

# Embryo-sac Development in Two Accessions of Giant Pangola *Digitaria valida* Stent

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## INTRODUCTION

Common Pangola, *Digitaria decumbens* Stent, is an excellent forage plant, being one of the most important species in Puerto Rico, but it is highly sterile. It has been found that it is male sterile as well as female because of meiotic irregularities (2)<sup>2</sup>. Giant Pangola, *Digitaria valida* Stent, Kob grass, or Kob Hill Grass (4,5) grows taller and with broader leaves than the common Pangola. Because it possesses some desirable forage characteristics it appeared to have usefulness in a breeding program, provided it was sexually reproduced. Before selecting it as parental material for breeding, its reproductive behavior had to be studied. Preliminary observations of very low seed set and meiotic irregularities raised questions about its potential value for breeding (3). Eleven South African *Digitaria valida* accessions have been kept under observation at the Agricultural Experiment Station in recent years. Two of them show a high percentage of pollen stainability, 77 to 94 percent, respectively, a regular meiosis in PMC, and have produced a progeny that revealed genetic recombination (6). The present investigation was conducted to obtain information about the embryo-sac development in these two accessions and its probable relation with the low seed set.

## MATERIALS AND METHODS

The accessions in question bear the labels 1950C and 1953A, corresponding to USDA plant introduction Nos. 209177 and 209372, respectively. Both are from Transvaal, 1950C originating from Krüger National Park, 1953A from an unnamed locality between Zeerust and Ottohoop.

Whole spikes were killed and fixed in CRAF solution. Ovaries were dissected and dehydrated with the n-butyl alcohol series, and embedded in paraffin. Sections were cut at 10 to 12  $\mu$ , and stained with propionic-carmin. By sectioning progressively older ovules, it was possible to follow the developmental sequence.

Photomicrographs were taken with a Leitz Makam camera mounted on a Leitz Ortholux research microscope. Final magnification is 820 $\times$ .

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<sup>2</sup> Italic numbers in parentheses refer to Literature Cited, pp. 482-3.

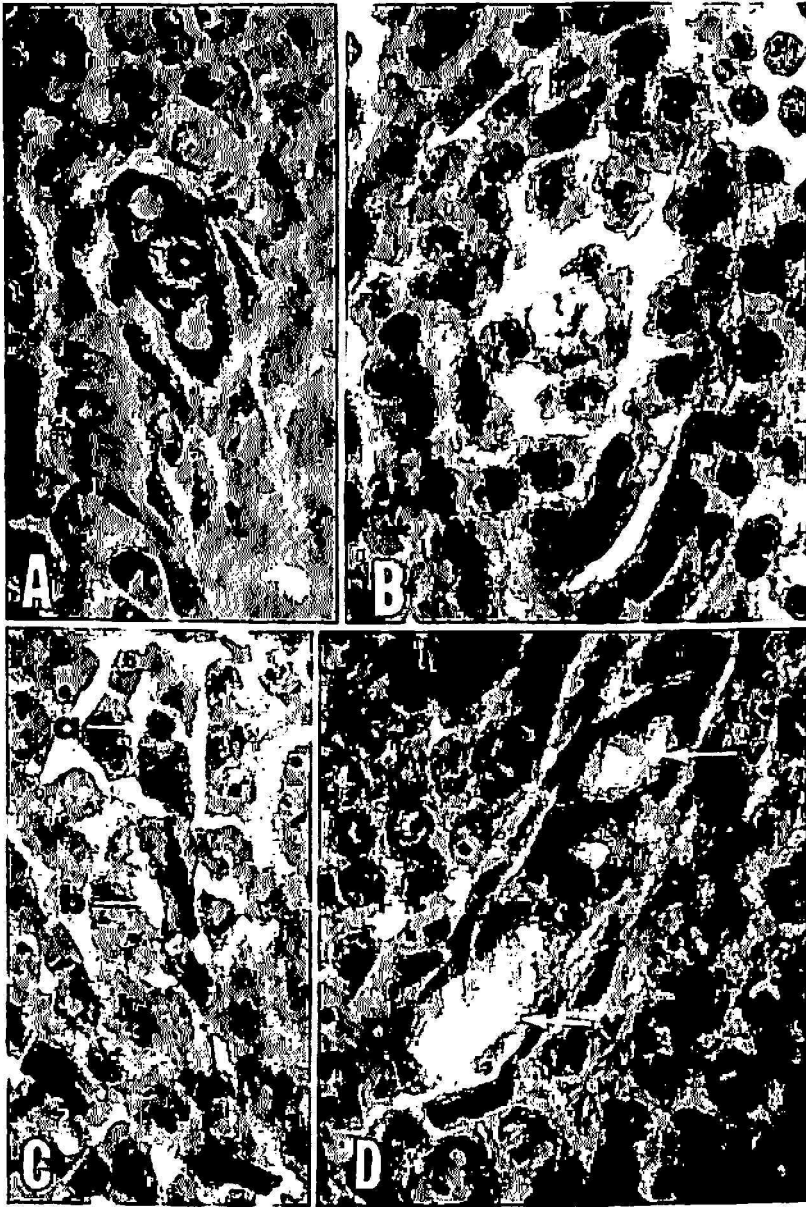


FIG. 1.— A, Archesporial cell differentiated from a single nucellar cell; B, pachytene stage in the MMC first meiotic division; C, a, functional cell and b, the 3 megaspores nearest the micropyle disintegrating; D, the enlarged megaspore with 1 vacuole (v) on either side of the nucleus.

## OBSERVATIONS

### NORMAL EMBRYO-SAC

The ovary of *Digitaria valida* has a single anatropous ovule, and the embryo-sac is of the *Polygonium* type (1).

The archesporial cell differentiates from a single hypodermal cell of the nucellus, (fig. 1,A) and undergoes two meiotic divisions forming a linear tetrad of megaspores. The meiosis is supposedly normal, but the actual division stages were not observed except for the pachytene stage in the

MMC first meiotic division, (fig. 1,B). After the meiosis the three megaspores nearest the micropyle disintegrate, leaving the chalazal member as the functional cell (fig. 1,C).

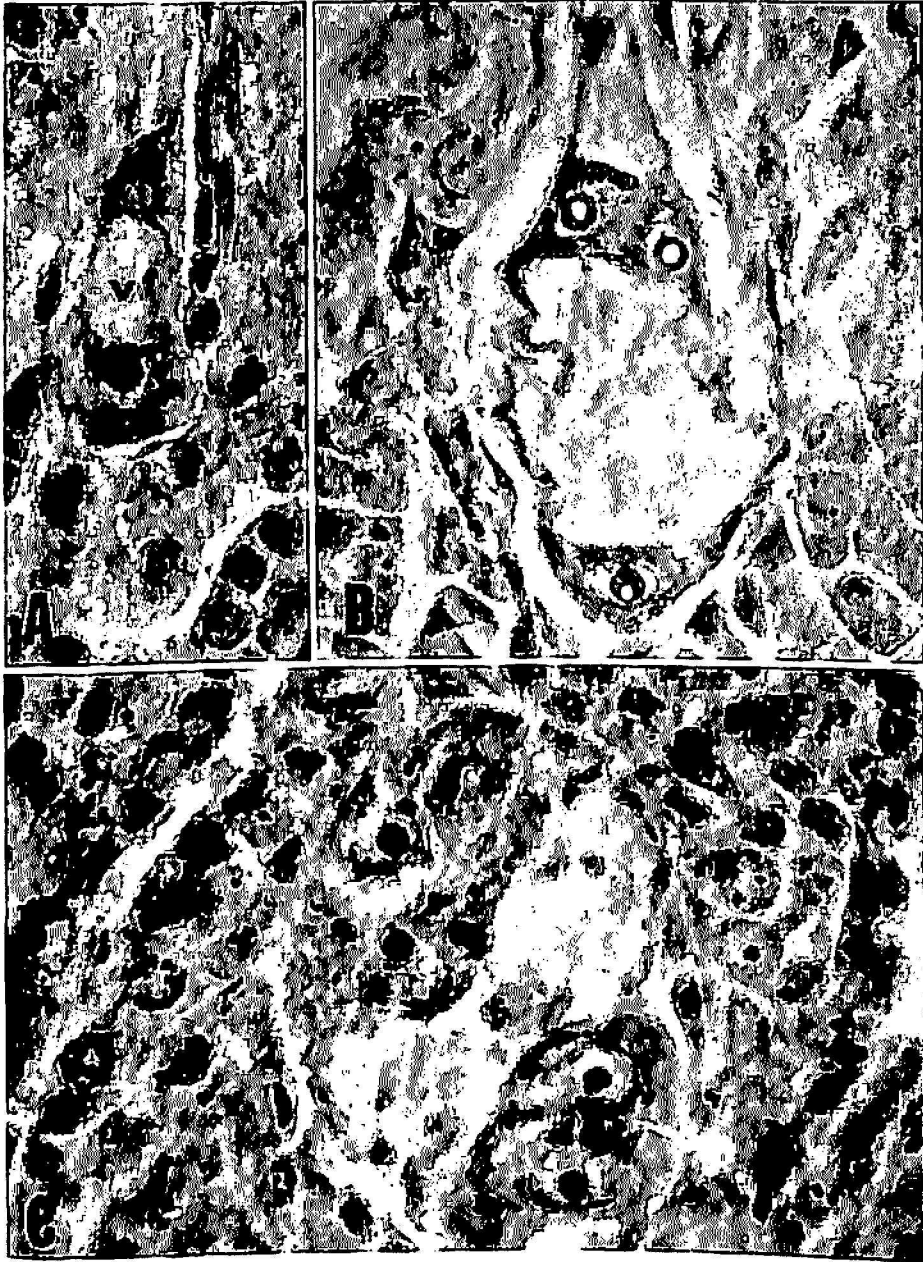


FIG. 2.—A, The 2-nucleate stage, a large vacuole (v) between the 2 daughter nuclei; B, the 4-nucleate stage; C, the 8-nucleate stage.

The megaspore nucleus enlarges and its enlargement is accompanied by increased vacuolation of the cytoplasm, one vacuole appearing on either side of the nucleus (1) (fig. 1,D). After the first division has taken place, the two daughter nuclei migrate to opposite poles leaving between them a large vacuole (fig. 2,A). The next division gives rise to a 4-nucleate stage (fig. 2,B) and the following division to the 8-nucleate stage (fig. 2,C).

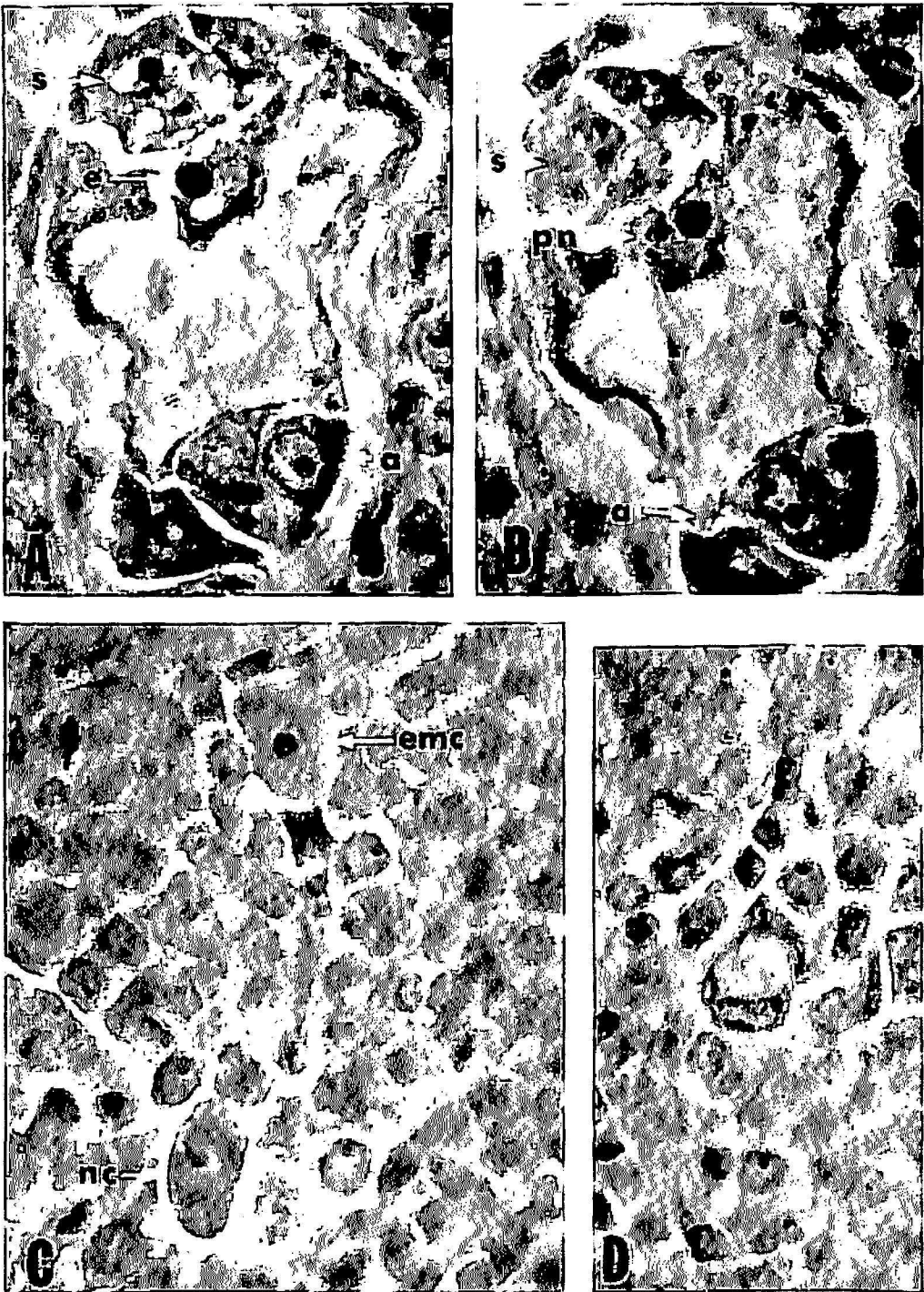


FIG. 3.— A, B, The complete embryo-sac. A, s, synergids; e, egg; a, antipodals. B, s, synergids; pn, polar nuclei; a, antipodals. C, Nucellar cell (nc) growing along with the functional megaspore (emc). D, A 4-nucleate apomictic embryo-sac.

The normal embryo-sac is always composed of more than eight nuclei. This condition arises by the occurrence of secondary divisions of the antipodals since sacs with up to seven and eight antipodals were encountered.

The mature embryo-sac consists of an egg apparatus, two polar nuclei, and the antipodals, ranging in number from 4 to 8, (fig. 3,A,B). The egg

apparatus consists of two synergids and the egg. The antipodals are usually present until the development of the embryo is well under way.

#### APOMICICTIC SAC

There is a very strong tendency to apomixis. The apomictic nucellar cells are encountered in about 50 to 60 percent of the ovules. They can be seen

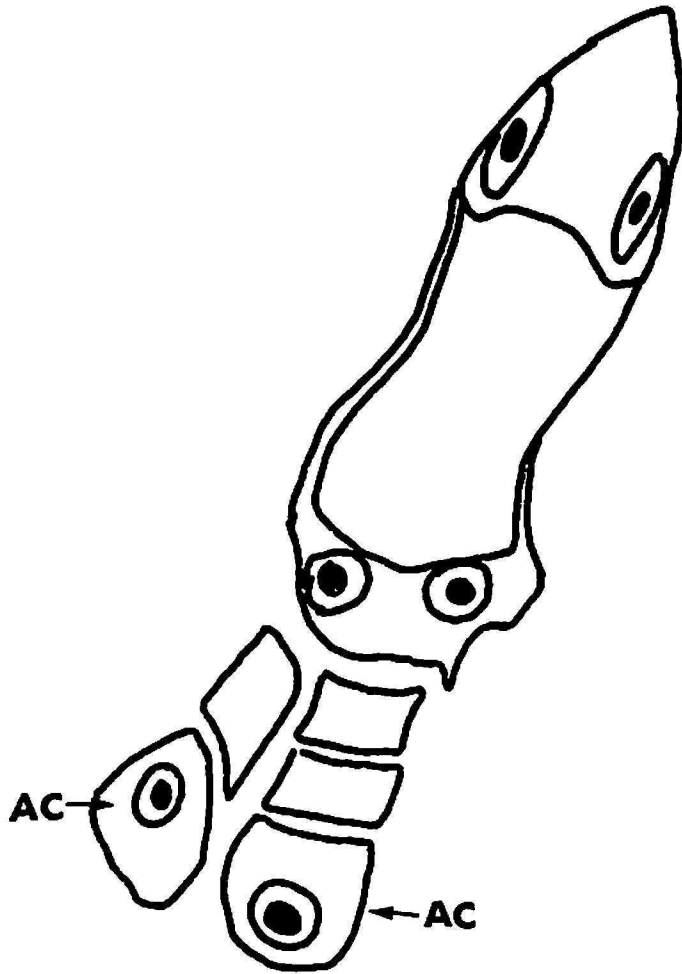


FIG. 4.—Apomictic nucellar cells (AC) growing along with the maturing normal embryo-sac.

growing along with the functional megaspore (fig. 3,C) or enlarging while the normal embryo-sac is almost mature. These apomictic nucellar cells, when present, are usually located near the antipodal region of the sac, (fig. 4). However, they may be found at the micropylar region, too. From all the material studied only one case of a small apomictic embryo-sac was encountered, (fig. 3,D). However, whenever apomictic nucellar cells were present the normal embryo-sac development did not proceed at normal pace. In many cases there was a complete arrest of the developing em-

bryo-sac; in other cases a mature normal sac might be encountered with one or more apomictic nucellar cells.

#### DISCUSSION

As long as an ovule is free from apomictic cells the development of the embryo-sac is normal. It is probable that the low seed set is due to apomictic cells which prevent the development of the normal sac, though there is no significant evidence of the formation of apomictic embryo-sac either. Apparently one inhibits the other. The percentage of ovules that presented apomictic nucellar cells included the second flower from every spikelet. According to Sheth *et al.* (2) the Pangola grass spikelets contain a lower sterile and an upper perfect floret. If this condition holds true for *Digitaria valida* as it does for *Digitaria decumbens*, then the future looks hopeful for a breeding program using it. Apparently it does hold true because a marked morphological difference is apparent and can be noticed by just looking at the flower. The lower floret is usually smaller in size and looks as if it were empty.

Contrary to what Sotomayor Ríos, *et al.* (3) found in other *Digitaria valida* accession, no significant evidence of meiotic irregularities in the PMC was encountered in these accessions, 1950C and 1953A, and the number of chromosomes was lower than that informed by them,  $2n = 38$  and  $2n = 36$ , respectively. Apparently the difficulty for seed production is the presence of the apomictic nucellar cells aside from other inherent characteristics.

#### SUMMARY

The embryo-sac development of two accessions of *Digitaria valida* Stent has been studied in an effort to clarify their reproductive behavior.

Even though irregular competition of nucellar cells was observed, a relatively high degree of normality was found in the development of the embryo-sac. It is concluded that these two accessions of Giant Pangola may be used for a breeding program and promising results may be expected.

#### RESUMEN

Se estudió el saco embrionario de dos especímenes introducidos de *Digitaria valida* Stent. (Pangola Gigante) para entender mejor el proceso de su desarrollo.

Aun cuando se observó cierta competencia irregular entre las células nucleares, se evidenció una relativa normalidad en el desarrollo del saco embrionario. Por tanto, si se usan estos dos especímenes de Pangola Gigante en un programa de cruzamientos, los resultados podrían ser prometedores.

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