum coloratum*, and *Setaria sphacelata* are good examples of cultivated pastures used in these transitional climate zones since they tolerate low temperatures during the winter and can be cultivated at higher latitudes in comparison with other warm-season perennial grasses. Recurrent phenotypic selection is regularly used to breed this group of species.

Apomixis, asexual reproduction through seed, is very common in *Brachiaria*, *Panicum*, *Cenchrus* and *Paspalum*. This type of reproduction offers the unique opportunity of developing seed propagated F<sub>1</sub> hybrids (Hanna and Bashaw 1987). The large variability present in apomictic species (Brugnoli et al. 2014) allow

breeding for the limiting aspects typical of subtropical environments, such as cold tolerance and cool-season growth (Acuña et al. 2019). However, specific breeding technics need to be used for breeding apomictic species.

The purpose of this paper is to describe the general aspects related to breeding warm-season perennial forages in subtropical South America, and provide examples of breeding activities directly related to the adaptation of new cultivars to these transitional climate zones.

## Results

### ***Breeding polyploid species with reduced seed fertility***

*Acroceras macrum* was introduced in Argentina from South Africa about 30 years ago, and has been cultivated as a vegetatively-propagated forage since then. It is usually planted in waterlogged soils between 28 to 31° latitude. The introduced germplasm was variable and two ploidy levels were present (4x=36 and 6x=54) (Ferrari Usandizaga et al. 2015). The species is sexual and cross-pollinated due to self-incompatibility (Ferrari Usandizaga et al. 2020). New hybrids were generated by controlled crosses, and heterosis was observed for cold tolerance and drought tolerance. A few selected hybrids are being evaluated in different locations in Northeastern Argentina aiming to release a new tetraploid cultivar in the near future. A cross-pollinated population has also been created with the purpose of developing a seed-propagated cultivar after a few cycles of selection.

The genus *Paspalum* L. has several species that grow naturally in swampy areas within the subtropics. Examples of these species are: *Paspalum modestum*, *P. repens*, *P. palustre*, and *P. oteroi* (Novo et al. 2017). All of them are prostrate growing species that can be vegetatively propagated. An on-farm experiment is underway in order to determine the adaptation of these species to a cultivated environment with seasonal floods. The challenge is to identify germplasm that can tolerate alternating periods of drought and flooding.

About 20 years ago several cultivars of *Hemarthrya altissima* were introduced in northern Argentina and southern Brazil. The limited adoption of these cultivars was attributed to the lack of seed and the need for vegetative propagation. However, the need to incorporate cultivated pastures in waterlogged soils has renewed the interest for this species. The two better adapted cultivars were Bigalta and Floralta. Two new cultivars were recently developed by the University of Florida named Kenhy and Gibtuck by crossing Bigalta and Floralta (Quesenberry et al. 2018). Since the new cultivars combined the grazing tolerance of Floralta with the high digestibility of Bigalta, they were introduced in Argentina in 2019 as part of an agreement between National University of the North East (UNNE) and University of Florida (UF). They are now being propagated and evaluated in three locations for seasonal forage yield and ground cover.

### ***Breeding polyploid species without limiting seed fertility***

Several cultivars of *Setaria sphacelata* originally developed in Australia have been introduced in Subtropical South America, including Narok, Kazungula, Splenda, and Solander. Improved populations have been recently developed for *S. sphacelata* in northern Argentina by INTA (National Agricultural Technology Institute, Mercedes Experimental Station) and northern Uruguay by INIA (National Institute of Agricultural Research, Tacuarembó) with the objective of releasing cultivars specifically adapted to the local environmental conditions. These breeding programs have used as the original germplasm the cultivars introduced from Australia, and the main breeding objectives were related to increasing cold tolerance, winter regrowth and seed yield. Recurrent phenotypic selection, and recurrent selection based on combining ability is being used in the program located in Argentina (McLean 2018).

Similar to *S. sphacelata*, a few cultivars of *Panicum coloratum* were brought to South America several decades ago. The species is well adapted to heavy soils in the transition between the subtropics and the temperate region of Argentina. A breeding program was established in central Argentina by INTA (Rafaela Experimental Station, 31°11'S, 61°29'W), with the main breeding objectives of developing cultivars with higher seed yield and tolerance to salinity. A cultivar named Kapivera has been developed using recurrent phenotypic selection and released in 2018 and is already in the market (Giordano et al. 2019).

*Chloris gayana* is also a cross-pollinated species native to Africa as *S. sphacelata* and *P. coloratum*. The species is well known because of its tolerance to drought and salinity. Since salinity is a major limitation in north central Argentina (the Chaco Region) a breeding program was developed by INTA, and diploid and tetraploid cultivars resulted (Ribotta et al. 2020). Diploid cultivars are better adapted to greater latitude in comparison to tetraploid ones. *C. gayana* and *P. coloratum* have great potential for advancing into high latitudes, today temperate zones, as a consequence of global warming.

### **Breeding apomictic species**

The main breeding programs for *Brachiaria* and *Panicum maximum* are located in the tropical area of South America, i.e. Colombia (CIAT, Cali) and Brazil (EMBRAPA, Campo Grande), so most developed cultivars are cold sensitive, and not well adapted to the subtropics. However, *Brachiaria brizantha* and *Panicum maximum* are the most important perennial warm-season grasses in the South American seed market (Jank et al., 2014), and cultivars as *B. brizantha* cv. Marandú and *P. maximum* cv. Gatton are among the most commonly sown in the belt between 27 and 28 ° South latitude. Expansion of the cultivation area of these species will be related to the establishment of new breeding programs or evaluation sites (for early evaluations) within higher latitudes (between 29 and 31 ° South).

Apomixis predominates among the more than 300 species of *Paspalum* (Quarin et al. 1992). There are several forage species native to southern Brazil, Northern Argentina, and Uruguay (Acuña et al. 2019). Cultivars have been developed for *P. notatum*, *P. atratum*, *P. guenoarum*, *P. nicorae*, *P. dilatatum* and *P. lenticulare*. The cultivated species better adapted to the subtropics are *P. dilatatum*, *P. notatum* and *P. guenoarum*. Efforts have been made by UNNE (Corrientes, Argentina), University of Buenos Aires (Buenos Aires, Argentina), Federal University of Rio Grande do Sul (Porto Alegre, Brazil) and INIA (Tacuarembó, Uruguay) to improve these species. Several cultivars resulted from these programs, such as *P. notatum* cv. Boyero UNNE, *P. notatum* cv. INIA Sepé, and *P. dilatatum* cv. Relincho UBA.

The technique used to develop most available apomictic cultivars is based on selection among germoplasm collections. Examples are *Brachiaria decumbens* cv. Basilisk, *P. maximum* cv. Mombasa, and *P. dilatatum* cv. Relincho. Hybrids generated by crossing induced sexual and apomictic tetraploid plants have been also released as cultivars, such as *Brachiaria* spp. cv. Mulato II, *Panicum maximum* cv. BRS Tamani and *P. notatum* cv. Boyero UNNE. Novel approaches have been developed with the aim to accumulate mainly non-additive genetic effects within syntetic sexual tetraploid populations, which are crossed in each cycle to apomictic male parents to generate improved apomictic hybrids (Miles et al. 2007; Marcón et al. 2020). These novel techniques combine classical and molecular methods as it is described in Figure 1.

### **Conclusions**

Several forage breeding programs are active in South America with the objective of developing cultivars adapted to the transition zone between the tropical and temperate regions. Vegetatively propagated hybrids are mainly developed for areas with waterlogged soils. *P. coloratum*, *C. gayana*, *P. notatum*, and *P. dilatatum* are among the seed propagated species with greater potential to advance into highest latitudes within the subtropics or even into the temperate regions. Cold tolerance and cool-season growth are the most limiting traits, however drought and salinity are also major limitations mainly in semiarid regions. Variations of recurrent phenotypic selection are preferentially used for breeding cross-pollinated species. Although ecotypic selection and direct generation of apomictic hybrids are the commonly used techniques for improving apomictic species, new population based methods are being developed and used.

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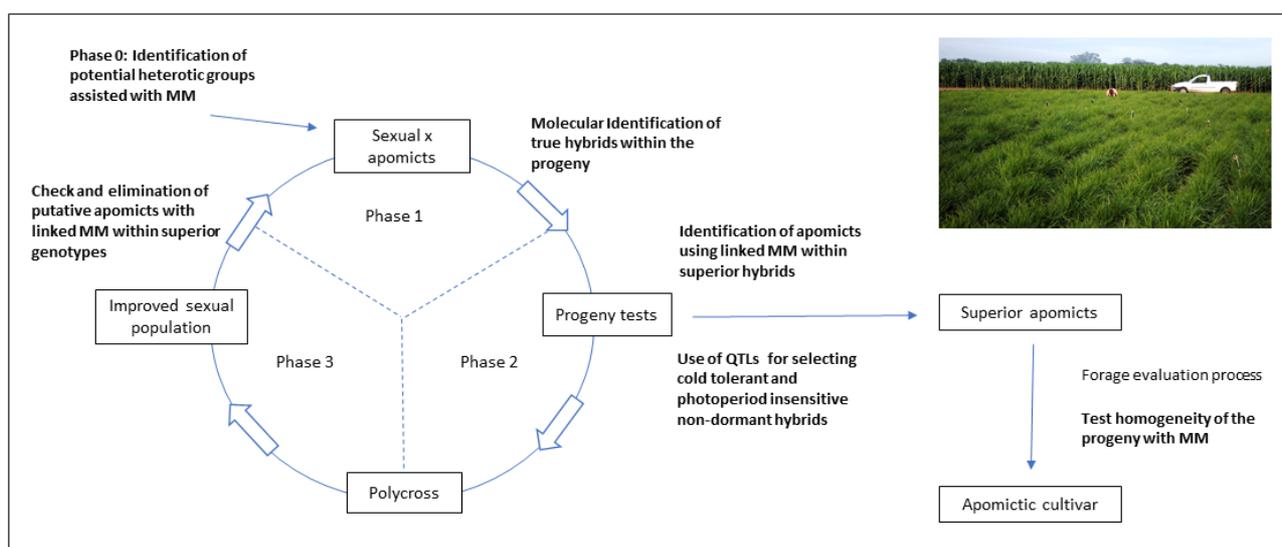


Fig. 1. Theoretical scheme of recurrent phenotypic selection based on a combination of classical and molecular techniques for forage tetraploid *Paspalum notatum*. The molecular markers (MM) use scenarios are indicated at each phase of the process. An image of an experimental/seed production plot of an improved apomictic hybrid cultivar of *Paspalum notatum* (cv. Boyero UNNE) is shown at top right (extracted from Ortiz et al., 2020. *Genes*, 11:1-27).