GENETIC DIVERSITY OF MUNGBEAN [(Vigna radiata (L.) WILCZEK)]
GENOTYPES BASED ON SDS-PAGE OF ALBUMIN SEED STORAGE PROTEIN

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ABSTRACT

41 test genotypes including 34 improved varieties, five local land races, one wild accession of mungbean; and one local variety of urdbean were analysed for albumin seed storage proteins by SDS-PAGE. The electrophoregrams revealed eleven polypeptide bands for albumin which has shown a great array of polymorphism in presence/ absence of bands as well as the intensity/ thickness of bands. The test genotypes were distributed over six clusters. The cluster analysis based on, albumin polypeptide banding patterns revealed different separate clusters for genotypes e.g., Pant M-5, RCM 15, OGG 57, OUM 7, COGG-912 and ML 613 indicating their genetic divergence from rest of the genotypes. These genotypes could serve as valuable breeding material for further crop improvement.

Key words: Genetic diversity, Albumin seed storage proteins, SDS-PAGE, Mungbean [Vigna radiata  (L.)].

INTRODUCTION

Mungbean is an important pulse crop of India and Orissa in particular. India leads the list of mungbean growing countries with an area of 2.98 m. ha. and production of 1.29m. tonnes. But, its productivity is very low due to low yield potential of the existing varieties, vulnerability to cold stress and diseases(YMV and powdery mildew) and usual practice of minimal cultivation in rice follow. Among different pulses, mungbean harbors high vit-B content and possesses high degree of digestibility. It bears traces of antinutritional factors without any health hazards, compared to other legumes. Besides, mungbean bears moderately high protein content ranging from 17.2 to 29.9% in seeds with an average of 22.8%(Naik 1998). However, available literature does not reveal any work for amelioration of protein quality in mungbean. Thus, there is an urgent need to breed suitable genotypes with desirable traits for increasing crop production as well as quality. In the present pursuit, a set of mungbean germplasm lines were analysed for albumin seed storage proteins through SDS-PAGE to assess relative genetic distance of the genotypes in terms of protein quality.

MATERIAL AND METHODS

The experimental materials comprised 41 test genotypes including 34 improved varieties, five local land races and one wild accession of mungbean; and one local variety of urdbean. Albumin seed storage proteins were extracted with pre-chilled distilled water, denatured with an equal volume of cracking buffer (0.125M Tris HCl pH 6.8, 4% SDS, 20% glycerol, 10% 2-mercaptoethanol, 0.1% bromophenol blue) at 80°C in hot water bath. Seed proteins were analyzed through vertical slab gel (12.5% polyacrylamide gel) SDS-PAGE following Laemmli(1970) with minor modifications at 220v voltage and a constant current of 60mA for four hours. To check up the reproducibility, each set of experiment comprising 10-11 genotypes was carried out on two separate gels under similar electrophoretic conditions. After electrophoresis, gels were stained with 0.125% w/v Coomassie brilliant blue R 250, 50% v/v methanol, 10%v/v glacial acetic acid for four
Fig. 1: Dendrogram showing genetic diversity of genotypes (Sl. No. 1-41) based on albumin polypeptide banding pattern
hours with intermittent shaking followed by
destaining overnight in 50% methanol and 10%
Acetic acid; and finally several washing with
5% methanol and 7% acetic acid. The
molecular weight of the dissociated polypeptides were determined by using
molecular weight markers or protein standards
with known molecular weights i.e, bovine
plasma albumin(66kd), egg albumin(45kd),
glyceraldehydes-3-phosphate dehydrogenase
(34.7kd) and bovinepancreas
tripsinogen(24kd).

In the present investigation, genetic
diversity among the test genotypes was
assessed based on similarity coefficients
calculated using eleven polypeptide bands of
albumin following Jacord(1908) and UPGMA
(unweighted pair group method with arithmetic
average) dendrograms were constructed as per
Sneath and Sokal(1973).

RESULTS AND DISCUSSION

The dendrogram showing clusters at
appropriate phenon levels is presented in fig
1. The albumin seed protein profile analysis
at comparatively lower phenon level revealed
distribution of genotypes over six divergent
clusters. Ghafoor et al .(2002) revealed four
clusters of genotypes in a set of diverse
germplasm lines of Vigna mungo and V . radiata
resembling to V . mungo for seed characters.
Gallab et al .(2007) delineated the genetic
relationship among a set of mungbean
genotypes from a dendrogram which showed
three different genetic clusters. Ashghar et
al.(2003) reported intra- and interspecific
variation of seed protein in 29 accessions of
Cicer L. following SDS–PAGE analysis. They
classified all the genotypes into five distinct
genetic clusters based on similarity in banding
pattern and delineated the genetic relationship
among the accessions.

In the present investigation, the
multivariety cluster(s) was dissociated rapidly
into several distinct subclusters, each
containing a few genotypes. Thus, it was
possible to discern subtle difference between
genotypes grouped in different cluster(s) and/or
subcluster(s) at different phenon levels. At
lower phenon level, the test genotypes were
distributed into six divergent clusters e.g., IA₁,
IA₂, IIA₁, IIA₂ and IIB, each containing one or
a few genotypes; and IIB₂ which was a large
multivariety cluster. Cluster IA₁, IA₂ and IIA₂
included one variety each i.e., RCM 15, Pant M
5 and OGG 57 respectively. Cluster IIA₁
included var. ML 613, COGG 912 and OUM
7; and cluster IIB₁ comprised Keonjhar local
and LGG 460. At moderate phenon level, the
multivariety cluster IIB₂ was divided into a
small cluster IIB₂₁ with five genotypes including
the wild mungbean accession TCR 213 and a
large cluster IIB₂₂ which contained rest 28 test
genotypes. The wild accession being placed in
a separate small cluster, exhibited appreciable
divergence with other genotypes. However,
more polymorphic bands needed to be scored
to reveal its distinctiveness at lower phenon
level. The large cluster IIB₂₂ again dissociated
sequentially to form several subclusters which
would have less genetic divergence inter se. The
mayurbhanj local, a popular urdbean local land
race was included in a subcluster of IIB₂₂
which revealed a greater homology between
mungbean and urdbean. The above findings
envisaged large genetic diversity and genotype-
specific protein type among the test genotypes.
However,Roy (2003) observed similarity
coefficient values ranging from 68-100% with
high average coefficient value of 90% and
consequently obtained very restricted level of
polymorphism among some Indian cultivars
and wild accession of mungbean. Tomooka et
al. (1992) carried out electrophoretic
assessment of 581 local strains of mungbean
collected from different regions of Asia. They
observed 8 protein types on the basis of
combination of four albumin and two globulin
bands. The frequency of each protein type
strain showed a clear geographical cline.

Pant M-5 and RCM 15 constitute
separate cluster owing to their typical
characteristic polypeptide banding pattern. A
few genotypes namely, OGG 57, OUM 7,
COGG-912 and ML 613 were also clubbed in
separate clusters at lower phenon level. These
genotypes are considered to be quite divergent and could serve as valuable breeding material for improvement in protein quality.

REFERENCES