INVESTIGATION OF MALE STERILITY IN SUGARBEET POPULATIONS

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Abstract

The main aim of this research was to select male sterile parents for breed triploid monogerm sugarbeet (*Beta vulgaris* subspp saccarifera) cultivars. Monogerm triploid hybrid sugarbeet cultivars that obtained using cytoplasmic male sterility have been grown over large areas throughout the world. Identification of male sterile plant is easy; however, O-type plants need progeny test for identification. The experiment was conducted in the fields of seed production of Sugar Institute in Adapazarı and Ankara during 1984-1996. Diploid and tetraploid plants were selected by using chromosome counts from 21 anisoploid populations. During the flowering season, 231.424 plants were examined for fertility, and 190 plants were identified as male sterile. The seeds obtained from these plants were used to grow seedlings which were bolted and checked for the extent of sterility after vernalization. Male sterile progeny ratios of these plants varied between 3.17 % and 18.84 % from population to population. A possitive and strong correlation was found between male sterile plant ratio and O-Type plant ratio within the same population. Based on this correlation, numbers of progeny test were calculated as between 24-148 for the populations.

Keywords: Male Sterility, Vernalization, Progeny Test

Şeker Pancarı Populasyonlarında Erkısırlık Araştırması

Özet

Araştırmanın amacı, monogerm triploid hibrit şeker pancarı çeşidi ıslah etmek için erkısır ebeveynleri seçmektir. Erkısırlıktan yararlanılarak elde edilen monogerm triploid hibrit şeker pancarı çeşitleri (*Beta vulgaris* subspp saccarifera), dünyada geniş alanlarda ekilmektedir. Populasyonda, erkısır bitkilerin morfolojik olarak ayırt edilmesi oldukça kolay olduğu halde, O-Tip bitkilerin belirlenmesi için döl testi melezi yapmak gerekir. Araştırma, 1984-1996 yılları arasında, Şeker Enstitüsü'nün Ankara ve Adapazarı'ndaki tohum üretim tarlalarında yapılmıştır. 21 adet anisoploid populasyondan, kromozom sayımı ile, diploid ve tetraploid bitkiler seçilmiştir. Çiçeklenme döneminde, populasyonlara ait toplam 231.424 bitki, erkısırlık bakımından kontrol edilerek toplam 190 adet erkısır bitki tespit edilmiştir. Bu bitkilerden alınan tohumlar ekilmiş ve yetiştirilen fideler vernelizasyona tabi tutulduktan sonra sapa kaldırılmıştır. Bu bitkilerin dölleri arasında erkısırlık oranları belirlenmiştir. Erkısır bitki oranlarının populasyonlara göre % 3.17-18.84 arasında değiştiği saptanmıştır. Aynı populasyondaki erkısırlık oranı ile O-Tip bitki oranı arasındaki korelasyonlara bağlı olarak yapılan hesaplamalarla, populasyonlarda döl testine tabi tutulması gereken bitki sayılarının 24-148 arasında değiştiği bulunmuştur.

Anahtar Kelimeler: Erkısırlık, Vernelizasyon, Döl Testi

1. Introduction

Commercial triploid seed production is only achieved by use of cytoplasmic male sterility (CMS) in F_1 progenies (Owen, 1946; Schoroter, 1967; Kleon, 1967; Koç, 1989a). For this production, one of the parents must be monogerm (2n or 4n) CMS (Sxxzz) (Owen, 1948, 1950; Hagihara *et al*., 1999), the other one must be O-Type (Maintainer has alternative ploidy of CMS) (Nxxzz) (Koç, 1988c; Panella and Hecker, 1995; Orlov, 2000). It is very easy to identify CMS parent based on morphological traits in the populations during the flowerin period. But the O-type plants could be identified by only progeny test method (Owen, 1948; Margara, 1954; Cleij, 1967; Schoroter, 1967; Hogaboam, 1967; Zinecker, 1975; Gerald and Stewart, 1977; Koç, 1983, 1989a, 2001). There is a strong and positive correlation between the frequency of male sterile and O-Type plant ratios within a population (Schoroter, 1967; Koc, 1989a). In order to exploit this relationship, first of all, the male sterile plants are identified and selected and

the ratios of male sterility are calculated (Savitsky, 1952; Koç, 1988b). Consequently, the breeders have to find at least two O-Type plants in each population (Koç and Kandemir, 1992; Koç, 1983, 1988b, 1989a and 2001). Numbers of the progeny test can be calculated using the ratio of male sterile plant in the same population (Tatlıoglu, 1978; Koç, 1989a). The main aim of this experiment was to select male sterile parents for breed triploid monogerm sugarbeet cultivar (*Beta vulgaris* subspp saccarifera).

2. Material and Method

Twenty one ansoploid populations were used in this study. They were collected from various countries to improve new cultivar(s) during 1984-1986. The development of new triploid hybrid cultivar(s) was planned to use these lines in the following years. For this purpose, O-Type and male sterile plants were identified in each population (Koç, 1989c).

During course of elite seed production, each plant was periodically controlled for sterility throughout the flowering season. After identifiying male sterile plants, they were open pollinated to produce F_1 hybrid seeds (Koç, 1989b). The seeds were sown in the boxes and later germinated seedlings were transferred to the pots (Koç, 1989d).

The seedlings were transferred to the environmental chamber at the 6-8 leaf stage with maintenance of temperature at 4-8 ˚C during 10 weeks for vernalization. Thereafter, the seedlings were transferred to controlled conditions of 19$\pm$1 ˚C and 18 h/day light to stimulate growth (Cucrth, 1967; Koç, 1983 and 1989d; Guan *et al*., 1994; Koç, *et al*., 1995). Bolted plants were examined for male sterility 3 times, every 10 days. Fertility status of each genotype in F_1 progeny was identified by accounting for their ratios (Tatlıoglu, 1978; Koç, 1983, 1988a and 1989a). Atleast two O-Type plants had to be found in a population for maintaining of O-Types. For this purpose, fertility-sterility status of F_1 progenies were used, and calculated with Binomial Dispersion (Tatlıoglu, 1978; Koç, 1989a).

3. Results and Discussions

The male sterile plants were determined during the flowering period in the elite parcels. Total number of examined plants, identified male sterile plants, their status for fertility or sterility, and ratios in F_1 progenies were given in Table 1. Out of 231.424 plants examined during experimental years in the original population, 190 of them were male sterile. Number of the male sterile plants (1-19 pieces for populations) and their ratios (0.010-0.143 %) were low. Only one male sterile plant was found in each of the KMP and R populations. Low ratio of male sterility was reported by Owen (1945) as 6 % and 0.1 %, by Bandlow (1964) as 0.1-1.0 %, by Kleon (1967) as 0.15 % and by Lecochec (1969) as 1.0 %. Koç (1989a) found no male sterile plants in his two breeding lines, and 0.01-1.0 % male sterility in other breeding lines. These results supported that low number of male sterile plants found in sugarbeet populations of this study.

Male sterility in the CMS plants (Sxxzz) is determined by 2 chromosomal genes and sterility factors of cytoplasm (Owen, 1948; Hagihara *et al*., 1999). Plant with the S cytoplasma is completely male sterile, provided its chromosomal genes are homozygous recessive (Sxxzz). If one of these two genes are dominant (X.zz or xxZ.), then this plant is semi-male sterile Type-I, and if both genes are dominant (XxZz) then it is semi-male sterile Type-II. Each of male sterile and fertile plants produces 4 different gamets with these genes combinations (xz, Xz, xZ and XZ) (Tatlıoglu, 1978; Koç, 1989a). Expected genotypic ratio in F_1 progeny after open pollination (AJ-3 population) was given in Table 2. Table 2 shows alternative gamet combinations with occurance of 16 different genotypes (Tatlıoglu, 1978; Koç, 1989a) which are given in Table 3.

Table 1. Genotypic dispersions of male sterile plants in F_1 progenies

Cultivars	Observed plants	Male sterile plants		Frequency of the sterility and fertility in the F_1 progenies								Observed F_1 progenies
		Numbers	%	(xxzz)(a) Numbers	%	(Xxzz) Numbers	%	(xxZz) Numbers	%	(XXZZ) Numbers	%	
AJ-3	7916	6	0.076	87	39.19	46	20.72	53	23.87	36	16.22	222
KWE	13283	19	0.143	190	25.88	251	34.20	192	26.16	101	13.76	734
Max	9447	11	0.116	143	34.71	87	21.12	143	34.71	39	9.46	412
PR	8654	9	0.104	126	33.87	109	29.30	82	22.04	55	14.79	372
Px	16719	12	0.072	103	24.47	73	17.34	128	30.40	117	27.79	421
PK	12196	17	0.139	224	26.63	251	29.85	196	23.31	170	20.21	841
PO	14266	4	0.028	-		-		143	68.75	65	31.25	208
P-3	10095	7	0.069	83	17.77	165	35.33	128	27.41	91	19.49	467
KWP	12453	16	0.128	115	18.34	243	38.76	160	25.52	109	17.38	627
KMP	9814	1	0.010	-		-		14	42.42	19	57.58	33
KW-231	11346	4	0.035	89	43.42	16	7.80	76	37.07	24	11.71	205
KW-S	10459	8	0.076	137	37.13	114	30.89	73	19.78	45	12.20	369
RP	12538	13	0.104	104	35.86	99	34.14	64	22.07	23	7.93	290
MP	10187	5	0.049	126	31.50	117	29.25	75	18.75	82	20.50	400
MMP	8798	2	0.023	-		-		29	61.70	18	38.30	47
ZP	14631	18	0.123	76	40.64	54	28.88	41	21.93	16	8.56	187
R	7493	1	0.013	-		-		9	56.25	7	43.75	16
SR	10549	5	0.047	114	32.20	103	29.10	81	22.88	56	15.82	354
H-5117	9157	10	0.109	65	38.24	42	24.70	45	26.47	18	10.59	170
BM	11286	15	0.133	51	33.12	37	24.03	42	27.27	24	15.58	154
Tri	10137	7	0.064	19	18.81	52	51.49	21	20.79	9	8.91	101
Total	231424	190		-		-		-		-		6630

Table 2. Probable genotypes and frequencies of the male sterile plants in AJ-3 cultivar

	xz (0.3919)	Xz (0.2072)	xZ (0.2387)	XZ (0.1622)
xz (0.3919)	xxzz (a)(0.1536)	Xxzz (0.8120)	xxZz (0.9355)	XxZz (0.6357)
Xz (0.2072)	Xxzz (0.8120)	XXzz (0.4293)	XxZz (0.4946)	XXZz (0.3361)
xZ (0.2387)	xxZz (0.9355)	XxZz (0.4946)	xxZZ (0.5698)	XxZZ (0.3872)
XZ (0.1622)	XxZz (0.6357)	XXZz (0.3361)	XxZZ (0.3872)	XXZZ (80.2631)

Table 3. Observed numbers of genotypes and phenotypes at the end of the open pollination

Genotypes	Phenotypes	Numbers
Xxzz	Completely male sterile	1
Xxzz	Semi-male sterile Type-I	2
XXzz	Semi-male sterile Type-I	1
xxZz	Semi-male sterile Type-I	2
xxZZ	Semi-male sterile Type-I	1
XxZz	Semi-male sterile Type-II	4
XXZz	Semi-male sterile Type-II	2
XxZZ	Semi-male sterile Type-II	2
XXZZ	Fertile	1
Total		16

One of the 16 genotypes had homozygous recessive (xxzz) chromosomal gene combination, six of them had one dominant gene (Xxzz or xxZz: Semi-male sterile Type-I), 8 of them had two heterozygous dominant genes (XxZz: semi-male sterile Type-II) and one of them had homozygous dominant (XXZZ: fertile) genes. Ratios of genotypes were determined and were given in Table 4 showing that among F_1 progenies of male sterile plants belonging to PO, KMP, MMP and R populations, male sterile progeny was not found. The ratios of the other populations varied between 3.17 %

Table 4. Genotypic dispersions of male-sterile parents (%) from F_1 progenies

Cultivars	xxzz (a)	Xxzz	XXZz	xxZz	xxZZ	XxZz	XXzz	XxZZ	XXZZ
AJ-3	15.37	16.21	6.70	18.74	5.70	22.60	4.88	7.74	2.67
KWE	6.70	17.72	9.42	13.57	6.83	25.06	11.68	7.20	1.90
Max	12.04	14.64	4.00	24.08	12.04	21.24	4.45	6.60	0.91
PR	11.48	19.86	8.68	14.92	4.84	22.93	8.59	6.52	2.18
Px	6.00	8.48	9.62	14.90	9.24	24.14	2.99	16.90	7.73
PK	7.09	15.90	12.07	12.42	5.43	24.68	8.91	9.42	4.08
PO	-	-	-	-	47.6	-	-	42.78	9.62
P-3	3.17	12.56	13.76	9.76	7.52	26.28	12.46	10.68	3.81
KWP	3.35	14.20	13.50	9.34	6.50	26.14	15.05	8.88	3.04
KMP	-	-	-	-	17.98	-	-	48.84	33.18
KW-231	18.84	6.78	1.82	32.20	13.76	15.94	0.61	8.68	1.37
KW-S	13.74	22.92	7.54	14.70	3.92	21.30	9.55	4.84	1.49
RP	12.80	24.48	5.38	15.86	4.88	20.80	11.63	3.50	0.60
MP	9.92	18.46	12.02	11.72	3.46	23.82	8.58	7.63	4.21
MMP	-	-	-	-	38.07	-	-	47.26	14.67
ZP	16.48	23.46	4.98	17.78	4.80	19.64	8.36	3.76	0.74
R	-	-	-	-	31.70	-	-	49.20	19.10
SR	10.37	18.74	9.20	14.74	5.24	23.51	8.47	7.04	2.51
H-5117	14.59	18.88	5.24	20.24	7.02	21.19	6.10	5.62	1.12
BM	10.96	15.88	7.48	18.08	7.46	23.42	5.76	8.52	2.44
Tri	3.54	19.37	9.16	7.82	4.33	24.76	26.53	3.70	0.79

Table 5. Numbers of the required progeny test for cultivars

Cultivars	(**a**) Degries	Progeny test	Cultivars	(**a**) Degries	Progeny test
AJ-3	0.1537	30	KW-S	0.1374	33
KWE	0.067	69	RP	0.128	36
Max	0.1204	38	MP	0.0992	46
PR	0.1148	40	MMP	-	-
Px	0.06	78	ZP	0.1648	27
PK	0.0709	66	R	-	-
PO	-	-	SR	0.1037	44
P-3	0.0317	148	H-5117	0.1459	31
KWP	0.0335	140	BM	0.1096	42
KMP	-	-	Tri	0.354	133
KW-231	0.1884	24	-	-	-

(P-3) and 18.84 % (KW-231). Tatlıoglu (1978) and Koc (1989a) did not find any male sterile generations in their some studied populations. However, Koç (1989a) found ratios of the male sterile plants between 24.4 % and 54.6 % in his some other populations. If the ratio of male sterility is high among F_1 progenies, the number of required progeny test is low for this population. If the male sterility ratio is low among F_1 progenies, number of the convenient progeny test is high (Schoroter, 1967; Koç, 1989a). To find two or more O-Type plants in a given population number of required progeny test could be found, using formula $(a + b)^n = 1$ bionomial dispersion (Tatlıoglu, 1978; Koç, 1989a). At this binomial dispersion: to be able to find two or more O-Type plants in a given population number of required progeny test could be found, using formula $(a + b)^n = 1$ bionomial dispersion (Tatlıoglu, 1978; Koç, 1989a). At this binomial dispersion:

a. Frequency of male sterile progeny among the F_1 progenies of male sterile parents.

b. Frequency of O-Type plants in the same population.

n. The number of the required progeny tests.

To be able to find atleast two O-Type plants, within 95 % probability level, n

number of the binomial dispersion is accounted following as:

For AJ-3 population:

$a + b = 1$

$a = 0.1537$ (from Table 4)

$b = 0.8643$ (1-0.1537)

$(a + b)^n = b^n + nb^{n-1}a\ n(n-1)b^{n-2}a^2 +.. + nba^{n-1} + a^n$

95 % 5 %

(Probability being O-Type) or

$0.05 \geq (1 - 0.1537)^n + n(1-0.1537)^{n-1}(0.1537)$

This n number is 30 for AJ-3 population. With the same principles, the numbers could be accounted for each population. Accounted numbers are given in Table 5, which is required progeny tests, these numbers varied from population to population. Moreover, required progeny test numbers were low in the populations which have high ratio of male sterile plant, and were high in thepopulations having low ratio of male sterile plant. The lowest number of the progeny test (with 24 pieces) was found in P3 population. Koç (1988a and 1989a) investigated O-Type plants in 18 breeding lines and varietied progeny test number from 14 to 76. In the following years, O-Type plants were investigated in these populations by the author and were reported in his anothers manustcript.

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