ASSESSMENT AND ANALYSIS OF AUTOPOLYPLOID GROSSULARIA RECLINATA MILL (= *RIBES UVA-CRISPA L. SUBSP. RECLINATA* (L.) RCHB.) TRAITS

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ABSTRACT

Autopolyploid method of breeding has great importance for hereditary variation increase in receipt of the primary breeding source. This method causes far-going versatile changes in traits and properties of plants. As experience shows, economic traits, which did not become apparent at diploid level could be increasingly revealed in passing to a new ploidic level, changing the norm of reaction, and stipulating biological benefits.

Key words: gooseberry, avtopoliploidy, the initial breeding material, colchicyny.

INTRODUCTION

The researches on polyploidy experiment, clarifying the distinctiveness of autopolyploids in comparison with primary diploids, make the basis for sustainable use of plant genebank as a primary source for selective breeding. Thereby, autopolyploidy should be treated as one of the most important selective breeding technique, which allows to get new primary genebank (Chuvashina 1980).

In the middle of the last century, induced autopolyploidy has been put in use and become effective with crop series. It is apparent that selective breeding at diploid level within the range of one type reaches a deadlock lately. It is hard to grow something new standing in marked contrast with parent. Selective breeding on a polyploidy level opens up opportunities in gaining of new traits and enhancement of desired traits (Trunin 1972).

The first steps in experimental autopolyploidy gaining in gooseberry family (*Grossulariaceae* Dumort) were taken by E.V. Velikanova (1937) in Central Genetic laboratory named after I.V. Michurin. In the 40s-60s of last century, the method of colchicination was used to induce autotetraploids (2n(4x) = 32) of different currant and gooseberry varieties.

In the 70s-80s of last century, autopolyploid series were gained and are successfully being used in selective breeding nowadays (Bavtuto 1980).

In recent years, the method of experimental autopolyploidy was put to use for gaining tetraploid forms of different wild and cultural varieties of black currant, red currant, golden currant, and gooseberry. Forms, resistant to fungal and viral diseases, currant gall mite, and with high winter hardiness have been selected from the mentioned material. During the selective breeding process, the competitive forms, combining tolerance to hostility with high productivity and good fruit quality have been segregated (Buchenkov 2013).

In spite of subfecundity, the study shows that autotetraploids are easily responsive to selective breeding improvement. Quadruplication of the same chromosomal complements sharply limits the scope for morphological and physiological core flaw expression that allows to gain high-yielding varieties (Sankin 1993).

Further successful autopolyploidy implementation into gooseberry culture (*Grossularia* Mill) is eventual in case of experimental polyploidy methods improvement. The most important among which is the study of different polyploidy factors efficiency of actions both separately and combined with growth-substances such as auxesis.

Gooseberry culture meets the requirements applicable to plants where colchicination is the most promising as it is true diploid (2n = 16), evolves only at diploid level, and vegetatively propagable. The latter peculiarity allows to consolidate polyploidy-caused hereditary changes.

MATERIAL AND METHODS

The studies were performed from 1998 to 2009 at an agrobiological station of BSPU named after M. Tank, and from 2009 to 2013 at an experimental ground in Polessky State University. The object of research – *Gr. reclinata* (= *Ribes uva-crispa L.* subsp. *reclinata* (L.) Rchb.) varieties – Russkiy, Slivoviy, Kolobok (agrobiological station of BSPU named after M. Tank); Belorusskiy sakharniy, Chernomor, Yubileyniy (experimental ground of Polessky State University).

To get the gooseberry autotetraploid forms the apical buds were treated in the budbreak phase by colchicine in water- and glycerine solution in concentration 0.1; 0.5; 1,0; 1.5% within 24, 36, 48 hours exposure. There were treated 40-60 buds in each example with each variety. There were used two application methods such as gelatin capsule and apical meristem dropping. After certain exposure effect, the buds were 0,001% heteroauxin-washed. The sprigs were cut and rooted under mist propagation.

At the end of the first growing period, the tetraploid selection was done according to morphological character, the next year – to the results of cytologic analysis (Sankin 1967). Chromosome count in rootlet tip cells was done on stained squash preparation (Ribin 1967).

RESULTS AND DISCUSSION

During the research period, 14 504 buds were treated in 48 options. 411 plants were selected (2.83% out of treated buds) according to morphological analysis, and 44 plants (0.30% out of treated buds) – to a cytologic analysis (see Table 1).

Table 1

Estimation of polyploidy methods of gooseberry varieties

Polyploidized solution	Application methods	Concentration, %	Exposure, hour +	Treated buds, in pieces	Selected tetraploids according to analysis, in pieces	
					morphological	citologic
Colchicine in water	apical meristem dropping	0.1	24	295	-	-
			36	297	-	-
			48	300	-	-
		0.2	24	301	-	-
			36	308	11	-
			48	303	12	-
		1.0	24	306	13	1
			36	305	20	5
			48	304	15	2
		1.5	24	302	13	1
			36	304	12	–
			48	306	11	-
	gelatin capsule application	0.1	24	296	-	-
			36	304	-	-
			48	302	-	-
		0.5	24	307	12	-
			36	304	15	-
			48	302	11	1
		1.0	24	301	13	1
			36	303	23	8
			48	300	15	2
		1.5	24	305	11	–
			36	307	13	1
			48	300	11	-
Colchicine in glycerine	apical meristem dropping	0.1	24	302	-	-
			36	301	-	-
			48	304	-	-
		0.5	24	305	-	-
			36	300	12	-
			48	300	13	1
		1.0	24	302	13	1
			36	303	22	5
			48	300	13	1
		1.5	24	301	12	1
			36	302	-	-
			48	300	-	-
	gelatin capsule application	0.1	24	299	-	-
			36	298	-	-
			48	300	-	-
		0.5	24	301	-	-
			36	304	12	–
			48	302	11	1
		1.0	24	305	12	1
			36	304	21	8
			48	302	13	1
		1.5	24	301	12	1
			36	304	14	1
			48	302	-	-

Source: own research

Summing up the findings of estimation of polyploidy methods, the most efficient method is gelatin capsule application, when the apical buds were treated in the bud-break phase by colchicine in water solution in concentration 1% within 36 hours exposure. Under given conditions 26 autotetraploid plants (59.09% of obtained polyploids) have been gained.

Morpho-anatomical analysis of the selected forms showed that autotetraploids *Gr. reclinata* are plants with compact shrub of heterosis type. The browses tend not to branch and slope upward. The axillary buds are connivent. The leaves are dark green, and twice as large as diploids with blistered surface of limb lamina. The blooms are bigger than their diploids, and with massive ovary. Rounded and fewseeded fruits a bit exceed diploids in size and mass (see Table 2, fig. 1).

Table 2

Morphological characteristic of diploid and autotetraploid forms of gooseberry

Trait	2n = 16	2n = 32
Shrub	medium-grown	heterosis
Browse		
colour	taupe	taupe
branching	ample	weak
Buds		
shape	ovate-pointed	ovate-pointed
colour	dark brown	dark brown
position	right-angular	right-angular
Leaf		
length, cm	3.96 ± 0.18	5.84 ± 0.13
width, cm	4.52 ± 0.12	7.25 ± 0.26
colour	green	dark-green
edge	scalloped	sinuate
shape	5-lamina	asymmetric, cut-leaved
surface	weak leaf-wrinkling	blistered
Petiole		
length, cm	1.95 ± 0.21	2.28 ± 0.43
Flower		
length, mm	9.38 ± 0.16	12.07 ± 0.28
diameter, mm	4.32 ± 0.18	6.29 ± 0.31
time of flowering	first decade of May	9-10 days after diploids
Berry		
mass, gr	3.7	3.8
colour	yellow-green	yellow-green
shape	rounded	ovary
Seeds		
in pieces/ fruit	31	11

Source: own research

a b c g

Fig. 1. Fruits and leaves of gooseberry (Белорусский сахарный) autotetraploid (a,c) and diploid (b, g)
Source: own research

The study of leaf anatomy structure showed that the cells of the upper and lower leaf epidermis of tetraploids are bigger than diploid cells. It is typical for autotetraploids to have elongation of guard cells, chloroplast number and their size, reduced number of stomata and repugnatorial glands per unit of epidermis area, reduced layer of column shaped mesophyll and conducting bundle diameter in comparison with diploids (see Table 3).

Table 3

Comparative epidermis structure leaf analysis of diploid and tetraploid forms of gooseberry

Trait	2n = 16	4n = 32
Upper epidermis cell size (growth 7 x 20)**	10.8 ± 0.8	12.4 ± 0.9
Lower epidermis cell size (7 x 20)**	13.8 ± 1.1	8.1 ± 1.2
Guard cell size (10 x 20)**	6.9 ± 0.7	7.5 ± 0.9
Chloroplast size in guard cells (15 x 90)**	29.2 ± 1.4	31.2 ± 1.5
Number of stomata per visual field (10 x 20), in pieces	19.6 ± 1.1	28.4 ± 1.5
Number of chloroplast in guard cells (10 x 60), in pieces	21.6 ± 1.2	23.6 ± 1.3
Number of repugatorial glands per 1 cm^2 (10 x 20), in pieces	-	-

** in points of eyepiece [ocular] micrometer
Source: own research

It is common for induced autotetraploids to have better than diploids fertility. The study shows that the fertility tends to low 1.36 times on average when diploid gooseberry varieties are converted to the tetraploid level. The pollen fertility of diploid varieties *Gr. reclinata* (= *Ribes uva-crispa L.* subsp. *reclinata* (L.) Rchb.) is 38--42%. The percentage of big, eumorphic and sprouted autotetraploids pollen grains

was above 30% depending on the variety (see Table 4). Therefore, subfecundity of gooseberry autotetraploids in comparison with diploid varieties is caused by abnormal pollen development.

Table 4

Gooseberry pollen viability of various ploidy

Variety	Ploidy	Pollen grains in 5 microscopic fields		
		seen in total, in pieces	serminated	
			in pieces	%
Belorusskiy sakharniy	2n 4n	109 63	42 19	38.18 ± 0.22* 30.16 ± 0.12*
Chernomor	2n 4n	112 68	47 21	41.96 ± 0.33* 30.88 ± 0.15*
Yubileyniy	2n 4n	103 59	40 18	38.83 ± 0.55* 30.51 ± 0.18*
Russkiy	2n 4n	110 61	43 19	39.09 ± 0.25* 31.15 ± 0.12*
Slivoviy	2n 4n	114 64	49 20	42.98 ± 0.36* 31.25 ± 0.18*
Kolobok	2n 4n	107 58	44 18	41.12 ± 0.33* 31.03 ± 0.16*

* X ± xs

Source: own research

CONCLUSION

1. The optimum method of gaining autotetraploid *Gr. reclinata* (= *Ribis uva-crispa L.* subsp. *reclinata* (L.) Rchb.) is treating the apical buds in the bud-break phase by colchicine in water solution in concentration 1% within 36 hours exposure.
2. Mutuality of ploidy level and morphology of vegetative organs, and tendency to enlargement of autotetraploid epidermal structures allows their primary identification at the initial period of plant development.
3. Induced autotetraploids *Gr. reclinata* (= *Ribis uva-crispa L.* subsp. *reclinata* (L.) Rchb.) is a primary source that can be used in selective breeding of varieties with elevated shape of shrub and big fewseeded fruit.

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SUMMARY

Obtained and studied Fund avtotetraploidov gooseberry six varieties. The optimal way to obtain avtotetraploidov gooseberry is the Treatment of the apical buds start blooming stage in a 1% aqueous solution of colchicine for 36 hours. Interdependence ploidy level and morphology of vegetative organs, as well as a tendency to increase the size of epidermal struc-tures in avtotetraploidov allows their identification in the primary period of plant development. Avtotetraploidy gooseberries represent a new source, which can be used in breeding for varieties with elevated form of a bush and large fruits.