RESEARCH PAPER | Botany | Volume : 4 | Issue : 12 | Dec 2014 | ISSN - 2249-555X

SDS-PAGE Analysis of Proteins in Sterile and Fertile Anthers in Sorghum.

KEYWORDS: Sorghum, male sterile, male fertile, Electrophoresis.

R.D. BORSE

PG-Department of Botany, Padmashri Vikhe Patil College of Arts, Science and Commerce, Pravaranagar, At/P: Loni, Tal: Rahata, Dist: Ahmednagar, Maharashtra

ABSTRACT *The present investigation employing Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis (SDS-PAGE), analysis of protein profiles of fertile and sterile lines of sorghum. Polyacrylamide gel were analyzed on their protein banding patterns (profiles). The A line (male sterile) differed significantly with B (male fertile) regarding composition of certain bands. Molecular weights of these proteins were determined employing the protein molecular weight marker, PMW-M of Genei, Bangalore, India Ltd. Difference in polypeptide profiles has been shown between male-sterile and fertile anthers. Whether the lack of a number of other polypeptides is directly responsible for pollen degeneration. Results indicate that the fertile and sterile anthers differed in their polypeptide band composition.*

Introduction

Electrophoresis is widely used to separate and characterize proteins by applying electric current. Electrophoretic procedure is rapid and relatively sensitive requiring only micro-weight of proteins. Electrophoresis in the polyacrylamide gel is more convenient than in any other medium such as paper and starch gel. Separation of proteins in SDS-free polyacrylamide gels relies on both the charge and size of the protein, whereas it depends only upon the size in the SDS-gels. Analysis and comparison of proteins in a large number of samples is easily made on polyacrylamide gel slabs. SDS is an anionic detergent, which binds strongly to, and denatures proteins. The number of SDS molecules bound to polypeptide chain is approximately half the number of amino acid residues in that chain. The protein-SDS complex carries net negative charges, hence polypeptides move towards the anode and separation is based on the size of the protein.

Materials and Methods

The experimental plant material used in the present investigation is sorghum (*Sorghum vulgare* Pers [Syn. *Sorghum bicolor* L. Moench]). The sterile and fertile anthers of sorghum, containing sterile and fertile pollen, respectively, were analyzed for their protein profiles, SDS-PAGE technique of Devis, (1964), modified by Dadalani, et al., (1993). In this plant germplasms of these A, and B lines were procured from the Sorghum Improvement Project of Mahatma Phule Krishi Vidyapeeth (MPKV), Rahuri, (District Ahmednagar, Maharashtra state, India). The germplasms thus collected were sown in the experimental fields of the Padmashri Vikhe Patil College of Arts, Science and Commerce, Pravaranagar, and the crop was reared during the kharif and rabi seasons of 2002-03. SDS-PAGE was carried out following the methods as suggested by Davis, (1964), Laemmli, (1970) and Dadlani and Varier, (1993). Bhadula and Sawhney (1991)

Result and Discussion

Protein profiles of male sterile and male fertile lines (104A, 104B) are observed. Variation could be observed in the number, position and molecular weight of polypeptides between male fertile and male sterile lines. The eletrophorogram has revealed the presence of four polypeptide bands in the anther proteins of male fertile. Molecular weights of these polypeptides are 54,850, 30,230, 23,240 and 10,230, Daltons (Table No 1). The male sterile lines showed only three polypeptide bands. Molecular weights of these polypeptides are, 54,850, 23,240 and 10,230, Daltons. Thus the two male sterile lines (104A) showed a missing polypeptide band which has a molecular weight of 30,230 Daltons. This band was prominent in B line. The male fertile line were characterized by the presence of dense bands and the male sterile lines was characterized by the presence of narrow and light bands. The SDS-PAGE analysis of male sterile and maintainer lines revealed differences in the number, position and molecular weight of polypeptides. The male fertile lines (B lines) showed the presence of 4 polypeptide bands while the male sterile lines (104A) showed the presence of only 3 polypeptide bands.

Table 1. Total Protein profile of sorghum lines

Line	Band No.	Rm Value	Molecular Weight in Daltons
104 B	1	0.5714	54,850
	2	0.0952	30,230
	3	0.0476	23,240
	4	0.0238	10,230
104A	1	0.5714	54,850
	2	--------	--------
	3	0.0476	23,240
	4	0.0238	10,230

Difference in polypeptide profiles has been shown between male-sterile and fertile anthers. Whether the lack of a number of other polypeptides is directly responsible for pollen degeneration. Results indicate that the fertile and sterile anthers differed in their polypeptide band composition. Male-sterile anthers contained fewer polypeptide bands than the fertile anthers. The polypeptide band with a molecular weight of 30,230 Daltons, present in male fertile line anthers and is absent in male sterile anther polypeptides. Similar reduction of polypeptide bands in sterile anthers was reported by several authors (Alam and Sandal, 1969; Banga et al., 1984; Sawhney and Bhadula, 1987); (Markova and Daskaloff (1974). This indicates disturbed and derived protein metabolism in the male sterile anthers. Thus assuming the male sterile gene action starts during the pollen development, one could suppose that it may encode a product required for the normal development of both vegetative and generative

Volume : 4 | Issue : 12 | Dec 2014 | ISSN - 2249-555X

tissues of the anther. Thus our results clearly indicate that, in sorghum reproductive failure and fertility restoration are gametophytic, occurring during the starch–filling stages of pollen development.

Acknowledgement
The authors are grateful to Dr. S.R. Walunj, Principal, P.V.P. College, Loni, for providing necessary laboratory facilities and UGC providing financial assistance.

REFERENCE Alam, S and Sandal, P. C. (1969). Electrophoretic analysis of anther proteins from male fertile and male sterile sudangrass Sorghum vulgare var sudanensis (Piper). Crop Sci 9:157-159. | Banga, S.S., Labana, K.S and Banga, S.K. (1984). Male sterility in Indian mustard (Brassica juncea (L.) Coss: a biochemical characterization. Theor. Appl. Genet.68: 515-519. | Bhadula, S.K and Sawhney, V.K.(1991). Protein analysis during the ontogeny of male sterile stamenless-2 mutant stamens of tomato (Lycopersicon esculentum Mill.). Biochem. Genet.29: 29-41. | Davis, B.J. (1964). Disc electrophoresis method and application to human serum. Ann. NY. Acad. Sci. 122: 404-429. | Dadlani, M and A. Varier. (1993). Electrophoresis for variety identification. Technical Bulletin published by Division of seed science and Technology. IARI, New Delhi. | Laemmli, H. (1970). Cleavage of structural protein during the assembly of the head of bacteriophage T4. Nature. 227:680-685. | Markova, M and Daskaloff , S. (1974). Electrophoretic studies of protein in reproductive tissues of the cytoplasmic male sterile form of pepper (Capsicum annuum L.). In: Proc. Eucarpia Meet Budapest, pp 105-114. | Sawhney, V.K and Bhadula, S.K. (1987). Characterization and temperature regulation of soluble proteins of a male sterile tomato mutant. Biochem.Genet. 25:717-728. |