Assessment Of Genetic Diversity of Cyamopsis Tetragonoloba and Identification of Stress-Resistant Genotype

Shumaila Khan, Alona Cuevas Linatoc
Panel Bidang Teknologi Warisan, Uthm, Malaysia
*E-mail: Shumylakhan74@yahoo.com

ABSTRACT

Introduction: *Cyamopsis tetragonoloba* (Guar) is known as Cluster bean is a leguminous vegetable. Guar belonging to the Family Fabeaceae. Guar is a vigorous and herbaceous legume. It also one of the minor crops of Pakistan.it grown on arid and semi rid soil usually in kharif season. Guar is important and useful crop because it is highly valued for its yield and vegetable protein. It has great nutritional value and also known as drought tolerant crop. Cluster bean is a traditional plant due its use in traditional medicine as well. It acts as a cooling agent, gastrointestinal aid, and is beneficial in indigestion and anorexia. It is also useful as anti-hyperglycemic, anti-ulcer, stimulate mucus production, inhance blood flow throughout the lining of the gastrointestinal tract and hypolipidemic. As a leguminous plant it defense the body from various cardiovascular diseases. Moreover, this plant is used in the treatment of cancer diseases specially in colon cancer and also beneficial for diabetic patient. Seed gum (galactomannan gum) is the major importance of guar is the by commercial value. The gum contented means concerning 31-43. About 16% on a whole seed basis. Pakistan is the major supplier of guar and account for the 15% of total guar produced in the world.

Objectives: The main objective is to assess and access the presence of genetic changeability among indigenous guar concurrences also access the genetic diversity in stress resistance genotype and to breed new diversities with necessary characters related to seed yield and gum quality. Morphological and biochemical markers are not much reliable for diversity examination as the impression of environmental factors. For consistent and effective analysis of genetic variation in the germplasm and to explain the intra- and interspecific relationships, the use of molecular markers is required. Likewise, molecular markers increase the efficacy of traditional plant breeding. Guar production should be promoted by prioritization and improve their varieties to increase its yield.it is necessary to keep this crop from weeding as weeding is very crucial crop for this reason guar crop kept weed-free up to 30-45 days of seeding. In addition to this thought field experimentation, it is quite possible to increase this crop production. Quality of seed must be addressed for increasing crop production especially for farmers. Guar seed variety and their suitability for different region established by research work.

Methodology: A total of 112 accessions representing better agronomic types will be used for the present study. Optimal agronomic practices will be followed through various stages of crop growth. Morphological characterization will be supported for assessing of genetic diversity among the healthy plant of *Cyamopsis tetragonoloba*. In quantifiable traits of leaf length, leaf width, fruit length, fruit width, Petiole length, 100 Seeds weight, and branches in number, number of pods per plant, number of clusters will be studied. PCR amplification will be performed to find genetic diversity and best yield genotype.

For biochemical characterization leaves are used from each sample of different varieties. About 5 leaves from each sample are going to be grinded for making very fine powder for SDS-PAGE. When leaves convert into powder form, we used 0.02g of this fine flour for extraction of crude protein.400ul of PEB must be added in each sample. After that each sample is going to be configurator at 12000rpm for 30 minutes under temperature of 40 degree. By using electrophoretic technique separation gel that is polyacrylamide gel about

12.5% and staking gel about 4.5% can be used. The crude protein in the supernatant and the cell- lysate precipitated at the bottom of each E-tube is present. The E-tube vortexes and then centrifuged at 14000 rpm for 30 minutes at 25 0C.

By using electrophorese technique 10ul of each sample is added to well from left to right. In electrode buffer solution the gels and run it on 12V.In the staining solution gel added to stained the protein for 180 minutes. For de-staining of non-protein part of the gel added solution having methanol 2%, acetic acid 5% and distal water 75% with a ratio of (4:1:15) for analysis of all genotype. using 1 for the presence of a protein band and for absent of protein band 0is used for analysis of data. For all genotypes match coefficients is used to find out the relationship among study genotype by using (UPGMA) and a dendrogram is constructed by using PECORD software.

Polymerase Chain Reaction (PCR) accomplished 20L reaction mixture that consist of 0.5-unit Taq DNA polymerase.200mm each of dNTP,0.2M primer and about 50 mg of genomic DNA. Amplification is also performed in PTC-100 programmable thermal cycle. PCR is performed after initial denaturation at 94 degree just for five minutes. The cycle run for 30-35 cycles comprising of denaturing step for 1min and extension at 72 ºC for 2 min. Finial extension also appended for 10 min. Amplication products also be detached on 1.5 % agrose gel in TBE BUFFER using gel documentation system

Conclusion: The outcome of the current study will display molecular marker as a corresponding tool should be used in conjunction with morphological characterization for a improved description of the level and arrangement of genetic diversity and crop development. This work will also give to the development of guar species workflow, and progress Gene bank to make them accessible for breeding and research. Such measurable will be safely conserved under conditions that ensure genetic integrity and viability in the long.

The current study determined that molecular marker as a complementary tool that use in mixture with morphological characterization for better description of the pattern of genetic diversity and for the development of guar crop. This work will also contribute to the enhancement of guar species workflow, and develop Gene bank to make them accessible for breeding and research. Such material also safely preserved under conditions that ensure genetic integrity and viability in the long.

Keywords: accessions, guar, PCR, traits