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## GENETIC ANALYSIS OF ANOTHER CULTURE RESPONSE IN WHEAT GENOTYPES\*

BRANKA LJEVNAIĆ-MAŠIĆ<sup>1</sup>, ANKICA KONDIĆ-ŠPIKA<sup>2</sup>, LJILJANA BRBAKLIĆ<sup>2</sup>, DRAGANA TRKULJA<sup>2</sup>, NIKOLA HRISTOV<sup>2</sup>

SUMMARY: Androgenic responses of  $10~F_{_{\parallel}}$  wheat hybrids and 13~parental genotypes were studied. It was observed that the studied androgenic traits (androgenic capacity, callus yield, and frequency of green plants) were genotype dependent. On average for all genotypes, 10.6% of the anthers were responsive and 12.2 calluses were produced per 100 anthers. Line 9D-27-262 and the hybrid 9D-27-262/Posavka-2, had the highest frequencies of green plants, 11% and 12% respectively. It was found that 30.4% of the genotypes produced 2 or more green plants per 100 anthers, which can be considered as sufficient number for use in breeding. Heterotic effects as well as additive and dominant gene actions were found to control the inheritance of all characters.

**Key words**: androgenesis, inheritance, breeding, wheat

## **INTRODUCTION**

Production of doubled haploid lines (DHL) is commonly used by plant breeders in order to obtain pure lines in the shortest possible time. The anther culture method, in addition to wide hybridization, is a usual way to produce DH plants in cereals (Maluszynski et al. 2003; Ljevnaić and Kondić, 2008). Success in anther culture has been shown to be highly dependant on genotype of anther donor plants (Bruins and Snijders, 1995). Androgenic capacity in hexaploid wheat is controlled by independently inherited traits:

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<sup>&</sup>lt;sup>1</sup> M.Sc. Branka Ljevnaić assistant, Faculty of Agriculture, Trg D. Obradovica 8, 21 000 Novi Sad.

<sup>&</sup>lt;sup>2</sup> Ph.D. Ankica Kondić-Špika sci. associate, M.Sc. Ljiljana Brbaklić associate researcher, M.Sc. Dragana Trkulja associate researcher and Ph.D. Nikola Hristov sci. associate, Institute of Filed and Vegetable Crops, Novi Sad, Serbia.

Corresponding author: Ankica Kondić - Špika, Institute of Filed and Vegetable Crops, Maksima Gorkog 30, 21 000 Novi Sad. Tel.:+381214898-224; e-mail: ankica@ifvcns.ns.ac.rs

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callus induction, plant regeneration and the ratio of green plants. In determination of these parameters, nuclear gene effects were reported to be most important, including mainly additive effects besides dominance, epistasis and heterosis (Ekiz and Konzak, 1994; Chaudhary et al. 2003; Zamani et al. 2003).

The objective of this study was to investigate the androgenic response of  $F_1$  wheat crosses and their parents as well as the genetic control of the component traits of anther culture.

### MATERIAL AND METHODS

The plant material used in this experiment included 10 randomly selected single cross F<sub>1</sub> hybrids of wheat (*Triticum aestivum* L.) and their 13 parental genotypes (Obrij, NSP1, Szegedi-746, Žitnica, 9D-27-262, Posavka-2, Balkan, Košuta, Pobeda, Rodna, Sremica, NS-0-694 and Fundulea-4). The F<sub>1</sub> generation and parental lines were grown at the experimental fields of the Institute of Field and Vegetable Crops, Small Grains Department during the season 2005/2006. The sampled spikes were cold pretreated at 4-6°C for 5-10 days.

After the surface sterilization of spikes in 1.3% NaOCl, anthers were plated onto a modified Potato 2 (Chuang et al. 1978) inductive medium. Four to six weeks after isolation, the developing embryogenic calli were transferred to the 190-2 (Zhuang and Jia, 1980) regeneration medium. The number of responding anthers, calluses/100 anthers, and green plants/100 anthers were recorded.

The experimental design was completely randomized, with four replications. Statistical analyses were carried out in order to determine the effect of genotype on the traits studied. In order to normalize the distributions, all data were transformed before statistical analysis. LSD was used for comparing means for the analyzed traits. Inheritance of androgenic component traits was estimated by using t-test (Kraljević-Balalić et al., 1991).

### RESULTS AND DISCUSSION

Data obtained with respect to androgenic capacity, calli and green plantlet development were recorded on per cent basis and the performance of parents and  $F_1$  hybrids is presented in Table 1. The average androgenic capacities in parental genotypes and  $F_1$ s were 8.2% and 13.8%, respectively. Callus yield ranged between 0.6% (NS-0-694) and 43.3% (NSP 11) in homozygous genotypes, and between 3.9% (NS-0-694/Košuta) and 40.0% (Szegedi-746/Zitnica) in heterozygous  $F_1$  hybrids. Line 9D-27-262 and the hybrid which involved this line, 9D-27-262/Posavka-2, had the highest frequencies of green regenerants, 11% and 12%, respectively.

It was found that 30.4% of the genotypes produced 2 or more green plants per 100 anthers, which can be considered as a sufficient number for use in breeding. Three homozygous genotypes (Obrij, Szegedi-746 and NS-0-694) and one heterozygote (NS-0-694/Košuta) had no regenerated green plants.

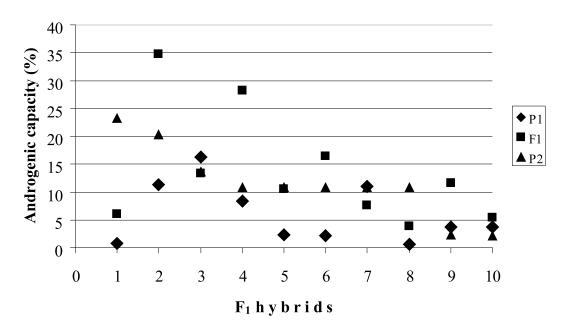
Out of the 10 F<sub>1</sub> combinations, three combinations (Obrij/NSP11, Szegedi-746/Žitnica and NS-0-694/Košuta) had the androgenic capacity close to the mid-parental values, and two combinations (9D-27-262/Posavka-2 and Pobeda/Košuta) had the ca-

pacity at the level of the parent value. Four combinations (Balkan/Košuta, Rodna/Košuta, Fundulea 4/Pobeda and Fundulea 4/Rodna) had a higher number of responding anthers than the better parent, while only one combination (Sremica/Košuta) had the value lower then both parents (Graph. 1).

The  $F_1$  combinations Obrij/NSP11, NS-0-694/Košuta and Fundulea 4/Rodna had callus yield close but significantly different from the parent value, indicating partially dominant inheritance and additive gene action. Callus yield values of the  $F_1$  hybrids 9D-27-262/Posavka-2 and Pobeda/Košuta were not significantly different from the parent value, indicating dominant inheritance. All other  $F_1$  combinations had higher callus yield than the better parent, except Sremica/Košuta, which had a lower value than the parent with low callus yield (Graph. 2).

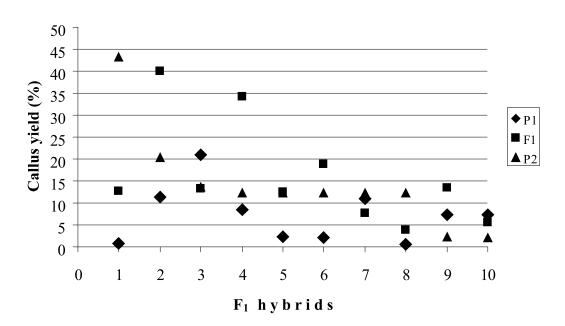
Table 1. Anther culture response of  $F_1$  wheat hybrids and their parents Tabela 1. Reakcija u kulturi antera  $F_1$  hibrida pšenice i njihovih roditelja

	Androgenic capacity	Callus yield	Green plants
Genotype/cross	Androgeni kapacitet	Prinos kalusa	Zelene biljke
Genotip/Ukrštanje	(%)	(%)	(%)
Parents/Roditelji:			
1. Obrij	0.7	0.7	0.0
2. NSP11	23.3	43.3	0.7
3. Szegedi-746	11.3	11.3	0.0
4. Žitnica	20.3	20.3	4.7
5. 9D-27-262	16.3	21.0	11.0
6. Posavka-2	13.7	13.7	4.3
7. Balkan	8.4	8.4	0.6
8. Košuta	10.8	12.3	1.0
9. Pobeda	2.3	2.3	0.4
10. Rodna	2.1	2.1	0.6
11. Sremica	11.0	11.0	1.8
12. NS-0-694	0.6	0.6	0.0
13. Fundulea-4	3.7	7.3	0.4
Mean/Prosek (parents):	9.6	11.9	2.0
F1 hybrids/F1 hibridi:			
1. Obrij/NSP11	6.0	12.7	0.7
2. Szegedi-746/Žitnica	34.7	40.0	3.0
3. 9D-27-262/Posavka-2	13.3	13.3	12.0
4. Balkan/Košuta	28.2	34.3	5.1
5. Pobeda/Košuta	10.5	12.5	1.6
6. Rodna/Košuta	16.5	18.8	3.1
7. Sremica/Košuta	7.6	7.6	0.6
8. NS-0-694/Košuta	3.9	3.9	0.0
9. Fundulea 4/Pobeda	11.6	13.5	0.6
$10.\ $ Fundulea 4/Rodna	5.5	5.5	0.2
Mean/Prosek (F1s):	13.8	16.2	2.7
LSD/NZR 0.05	0.49	0.52	0.36
0.01	0.65	0.70	0.48



Graph. 1. Relative performance of F<sub>1</sub> wheat hybrids and their parents with respect to androgenic capacity

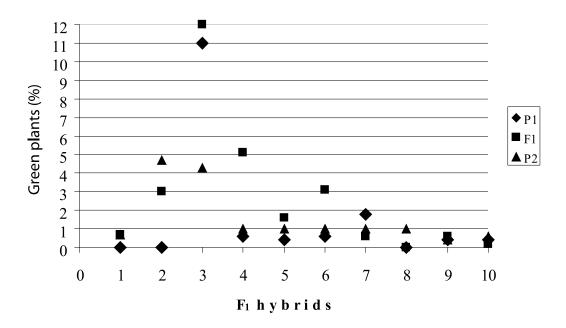
 $Graf.\ 1.\ Relativni\ odnos\ F_{_l}\ hibrida\ pšenice\ i\ njihovih\ roditelja\ u\ pogledu\ androgenog\ kapaciteta$ 



Graph. 2. Relative performance of  $F_1$  wheat hybrids and their parents with respect to callus yield

 $Graf.\ 2.\ Relativni\ odnos\ F_{_l}$  hibrida pšenice i njihovih roditelja u pogledu prinosa kalusa

In terms of green plant frequency, one cross (Szegedi-746/Žitnica) showed partially dominant inheritance, two combinations (Obrij/NSP11 and NS-0-694/Košuta) showed dominance, five combinations (9D-27-262/Posavka-2, Balkan/Košuta, Pobeda/Košuta, Rodna/Košuta and Fundulea 4/Pobeda) exhibited positive and two (Sremica/Košuta and Fundulea 4/Rodna) negative heterosis in green plant yield (Graph. 3).



Graph. 3. Relative performance of F<sub>1</sub> wheat hybrids and their parents with respect to per cent green plant formation

Graf. 3. Relativni odnos  $F_1$  hibrida pšenice i njihovih roditelja u pogledu formiranih zelenih biljaka

Considerable variation was obtained for all component traits of androgenesis, indicating genotypic specificity of wheat androgenic response. It is in agreement with our earlier results (Ljevnaić et al. 2007; Kondić-Špika et al. 2007), as well as with the results of other authors (Tuvesson et al. 2000; Tersi et al. 2006). Genotypes with high per cent responding anthers and calli induction mostly did not respond favorably to green plants regeneration. This suggests that three component traits of androgenesis are governed by different genes leading to independent inheritance of the parameters. The results are in agreement with earlier reports of Chaudhary et al. (2003), Zamani et al. (2003), Vukosavljev (2009).

Significant differences between the average values of the F<sub>1</sub> hybrids and their parents were found for all characters. The potential heterotic cross combinations identified in this study are Balkan/Košuta and Rodna/Košuta for androgenic and 9D-27-262/Posavka-2 for green plantlet regeneration ability. In the earlier studies (Ekiz and Konzak, 1994; Kondić and Šesek, 1999; Chaudhary et al. 2003) similar results were proposed suggesting that heterozygosity is important for androgenic response in wheat.

## **CONCLUSSION**

As one parent with a high androgenic response lead to the production of sufficient number of green plants from the F<sub>1</sub> hybrids, it was concluded that screening inbred lines for anther culture response could increase the efficiency of this method if used in combination with well-responsive parent lines.

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# GENETIČKA ANALIZA REAKCIJE GENOTIPOVA PŠENICE U KULTURI ANTERA

## BRANKA LJEVNAIĆ-MAŠIĆ, ANKICA KONDIĆ-ŠPIKA, LJILJANA BRBAKLIĆ, DRAGANA TRKULJA, NIKOLA HRISTOV

### Izvod

U radu je ispitana androgena sposobnost kod 10 F<sub>1</sub> hibrida pšenice (*Triticum aestivum* L.), kao i 13 roditeljskih genotipova.

Utvrđeno je da su ispitivana androgena svojstva (androgeni kapacitet, prinos kalusa i frekvencija zelenih biljaka) u velikoj meri zavisila od genotipa. U proseku za sve ispitivane kombinacije ukrštanja androgeni kapacitet je iznosio 10.6%, sa prosečno formirana 12.2 kalusa na 100 izolovanih antera. Linija 9D-27-262, kao i hibrid koji uključuje ovu liniju (9D-27-262/Posavka-2) imali su najviše frekvencije zelenih biljaka (11% i 12%). Rezultati su pokazali da je kod 30.4% genotipova dobijeno 2 ili više zelenih biljaka na 100 izolovanih antera, što se smatra dovoljnim brojem za korišćenje u oplemenjivanju.

Heterotični efekat, kao i aditivno i dominantno delovanje gena utvrđeno je u kontroli ispitivanih androgenih svojstava.

Ključne reči: androgeneza, nasleđivanje, oplemenjivanje, pšenica

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