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A RECENTLY DISCOVERED ALBINO SEEDLING MUTANT IN MAIZE

JELENA VANČETOVIĆ*, SOFIJA BOŽINOVIĆ, DRAGANA IGNJATOVIĆ-MIČIĆ, KSENIJA MARKOVIĆ and VIOLETA ANDJELKOVIĆ

Maize Research Institute, Zemun Polje, 11000 Belgrade, Zemun, Serbia

Abstract – A lethal albino seedling mutant of maize that is resistant to imazethapyr was found in the material from the Maize Research Institute, Zemun Polje, gene bank, The mutation was characterized by a normal development of the first two leaves, with white coloration appearing from the third to the fifth leaves, at the base of the leaves spreading towards their end, resulting in the death of the whole plant. While the upper leaves were dying, the first two were still green for eight to ten days. It seemed that the mutation was conditioned by one recessive allele.

Key words: Albino, lethal, maize, mutant

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INTRODUCTION

Albino mutants are well known in plant and animal species. They are usually caused by one recessive allele, which in a homozygous state leads to a total or partial absence of pigment in the organism. In maize, these are so-called white genes, and these are abbreviated by the letter *w*. The following alleles exist: *w1*, characterized by yellow kernels that germinate normally to produce lethal off-white seedlings; *w2* - allelic to *dek21*, white seedlings, endosperm pitted and mosaic for colored and colorless aleurone when colored aleurone genes are present, plastid DNA reduced; *w3* - kernels are viviparous, embryo, scutellum and endosperm are white, aleurone with *A1*, *C1*, *R1*, etc., is colored but with reduced pigment intensity, seedlings are white, a pale yellow kernel appearance in some backgrounds seems to be due to an unidentified flavonoid pigment; allele *w3-8686* is dormant–with white endosperm, pale green seedlings in dim light, blocked in the carotenoid biosynthetic pathway; *w11*, *w14*, *w16* and *w17* - lethal seedlings like *w1*; *w15*, like *w1*, which fail to convert protochlorophyllide to chlorophyllide; *w18* - white seedlings, pale green streaks in some backgrounds; and *w19* - white plant tissue in leaf and sheath chimeras (Neuffer et al., 1997). A similar group of genes, so-called white luteus (abbreviated as *wlu*: *wlu1*, *wlu3*, *wlu4* and *wlu5*) have pale yellow lethal seedlings, varying from nearly white to yellow depending on genetic background; *wlu2* has white seedlings with faint yellow green on the midrib and leaf sheath. A series of similar mutations, but some of them with mostly yellow seedlings, are designated as *cl*, *l*, *lw*, *grt*, *oy* and *ppg*. The mutant we found was not like any of those given above. Here we only describe it, but for a definite genetic description a test for allelism with similar ones would be needed.

MATERIALS AND METHODS

In 2001, eleven total and broad-spectrum herbicides were applied to the whole material of the Gene Bank of the MRI Zemun Polje-Belgrade, in order to search for potential rare mutants resistant to some of them. Only the resistance to the herbicide imazethapyr (Pivot) was unequivocally observed (Vančetović et al., 2004). Six of about 30 plants that showed the full resistance to Pivot were out-crossed to the non-resistant testers. In the year of 2002, testcrosses of the six resistant plants to non-resistant testers were selfed in order to investigate the mode of inheritance of this trait. In 2006 (the

material was planted in 2003, but a strong storm destroyed everything), F_2 generations obtained in this way were sown with as many plants as possible. They were sown ear-to-row, and unusual albino plants were observed in the progenies of some of the selfed ears of two of the resistant plants (Vančetović et al., 2007).

RESULTS AND DISCUSSION

The first two leaves in the albino plants developed normally, but a white coloration appeared from the third to fifth leaf, starting at the bottom of the leaf and spreading towards the edges, causing the death of the white leaves. The first two leaves lived for another eight to ten days. We called this phenomenon "delayed albinism" (Figs. 1 to 8). The field trial was treated only with Banvel-DP (dicamba + dichlorprop) herbicide for broad-leafed weeds which did not affect maize, and so the phenomenon was considered to be of a purely genetic nature. There is, however, a possibility that Banvel-DP (dicamba + dichlorprop) was a sort of a trigger for the expression of this genetically determined trait. However, the proof against this statement is that the plants were in different stages during the treatment with Banvel-DP (dicamba + dichlorprop), as sowing had been done by hand and the time of plant emergence varied due to a very cold spring (plants were in the 3-5-leaf stage), but all the plants showed the same symptoms (occurring at the same spot on the plant).

In order to investigate the mode of inheritance of the trait, the albino and normal green plants in the F2 generations were counted, and the χ^2 test for the segregation of a 3:1 ratio of green:albino plants was performed. The obtained results were perplexing (Table 1).

Since not all progenies of the selfed ears in the F2 generations of plants no. 2 and 5 (resistant to imazethapyr (Pivot)), showed segregation for albino plants (four not segregating ears versus five segregating ears for plant no. 2, and two not segregating ears versus three segregating ears for plant

Fig. 1.

Fig. 2.

Fig. 3.

Fig. 4.

Fig. 5.

Fig. 6.

Fig. 7.

Fig. 8.

no. 5), and since the involved sister cross-testers and their components never showed albinism, it was presumed that plants 2 and 5 were heterozygous for the respective albino gene(s) which expressed their phenotype in the examined F2 generations. As the phenomenon was the same for the progenies of plants 2 and 5, they had to be genetically closely related, which was also supported by their mutual resistance to the herbicide (Vančetović et al., 2007). These plants probably originated from the same line or population (mixtures of our gene bank accessions comprised from 20 seeds of each population and 10 seeds of each inbred line, for the test in 2001 (Vancetović et al.,

Table 1. χ^2 test for the segregation of 3:1 of green : albino plants in segregating F2 populations.

Genot. [a]	Green plants(no.)	Albino plants(no)	Total no.	χ^2	Signifi-cance[b]	Actual segr.	Plants sawn	Emerg. (%)
(T31x2)/1	206	34	240	15.02	**	6:1	288	83.3
(T31x2)/2	189	45	234	5.01	*	5:1	352	66.5
(T31x2)/4	229	75	304	0.00	NS	3:1	384	79.16
(T31x2)/5	49	17	66	0.0605	NS	3:1	96	68.75
(T32x2)/1	56	11	67	3.596	NS	3:1	384	17.45
(T30x5)/1	282	118	400	3.894	*	2:1	448	89.93
(T30x5)/3	135	36	171	1.603	NS	3:1	352	48.58
(T30x5)/4	237	79	316	0.00	NS	3:1	384	82.29
(T31x5)/1	343	81	424	9.53	**	5:1	512	82.8
Total	1726	496	2222	9.189	**	9:2	3200	69.44
Total χ^2				38.71	**			
Consisten.				29.521	**			

*,** - significant at 0.01 and 0.05 probability level, respectively.
NS – statistically non-significant.
[a] Genotypes are obtained by out-crossing plant no. 2 and plant no. 5 resistant to Pivot to the non-resistant sister-cross testers (T), and /1, 2…is a designation of the selfed plant.
[b] χ^2 values for 1 DF: probability 0.05=3.841; 0.01=6.635.

Table 2. χ^2 test for the segregation of 3:1 ratio of green : albino plants in segregating F2 populations separately for plant 2 and 5, respectively.

Genot-ypes[a]	Green plants(no)	Albino plants(no)	Total no.	χ^2	Signifi-cance[b]	Actual segr.	Plants sawn	% of emerg.
Plant 2	729	182	911	14.371	**	5:1	1504	60.57
Plant 5	997	314	1311	0.792	NS	3:1	1696	77.3

** - significant at 0.01 probability level.
NS – statistically non-significant.
[a] Genotypes for plants no. 2 and 5, summing over all cobs and F2-s, respectively.
[b] χ^2 values for 1 DF: probability 0.05=3.841; 0.01=6.635.

2004)). It was also assumed that the seeds of the albino plants could have a lower percent and/or emergence energy in comparison with the seeds of the normal green plants. This could be the reason for a lower number of albino plants in the progenies of plant no. 2 than theoretically expected. For this reason the emergence for each segregating F2 population was calculated, but no obvious correlation between emergence and the obtained segregation ratio (Table 1) was found.

Furthermore, the χ^2 test was separately performed for plants 2 and 5, summing over all cobs and F2s. Results are given in Table 2. The expected ratio of 3:1 of green : albino plants was registered in plant no. 5, while this was not the case with plant

Table 3. Results of χ^2 tests for the 3:1 segregation ratio in selfed progenies *per se*, and also for the sum of them in a particular F2

Genotype Progeny	(T31x2)/1	(T31x2)/2	(T31x2)/4	(T31x2)/5	(T32x2)/1	(T30x5)/1	(T30x5)/3	(T30x5)/4	(T31x5)/1
1	0.244	0.1633	0.0208	0.0083	0.0033	0.1056	0	0.0357	0.0313
2	0.0025	0.0988	0.2664	0.0679	0.5568	0.1746	0.0048	0.2978	0.0225
3	0.1056	0.0022	0.2801	0.1429	0.1667	0.2083	0.2978	0.0679	0.1746
4	0.0208	0.0179	0	0.0023	0.0865	0.0679	0.0625	0.0417	0.0679
5	1.2868	0.6667	0.003	0.2201		0.0181	0.45	0.2201	0.2608
6	0.075	0.0083	0.0473	0.1134		0.002	0	0.1023	0.6667
7	0.0161	0.0679	0	0.3912		0.0139	0.4225	0.7788	0.0417
8	0.8897	0.5	0.1667			0.4849	0.0096	0	0.0096
9	0.0027	0.0181	0.1134			0.6667	0.003	0.0988	0
10	1.1635	0.0417	0.0022			0.0357	0.0225	0.0357	0
11	0.3214	0.0225	0.0245			0.0579	0.2841	0.5559	0.0083
12	0.0023	0.0139	0.1746			0.05	0.2	0.1429	0.0357
13	0.4592	0.2404	0.0865			0.1746	0.0268	0.0296	0.1746
14	0.0679	0.1746	0.375			0.0181	0.2411	0.0179	0.0357
15	0.0096	0.0194	0.2201			0.0357	0.0179	0.1023	0.1429
16	0.0301	0.0865	0.0022			0.4592	0.0069	0	0.2404
17	0.0023	0.2801	0.375			0.0181	0.5042	1.4698	0.0473
18	0.5	0.0194	0.7278			0.0025	0.5817	0.0245	0.075
19	0.0662	0.2841	0.1875			0	0.4712	0.3214	0.4083
20	0.8929	0.0357	0.002			0.0096	0.025	0.0057	0.0662
21	0.4083	0.0357	0.1667			0.1332	0.0042	0.2201	0.0417
22	0.0357	0	0.0208			0.0023	0.4605	0.1023	0.244
23	0.6228	0.0027	0.1458			0	0.9504	0.0313	0
24	0.0083	0.125	0.0096			0.0988	0.0114	0	0.1134
25	0.1134	0.075	0.5568			0.0083	0.0296	0.0579	0.0625
26	0.1332	0.1023	0.3288			0.0096	0.0804	0.0988	0.0662
27	0.0579	0.1429	0.0181			0.0822	0.0625	0.0417	0.2404
28	0.0022	0.0579	0.0539			0.003	0.0988	0.7225	0
29	0.1225	0.0083	0			0.3214	0.0023	0.0417	0.0074
30	0.0225	0.0025	0			0.2411	0.25	0.0245	0.2801
31	0.075	0.0194	0.3642			0.0296		0	0.675
32	0.0025	0.0417	0.3214			0.1667		0.125	0.0179
33	0.1746	0.0539	0.1633			0.675		0.003	0.1667
34	0.125	0.1023	0.0539			0.3288		0.05	0.0357
35	0.4849	0.0018	0.0625			0.0919		0.0139	0.3214
36	0.1667	0.1429	0.002			0.2813		0.0139	0.4536
37	0.5	0	0.0313			0.0296		0.0865	0.1332
38	0.0023	0.5714	0.0027			0.003		0.0096	0.0473
39	0.3642	0.1023	0.0225			0.1056		0.0225	0.0066
40	0.2083	0.0679	0.0194			0		0.0268	0.3214
41		0.4712	0.0625					0.0074	0.0539
42		0.0865	0.2083					0.0313	0.002
43								0.0096	0.4592
44								0.017	0.4849
45									0.1134
46									0.244
Σ^a	0.0516	0.035	0.2444	0.076	0.0831	0.2197	0.6897	0.0232	0.146

[a] - χ^2 for the sum of plants of all the progenies of a particular selfed F2 generation.

Table 4. χ^2 values for 3:1 segregation ratio separately for plant number 2 and 5, respectively.

Plant	Total no. of plants	Σ of green plants	Σ of albino plants	χ^2 values for 3:1 segr.
2	3622	2743	871	0.2276 NS
5	3975	3035	940	0.7268 NS

Table 5. χ^2 values for segregation ratio of 2:1 of segregating:all-green selfed progenies in particular F2s

Genotype	χ^2 for 2:1 segr.	Σ of families	Σ of segregating families	Σ of all-green families
(T31x2)/1	0.47927 NS			
(T31x2)/2	0.12053 NS			
(T31x2)/4	0.72673 NS			
(T31x2)/5	0.38889 NS			
(T32x2)/1	1.0101 NS			
Σ for plant no. 2	1.13778 NS	225	134	91
(T30x5)/1	0.32504 NS			
(T30x5)/3	0.31373 NS			
(T30x5)/4	0.00663 NS			
(T31x5)/1	0.28655 NS			
Σ for plant no. 5	0.75096 NS	261	160	101

no. 2. Given that the total number of plants and the emergence percentages were much higher for plant no. 5 than for no. 2, the obtained results for plant no. 5 are probably more statistically accurate than for plant no. 2.

In 2006 as many green plants as possible were selfed in the segregating populations and in 2007 the expression of this trait was observed. The selfed material was sown ear-to-row with 36 plants in each row for all selfed F2s. It was sowed on an area without herbicide treatment, so that a conclusion on the effects of Banvel herbicide on this phenomenon could be drawn.

As in the previous year the albino plants appeared in some of the selfed progenies, so the influence of Banvel was excluded. Green versus albino plants in the progenies, as well as segregating versus all-green progenies in all selfed F2s, were counted. Results of χ^2 tests for the 3:1 segregation ratio in the selfed progenies *per se*, and also for their sum in a particular F2 (because of a small number of plants in the selfed progeny *per se*) are given in Table 3. Table 4 shows the χ^2 values for the 3:1 segregation ratio separately for a plant number of both 2 and 5, for all selfed F2 generations, while Table 5 presents χ^2 values for the segregation ratio of 2:1 of segregating versus all-green selfed progenies in a particular F2. None of the χ^2 values was significant. Based on these results it is clear that this trait is conditioned by one recessive allele of an actually dominant gene.

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