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ABSTRACTS

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BRD SCHOOL OF BIOSCIENCES
SARDAR PATEL UNIVERSITY
VALLABH VIDYANAGAR - 388 120
GUJARAT

MBBS-1: DETERMINATION OF LIPOXYGENASE ACTIVITY USING HPTLC

Amit Kumar N.Thakkar and Vasudev R.Thakkar
BRD School of Biosciences, Sardar Patel University, Vallabh Vidyanagar-388120

Lipoxygenase (LOX) (EC: 1.13.11.12) catalyses the dioxygenation of polyunsaturated fatty acids. Lipoxygenases are widely distributed in plants, fungi and invertebrates and in mammals. A variety of assay methods have been used to measure the LOX activity. However all these methods have been limited by the non specificity. In present work we have measured LOX activity by estimating the consumption of linoleic acid by the LOX enzyme using HPTLC (High performance thin layer chromatography). This method seems to be more specific than earlier methods for measuring LOX activity. The reaction products from the reaction mixture were extracted using mixture of chloroform : methanol and the linoleic acid was separated on precoated silica gel plates 60 F_{254} plate using appropriate solvent system (hexane: diethyl ether, acetic acid). The remaining linoleic acid after the reaction was measured using the calibration curve of standard linoleic acid with the help of Wincats planar chromatography software. The presence of LOX protein was confirmed on 10 % native gel activity using O-dianisidine as stain and was found to be of 73 Kilo Dalton.

MBBS-2: ISOLATION, PURIFICATION AND CHARACTERIZATION OF L-ASPARAGINASE FROM *ASPARAGUS OFFICINALE* L.

Sushil kumar[1], Vishal Oza[2] and R.B. Subramanian[2]
[1]AMCOST, Anand
[2]BRD School of Biosciences, Sardar Patel University, Vallabh Vidyanagar-388120

L-asparaginase (L-asparagine amidohydrolase, EC 3.5.1.1) is one of the basic drugs in the treatment of acute lymphoblastic leukaemia (ALL). In recent years plant L-asparaginases have gained importance due to significant side effects of L-asparaginase obtained from microbial sources. Various plant parts from different plant species were screened for L-asparaginase activity by Nessler's method. *Asparagus officinalae L.* was identified as a potential source of L-asparaginase. The protein was purified using different purification methods. Each step of purification was monitored by activity assay and SDS – PAGE. Various parameters such as optimum pH, Temperature, K_m and V_{max} were studied using activity assay. Molecular weight (approximately 36 KD) of enzyme subunit and PI value (Between 5 to 5.5) were determined by SDS PAGE and 2D PAGE respectively. The specific activity found was 106.66 IU/mg. optimum pH and optimum temperature were found to be 7 and 37^{0}C, respectively. The plant therefore, holds promise as a potential source of this therapeutically important protein.

MBBS-3: RAPD AS A MOLECULAR TOOL FOR IDENTIFICATION OF CHILLI (*CAPSICUM ANNUM.* L) GENOTYPES

Diwakar Singh, Reetu Mehta and J.G. Talati
Department of Biochemistry, Anand Agricultural University, Anand – 388 110, Gujarat, India

The Chilli (*Capsicum annum* L.) genotypes were identified using Polymerase Chain Reaction (PCR) with arbitrary oligonucleotide primers by the Randomly Amplified Polymorphic DNA (RAPD) method. PCR with 10 arbitrary primers applied to 11 promising genotypes, out of which, at least 5 primers amplified the DNA and produced a total of 37 useful markers, where 18.91 % (7 bands) were common across all varieties. The remaining 30 bands (81.08 % of the total products scored) were polymorphic among the genotypes tested. This accounts to an average of 6 polymorphic bands per primer. Few genotypes were identified on the basis of specific banding pattern through visual examination of

electrophoresis gels. Thus, RAPD as a molecular tool offers a potentially simple, rapid and reliable method for Chilli genotype identification.

MBBS-4: ELECTROPHORETIC CHARACTERIZATION OF AROMATIC RICE VARIETIES USING ISOZYMES AND SDS-PAGE

Rallapalli Ravikiran, Vinit Patel S, Vipulkumar B. Parekh and Subhash N.
Department of Agricultural Botany, B.A.College of Agriculture, A. A.U., Anand 388110
E-mail: ravi_aau@yahoomail.co.in

Aromatic rice is of great value in the Indian sub continent and its cultivation is characterized by the use of distinct varietal profile as these varieties are predominantly grown in the Himalayan foothill regions. In order to ensure that genetically distinct varieties are available to promote agricultural productivity, it is necessary to characterize the commonly grown varieties in the region. In the present study 30 aromatic and 5 non-aromatic locally adapted rice cultivars were studied for variation at the isozyme level. Rice leaves were collected at the growth stage of 3-4 leaves. The analysis was carried out as per the standard protocol. Electrophoretic studies on Polyphenol oxidase (PPO) and Esterase (EST) isozymes were conducted. The bands of Polyphenol oxidase and Esterase isozymes on gel after electrophoresis varied among 35 varieties of rice cultivars. In the present study SDS-PAGE analysis was carried out. The aromatic genotypes differed from each other with respect to the total number of bands which ranged from 4 to 14. A dendrogram was generated on the principle of unweighted pairwise method using arithmetic average (UPGMA) and the genotypes were grouped in to clusters. The similarity index value ranged 0.08 between Basmati-370 and IET-19230 to 0.75 between IET-16313 and IET-18990. The average similarity index was 0.27. The studies indicate good possibility of using isozyme and SDS-PAGE profile for identification of rice cultivars.

MBBS-5: DIVERSITY ANALYSIS OF CMS AND RESTORER LINES OF PEARL MILLET (*PENNISETUM GLAUCUM* (L.) R. BR. EMEND. STUNTZ) USING RAPD AND MICROSATELLITE MARKERS

Vipulkumar B. Parekh[1], G. C. Jadeja[1], R. Ravikiran[1] and N. Subhash[2]
[1]Department of Agricultural Botany, B. A. College of Agriculture, A.A.U., Anand, Gujarat
[2]Plant Tissue Culture Laboratory, A.A.U., Anand, Gujarat
E. mail: jadeja_gc@yahoo.co.in

Two PCR-based molecular fingerprinting techniques, *viz.* Randomly Amplified Polymorphic DNA (RAPD) and Simple Sequence Repeats (SSR) were applied in pearl millet genotypes to compare suitability for quantifying genetic diversity. Nineteen pearl millet genotypes having important agronomic traits were assayed for polymorphism using 18 RAPD primers and 18 CT based microsatellite (SSR) markers. Average Polymorphism for RAPD analysis was 89.86%, while for SSR analysis, it was 50%. Polymorphism Information Content (PIC) values for RAPD analysis ranged from 0.84 to 0.94 and for SSR analysis it varied from 0.00 to 0.68. For RAPD analysis, maximum band size was found to be 4526 bp and minimum was 117 bp; whereas in SSR, maximum band size was 304 bp and minimum was 200 bp. The results indicated that RAPD markers were more polymorphic than microsatellite markers. Comparison of cluster results obtained from the RAPD and SSR; restorer IPC1658 formed major distinct cluster in both analysis, proving that it was the most diverse line among all the parental genotypes. Pooled RAPD analysis grouped CMS and restorer lines into two distinct clusters. However, microsatellite markers did not separate CMS and restorer lines into distinct clusters.