



Cytochemical Differences on Male Sterile and Fertile Pollen in Sorghum (*Sorghum Bicolor* Moench)

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ABSTRACT

Cytochemical differences between the pollen of male sterile (296-A) and fertile (296-B, maintainer) lines of Sorghum (*Sorghum bicolor*. Moench.), were investigated employing cytochemical test. Male sterile pollen do not show any signs of synthesis of RNA and proteins. The present investigation the microspores do not enter the M phase of cell cycle, Due to non-synthesis of RNA and proteins, metabolism of sterile microspores suffers and the microspores start degeneration. All these events together, explain the probable reason for the observed reproductive failure in sorghum especially in the male sterile line, 296-A.

KEY WORDS: Sorghum, Male sterility, 296-A, B.

INTRODUCTION

Male sterility is the inability to produce functional male gametes, due to impaired male sex organ development or differentiation and /or abnormal male meiosis, without impairing the female sex [1]. It involves prevention of male gametophytic function while the female gamete formation is normal and functional [2]. Knowledge regarding the expression of male sterility genes greatly helps in unraveling how the fertility of pollen is genetically controlled. The mechanism of action of cytoplasmic male sterility (CMS) for Sorghum is presently unknown [3]. The present investigation is undertaken with an objective of exploring the probable cytochemical reason/s of male sterility in sorghum male sterile line, 296-A.

MATERIALS AND METHOD

Germplasm of fertile and male sterile lines of Sorghum (296-A and 296-B) were procured from M.P.K.V. Rahuri, (M.S.) and sown in the experimental fields of the Padmashri Vikhe Patil College of Arts, Science and Commerce, Loni. Crop was reared during the kharif season of 2009. Developing spikes containing different stages of anther development were collected and fixed in Carnoy's fixative (3:1 ethanol, glacial acetic acid). This material was used for cytochemical localization of RNA and total proteins was carried out following the methods [4, 5] respectively. Starch was detected using I2KI reagent.

RESULTS AND DISCUSSION

The morphological and cytochemical differences observed between the sterile and fertile pollen of Sorghum variety 296 is shown in Table 1. The male sterile pollen are smaller in size (Area = 5782) and the fertile ones are larger (Area = 8782). The sterile pollen is shreveled, highly vacuolated and possessed little or no cytoplasm, while the fertile pollen is round, less vacuolate and possessed dense cytoplasm.

The aceto-caramine staining has revealed that the sterile pollen is uninucleate and the fertile ones are binucleate. The cytochemical detection of RNA with pyronin-G reagent has revealed presence of no RNA in the sterile pollen, while the fertile pollen showed abundant presence of RNA (Table 1).

Similarly the sterile pollen did not stain positive with mercuric bromophenol blue reagent, indicating that there is little or no proteins are present in the sterile pollen. On the other hand the fertile pollen is darkly stained with bromophenol

blue indicating that they are rich in total protein content. The sterile pollen is I₂KI negative while the fertile pollen is strongly I₂KI positive. This indicates the fertile pollen synthesis starch in their cytoplasm, while the sterile ones do not. Results indicate that the reproductive failure leading to male sterility in Sorghum is gametophytic, occurring during macromolecular syntheses and mitosis stages of pollen development. The microspores of fertile anthers show synthesis of macromolecules like RNA and proteins and pollen mitosis leading to the formation of vegetative and generative nuclei. The microspores of sterile pollen do not show any signs of synthesis of RNA and proteins, nor undergo mitotic (pollen mitosis) division. [6] have reported that non-syntheses of nucleic acids and proteins in the sterile microspores lead to male sterility in *Caesalpinia pulcherrima*. In the present investigation also the microspores of sterile Sorghum plants do not synthesize the RNA and proteins.

The results indicate that non-availability of the respiratory substrate (starch) in the microspores leads to poor growth of microspores. They remain smaller in size. Due to non-synthesis of RNA and proteins, metabolism of sterile microspores suffers and the microspores start degeneration. All these events together, explain the probable reason for the observed reproductive failure in Sorghum especially in the male sterile line, 296 A.

Table 1 Morphological and Cytochemical differences between the male sterile and fertile pollen in Sorghum 296

Parameter	Male Sterile Line 'A'	Male Fertile Line 'B'
Shape	Shrunken	Round
Size (Area in μ^2)	558	898
Starch	Absent	Present
Protein	Absent	Present
RNA	Absent	Present

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REFERENCES

- [1]. Kaul, M.L.H. (1988). Male sterility in Higher plants. (Monographs of Theoretical and Applied Genetics, vol. 10) Springer Verlag, Berlin-Heidelberg. New York. pp. 1005.
- [2]. Sharnagpani, P.R. and Shirke, D.R. (2001). Histochemical changes during anther development in genic male sterile and fertile lines of pigeon pea (*Cajanus cajan* L. (Mill sp.) MS 3783). *Ad. Plant Sci.*, 14 (2):535-542.
- [3]. Mc Vetty, P.B.E. (1997). Cytoplasmic male sterility. In: *Pollen biotechnology for crop production and improvement*. (Ed.) Shivanna, K.R. and Sawhney, V.K. Cambridge University Press. p. 155-198.
- [4]. Tepper, H. B. and Gifford Jr., E.M. (1962). Detection of RNA with pyronin. *Stain Technol.*, 37: 52-53.
- [5]. Mazia, D., Brewer, P.A and Alfert, M. (1953). A cytochemical staining and measurement of protein with mercuric bromophenol blue. *Biol. Bulletin.*, 104:57-67.
- [6]. Apparao, B.J. and Shah, C.K. (1988). A cytochemical study of cytoplasmic male sterility in *Caesalpinia pulcherrima* L Indian bot. Reprtr., 7(1&2): 1-16.

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