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DEVELOPMENT OF PVX RESISTANT POTATO BREEDING LINES USING MARKER-ASSISTED SELECTION

Ercan OZKAYNAK[*]

**Yuksel Seed Company. Antalya, TURKEY*
**Corresponding author: eozkaynak@yukseltohum.com*

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ABSTRACT

Potato breeding requires a long and laborious study as a based on phenotypic and genotypic selection studies due to its tetraploid and heterozygous plant structure. This research was made to show that the variety development can be faster with molecular assisted breeding. In this research, a short breeding program was applied in order to develop new varieties by taking high dry matter content and PVX resistance as the main criterion. The research was carried out between 2014-2019 and 16.000 F1 potato seeds were used as a genetic material. A hard and effective selection breeding, breeder field observation and PVX markers based on tuber and plant characteristics were selected. In this research, fresh market, French fry and chips variety candidates which have high dry matter content, PVX resistant and higher tuber yield compared to commercial varieties have been developed. According to the results of the research; special breeding programs with two selection criteria can be used to shorten the potato breeding time and commercial candidate varieties with superior properties can be developed. Superior lines that showed to be carrying the markers are strongly recommended to be used by breeders to develop new PVX extreme resistance potato varieties.

Keywords: Advanced line, genetic advance, potato clones, *Solanum tuberosum* L., tuber yield

INTRODUCTION

Potato varieties are developed by crossing hundreds of genotypes and then selection of superior progenies throughout several successive generations in all breeding programs. After crossing, large populations of F1 seedlings are generally grown for visual selection. Single tuber is usually obtained from each F1 plant selected for planting in the next growing season. During the next two to three selection seasons, potato breeding lines are grown and screened for further assessments and propagated to increase seed tuber number. After several years, advanced lines are tested in replicated experiments in several locations to estimate the line x environment interaction (Carputo et al., 2011; Ozturk and Yildirim, 2011; Ozturk and Yildirim, 2014; Ozkaynak et al., 2018).

The normal breeding period in potato can take over 10 years (Jansky, 2009). Due to its autotetraploid and heterozygous nature, breeding is difficult and continues for a long time. In this heterozygous and autotetraploid genome structure has enabled potato to ready adapt to a wide chancing of environmental conditions through the development of small regionally adapted varieties (Ozkaynak et al., 2018). High levels of genetic heterogeneity have also made possible the development of varieties for different industrial options and applications, such as fresh and traditional market use, French fry, crisp processing and starch production (Slater et al., 2014). Recent ten years significant advances in molecular genetics and the analysis of highly complex quantitative traits like tuber yield, starch content, can be exploited by potato breeders to accelerate genetic gains, thus enabling more rapid improvements in potato varieties (Slater et al., 2014).

The main criteria for selection in industrial potato are tuber shape and size, eye depth, dry matter content, starch content and reducing sugar content. Selection of varieties were made according to the desired product. French fries varieties should be long tuber, while chips varieties should be round tuber (Gegov et al., 2007; Kirkman, 2007). The dry matter content is a first and important component in industrial processing and determines the internal quality of the product. Dry matter content of French fry varieties should not be below 19.5% (Kirkman, 2007). In potato chips, the lower limit is around 20%. In addition, the rate of dry matter over 25% of potato is not suitable. Dry matter content was affected by solar radiation, soil temperature, soil moisture, location, harvest date, fertilization and haulm killing during growing period (Gegov et al., 2007; Haverkort, 2007; Mehta et al., 2011). The accumulation of dry matter content was began during the vegetative growth of the plant and continued until harvest. The root system continues to work as long as there is moisture in the soil. With the defoliation of the haulm there is a sudden decrease in the dry matter in the tuber (Kumari et al., 2018; Storey, 2007). Last fifteen years, potato processing industry is emerging as a fast growing industry that more entrepreneurs are investing to this sector while existed ones are increasing their processing capacity (Mehta et al., 2011; Pandey et al., 2009).

Although, last ten years important developments and achievements in molecular tools supply opportunities for rapid genetic income (Slater et al., 2014). But, the use of molecular approaches entail quantitative genetic analysis of the deeply heterozygous breeding populations for development of the complex quality and yield traits with low heritability. Thus, phenotypic selection in potato still remains the common practice in breeding programs. Potato virus X (PVX) is one of the main potato viruses infecting potato all around the world (Gebhard et al., 2006). PVX can infect trade stocks and cause up to 20% crop loss (Ahmadvand et al., 2013). Resistance is the most effective method to control this virus. Dominant resistance gene *Rx1* and *Rx*2 was determined in *Solanum tuberosum* subsp. *andigena* (Ahmadvand et al., 2013). Ohbayashi et al. (2010) developed an STS marker linked to *Rx1*, and the results showed that the recombination frequency was 1.3%. New PVX molecular markers that are tightly linked to the genes were developed for breeding programmes and extreme resistance to PVX (Ahmadvand et al., 2013).

This study was conducted to show that the short-term breeding study can be used in potato for specific purpose (PVX resistant and high dry matter content).

MATERIALS AND METHODS

Genetic materials

For selection program, 16 families were created by crossing 24 commercial varieties and breeding lines having different dry matter content and PVX resistance level. List of parents with main characteristics were given in Table 1. Nine thousand two hundred true seeds per family (1200 seed per family, 19.200 in total) were sown into pots for seedling selection in greenhouse in 2014. No selection was applied at the seedling stage. It was not germinated all seeds and at the end of the season, 16.000 single tuber were harvested. At harvest, one tuber per seedling were selected for selection in the first field generation in next season. Hence field selections were started with 16.000 single hills in 2015. After harvest of the first field generation, 800 breeding lines were selected according to main tuber characteristics (tuber shape, tuber size etc.) as visual evaluation. In 2016, 800 breeding lines were planted as a two row (one row 3-5 plant). 125 advanced lines were selected for plant (plant growth and development, earliness etc.) and tuber characteristics (tuber shape, plant tuber yield etc.) as a breeder eye in 2017 (Table 2). The Rx1 marker (for PVX) was used in parents, breeding lines and selected advanced lines. In 2016, 800 lines tested Rx1 molecular marker with one plant fresh leaves. Selected advanced lines (125 lines) in 2017, marker test was made secondly and confirmed PVX marker results. Generally, PVX resistant lines were selected, but some lines of having good tuber features and not resistant PVX also selected. At the end of 2017, 16 superior potato lines were selected. In 2018, 16 advanced breeding lines were planted in two locations using 30-40 tubers in each location as a two rows. In each growing season, commercial important varieties were used as a control variety. VR-808, a crisp processing potato control variety, Madeline and Orchestra, fresh market potato control varieties; Agria and Lady Olympia as a French fry type potato control varieties were used. Field experiments were conducted at highlands of Antalya/Korkuteli (30° E, 37° N, 1100 m above sea level) in the West-Mediterranean region of Turkey. In 2019, 5 commercial candidates were tested extensively in two locations (Antalya/Korkuteli and Afyonkarahisar/Sandıklı; 30° E, 38° N, 1120 m above sea level) with control commercial varieties. The experiments were laid out in a Completely Randomized Block Design with four replication. 54 tubers of each selected lines/commercial varieties were planted in two rows in each replication. Tuber yield, starch and dry matter rate were subjected to analysis of variance and differences were compared with LSD tests using MSTAT-C statistic program (Freed et al., 1989).

Table 1. Resistance to PVX and association of *Rx1* marker and dry matter content of 24 genotypes used as parents in crossings

Variety	**DM (%)**	**PVX**		**Advanced Line**	**DM (%)**	**PVX**	
Verdi	22	S	Crisp processing	YP-Ali-13	21.4	R	French Fry
Maradona	20. 1	R	Fresh market	13-73-26	20.8	R	Fresh market and French Fry
Madeline	19.5	S	Fresh market	12-53-02	19.1	S	Fresh market
Desiree	21. 6	S	Fresh market and French Fry	22-102-71	21.2	R	French Fry
Florice	17. 6	S	Fresh market	12-189-06	19	S	Fresh market
Granola	21.2	S	Fresh market	12-217-03	22.3	R	French Fry
VR-808	23	S	Crisp processing	12-40-08	21.2	S	Fresh market and French Fry
Orchestra	18.5	S	Fresh market	12-52-96	18.3	R	Fresh market
Volumia	19.1	R	Fresh market	11--03--38	22.4	R	French Fry
Touareg	21	S	Fresh market and French Fry	12-01-128	20.2	S	Fresh market
Victoria	21.4	S	Fresh market and French Fry	12-68-42	20.4	S	Fresh market
Margit	20.6	R	Fresh market	12-245-11	18.8	S	Fresh market

DM: Dry matter content (%)

Table 2. Description of breeding program used in this project

Breeding Stage	No. of year			
Crossing	1	19.200 potato seed spring and 16.000 one tuber autumn season (each population 1000 seed)		2014
Primary individual selection of seedling	2	16.000 single hills in field		2015
Secondary individual clonal selection (4-10 tubers)	3	% 5 hard selection 800 breeding line planted	800 PVX molecular marker test	2016
Secondary individual clonal selection (2-6 kg total tuber)	4	125 superior breeding line (good tuber shape, yield, skin and flesh colour, earliness etc.)	125 superior breeding line dry matter and PVX test	2017
Preliminary performance yield test (60-80 tubers)	5	16 candidate variety selected (minituber production with tissue culture and large-scale yield test in two location)	16 candidate variety	2018
Commercial Candidate Lines	6	5 variety		2019

16 Cross Population

Cultural practices and measured traits

Seed tubers of breeding lines were planted with 30 x 70 cm planting distance in field conditions. Fertilizers was broadcast at 60-80 kg ha^{-1} N, 40-50 kg ha^{-1} P_2O_5 and 80-100 kg ha^{-1} K_2O in different experiment fields. Weeds were controlled by hand after emergence. Disease control like PVX, *Alternaria spp.*, *Phytophthora infestans* and sprinkler irrigation was carried out according to practice. The traits such as maturity, yield, tuber shape, skin and flesh colour, cooking type etc. were determined as a breeder selection. Dry matter content (%) was measured by Zeal potato hydrometer and starch content (%) was determined with a polarimetric procedure (Haase, 2003).

DNA isolation and PCR analysis

Genomic DNA was isolated from young fresh leaves of potato lines using the Wizard Magnetic Kit (Promega) following the manufacturer's instructions. One primers was used for molecular analysis. *Rx1* gene resistance to PVX was screened using the RxSP-S3 and RxSP-A2 primer sets (Mori et al., 2011; Ohbayashi et al., 2010). The PCR products were separated on a 2% agarose gel containing TAE buffer at 110 V for 2 h and visualized under UV light after staining with ethidium bromide. For PVX resistance, the lines having presence *Rx1* marker were selected in 800 lines stage 2016, generally.

RESULTS AND DISCUSSION

Breeding studies are carried out for potato by taking into consideration around 50 different characteristics before the potential is established for new commercial varieties. A breeder will typically select parents for potential pair-wise crossing on the basis of complementary phenotypic characters (Bradshaw and Mackay, 1994; Bradshaw, 2007). These parents will generally be high-performing varieties that require additional improvement, or breeding lines that were proven elite parents and produce higher numbers of superior offspring. It is often important to evaluate new varieties over a number of years and environments, a process that can take over 10 years (Jansky, 2009; Slater et al., 2014). Tarn et al. (1992) reported that at the early stages of selection, parents and crosses play a significant role in breeders' decisions at the later stages of selection when characteristics of the developed individual clones are important.

In this study, twelve special commercial variety and twelve superior advanced line were used as a parent for produce higher numbers of superior lines with high combining ability (Table 1). The varieties Maradona, Volumia and Margit, and advanced breding lines YP-Ali-13, 13-73-26, 22-102-71, 12-217-03, 11-3-38 and 12-52-96 were used as parents in the study due to their resistance to PVX with *Rx1* marker. In 2015, potato lines were selected among 16.000 tubers with a hard selection rate. In 2016, 4 to 12 advanced lines were selected in 16 populations compared to the population. The maximum line was selected from the P2 (Crisp processing x Crisp processing), P9 (French fry x Fresh market), P10 (French fry x Fresh market and French fry), P12 (Fresh market x Fresh market), P13 (Fresh market x French fry), P14 (Fresh market x Fresh market) and P16 (Fresh market x Fresh market) populations, 9 lines and above (Table 3, 4). In 2017, 2-6 kg tubers of 125 lines were planted in double rows in the field. The selection was made in terms of tuber shape, tuber quality, tuber size, earliness, tuber skin colour and tuber flesh colour and yield characteristics (Table 3).

Table 3. Resistance to PVX and association of *Rx1* marker and dry matter content of advanced breeding lines (best sample lines of 125 advanced lines)

No	Advanced Line	DM (%)	PVX	Parents	Important Traits	Candidate Variety
1	62-55-07	21. 5	S	Margit x VR-808	Good tuber shape, high dry matter	
5	62-55-57	19.5	R	Margit x VR-808	Good tuber shape, high yield,, high quality, white flesh	
9	62-59-23	20.6	S	Verdi x VR-808	Long, big tuber, French fry	
10	62-59-34	23	S	Verdi x VR-808	High yield, good tuber shape	Selected Crisp processing
11	62-59-42	21	S	Verdi x VR-808	good tuber shape, French fry	
12	62-59-12	21.7	S	Verdi x VR-808	Tuber shape, quality super	Selected French Fry
16	62-66-05	20.5	R	Maradona X Madeline	Good tuber shape, high yield	
18	62-66-23	19.5	R	Maradona x Madeline	Very high yield, standard, good tuber shape, quality tubers	Selected Fresh market and French Fry
22	62-66-39	20.9	R	Maradona x Madeline	High yield and quality good tuber shape, medium dry matter	
27	62-46-20	17.7	R	Maradona x Desiree	High yield and quality, good tuber shape, super yellow flesh colour	Selected Fresh market
28	62-46-56	21.6	S	Maradona x Desiree	High dry matter, Crisp processing	
33	62-64-03	22.8	R	12--40--08 x 12-52-96	High yield, good tuber shape, high dry matter, Crisp processing	
35	62-64-39	20.1	R	12--40--08 x 12-52-96	Long French fry, good tuber shape, high quality	
36	62-17-64	18. 5	R	12--189--06 x Yp-Ali-13	Big tuber size, medium yield, long tuber shape like Spunta variety	Selected Fresh market
40	62-17-S6	22.3	R	12-189-06 x Yp-Ali-13	Big size, early, good tuber shape, high dry matter, French fry and Crisp processing	
42	62-50-04	21.7	R	12-01-128 X Margit	High yield, good tuber shape, big tuber size, oval-long tuber	Selected French Fry
44	62-50-17	17.5	S	12-01-128 x Margit	good tuber shape, high quality, long tuber	
49	62-61-55	23	R	12-68-42 x Volumia	High yield, oval tuber shape, high dry matter, French fry and Crisp processing	Selected French Fry and Crisp processing
53	62-115-64	16.4	S	12-217-03 x Touareg	Brown tuber skin colour, low dry matter	
56	62-115-34	20.7	R	12-217-03 x Touareg	Brown tuber skin colour, medium dry matter, French fry, and Crisp processing	
57	62-115-25	19.5	R	12-217-03 x Touareg	Very high yield	Selected Fresh market
58	62-115-52	21.1	S	12-217-03 x Touareg	Brown tuber skin colour, white flesh colour, highly dry matter	
61	62-21-06	19.7	S	11-3-38 x Victoria	Very high yield, medium size tubers, yellow flesh colour, French fry	
64	62-21-33	23.0	R	11-3-38 x Victoria	Oval-long tuber, good tuber shape, high yield, yellow flesh colour	Selected French Fry and Crisp Processing
68	62-21-19	23	S	11-3-38 x Victoria	High yield, good tuber shape, Crisp processing	
73	62-83-86	22	R	11-03--38 x Orchestra	Super, long tuber shape, White French fry, White flesh colour	Selected French Fry
76	62-83-44	21.7	S	11-03--38 x Orchestra	good tuber shape, long tuber size, high dry matter, French fry	
79	62-83-52	21.6	R	11-03--38 x Orchestra	Brown tuber skin colour, like Russet Burbank type, long tuber	Selected French Fry
80	62-78-117	19.3	S	Florice x Orchestra	Very high yield, high quality, medium size tubers, good tuber shape	
83	62-78-16	21.5	S	Florice x Orchestra	High yield, good tuber shape, Crisp processing	
85	62-78-23	23.0	S	Florice x Orchestra	High yield, good tuber shape, high dry matter, French fry and Crisp processing	
89	62-78-24	20.5	S	Florice x Orchestra	High yield, good tuber shape, medium dry matter, Crisp processing	
91	62-91-04	18.5	R	13-73-26 x Orchestra	High yield, good tuber shape, Fresh market	Selected Fresh market
94	62-91-53	22.5	R	13-73-26 x Orchestra	High yield, round tuber, yellow flesh colour, good tuber shape	Selected Crisp processing
95	62-91-67	21.5	S	13-73-26 x Orchestra	High yield, good tuber shape, medium dry matter	
100	62-91-52	23	S	13-73-26 x Orchestra	Good tuber shape, high dry matter, French fry, Crisp processing	
103	62-35-76	20.5	S	12-53-02 x Orchestra	Russet Burbank type, long tuber, medium dry matter	
105	62-35-11	19.3	S	12-53-02 x Orchestra	Good tuber shape, Fresh market, low-medium dry matter	
110	62-43-02	22.5	R	22-102-71 x Orchestra	Oval, good tuber shape, high yield, long tuber	Selected French Fry
112	62-43-S9	18.5	S	22-102-71 x Orchestra	Early, good tuber shape, Fresh market	
113	62-43-10	23	R	22-102-71 x Orchestra	Tuber shape and quality super, oval-long tuber, high dry matter	Selected French Fry
118	62-32-20	19.7	S	12-245-11 x Granola	Good tuber shape and quality, low-medium dry matter, Crisp processing	
121	62-32-09	21.5	S	12-245-11 x Granola	Long tubers, high yield	Selected French Fry
125	62-32-76	20.9	S	12-245-11 x Granola	High yield, good tuber shape, medium dry matter, French fry/ Crisp processing	

Table 4. Number of selected advanced lines in sixteen potato families

Family	Crossings	Number of selected advanced line	Family	Parents	Number of selected advanced line
P1	Margit x VR-808	5	P9	12-217-03 x Touareg	9
P2	Verdi x VR-808	10	P10	11-3-38 x Victoria	12
P3	Maradona x Madeline	7	P11	11-3-38 x Orchestra	7
P4	Maradona x Desiree	8	P12	Florice x Orchestra	11
P5	12-40-08 x 12-52-96	5	P13	13-73-26 x Orchestra	10
P6	12-189-06 x Yp-Ali-13	5	P14	12-53-02 x Orchestra	9
P7	12-01-128 x Margit	7	P15	22-102-71 x Orchestra	7
P8	12-68-42 x Volumia	4	P16	12-245-11 x Granola	9
				Total	125

After harvesting, dry matter content of the tubers were determined. High levels of genetic heterogeneity have enabled the development of varieties for multiple applications, such as fresh market use, French fry and crisp processing, and starch production. Although some selection and breeding may be achieved without detailed genetic knowledge, effective breeding programs are now enhanced by both understanding and utilization of the underlying genetics of the target breeding traits (Slater et al., 2014)

In research, 16 advanced lines having super features were selected in 12 populations in 2017. The majority of the lines selected are candidates for varieties suitable for the French fry potato industry (Table 5). At the end of the experiment in two location, 62-66-23 (Fresh market - French Fry), 62-46-20 (Fresh market), 62-61-55 (French Fry-Crisp processing), 62-91-53 (Crisp processing) and 62-43-10 (French fry) lines were selected as commercial candidates. Mini-tuber production will be provided in 5 selected commercial candidates and 11 advanced potato lines. Knowledge of the genetic and other environmental influences on the expression of the target traits is important, and will influence methods for the identification of superior parents, screening of the derived populations and effective selection methods to identify superior phenotypes (Slater et al., 2014).

Table 5. PVX resistance and dry matter content of selected advanced breeding lines

Family	No	Parents	Line Name	PVX^a	DM (%)	Use
P2	YT-1	Verdi x VR-808	62-59-34	S	23	Crisp processing
	YT-2	Verdi x VR-808	62-59-12	S	21.7	French Fry
P3	**YT-3**	**Maradona x Madeline**	**62-66-23**	**R**	**19.5**	**Fresh market-French Fry**
P4	**YT-4**	**Maradona x Desiree**	**62-46-20**	**R**	**17.7**	**Fresh market**
P6	YT-5	12-189-06 x Yp-Ali-13	62-17-64	R	18.5	Fresh market
P7	YT-6	12-01-128 x Margit	62-50-04	R	21.7	French Fry
P8	**YT-7**	**12-68-42 x Volumia**	**62-61-55**	**R**	**23**	**French Fry- Crisp processing**
P9	YT-8	12-217-03 x Touareg	62-115-25	R	19.5	Fresh market
P10	YT-9	11-3-38 x Victoria	62-21-33	R	23	French Fry- Crisp processing
P11	YT-10	11-3-38 x Orchestra	62-83-86	R	22	French Fry
	YT-11	11-3-38 x Orchestra	62-83-52	R	21.6	French Fry
P13	YT-12	13-73-26 x Orchestra	62-91-04	S	18.5	Fresh market
	YT-13	**13-73-26 x Orchestra**	**62-91-53**	**R**	**22.5**	**Crisp processing**
P15	YT-14	22-102-71 x Orchestra	62-43-02	R	22.5	French Fry
	YT-15	**22-102-71 x Orchestra**	**62-43-10**	**R**	**23**	**French Fry**
P16	YT-16	12-245-11 x Granola	62-32-09	S	21.5	French Fry

DM : Dry matter content (%), PVX$^{a:}$ *Rx1* marker

Tuber yield is the most important character besides the plant, tuber, agronomic and quality characteristics of potato. Tuber yields were given in Table 6 on two-location basis. If Table 6 were examined, the highest yields were obtained for 62-66-23 and 62-46-20 candidate varieties under combined of two conditions, and variety Madeline, among the commercial control variety, was statistically included in the same group as 62-46-20. The lowest yields were obtained with the commercial varieties Orchestra and Lady Olympia, and among the candidate varieties, 62-91-53 was included in the same group as Orchestra variety. But, 62-91-53 line was crisp type potato and average tuber yield (48.6 ton/ha) was higher than commercial variety VR-808. All candidate varieties presented higher tuber yield than the control varieties except Madeline. Many traits contributing to the phenotype of a potato plant can be highly influenced by the growing environment, like tuber yield, tuber number, tuber size, specific gravity and processing quality (Jansky, 2009; Slater et al., 2014).

The dry matter content and starch content are the two most essential quality criteria for potato. In terms of dry matter, values of 17.9-23.1% were obtained based on commercial varieties and candidate varieties of the regions of Antalya and Afyonkarahisar (Table 7). It is reported that dry matter content of most potato varieties for commercial usage was ranged about 18-26 (Storey, 2007). The highest dry matter content was provided in the variety candidates 62-43-10 and 62-61-55. Starch is the main component of the tuber dry matter content (Storey, 2007). The distribution in terms of starch content varied between 12.2 % and 14.6%. Of the candidate varieties, the varieties 62-91-53 and 62-43-10 were given higher starch content than other candidates. In general, higher starch and dry matter content was determined in the Afyonkarahisar region compare to Antalya.

Table 6. Tuber yield results of candidate lines with comparing commercial varieties at two locations in 2019

Variety/Candidate Line	**Tuber Yield (ton/ha)**			
	Antalya	**Afyonkarahisar**	**Mean**	**vs**
Madeline	53.5 b	61.9 ab	57.7 b	3
Lady Olympia	43.9 d	47.8 f	45.8 e	9
VR-808	42.9 d	49.6 e	46.2 de	8
Agria	57.9 a	37.7 g	47.8 d	7
Orchestra	40.8 de	50.5 e	45.6 e	10
62-61-55	51.0 bc	62.2 a	56.6 b	4
62-66-23	57.7 a	63.8 a	60.8 a	1
62-46-20	59.9 a	57.3 c	58.6 b	2
62-91-53	49.4 bc	47.9 f	48.6 d	6
62-43-10	51.7 bc	50.7 e	51.2 c	5
General Mean	50.9	52.9	51.9	
Mean of control varieties	47.8	49.5	48.9	
% CV	13.45	12.73	13.09	
F	21.18**	20.43**	24.37**	
LSD	3.28	3.17	2.58	

**: Within columns, means followed by the same letter are not significantly different by ANOVA protected LSD test (p<0.01).

Table 7. Dry matter content and starch content results of candidate lines with comparing commercial varieties at two locations in 2019

Variety/Candidate Line	**Starch (%)**		**DM (%)**	
	Antalya	**Afyonkarahisar**	**Antalya**	**Afyonkarahisar**
Madeline	12.7 bc	13 b	19.4 bc	19.2 bc
Lady Olympia	14.2 a	14.6 a	20.9 b	20.4 b
Agria	12.8 bc	13.3 b	19.6 bc	20.2 b
Orchestra	12.2 c	12.6 c	18.8 c	18.5 c
VR-808	14.4 a	14.6 a	23 a	22 ab
62-61-55	13.3 b	13.8 ab	22.4 ab	23 a
62-66-23	12.6 bc	13.2 b	19.4 bc	19.9 b
62-46-20	12.5 c	12.7 c	17.9 d	18.5 c
62-91-53	14.2 a	14.6 a	22.2 ab	22.7 ab
62-43-10	13.9 a	14.4 a	22.8 a	23.1 a
% CV	11.23	10.58	12.03	11.76
F	13.16*	14.62*	18.26*	19.36*
LSD	0.78	0.71	1.95	1.84

*: Within columns, means followed by the same letter are not significantly different by ANOVA protected LSD test (p<0.05).

The dry matter content is closely related with the efficiency of transmission of the assimilation products to the tuber (Parlar et al., 2001; Tekalign and Hammes, 2005) and is largely controlled by the genetic structure. Dry matter content is a character that can be affected by ecological conditions, duration of vegetation, soil and plant nutrition characteristics, soil temperature and moisture, solar radiation, cultural treatments besides genetic structure (Slater et al., 2014; Storey, 2007; Yilmaz et al., 2002). The results obtained from the candidate and commercial varieties share similarity with the results of (Asmamaw et al., 2010; Ekin, 2009; Jansky, 2009; Pawelzik et al., 1999) who reported that the dry matter content is affected by environmental factors as well as the genetic structure and that generally the varieties with industrial characteristics have a higher dry matter content.

Resistance to disease and pests is important in potato breeding, and the testing of resistance is performed with bioassays and molecular markers. Bioassays are used in the greenhouse or field. However, they are space-consuming and more time is necessary. Therefore, molecular markers that are tightly linked to resistance genes can be used without special facilities for biological evaluation and are not influenced by growth stages or growing conditions (Fullodolsa et al., 2015; Mori et al., 2011). Additionally, molecular markers may reduce costs and increase the precision and efficiency of the selections (Barone, 2004; Fullodolsa et al., 2015; Peleman and der Voort, 2003). In this study, we used molecular markers linked to the *Rx1* gene in potato breeding. PVX occurs commonly worldwide and causes losses of 10–40 % in single infections and is particularly damaging in combination with PVY or PVA (Kreuze et al., 2020).

The introgression of PVX resistance genes into potato varieties are used for controlling the virus and provided extreme resistance to this virus. In the present study, parents, breeding lines and advanced lines were screened with the *Rx1* marker. In 2016, 800 potato lines were tested with the *Rx1* marker. In terms of *Rx1* marker, 446 were found to be resistant according to marker test (unpublished data). Ahmadvand et al., (2013) were obtained that in two F1 populations of crosses showed a 1:1 segregation for PVX resistance (39:36 and 54:42 resistant to susceptible genotypes, respectively). Among 125 advanced lines, 77 lines were found PVX resistant according to *Rx1* marker. While the other lines were not carrier of *Rx1* marker, it cannot be a 100% warrant for their PVX susceptibility, a fact that would rather be important to consider on further investigations, including virus inoculation test under field conditions and breeding new varieties. The marker produced the expected DNA band in 62-61-55, 62-66-23, 62-46-20, 62-91-53 and 62-43-10 commercial candidate varieties for the *Rx1* gene and these candidates were evaluated as high PVX tolerant varieties according to marker test. Our results were in concordance with previous studies (Ahmadvand et al., 2013; Ohbayashi et al., 2010). Nie et al. (2018) were tested 12 segregating populations (642 plants of progeny) demonstrated its performance with HRM (high-resolution DNA melting) assay. In research, 371 progeny that have possessed Rx1. Shaikhaldein et al. (2018) were tested *Rx1* marker with 28 breeding lines. Among them, three contained Rx1 genes which is extreme resistance to PVX. They reported that these lines are resistance to PVX and have a great potential for gene introgression.

Many characteristics, such as high yield, good quality, and resistance to diseases and pests, are desirable in potato varieties (Asano et al., 2012). From a practical point of view, the introgression of PVX extreme resistance genes into potato varieties would be beneficial both for producer and consumer. Potato varieties are bred by the intercrossing of hundreds of genotypes/lines/varieties of various pedigrees resulting in different gene pools in the different breeding programs, where markers may be inherited, linked to a locus in one pool, but this would not be implicit for another. In the study, the selected lines were classified as resistant or susceptible to PVX. In the study, the 16.000 specific true potato seed lines were propagated and selected 2014 to 2019 with PVX resistance, an acceptable tuber shape, tuber yield and dry matter content continued in the variety development process for subsequent years for the evaluation of agronomic, quality and other disease resistance traits and/or used as parental lines in our selection program. Superior advanced lines were selected from the specific breeding program. Five PVX resistant advanced superior lines were submitted for commercial variety registration. These advanced lines are resistance to PVX and have a great potential for gene introgression. Therefore, it should be recommended that *Rx1* containing lines be considered as a contribution in potato crop improvement.

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