

**MAIZE (*ZEA MAYS* L.) GENE BANK FOR AGRICULTURE AND FOOD INDUSTRY****PÁL PEPÓ<sup>1</sup>, CSILLA BOJTÉ<sup>1</sup>, SZILÁRD TÓTH<sup>2</sup>**

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**ABSTRACT**

Corn breeding can be successful only on a broad base of genetic material. The expansion of breeding aims includes the research of germplasm materials. In our experiments, we examined 11 blue, purple and red exotic corn varieties on two locations in Hungary. We conducted a complex study to obtain more information. We studied several morphological and phenological features and determined the most important qualitative parameters (protein, fat, ash). Results show that there are great opportunities in exotic corn varieties. Their quality exceeds that of the yellow ones in many cases. There are significant differences in yield and nutritional parameters. The favourable nutritional composition is not coupled with great productivity. Based on their flowering time and their agronomical features, they can be cultivated under Hungarian conditions as well. They match the new breeding aims, so they can be used as functional food or energy plants. The cultivation of alternative crops has an important role in world agriculture. Their market share is continuously growing in the food industry sector.

**Keywords:** maize (*Zea mays* L.), gene bank, agricultural and food industrial application

**INTRODUCTION**

Maize is the most important feed crop the use of which is very versatile. In many countries it is widely applied as an important food stuff for the population. In recent years modern industrial and agricultural countries have a developed corn processing industry. Nowadays, most research for improvement of corn production has focused on grain yield (ear, LAI, plant density, fertilization, etc.) traits because of agronomic importance (EL HALLOF AND SÁRVÁRI 2006; MOLNÁR AND SÁRVÁRI, 2006). Maize breeders are mainly concerned with combining ability and grain yield, but resource allocation into male and female fitness is unknown (VIDAL-MARTINEZ ET AL., 2004). Furthermore, artificial selection for higher grain yield has indirectly led to poor pollen production of negative correlations between male and female component fitness (GARNIER ET AL., 1993; VIDAL-MARTINEZ ET AL., 2001). In maize selection, it is of great importance to know the performances and combining ability of crossing partners. It is also relevant in seed production and basic research to study differences between single and reciprocal crosses. A few Hungarian literature references draw attention to this field (BERZY ET AL., 2005). KOVÁCS (1963) investigated direct and reciprocal single crosses derived from Martonvásár lines and found no difference between the productivity of two hybrids. Transportation of parents can be accounted by the fact that the cytoplasm of certain plants does not take part equally in the development of agronomical features. With the transposition of parents, more economical seed production is possible (NAGY, 1982). NAGY (1985) examined leaf number, ear height ear length, cob weight, 500 kernel weight, shelling %, and oil content in 14 early SC hybrids and their reciprocal varieties. In one quarter of compared data pairs, significance difference was found at  $P > 5\%$ . The main areas of further development in maize production

are yield safety, chemical composition and better utilization. Mutant population of richer varieties provides considerable help in solving these problems (PÁSZTOR ET AL., 1980). We have continued our research of the previous years into the selection of maize mutants as well as our studies of combining abilities and chemical characteristics of hybrids. The aim of these experiments is to determine the important values of mutant lines both for chemical characters and for yield components. The development of animal husbandry and maize production necessitated the improvement of the chemical composition of maize (MARÁZ ET AL., 1993; PEPO AND PEPO, 1993; PEPO, 1995). We produced genetically highly variable mutant population with fast neutron treatment of seeds from American maize hybrid material (F<sub>1</sub>). From the starting basic material, with the use of radio mutation we created perspective hybrid-combinations by different genetic and breeding methods (PEPO ET AL., 1989).

### MATERIAL AND METHOD

The experimental setup was randomized block design with four replication, furthermore, the distance between rows was 70 cm and among plants 20 cm. We investigated 32 morphological characters based upon CPVO TP2/2 standards and conducted zein analysis with isoelectric focusing based on ultrathin polyacrylamid gel electrophoresis. We isolated chromosomal DNA using QIAGEN DNeasy Plant Mini Kit. Extracted genome DNA was digested with 10 U EcoRI restriction enzyme, incubated with 1 U TruII at 67 C° and inserted AFLP adapters into it. The DNA was multiplied with PCR and the DNA fragments were separated with capillary electrophoresis.

### RESULTS AND DISCUSSION

There is large genetic variation in the germplasm utilised, the exploitation of which is only possible using suitable methods of selection and evaluation. In 1985, we applied a physical mutant agent (fast neutron) for irradiation of F<sub>1</sub> hybrid seeds by 7.5-12.5 Gy dose. After selection, mutant lines were used for self-pollination over many years. As a result of this selection process, P26, P61 and P62 lines have been governmentally released by the OMMI (2001). These lines serve as a basic material in our future breeding programs. Identification and the origin of these genotypes are given in *Table 1*.

**Table 1. Pedigree of registered inbred lines**

Lines	Hybrids	Type of irradiation	Dose [Gy]
P26	F <sub>1</sub> (Pi 3747 SC) M <sub>2</sub>	fn	7.5
P61	F <sub>1</sub> (Pi 3901 SC) M <sub>2</sub>	fn	12.5
P62	F <sub>1</sub> (Pi 3901 SC) M <sub>3</sub>	fn	7.5

*f<sub>i</sub>* : First generation after crosses

*m<sub>n</sub>* : n<sup>th</sup> mutational generation

*fn* : Fast neutrons (produced in a cyclotron)

As a result of mutagenic treatment, morphologically very different mutant populations were obtained. Radiation generated conspicuous changes in the plant characteristics described by UPOV in comparison with the basic material. The most pronounced aberrations were observed for the expression of anthocyanin coloration in various plant organs. The variability manifested in the changes of pollination interval. Flowering time is considered to be quantitatively inherited, and different studies have identified loci that affect this trait in maize (BEAVIS ET AL., 1991; CIT. OLIVEIRA ET AL., 2004). Mutation treatments (fast neutron) induced earliness in flowering time of different inbred lines. Using these lines as crossing parents, they could cause earliness in hybrids. With earlier flowering time, we can avoid frequent drought periods, which reduce fertilization in maize. Earlier maturity could reduce grain moisture in harvest time, drying energy and fungi diseases (e.g. *Fusarium* ear rot). These characteristics are suitable for sustainable agriculture. We concluded that the cyclotron can be successfully applied in widening genetic variability. We produced a number of inbred lines with wide genetic variability using mutation breeding. We can use this information to develop maize hybrids, which can be useful in our breeding program.

Genotypes of high antioxidant content, BM (Black Mexican) and ANB1, in addition to our own inbred lines (S1 and P49/1), and those back-cross hybrids were examined. Antioxidant and flavonoid content were determined by Folin-Ciocalteu method. Significant differences were observed among the antioxidant content of maize genotypes. Antioxidant and flavonoid contents of our maize hybrids (60.29% and 53.11%) were lower than that of the blue maize hybrids. The highest antioxidant content was detected in the BM line (168.0 mg), while the lowest antioxidant content was measured in the S1 parental line. Increase of antioxidant and flavonoid contents were observed in the first generation. In the second generation antioxidant content decreased in a small scale, but it was still high (*Table 2*).

**Table 2. Flavonoid and antioxidant content**

	Antioxidant			Flavonoid		
	content	relative %	absolute value	content	relative %	absolute value
S1	91.6	100.00	-	18.81	100.00	-
ANB1	134.0	146.28	42.4	21.17	112.54	2.36
(ANB1xS1)F <sub>1</sub>	121.0	131.00	29.4	25.42	135.14	6.61
SzD <sub>5%</sub>	6.5			2.73		
P49/1	96.8	100.00	-	18.81	100.00	-
BM	168.0	173.55	71.2	36.44	193.72	17.63
(BMxP49/1)F <sub>1</sub>	160.0	165.28	63.2	32.44	172.46	13.63
(BMxP49/1)xP49/1F <sub>2</sub>	141.0	145.66	44.2	27.35	145.01	8.54
SZD <sub>5%</sub>	4.34			3.22		

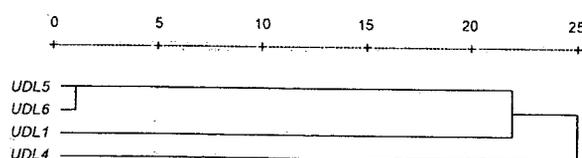
Corn breeding can be successful only on a genetic material of broad base. The expansion of breeding aims includes the research of germplasm materials. In our experiments, we examined 11 blue, purple and red exotic corn varieties on two locations in Hungary. We conducted a complex study to obtain more information. We studied several morphological

and phenological features and determined the most important qualitative parameters (protein, fat, ash). We applied the Tassel Area Index (TAI) under Hungarian conditions, and compared it with the pollen producing ability of different genotypes. Results show that there are great opportunities in exotic corn varieties. Their quality exceeds that of the yellow ones in many cases (Table 3). There are significant differences in yield and nutritional parameters. The favourable nutritional composition is not coupled with great productivity. Based on their flowering time and their agronomical features, they can be cultivated under Hungarian conditions as well. They match the new breeding aims, so they can be used as functional food or energy plants. With their high pollen producing ability and TAI, they can be utilized as markers in future pollen research.

**Table 3. Protein, fat and ash contents (%) of investigated blue and purple kernel corn genotype (1), protein content (2), fat content (3), ash content (4), check yellow corn (5), significant at P=5% probability levels for comparison with control**

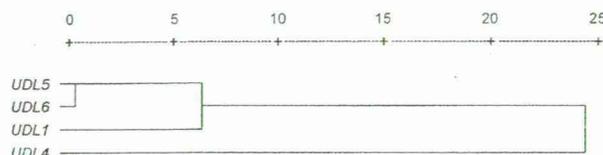
Genotype (1)	Kernel colour (5)	Protein (2)	Fat (3)	Ash (4)
Hopi Blue	blue	10.62*	4.73*	1.59*
Blaumais	blue-brown	11.69*	3.22	1.53*
Taos P. B.	blue	8.65	6.11*	1.42
Alamo N. B.	blue	8.86	6.51*	1.50*
Purple Red Flour	blue	9.79*	6.33*	1.35
Sandia P. Black	blue	8.13	5.11*	1.33
Santo D. Blue	blue-purple	9.81*	4.68*	1.36
Hopi Turquoise	red	11.26*	-	1.61*
DK 471 (Kontroll)(5)	red	7.36	3.12	1.23

The primer combinations were used as AFLP markers producing 207 bands, 70 of them being polymorphic. We used IEF (Isoelectric Focusing) of zein to determine the genetic similarities between lines. We found 15 bands with this technique. The dendrogram based on genetic similarities (GS) and morphological description separated the four studied inbred lines into well-defined groups. Morphological description, AFLP and zein analysis revealed the same reliable results (Figures 1-2).



**Figure 1. Dendrogram obtained by cluster analysis based on the zein patterns of four maize inbred lines. (1) Distance, (2) Line**

Our results suggest that AFLP and analyses seem to be efficient techniques to determine the genetic similarities/differences between different genotypes. Considering the genetic distance values, the UDL1 line and its hybrid with UDL6 in both directions showed significant heterosis effect which was confirmed by heterosis calculation based on grain yield.



**Figure 2. Dendrogram obtained from cluster analysis based on the AFLP amplification patterns of four maize inbred lines. (1) Distance, (2) Line**

Knowledge of genetic diversity among available parental inbred lines is fundamental for successful hybrid maize breeding. Clustering of inbred maize lines according to their genetic background is important for breeding, as a certain genetic distance between the parental lines is essential to predict the heterosis effect. The aims of our study were to estimate (1) genetic similarity (GS) of four maize inbred lines, (2) to classify the lines according to their GD and GS values, (3) to determine their hybrid performance based on CPVO TP2/2 AFLP and zein patterns to estimate genetic polymorphism of four maize inbred lines. We estimated the applicability of genetic similarity values in SC and reciprocal hybrids to predict their performance in a complete diallelic crossing.

## CONCLUSIONS

In the past, conventional breeding methods have been quite successful in improving crop yields throughout the world. Conventional breeding methods, however, are laborious and time and space consuming. Successful employment of additional methods and biotechnology could accomplish development of plants, this making breeding process more efficient in the use of critical resources. Hungarian maize production utilises many foreign hybrids that are derived from only a few inbred lines. Because of this genetic vulnerability, production can be in a serious danger and quick gene erosion is a possibility. During the past ten years, maize production level has remained unchanged and opportunity of further development would develop new inbreds with desirable agronomic attributes.

## REFERENCES

- BEAVIS, W.D., GRANT, D., ALBERTSEN, M., FINCHER, R. (1991): Quantitative trait loci for plant height in four maize populations and their associations with qualitative genetic loci. *Theoretical and Applied Genetics* 83(2): 141-145.
- BERZY, T., HEGYI, ZS., PINTÉR, T. (2005): Correlations between the seed quality and yield parameters of maize hybrids developed on different parental lines. *Növénytermelés* 54(3): 159-167.
- EL HALLOF, N., SÁRVÁRI, M. (2006): Relationship between fertilization, leaf area index, photosynthetic activity and yield of hybrids. *Cereal Research Communications* 33(1): 181-185.

- GARNIER, P., MAURICE, S., OLIVERI, I. (1993): Costly pollen in maize. *Evolution* 47(3): 946-949.
- KOVÁCS, I. (1963): The productivity of hybrid maize affected by the reciprocal maternal single cross. *Növénytermelés* 12(4): 425-438.
- MARÁZ, A., PEPÓ, P., PEPÓ, P., TÓTH, SZ. (1993): A kukoricavonalak és populációk variabilitásának növelése mutáció segítségével. I. NTN. Pp. 63.
- MOLNÁR, ZS., SÁRVÁRI, M. (2005): Effect of plant density on the maize yield. *Cereal Research Communications* 33(1): 275-279.
- NAGY, L. (1982): Investigation of productivity and some standard features in single-cross reciprocal maize hybrids produced with lines of diverse vegetation-period. *Növénytermelés* 31(1): 11-20.
- NAGY, L. (1985): Comparative study of reciprocal single crosses in early maize. *Növénytermelés* 34(4): 307-312.
- PÁSZTOR, K., RABBIE, H., ELEK, M. (1980): An evaluation of the chemical composition of maize mutants and of their hybrids. *Növénytermelés* 29(1): 9-20.
- PEPÓ, P. (1995): Genetic improvement for sustainable maize (*Zea mays* L.) production in Hungary. *Int. Conference on Sustainable Agriculture, Hisar, India. Abstracts: 114-122.*
- PEPÓ, P., PÁSZTOR, K., PALIJ, A.F. (1989): Kukorica mutánsvonalak genetikai analizise. *Növénytermelés* 38(3): 193-199.
- PEPÓ, P., PEPÓ, P. (1993): Biological background on sustainable maize (*Zea mays* L.) production. *J. of Landscape and Urban planning* 27: 179-184.
- VIDAL-MARTINEZ, A.V., CLEGG, D.M., JOHNSON, E.B. (2001): Genetic Studies on maize pollen and grain yield and their components. *Mayidica* 46: 35-40.
- VIDAL-MARTINEZ, A.V., CLEGG, D.M., JOHNSON, E.B., DUVICK, D.N., CASSMAN, K.G. (1999): Post-green revolution trends in yield potential of temperate maize in the North-Central United States. *Crop Science* 39: 1622-1630.