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PASPALUM ATRATUM AND P. MALACOPHYLLUM POLLEN CRYOPRESERVATION

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The genus Paspalum belongs to the Poaceae family, is native from Americas, and 214 species are found in Brazil under different ecological conditions, presenting high genetic diversity. Among these species, two stand out: P. atratum, which is a species with excellent forage production, great speed of establishment and regrowth, good acceptance by cattle and horses and has a small requirement in soil fertility, besides having a high production of seeds; and *P. malacophyllum*, that presents high palatability, being well accepted by the animals and presents tolerance to flooding. The majority of Paspalum cultivars that had been released as forage grasses were based on selection of apomictic ecotypes. Paspalum synthetic sexual tetraploid genotypes were obtained in breeding programs in Argentina and USA aiming hybridizations. Besides the countries previously mentioned, Brazil has also a *Paspalum* breeding program for forage and lawn purposes. However, the hybridization between parents just occurs if the flowering time is synchronized, which sometimes does not occur. Flowering time of different genotypes can be synchronized by manipulating photoperiod and temperature, however, the pollen preservation until the female parent starts to flower is the more feasible approach. The pollen cryopreservation success depends on different factors, like the cell water content, and, when it is high, pollen dehydration is necessary. This study aimed to determine the P. atratum and P. malacophyllum pollen stainability after 12month storage in liquid nitrogen. Pollen was collected under greenhouse conditions at Embrapa Pecuária Sudeste and exposed to two dehydration agents (saturated solution of LiCl (75%) and silica gel) as the humidity of the material should be around 20% to a successful freezing. Pollen dehydration was performed in a gerbox using the dehydration agents placed in an open petri dish with a grid supporting another open petri dish containing the pollen grains. The gerbox were kept closed during three different periods (30, 60 and 120 minutes), in three replications, at 25 °C. After this time, the pollen grains were placed in gelatine capsules inside cryotubes and immersed in liquid nitrogen. The non-dehydrated treatment was also evaluated. After slow thawing, which consists of pollen thawing by the maintenance of the samples for 30 minutes in freezer, then 30 minutes in refrigerator and then 30 minutes at room temperature, pollen stainability was evaluated using tetrazolium solution 0,25%. Fresh pollen was also evaluated as control. The non-dehydrated pollen samples presented 34% of stainability after 12-month storage. Pollen that was dehydrated with LiCl for 30 minutes and silica gel for 120 minutes and stored for 12 months into liquid nitrogen had the same stainability (70%) as the fresh pollen, which can be considered a well succeeded way of *Paspalum* pollen long term conservation.

<u>PRESENTER BIO</u>: Dr. Vigna is a Researcher at Embrapa and has more than 10 years of experience with genetic resources and molecular genetic breeding of various forage species (*Urochloa* spp., *Paspalum* spp., alfalfa).