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GENETIC DIVERSITY STUDIES IN SOME CHILI HYBRIDS FROM KHANDESH REGION USING SDS - PAGE

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ABSTRACT
Present study was undertaken to estimate genetic diversity in 12 hybrids of chili (Capsicum annm L.)from Khandesh region,using SDS-PAGE. Storage seed protein banding pattern studied. Total seed storage protein ware extracted and separated on 10 % polyacrylamide gel using standard protocol. The data generated from scoring of bands of proteins separated in 12 chili hybrids ware analyzed using NTsys& UPGMA dendogram developed. Dendogram analysis provide 2 major clusters. Hybrid 12(Pikato green) & Hybrid 1(ARCH-82) Shows maximum similarity, indicates probable origin from same parent. Hybrids from clusterI & II shows considerable genetic distance.

Keywords: Capsicum annum L., Seed storage protein, SDS-PAGE, UPGMA dendogram

INTRODUCTION
Chili (*Capsicum annum L.*) Is the most important vegetable cum spice crop because of their pungency, flavor, color and aroma. This is belonging to the family Solanaceae.Number of capsicum species has been known as part of human diet science the beginning of civilization.There is little recorded genetic diversity among cultivated chili. However, general knowledge is that commercial verities and particularly hybrids verities of vegetable crops are based on increasingly narrower genetic base(Padma.J and Shiva K. 2013).Previously the data like color, shape, size, pungency, flavor and physiological traits are generally used to estimate the magnitude of genetic diversity present in chili germplasm. However with the increase in the number of verities in each crop, it is difficult to distinguish verities/ hybrids on the basis of data from morphological characters alone. Such data may not provide an accurate indication of genetic diversity because of environmental influences upon the expression of observed traits. This has leads to the development of new stable parameter such as use of genetic material (Nucleic acid and Proteins) as a tool to estimate genetic diversity. Recently genetic diversity in capsicum has been studied using cytological and biochemical system.(Gopinath et.al., 2006.) Seed storage protein used as genetic markers in the study of genetic variation because any change in coding sequence of a gene generally reflects the corresponding change in the primary structure of a protein(Srivilli et.al.,1999)Genetic and taxonomic relationship in the genus capsicum have been investigated with electrophoresis of seed storage protein banding patterns(Valdova et.al., 2000; Zubiada et.al., 2006).

The present work was carried out on total 12 chili hybrids. The 12 chili hybrids from Khandesh region of Maharashtra state were taken to evaluate seed storage protein variability and for their exploitation in successful crossing programs.

MATERIALS AND METHODS
Seed Material: Seed material used for present study includes 12 accessions (Hybrids) which ware locally available in Khandesh regionof Maharashtra. 10 seeds of each accession ware taken for total seed storage protein extraction.

EXTRACTION OF SEED STORAGE PROTEIN
10 seeds of each accession ware powdered separately. 10 mg seed powder of each line along with 5 ml of 0.01M Tris.HCL buffer (pH 7.5) ware taken & vortex thoroughly to homogenize. The resulting homogenate was centrifuged at 15,000 rpm for 10 min. at 4°C. The supernatant was collected and used as soluble protein for SDS-PAGE.

SDS-PAGE analysis: Extracted soluble protein ware separated by one dimension SDS-PAGE. Percent of running and stacking gel was kept 10% &% respectively. Electrophoresis was conducted at a constant current 85mA until the tracking dye reached the bottom of the gel. After electrophoresis the gel was stained overnight in 0.25% Coomasie Brillent blue R-250, followed by de-staining in Methanol and Acetic acid solution for 45 min.The gel was further de-stained until the background was clear enough for band scoring. Protein marker of Hi-Media was used as standard to score the sample bands. Molecular weight of protein band were estimated by their relative mobility. After de-staining the gel were photographed using gel documentation system 'UVtech'.

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Data analysis: For Genetic diversity analysis, every scoreable band was considered and scored 1 for presence and 0 for absence. The bivariate 1-0 data was used to estimatethe pair-wise genetic distancefollowing UPGMA procedure for dendrogram (Nei and Li., 1979).

RESULT & DISCUSSION

Genetic diversity among cultivars are pre-requisite to identify high performing parents (Padma J. and Shiva S. 2013). Similarity matrix was subjected to UPGMA clustering to generate dendogram. Cluster analysis of UPGMA is extensively used for genetic diversity studies in important plants.Electrophoresis analysis of 12 chili genotypes reveal not much more variation in the banding pattern of proteins (Figure 1). Twelve Capsicum hybrids ware characterized on the basis of seed storage protein using SDS-PAGE (Figure2). Studied cultivars were group into 2 major clusters. Cluster I corresponding 4 genotypes where as cluster II includes 8 genotypes. Dendogram depicted by UPGMA shows low genetic diversity as most of verities belong to single cluster. This may be due to genotype homozygosity (Odeigah P.G.et.al., 1999)which causes the narrow genetic base. Valdova et.al., (2000) and Zubiada et.al., (2006) also studied chili verities of India using seed storage protein. Our results shows less variability in among the studied samples except for some lanes (L1,L4,L8)where considerable variation observed. There line specific bands shows variation between 12 chili genotypes and themselves. This observation suggests that SDS-PAGE technique can be used to differentiate this genotypes from the rest of the genotypes. Pikato green and ARCH 82 were found to be closely similar. This may attributed to the same genetic background of them. Whereas Sizziling hot, Shama, Shimla red, Omega, Sitara and Local genotyps formed separate cluster in cluster II. This may due to their morphological differences between them. Also, cluster pattern of these genotypes revealed that these genotypes may be developed from same parental genotypes.

CONCLUSIONS

The present study exploited seed storage protein for identification of better parents. Hybrid 12(Pikato green) and Hybrid1 (ARCH-82) shows maximum similarity indicates probable origin from same parents.Parents of cluster I & cluster II hybrids can be used to produce improve hybrid from Khandesh region.

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REFERENCES

- Gopinath , K., N.V. Radhakrishnan and J.Jayaral (2006) Effect of propicinazole and difenocozole on the control od anthracnose of chili fruit caused by *Colletotrichum capsici* . Crop Port., 25: 1024-1031.
- Nei, M. and W.H. Li (1979) Mathematical model for studying genetic variation in terms of restriction endonucleases. Proc.Natl.Acad.Sci.USA. 76: 127-5273.
- Odeigah, P.G. C., B. Oboh and I. O Aghalokpe(1999). The characterization of Nigerian varities of peeper, *Capsicum annum* and *Capsicum frutescence* by SDS-PAGE of seed protein. Genetic Resources and Crop Evolution 46: 127-131.
- Padma,J. and Sivasubramaniam, K. (2013). Characterization of chili genotype using SDS-PAGE protein profile. Internat.J. agric.sci.,9(2):539-541
- Srivalli, T., N. Lakshmi and C.H.G. Gupta(1999). Analysis of seed proteins by PAGE in diploids, tetraploids and tetraplois hybrids of *Capsicum.* Capsicum and Eggplant Newsletter 18:48-51
- Vladova, R., K. Petcolicheva., R. Pandeva and V. Nikova(2000). Seed storage protein of *Capsicum annum* L., *Nicotiana Tabaccum* L. and *Lycopersicon escluentum* L. cultivars: Comparative electrophoretic study. Plant genetic and Breeding.30:29-33.
- Yatung Taso, Dubey rakesh Kr, Singh Vikas, Upadhay Garima and Sing Siddhartha (2014) Studies on seed protein profiling in chilli (*Capsicum annum* L.)Genotype of Northeast India,Austrilian Journal of Crop sci.,8(3): 369-377.
- Zubaida,Y., M. Shahib., S. k. Zabta., K.Mir Ajab and R.Ashiq(2006). Evaluation of taxonomic ststus of medicinal species of the genus *Solanum* and *Capsicum*based on PAGE. Pakisthan Journal of Botany 38(1): 99-106.

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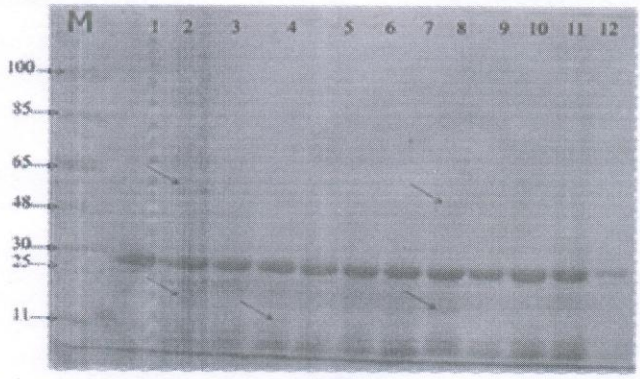

Figure – 1: Total seed storage protein profile of 12 Chilli hybrids. M= Molecular weight marker; 1-12 chili hybrids.

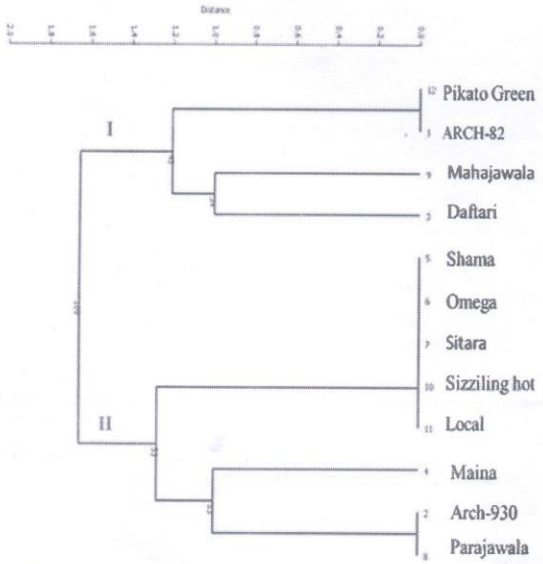

Figure - 2: UPGMA dendrogram of 12 Chili hybrids based on seed storage protein.