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Inheritance of Basal Branching in Sunflower

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Abstract: Collection of sunflower lines of the Institute of Oilseed Crops was studied based on its branching. Ten lines with the basal branching trait were identified. It was established that the inheritance of the basal branching is due to the recessive alleles of the genes. For lines InLD1240, Z1064, LD835, KG13, VIR130, KG13, LD4, basal branching trait is due to the recessive homozygote of one gene *b2*. In lines LD72/3, LD156, KG16, I2K2218, basal branching trait is due to the recessive homozygote of two genes *b3* and *b4*. It was established that the genes determining basal branching are distributed in the offspring independently of the genes controlling the top branching.

Keywords: gene, line, mutant, sunflower, trait, inheritance

Introduction

Sunflower is one of the most popular crops in Ukraine. Its branching trait is used when creating new hybrids. Studies of the genetic basis of this trait began in the last century. Primarily, branching types are divided according to branching location on the stem, namely, top, basal and all stem types of branching. Branching across the full stem height is controlled by the recessive allele of the gene *b* (Putt, 1964.) Recessive allele of the *b1* gene determines the top branching (Hockett and Knowles, 1970), and the *b2* gene-basal branching (Demurin and Tolmachev, 1986). Existence of two, and then seven genes, that when being recessive homozygous, were responsive for branching across the stem height was also reported. (Sandu *et al*., 1999).

Basal branching was studied by Gavrilova and Anisimova (Gavrilova and Anisimova, 2003), where lateral branches grew on both sides of the basal part of the main stem. This type of branching was also found in the line VIR 130. Inheritance of this trait was found to be dominant and caused by three dominant genes. At the same time, Nenov and Tsvetkova obtained different results where this type of

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branching character was due to a single recessive gene (Nenov and Tsvetkova, 1994). In our study, such branching was repeatedly found in different lines (Vedmedeva and Tolmaschov, 2005) including the VIR 130 line (Vedmedeva, 2005).

Due to the ambiguity of this issue, we tried to compile a collection of lines according to their branching and establish its inheritance.

Materials and Methods

Collection of sunflower (*Helianthus annuus* L.) lines from the Institute of Oilseed Crops was the material of the study. This collection from the Laboratory of genetics and genetic resources contained 503 samples. When describing the samples, we used the formula for the description of branches in the form of three gradations: apical, medial and basal. Also, individual samples were labeled as having all stem type of branching. For the genetic investigation of the basal branching trait, 15 lines were used. To establish the inheritance of the trait, we used lines with basal branching: I2K2218, LD156, LD835, VIR130, KG13, KG16, LD72/3, Z1064, InLD1240, LD4 and lines with top and all stem branching: AH70029Rf, LG3, In7034, InK235, HAR7. Crosses and studies of hybrids of the first and second generations were carried out in 2012–2017.

Hybridization was conducted as described by Plotnikov (1940) using manual castration. Study of genetics peculiarities was conducted according to “Tihomirova techniques” (Tihomirova, 1990).

Sunflower crops were seeded after barley as the predecessor. Sowing cultivation was carried out at a depth of 6–8 cm with simultaneous application of the “Herb” herbicide at a dose of 2.5 liters per ha. Sowing was carried out to a depth of 7–8 cm at the density of 40,000 plants per ha.

Results and Discussion

After describing collection with the branching types, a group with basal branching was identified. For plants belonging to its lines, development of lateral shoots in the basal part of the stem was observed, usually these were two shoots and the plant resembled a trident, sometimes there was a single shoot or several more thin and under-developed ones. Following lines were assigned to the basal branching group: I2K2218, LD156, LD835, VIR130, KG13, KG16, LD72/3, Z1064, InLD1240 and LD4. This type of branching manifested itself either in the form of two powerful side shoots, ending with heads, sometimes having practically the same diameter as the central one, or two or more weak lateral shoots in the basal part of the stem. Lines with this trait were crossed with single-head non-branched lines to establish their inheritance.

In all cross combinations, plants of the first generation had no branching, and in the second, segregation was observed. In seven combinations of crossing: VA1 × Z1064, LVO7 × Z1064, VA1 × InLD1240, LVO7 × KG13, LD835 × VK475, KG13 × SL2966, VIR130 × MV4 plants were divided with segregation ratio 3:1 between single-branched and branched plants. This indicates a monogenic recessive inheritance of basal branching trait in the lines InLD1240, Z1064, LD835, VIR130 and KG13 (Table 1).

At the same time, the type of branching studied was identified. Hybrids of VIR130 × KG13, KG13 × Z1064 were obtained. Hybrids obtained from crosses between themselves in the first and second generations had basal branching in all plants, which indicates identical genetic control of this trait. Demurin and Tolmachev (1986) studied line VIR130 and called this gene *b2*. We continued to call this gene *b2*.

LD72/3, LD156, and KG16 lines did not have two clear basal lateral branches; often they had more than two less pronounced branches with heads of considerably smaller diameter than in the previous sub-group of the lines. Results of their crossing with single-head lines and the segregation of second-generation offspring corresponded to 15 part non-branching to 1 part

Table 1: Inheritance of basal branching trait.

Source of branching trait	Cross combination	Phenotypic classes in F2		Total	Expected ratio	χ^2
		no branching	basal branching			
Z1064	VA1 × Z1064	302	91	393	3:1	0.72
Z1064	LVO7 × Z1064	263	76	339	3:1	1.2
InLD1240	VA1 × InLD1240	362	119	481	3:1	0.18
KG13	LVO7 × KG13	137	47	184	3:1	0.02
KG13	KG13 × SL2966	49	23	72	3:1	1.85
LD835	LD835 × VK475	71	24	95	3:1	0.0035
VIR130	VIR130 × MV4	106	45	151	3:1	1.85
LD72/3	LD72/3 × Temp1254	50	6	56	15:1	1.90
LD72/3	LD72/3 × SL2966	145	12	157	15:1	0.52
LD156	LD156 × KG111	110	11	121	15:1	0.41
KG16	KG16 × SL2966	84	4	132	15:1	0.43

$\chi^2_{0.5(k=1)} = 3.84$

branching, which corresponds to the control of the trait by the recessive alleles of the two genes.

To clarify the inheritance of basal branching trait, identification crosses between different groups of lines with basal branching were carried out, those that had monogenic and digenic inheritance of traits among themselves.

Hybrids of the first generation from crosses VIR130 × LD156, KG16 × VIR130, KG13 × I2K2218, I2K2218 × VIR130 had no branching. Combination of crossing KG16 × VIR130 showed segregation in the second generation between 85 non-branching and 47 basal branching plants. We tested the hypothesis: expression of the trait by one gene in a recessive allelic state - 3:1 ($\chi^2 = 7{,}92$), expression of the trait by the recessive alleles of two independent genes 9:7 ($\chi^2 = 3{,}55$), and the model combining the inheritance proven above, namely, the two-gene 15:1 and monogenic recessive 1:3. Latter model is presented in Table 2. According to the model, in second generation we have two classes of plants with the ratio of 45 (27 + 9 + 9) of non-branching to 19 (1 + 3 + 3 + 3 + 9) branching.

According to the model, two classes of plants were obtained in second generation, and according to Pearson criteria they corresponded to the presented three-gene model. To form a basal branch, either a recessive homozygous state of the *b2* gene or a double recessive state of two other genes, which we designated in the continuation of *b3* and *b4*, is necessary.

In sunflower the appearance of branches is described in scientific studies not only at the bottom of the stem, but also in the top part and throughout the

Table 2: Identifying basal branching type.

Parents	KG16 B2B2b3b3b4b4 basal branching	VIR 130 b2b2B3B3B4B4 basal branching
F1	B2b2 B3b3 B4b4 no branching	
F2	B2-B3-B4- no branching	b2b2B3-B4- branching
	27 expected plants	9 expected plants
	B2-B3-b4b4 no branching	b2b2B3-b4b4 branching
	9 expected plants	3 expected plants
	B2- b3b3B4-non branching	b2b2b3b3B4-branching
	9 expected plants	3 expected plants
	B2-b3b3b4b4 branching	b2b2b3b3b4b4- branching
	3 expected plants	1 expected plants
Expected ratio	45 non branching	19 branching
Obtained number of plants	93 non branching	39 branching
Obtained ratio	85 non branching	47 branching

$\chi^2 = 2.21 < \chi^2_{0.5(k=1)} = 3.84$

stem. It is known that these types of branching are caused both by the recessive alleles of one gene, and by the dominant alleles of other genes. In our collection there are lines with branching of these two types.

LG3, HAP7, and In7034 lines had upper branching due to the recessive alleles of one gene. Since the gene for which recessive allele causes the top branching was called *b1*, by Hockett and Knowles (1970) were also denote it with this symbol. In our collection, there were many lines with branching, in which this trait is due to the recessive alleles of one gene. Most of the lines are identified based on branching by studying offspring from crossing the first and second generations. To establish the interaction of their determining genes, we conducted a series of crosses of lines with basal branching and lines with top branching. In crossing combinations: KG13 x LG3, LD72/3 x HAR7, HAR7 x I2K2218, KG13 x In7034, the absence of branching was observed in hybrids of the first generation, and in the second generation segregation fit the ratio of 9:3:3:1, which corresponds to the difference of these pairs of lines in two independent genes. Recessive alleles of one of them cause basal branching, recessive alleles of another – top branching, and recessive homozygous for both genes causes all stem branching. (Table 3). LD72/3 line in Table 1 is shown as having two recessive genes causing basal branching. This fact does

Table 3: Inheritance of different types of branching (2009–2016).

Maternal form (trait)	Paternal form (trait)	Phenotypic classes in F2				Total	Expected ratio	χ^2
		No branching	Basal	Top	All Stem			
KG13 (basal branching)	LG3 (top branching)	78	21	21	8	128	9:3:3:1	1.25
HAR7 (top branching)	I2K2218 (basal branching)	107	34	43	10	194	9:3:3:1	1.78
KG13 (basal branching)	In7034 (top branching)	94	34	32	12	172	9:3:3:1	0.16
LD72/3 (basal branching)	HAR7 (top branching)	77	25	34	7	142	9:3:3:1	2.61
AH70029Rf (top branching)	VIR130 (basal branching)	29	8	85	25	147	3:1:9:3	0.53
AH70029Rf (top branching)	LD72/3 (basal branching)	30	16	102	31	179	3:1:9:3	2.66
InK235 (top branching)	LD4 (basal branching)	27	9	102	26	164	3:1:9:3	2.37

$\chi^2_{0.5(k=2)} = 5.99$

not contradict the obtained 9:3:3:1 segregation, since it is possible if the HAP7 line has one gene in a recessive state from a pair of genes causing basal branching.

In addition to the lines where branching is caused by recessive alleles, in our collection lines were studied where branching was due to the dominant gene. Two of these lines were covered in this study. Crosses involving AH70029Rf and InK235 lines, branching of which is caused by the dominant gene allele, with lines with basal branching, in the first generation showed the dominance of the top branching trait. In the second generation a class of plants with all stem branching appeared, and the class of plants with basal branching was in the minority. The resulting 9:3:3:1 segregation testifies to the presence of one gene whose dominant allele determines the presence of top branching, and the recessive allele determines its absence. Dominant allele of second gene causes emergence of all stem branching with the dominant allele of the first gene. Presence in both genes of recessive alleles in the homozygote causes emergence of a basal branching.

When working on this study, a number of other branching lines were identified, where its inheritance was determined by the dominant alleles of the genes. But morphological manifestation of these types of branching cannot be considered basal branching. It manifested itself either in the top part of the stem, or along its whole length including the basal part.

Conclusions

When summing up the analyzed genetic crosses it is clear that in our collection the basal branching trait is represented in 10 lines, among which two types of inheritance can be distinguished. For lines InLD1240, Z1064, LD835, KG13, VIR130 and LD4, the basal branching trait is due to the recessive homozygote of one gene *b2*. In lines LD72/3, LD156, KG16, I2K2218, the basal branching trait is due to the recessive homozygote in two genes *b3* and *b4*.

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