

POTENTIAL USE OF POLYMORPHISM OF ISOENZYMES IN SELECTIVE-GENETICAL PROGRAMS FOR SUNFLOWER (*Helianthus annuus* L.)

V.V.Kirichenko, N.R.Sharopova, V.N.Popov, E.N. Maklyak

*Yurjev Plant Production Institute, Moskovskiy prospect 142,
Kharkov 310060, Ukraine*

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SUMMARY

Within the diallel crossing of sunflower inbred lines we have studied a heterosis level for major agronomic traits in F_1 . A genetic diversity of lines for 11 isoenzymic systems has been defined. Genetic relationships between lines were investigated by the cluster analysis, which was performed on the base of a matrix of genetic distances. Five groups of lines were singled out by the cluster analysis. Significant correlation between genetic distances and seed yield of F_1 hybrids was shown. With the increase of genetic distances between lines, the heterosis level was also growing in F_1 hybrids.

Key words: Cluster analysis, genetic distance, heterosis, isoenzymes, sunflower

INTRODUCTION

Utilization of heterosis effect is one of the most prospective trends in selection. At present, when developing hybrids, some various crossing systems are used with subsequent testing of hybrid combinations in field conditions that will entail great expenditure of time and labor. In this connection there is a task to seek for methods by means of which it is possible to predict the level of heterosis effect in F_1 hybrids.

For prediction of heterosis effect a number of authors suggested the approaches based on the correlation of biochemical properties of parental lines with seed yield of F_1 hybrids and heterosis level (Shvetsova, 1978; Zazharskiy, 1978). The potential use of genetic diversity of original material for polymorphism of storage proteins, isoenzymes, DNA in the estimation of heterosis effect is of a great practical value. It has been established that seed yield of maize hybrids correlates with genetic diversity in parental lines as to catalase isoenzymes, esterase, acid phosphatase, alcoholdehydrogenase and peroxidase (Heidrich-Sobrino and Cordeiro, 1975). For estimation of maize hybrid yield potential, J.W. Dudley and his col-

leagues used the unified information on isoenzymes and polymorphism of DNA restriction fragment lengths and showed that the genetic distances between lines possessing remote relationship correlate with the yield of their hybrids significantly (Dudley *et al.*, 1991). Similar data for maize were obtained while using the products of DNA amplification (Verbitskaya *et al.*, 1998).

In this paper we present the results of our study relative to genetic relationships in 13 inbred lines of sunflower for 11 isoenzymic systems (esterase, acid phosphatase, alcohol dehydrogenase, leucine aminopeptidase, NAD⁺ malate dehydrogenase, catodal esterase, NADP⁺ malate enzyme, isocitrate dehydrogenase, catodal acid phosphatase, aspartate aminotransferase, 6-phosphogluconate dehydrogenase) and the ways of application of genetic diversity in original materials as to isoenzymes for prediction of heterosis effect.

MATERIAL AND METHODS

The collection of 13 sunflower lines developed in Plant Production Institute named after V.Ya. Yurjev was used as plant material.

Hybrid seeds were obtained from crosses in a complete diallel design. The hybrids were tested by means of randomized replications during 3 years (1994-1995).

The estimation of 156 hybrids and their parental forms for seed yield, plant height and seed oil content was conducted during vegetation.

The heterosis effect in hybrid combinations was determined in comparison with the best parental form (Riger and Mikhaelis, 1967). As regards to weather conditions, the vegetation period was dry in 1994. The conditions in 1995 were moderate as for temperature regimen and rainfall, however, the emergence-budding period was marked by high temperatures and insufficient water availability. The weather conditions of 1996 were defined by high average monthly air temperatures; the amount of rainfall was within the limits of long-term average during the vegetation period.

For the analysis we took 30 seeds from each line. The enzymes were extracted with 0.02 mM tris-HCl buffer (pH 7.5), containing 0.01 mM PVP, 0.006 mM EDTA, 0.01 mM DTT and 20% saccharose. The electrophoresis of enzymes was performed in vertical plates. The separating gel contained 7% acrylamide and 0.18% methylene-bis-acrylamide. For separating of Est, Mdh, Lap, 6-Pgd enzymes we used tris-EDTA-borate buffer system - 0.09M H₃BO₃; 0.0031 M EDTA (pH 8.3). For separation of Adh, Idh, Aat, Me, Acp enzymes we used tris-glycine buffer system (Maurer, 1971). For separation of isoenzymes cathodal Acp and cathodal Est we used aluminium-lactate system. The acrylamide concentration in the separating gel was 7% and in the concentrated one 5%. The bis-acrylamide concentration in the stock solution for the separating gel was 0.34%, and for the concentrated one 0.5%.

Genetic distances were used for the assessment of genetic diversity in sunflower lines. The genetic distances (D) were calculated according to Nei and Li (1979). The genetic distance matrix according to Nei and Li was used for the cluster analysis (UPGMA method, Sneath and Sokal, 1973).

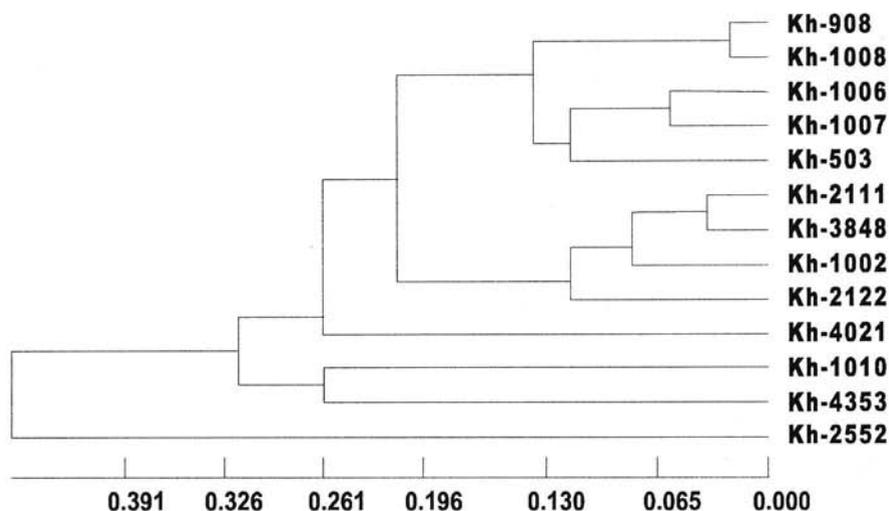


Figure 1: A dendrogram of genetic relationships in sunflower lines (*Helianthus annuus L.*)

RESULTS AND DISCUSSION

A dendrogram reflecting the genetic relationships between 13 sunflower inbred lines is given in Figure 1. Genetic distances between the lines were obtained by genetic diversity data for 11 isoenzymic systems. As the result of the cluster analysis the lines were divided into 5 groups. The first, most heterogeneous, cluster included Kh-908, Kh-1008, Kh-1006, Kh-1007 and Kh-2122 lines, the ancestors of which were local cultivar-populations and interspecific hybrids. The second cluster included Kh-2111, Kh-3848, Kh-1002 and Kh-2122 lines, the ancestors of which were interspecific hybrids. The third cluster was represented by 2 lines Kh-1010 and Kh-4353. Each of the two remaining clusters was represented by Kh-2552 and Kh-4021 lines, which were not included in any of the previous clusters. The cultivar-population Chernyanka 66 was the source of those lines. The genetic distance for separate pairs of lines varied from 0.078 to 0.615. Among the analyzed lines the nearest in the allelic composition of isoenzymic loci were Kh-908 and Kh-1008 lines. Those lines had the least genetic distance ($D=0.078$). The greatest genetic distance from the rest of the lines was observed in Kh-2552 line.

A correlation analysis was carried out in order to study the relationships between the genetic diversity among parental lines for isoenzymes and F_1 indices, i.e., seed yield, plant height and oil content in seeds. Table 1 shows the coefficients of pair correlation for the data of the 3-year study.

The analysis demonstrated that the correlation relationships between genetic distances/seed yield and genetic distances/plant height were significant. So the coefficients of correlation between genetic distances and the yield of hybrids in 1994-1996 were 0.53, 0.45 and 0.40, respectively. The yield of hybrids, the parental lines of which had the range of genetic distances 0.078-0.180, was 30.4 c/ha on the average, heterosis 64%. With the increase in the genetic distance to 0.262 the seed yield of F_1 hybrids went up to 32.3 c/ha on average, heterosis to 67%. The hybrids obtained from Kh-2552 line varied in yield from 30.1 to 36.1 c/ha, and heterosis was 82.6%.

The coefficients of correlation between genetic distances in parental lines and plant height during 1994-1996 were 0.37, 0.32 and 0.29, respectively. The taller hybrids originated from Kh-2552 line. The average height of those hybrids was 147.3 cm, heterosis was 34.7%.

According to the data of E.N.Maklyak (1998) there existed a prevalence of recessive alleles over dominant ones for plant height in Kh-2552 line. However, highly heterotic hybrids for plant height were obtained due to the contribution of that line.

During the development of Kh-2552 line from the cultivar-population Chernynka 66, the phenotypic selection for important agronomical traits led to choosing definite combinations of loci's alleles controlling some isoenzymes. In the process of the cluster analysis one could observe sufficiently clear separation of Kh-2552 into the individual cluster.

Table 1: Coefficients of correlation between genetic distances and agronomic traits, 1994-1996.

TRAIT	1994	1995	1996
Seed yield	0.53*	0.45*	0.40*
Oil content in seeds	0.70*	0.14	0.18*
Plant height	0.37*	0.32*	0.29*

* significant at 5% level

The coefficients of correlation between genetic distances and oil content in seeds varied considerably depending on environmental conditions (within the range of $r=0.14$ in 1995 to $r=0.70$ in 1994). The heterosis level for the 3-year investigation did not exceed 8%. The obtained data permit to assume that the selection of parental lines for this trait on the genetic distance basis between lines is not advisable.

CONCLUSION

The results obtained on the basis of the cluster and correlation analyses provided additional information, which complemented those received from the study of diallel crossing design, but also they permit to assume that the choice of pairs in heterosis breeding of sunflower is recommended to carry out both on the basis of the investigation of the pedigree data of lines and by means of the values of genetic distances among the lines. The most highly heterotic hybrid combinations were obtained in crosses of genetically heterogeneous forms.

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USO POTENCIAL DE POLIMORFISMO DE ENZIMAS EN LOS PROGRAMAS SELECTIVOS GENETICOS PARA EL GIRASOL (*Helianthus annuus* L.)

RESUMEN

Un grupo de cruces dialelicos de las lineas consanguineas de girasol fue investigado al nivel de heterosis para las propiedades agronomas mas importantes en la generacion F₁. Fue constatada la divergencia genetica entre las lineas investigadas para 11 sistemas isoenzimas. Las relaciones geneticas entre las lineas fueron investigadas por el analisis de grupos, realizado a base de la disposicion de distancias geneticas. El analisis distinguió cinco grupos de lineas. Fue constatada la correlacion importante entre las distancias geneticas y los rendimientos de semillas y la altura de planta hibrida F₁. Con la extension de distancias geneticas entre las lineas crecia el nivel de heterosis en los hibridos F₁.

UTILISATION POTENTIELLE DU POLYMORPHISME DES ISOENZYMES DANS LES PROGRAMMES DE SÉLECTION GÉNÉTIQUE DES TOURNESOLS (*Helianthus annuus* L.)

RÉSUMÉ

Un groupe de croisements dialleles de lignes inbred de tournesol a été étudié au niveau des hétéroses pour établir les traits agronomiques majeurs dans la génération F_1 . Une divergence génétique des lignes examinées pour 11 systèmes isoenzymatiques a été définie. Les liens génétiques entre les lignes ont été examinés au moyen de l'analyse des groupes, qui a été effectuée sur la base d'une matrice de distances génétiques. Cinq groupes de lignes ont été différenciés par l'analyse. Une importante corrélation entre les distances génétiques, le rendement de graines des hybrides F_1 et la hauteur des plantes a été démontrée. Le niveau d'hétéroses était en croissance dans les hybrides F_1 en même temps que les distances génétiques augmentaient.