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Utilization of Plant Genetic Resources in Peanut Breeding Program of Bulgaria

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Abstract

Based on study of some morphological indices, responsible for productivity in 97 breeding lines *Arachis hypogea* L. with Bulgarian origin, a linear regression model of the plant, that is characterized with high seed yield, was established. The breeding program requires the identification of direct and indirect effects on productivity. By application of the Path Coefficient Analysis, it has been found that the higher yield plants must have low and extensive bush (i> 2,0). The local accessions of peanuts are characterized by a lower index of the bush (i <2,0). For this purpose, 399 genotypes with different geographical origin were introduced from USDA (USA) in 2008. Based on the evaluation of foreign accessions by height and width of the plant, seed yield from one plant and resistance to *Fusarium*, four genotypes were selected. They were included in crosses with Bulgarian varieties and lines of Valencia type. The heterosis expressions and some genetic effects of progenies were studied.The results show that heterosis effect in F_1 at some of the progenies in connection with yield of the fruit and seeds are inherited without epistatic effect. The inbred depression in F_2 is an indication of homozygosity in respect of allelic pairs defining yield.

Key words: *Arachis hypogea* L., introduction, breeding, model of plant, hybrids

Introduction

Conservation of genetic diversity is a major task of the National Programme for Plant Genetic Resources in Bulgaria (Stoyanova, 1994). A large collection with original *Arachis hypogaea* L. germplasm from local and foreign origin is stored in the gene bank of IPGR-Sadovo (Maggioni et al., 2004; Krasteva et. al., 2007). The utilization of diverse genetic material in breeding programs determines the derivation of varieties with high yield and resistance to diseases (Pevicharova and Todorov, 2001; Krishna et al., 2004; Strano et. al., 2011).

The peanuts yield is in a complex polygenic character, as its components are in strict relations with each other. Dependencies between traits are measured by the coefficient of correlation, which does not allow to determine a relative contribution of each of them on the yield (Martynov, 1978).

Path coefficient analysis allows the separation of the direct and indirect effect on the yield and gives a realistic picture of the relations between the different indicators, which facilitates the selection process (Sumathi et al., 2007).

Introduction of new germ plasm in the genome ensures receipt of new promising forms and manifestation of heterosis for certain traits in F1 population. There is a positive heterosis in succession of the fruit weight and seeds mass in hybrid materials (Edilberto and Ricrdo, 1986). Dwivedi et. al (1989) show heterosis in the plant height, weight of the fruit and the yield in its hybrid materials. Isleib and Wynne (1983) obtain similar results, as the established heterosis effect on fruit yield reaches 19,2%. According to many authors this is due to the genetic distance of the parental pairs (Gowen and Frey, 1987).

The heterosis is a powerful method of increasing the yield of crop plants. The study is an integral part of almost every breeding program (Petr and Fray, 1966; Romero and Frey, 1973). As a result of the application of genetically regulated heterosis, it is used in a growing number of crops. Its use, however, is practically advantageous only in the first-hybrid generations. Complex seed production and the small coefficient of seeds propagating in some cultures, require to seek opportunities for fixing heterosis in F2 and subsequent generations. The solution of this issue would be of great practical importance in peanuts breeding (Georgiev, 1984; Petkova and others. 1999).

The aim of this study is evaluation of *Arachis hypogaea* L.

collection from different geographical origin to introduce selected accessions in hybridisation program for obtaining high-yielding peanuts resistant to economically important diseases.

Material and Methods

The status of Bulgarian peanutcollection covers 1137 accessions, including local (435) and foreign origin (702), consisting of varieties (54%) and breeding lines (46%). All stored materials are registered with catalog numbers in the National Register *PHYTO 2000*, in accordance with international standards of FAO, ECP/GR and Bioversity International. Bulgarian PGR collection (BGR National Inventory) is a part of the European catalogue on Plant Genetic Resources (EURISCO).

399 introduced genotypes from USDA (USA) with a different geographical origin and 97 Bulgarian breeding lines *Arachis hypogea* L. were studied in the experimental field of Institute of Plant Genetic Resources, Sadovo in the agroecological conditions of South Bulgaria (Fig. 1).

Fig.1.Experimental field of IPGR-Sadovo, Bulgaria
Eksperimentalno polje IPGR-Sadovo, Bugarska

The genotypes were evaluated by morphological characteristics: height of the bush, the bush width, mass of fruit, seed weight, yield and weight of 1000 seeds, and resistance to *Fusarium*.

In the period 2012-2014, hybrid materials were studied based on crosses made. The object of the present study are the parental components (P1, P2) and 110 hybrid generations from F1 and F2 generation,that have

shown heterosis in F1 by the harvesting of fruit and kernel. The retention and stabilization of the trait in the F2 is an indicator defined by the extent of the presence of an overly dominant and inbreeding depression.Analysis of the degree of dominance in F1 of the trait suggest that the major importance for the expression of heterosis is overly dominant positive, the indicator d/a is larger than 1. When the degree of overly dominant (h1) in the F1 is greater than the degree of overly dominant (h2) in the F2, there is an inbred depression. Conversely, where h1<h2, shows that the succession became with participation of between locus interactions (epistatic).

The heterosis and inbred depression are defined by Omarov (1975), and the degree of dominance in F1 and F2 by Romero and Fray (1973).

The obtained results were processed with statistical package *SPSS 13.0* by hierarchical cluster, correlation and Path coefficient analysis.

Results and Discussion

Based on the study of 97 breeding lines with germplasm of Bulgarian origin *Arachis hypogea* L., by morphological traits, responsible for the productivity, a linear model of plant, characterized with high seed yield, was established.

$$Y = -40,09 + 0,13x_1 + 0,1x_2 + 0,64x_3 + 0,47x_4 - 0,002x_5$$

x_1 - plant height
x_2 - width of the plant
x_3 - weight of the fruit
x_4 - weight of kernels
x_5 - mass of 1000 seeds

The breeding program requires the definition of direct and indirect effects on productivity. It has been found that the plants with higher yields are required to have low and spread bush (i> 2,0), and the local germplasm in peanuts is characterized by a lower index of the bush (i <2,0).

Based on assessment of foreign germplasm by characteristic width and height of the bush, seed yield of a plant (Fig. 2) and resistance to *Fusarium* are selected four accessions: A8000205 (South Korea), A8000243 (Brazil), A8000273 (Argentina) and A8000261 (Argentina), which are included in crosses with Bulgarian varieties and lines of type Valencia. Kalina, Stanko, Tsvetelina and Sadovo 3871 are Bulgarian breeding materials with high productivity.
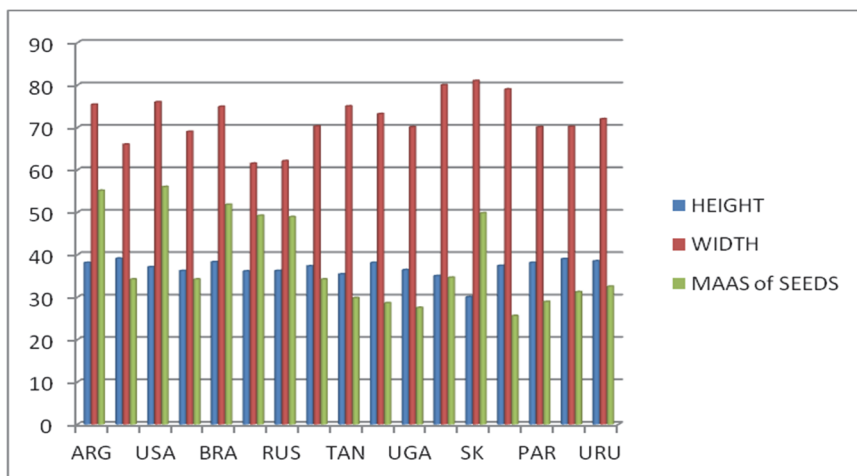
Fig. 2**.** Evaluation of introduced *Arachis hypogea* L. accessions
by morphological traits
Procjena potomstva uvedenih sorti Arachis hypogeal L.
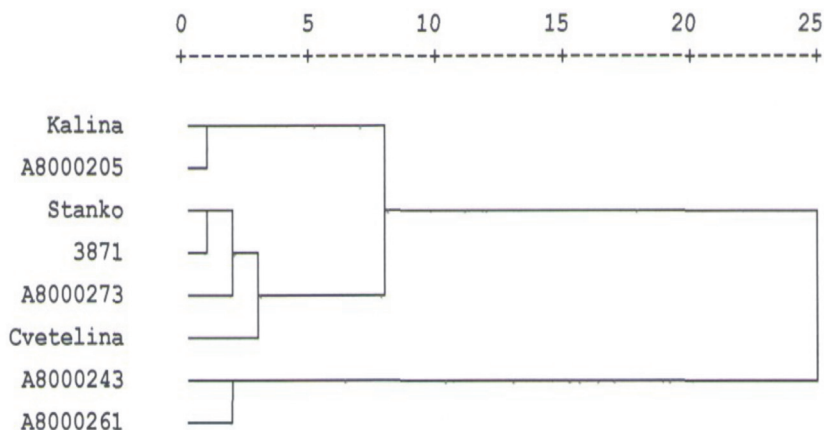*po morfoloskim svojstvima*

Fig. 3. Grouping of *Arachis hypogea* L. accessions based on morphological traits
Grupisanje potomstva Arachis hypogeal L. *na osnovu morfoloških svojstava*

The cluster analysis (Fig. 3) shows a genetic distance of the accessions by the studied characteristics and it forms four cluster groups. Kalina and Cat. № A8000205 fall in the first cluster with the smallest distance from the other germplasm accessions. Stanko, A8000273 and

Sadovo 3871 are includedin the second cluster group. Two accessions -
A8000243 and A8000261 fall in the third cluster at a sufficient distance
away from the others. Tsvetelina forms separate cluster group at the
greatest distance from the others.

The correlation analysis shows that the proven influence on the mass
of seeds of a plant has only the weight of the fruit. The correlation
coefficient (0,880) is mathematically proven at a significance level of 0,001
(Table 1).

Tab. 1. Correlation coefficients, reflecting the dependencies between the studied
morphological traits
*Koeficijenti korelacije koji odražavaju ovisnost između proučavanih
morfoloških svojstava*

Characteristics *Karakteristike*	Plant height *Visina biljke*	Plant width *Širina biljke*	Fruit weight *Težina ploda*	Mass of seeds *Masa sjemena*	Yield *Prinos*	Absolute mass of seeds *Apsolutna masa sjemena*
Plant height *Visina biljaka*	1	0,347	-0,671	-0,389	0,565	0,147
Plant width *Širinabiljke*		1	-0,046	0,111	0,263	0,296
Fruit weight *Težina ploda*			1	0,880**	-0,221	0,284
Mass of seeds *Masa sjemena*				1	0,264	0,287
Yield *Prinos*					1	0,028
Absolute mass of seeds *Apsolutna masa sjemena*						1

**Significance level p=0,001 / **Nivo značajnosti p=0,001*

Path-coefficient analysis complements the correlation and gives an
idea of the characteristics that indirectly influence the formation of yield
(Table 2). The weight of kernel of a plant is directly dependent on the mass
of fruit and seed yield.The Path coefficients are 0,987 and 0,479. Plant
height, width of the brush and absolute seed weightaffect indirectly on this
indicator.Reducing the height of the plant leads to an increase of the seed
yield from a plant by increasing the yield, the Path coefficient is 0,270.

Increasing the width of the bush leads to an increase of seed yield, the Path coefficient is 0,126, expressed by r_{25}. Indirectly, the fruit weight and the mass of seeds are influenced by the absolute seed mass, the Path coefficient is 0,281, expressed by r_{35}.

Tab. 2.Direct and indirect effects of studied characteristics on seed yield of plant
Direktni i indirektni efekti proučavanih karakteristika prinosa sjemena biljke

Characteristics *Karakteristike*	Path coefficients *Koeficijenti puta*					Correlation coefficients (r) *Koeficijenti korelacije (r)*
	Plant height *Visina biljke*	Plant width *Širina biljke*	Fruit weight *Težina ploda*	Yield *Prinos*	Absolute mass of seeds *Apsolutna masa sjemena*	
Plant height *Visina biljke*	**-0,008**	0,013	-0,663	0,270	-0,002	-0,389
Plant width *Širina biljke*	-0,002	**0,039**	-0,046	0,126	-0,005	0,111
Fruit weight *Težina ploda*	0,005	-0,001	**0,987**	-0,106	-0,005	0,880
Yield *Prinos*	-0,004	0,010	-0,219	**0,479**	-0,001	0,264
Absolute mass of seeds *Apsolutna masa sjemena*	-0,001	0,011	0,281	0,013	**-0,017**	0,287

*Dependent characteristics: Mass of seeds per plant / *Zavisna karakteristika: masa sjemena po biljci*

Based on the study on the genetic distance of parental pairs and model of the plant, characterized with a high yield of fruit and kernels of a plant, parental pairs are selected (Fig. 4, 5). The materials included in the hybridisation program in four straight crosses.

♀ Kalina x ♂ A8000205
♀ Stanko x ♂ A8000243
♀ Tsvetelina x ♂ A8000273
♀ Sadovo 3871 x ♂ A8000261

Fig. 4. Scheme of hybridization in peanuts
Shema hibridizacije kikirikija

Fig. 5. Hybrid materials *Arachis hypogea* L. from Bulgarian peanut collection
Hibridni materijal Arachis hypogeal L. *iz bugarskekolekcije kikirikija*

Six progenies are with manifestation of heterosis in F1, d/a>1 and confirmed inbreeding depression in F2, based on the weight of the fruit of a plant in crosses with parents ♀ Sadovo 3871 x ♂ A8000261. Inbred depression in these progenies is with values from 38,8 to 61,7 percent. The dominant allele couples in them, forming this indicator,are in the homozygous state. Seventeen progenies which show heterosis in F1 with coefficient of d/a>1 are confirmed without inbreeding depression. These progenies are in heterozygous state of the allele pairs and they will continue to disintegrate in the next generations. Other progenies that expressed heterosis effect in F1, do not show overly dominant in F2.

The index weight of kernel demonstrated that eleven of progenies which have shown heterosis in F1, have coefficient d/a>1 and inbred depression in F2 in them is confirmed with values from 14,05 %to 60,1%. These progenies are homozygous with respect to dominant allele pairs. In nine of them, the ratio is d/a>1, but there is not inbred depression. These progenies are heterozygous and they will continue to disintegrate in the next generations.

One progeny is with the manifestation of heterosis in F1, d/a>1, and confirmed inbreeding depression in F2 based on the weight of the fruit of a plant in crosses with parents ♀Kalina x ♂A8000205. Inbreeding value of depression is 12,4%. This progeny has dominant allele pairs in homozygous state by this indicator.

One progeny is characterized by manifestation of heterosis in F1, d/a>1, and no confirmed inbred depression in F2 with respect to the weight of the fruit from a plant.

Thirty-four progenies are with d/a<1, and they have inbreeding depression. In these progenies the recessive allele pairs, defining this attribute, is in the homozygous state.

The analysis on the seeds mass of a plant shows that one progeny is with the manifestation of heterosis in F1, the ratio d/a>1, and inbreeding depression of 42,9% in F2. The dominant allele pairs in this progeny are in homozygous state in terms of this indicator.

Another progeny is with a manifestation of heterosis in F1, the ratio d/a>1, and absence of inbreeding depression, which speaks about the heterozygosity with respect to this feature.

Two progenies are with the manifestation of heterosis in F1, d/a>1 and confirmed inbreeding depression in F2 on the weight of the fruit in crosses with parents ♀ Stanko x ♂A8000243. The inbred depression is with values of 45,51 % and 45,02%. These progenies show homozygosity at dominant state of the alleles for the harvesting of fruit from a plant. All other progenies of this crosses are in homozygous recessive condition of alleles on this indicator.

Two progenies, which show heterosis in F1 in terms of seeds mass of plant, have coefficient d/a>1 and confirmed inbreeding depression with values of 38,41% and 33,29% in F2. This suggests that the dominant alleles in them are in homozygous condition. All other progenies are with coefficient d/a<1, with confirmed depression, which indicates heterozygosity by this index.

Three progenies are the manifestation of heterosis in F1, d / a> 1 and confirmed inbreeding depression in F2 on the weight of the fruit in crosses with parents ♀ Tsvetelina x ♂ A8000273. They have dominant alleles in homozygous state. In others the indicator is d/a>1, and there is not depression. These progenis are in heterozygous state of the allele pairs.

Three progenis from this crosses have shown heterosis in F1, d/a>1, and the confirmed inbred depression in F2 with respect to the weight of the seeds. The values of inbreeding depression in each of them are from 33,66 to 45,41 percents. Only in one progeny the recessive state of the allele pairs is homozygous.

Conclusion

1. The peanut collection stored in National Bulgarian Genebank is characterized by great diversity in terms of the studied morphological traits, explained by the wide genetic base and diverse geographical

origin of the accessions.

2. On the basis of study of 97 breeding lines of Bulgarian origin *Arachis hypogea* L.by some morphological characteristics, responsible for productivity, a linear model of the plant characterized by high yield of seeds was established.

3. As a result of the evaluation of parents study their genetic distance and objectives of the breeding model the set parental couples were selected. The obtaining hybrids with high yield and total seed weight, low and spread shrub are of interest.

4. As a result of hybridization, interesting hybrid material that allows the acceleration of the breeding improvement in peanuts were obtained.

5. The heterosis effect in the F1 and F2 inbred depression of the production of fruit and kernels allow selection of homozygous and heterozygous state of the allele pairs, responsible for the manifestation of these traits.

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Korišćenje biljnih genetskih resursa u programu uzgajanja kikirikija u Bugarskoj

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Sažetak

Na osnovu proučavanja nekih morfoloških indeksa, koji su odgovorni za produktivnost u 97 uzgojnih linija kikirikija *Arachis hipogea* L. bugarskog porijekla, utvrđen je model linearne regresije biljke, koja se karakteriše visokim prinosom sjemena, Program uzgajanja zahtijeva identifikaciju direktnih i indirektnih efekata na produktivnost. Primjenom analize koeficijenta puta, utvrđeno je da biljke sa višim prinosom moraju imati nisku i široku krosnju (i> 2,0). Lokalne sorte kikirikija se odlikuju nižim indeksom grma (i <2,0). U tu svrhu, 399 genotipova različitog geografskog porekla je uvedeno od USDA (SAD) u 2008. Godini. Na osnovu procene stranih sorti, po visini i širini biljke, prinos sjemena od jedne biljke i otpornost na *Fusarium*, izabrana su četiri genotipa. Oni su bili uključeni u ukrštanjima sa bugarskim sortama i linijama tipa Valencia. Proucavane su manifestacije heteroze i neki genetski efekti potomstva. Rezultati pokazuju da su heterozni efekti u F1 nekih od potomstva, u vezi sa prinosom ploda i sjemena, nasliedjeni bez epistatskog efekta. Prirodjena depresija u F2 je pokazatelj homozigotnosti u odnosu na alelnih parova koji definišu prinos.