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Rice Breeding For Herbicide Resistance In Turkey

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Abstract

The objective of this study was to develop IMI (Imidazolinone) group herbicide resistant rice varieties to control weedy rice (red rice) and the weeds gained resistance against conventional rice herbicides in rice fields. For this, the thirteen cross combinations were done with an IMI resistant rice variety at Trakya Agricultural Research Institute in 2007. Using these crosses, some breeding activities carried out. As a results of these studies promising IMI resistant lines were developed and they were tested in the observation nurseries and the yield trials in 2012 and 2013. Also, a backcross program started in 2008, eight backcross combinations were obtained, seven of them reached to BC_6 and one to BC_4 at the end of 2013. As a preliminary results, some promising IMI resistant rice lines selected in 2013, and they are being tested in the regional trials and demonstration experiment under farmer's conditions in 2014. A few of them will be nominated for registration as commercial varieties in the end of 2014 crop season.

Key Words: Herbicide resistant, Imidazolinone herbicide, rice (*Oryza sativa* L.), rice breeding

Introduction

Rice domestication emanated from the Asian continent specially China about 8000 year ago through selection of agronomically desirable from wild *Oryza Spp* (Khush, 1997). With human influence on the evolutionary process and the intermittent genetic introgression from wild relatives to the crop, weedy relatives of rice come into being. On the other hand, Sato (2000) mentioned that weedy rice differentiated from natural hybridization between cultivars or between perennial wild and domestic types.

Weedy rice populations have been reported in many rice growing areas in the world where the crop is directly seeded. It is a serious problem in the temperate regions. In Asia, however, weedy rice is an emerging problem for example, weedy rice infestation were first reported in Malaysia in 1988, in the Philippines in 1990, and in Vietnam in 1994. They infest rice fields in almost all the rice-growing areas in south and southeast Asia, Sri Lanka and the Philippines (Chauhan, 2013). Sato (2000) pointed out that the weedy rice strains were recognized as harmful weeds in lowland fields, particularly in direct-seeded habitats in Thailand, Malaysia, Vietnam, China, Korea, and Japan. Also, it is a common weed in Bolivia, Chile, Colombia, Guyana, Brazil, United States, and Venezuela.

Weedy rice is a superior competitor to crop cultivars due to early vigor, greater tillering, and greater height of plats. It is a close relative of rice, and weedy rice is most easily recognized in the field at harvest by examination of panicle and grains. Most red rice biotypes are characterized by a red pericarp or outer covering, during processing, it reduces the quality of milled white rice, and shattering the grains before harvesting, it also reduces paddy yield. Burgos et al., (2008) estimated 274 US dollar loss per hectare due to red rice. Rice yield losses due to weedy rice depend on the amount of infestation, it caused rice yield loss by 50-60% with moderate infestation (15-20 weedy rice panicle m^{-2}), 70-80% under high infestation (21-30 panicle m^{-2}) (Chauhan, 2013). Volgsaroj (2000) reported yield losses ranging from 60% to 80% due to red rice infestation in Thailand. Kwon et al., (1991) determined reduction in head and total milled rice production, depending on duration of competition and red rice density, the reduction increased. On the other hand, Diarra et al., (1985) reported 22, 77, and 82 yield losses when the red rice densities 5, 108, and 215 plant/m^{-2}, respectively. Fischer and Ramirez (1993) determined a very strong competition between

cultivated rice an red rice plants, when 5 and 20 red rice plants/m^{-2} existed in rice filed, the yield reduction was 40% and 60%, respectively. Twenty red rice plants/m^{-2} shattered only 35 seeds/m^{-2} before harvest, but contaminated harvested rice with about 1100 kg/ha of red rice grain.

Weedy rice is a great problem in direct seeded area, if the rice is monoculturaly cultivated in the same areas without rotation for a long time, this situation makes the problem more serious. Weedy rice seeds can be dormant for a longer time, because of its shattering and dormancy traits, once a field is infested with red rice, the seed will remain viable and problematic for many years. Noldin et al., (2006) reported that the red rice seed longevity was greater with deeper burial. After a year, there was no viable seed at 5 cm depth, however, at 25 cm depth, there were more persistent, its longevity continues more than three years. Since commercial rice and red rice so closely related, it is difficult to control red rice with conventional herbicides without giving damage to the commercial rice crop.

In order to control red rice, Clearfield technology developed in the United States. It allows the control of red rice (Lincombe, 2004). This technology provides to use imidazolinone herbicides in rice production. The varietal resistance is as a result of induced mutations that were selected for this resistance. The genetic resistance in these mutations has been incorporated into agronomically viable varieties by conventional plant breeding techniques.

Lincombe (2004) IMI resistant rice developed by Dr Timothy Croughan at the Louisiana State University, Agricultural Rice Research Station in 1993 as ethyl methyl sulfonate induced mutation. Using this source, the first IMI varieties CL 121 and CL 141 were released in 1999 and then CL 161 released in 2000. Linscombe (2010) reported that the Clearfield technology was first used on a limited areas in 2002 in United States. This area steadily increased through the years and Clearfield rice was grown on more than 60% of the rice area in the southern United States. This technology is also used in some shout American countries such as Costa Rica, Columbia , and Brazil and European countries such as Italy, Spain, Greece, and Bulgaria. Imidazolinone-tolerant technology is not used only in rice crop (*Oryza sativa* L.), it is also used in the other crops such as maize (*Zea mays* L.), wheat (*Triticum eastivum* L.), oil seed rape (*Brassica naspus* L.), and sunflower (*Helianthus annus* L.) (Tan et al., 2005).

Imidazolinone herbicides provide a useful tool to control red rice and some weeds are resistant to conventional rice herbicides. Meins et al., (2003) applied imazamox from the 3- to 4- leaf up to panicle initiation, it provided excellent control of red rice population. Chin et al., (2007) used imazapic, imazapyr, imazapic+imazapyr and the result revealed that common weeds observed in the experiment field including *Echinochloa cruss-galli*, *Leptochloa chinensis*, *Cyperus iria*, *Cyperus difformis*, *Ludwigia octovalvis* and especially weedy rice (*Oryza sativa)* were controlled successfully by herbicides in Vietnam. On the other hand, in both conventional and reduced tillage systems conditions, 70 g ai/ha Imazethapyr applied by Levy (2004) at preemergence and postemergence stages, controlled red rice (*Oryza sativa* L.), barnyardgrass [*Echinochloa cruss-galli* (L) Beauv.], Amazon sprangletop [*Leptochloa panicoides* (Persl) Hitchc.]i and rice flatsedge (*Cyperus irina* L.) 87 to 99%, respectively.

Rice have been cultivated under continuous flooding irrigation systems for a long time in Turkey. The amount of water needed for rice irrigation depending on length of irrigation period and soil structure changes between 1500 and 2000 mm. Therefore, rice crop needs too much water, because of water shortage, rice growing area is very limited in Turkey. In addition, rice still is a very profitable crop in Turkey, thus, the farmers are very willing to grow rice, therefore, the rice is monoculturaly cultivated without rotation for a long time. On the other hand, after mechanization of rice cultivation or developing high yielding varieties, short plant height varieties have poor competition ability with red rice, are widely grown in Turkey. These situations aggravate the red rice problem, these conditions cause weed problems as well and the weed populations increase year by year in the rice fields. Also, some weed species or weed varieties are getting resistant to conventional rice herbicides in the last years. Thus, new alternative tools are needed for weed control. Imidazolinone group herbicides could be a new alternative to control herbicide resistant weeds.

The objective of this study was to develop IMI (Imidazolinone) group herbicide resistant rice varieties to control weedy rice (red rice) and the weeds gained resistance against conventional rice herbicides in the rice fields.

Material and Method

Material

An IMI resistant variety which has resistant gene against Imidazolinone group herbicide, used in this study as resistance source. This variety was crossed with commercial varieties such as Osmancık-97, Durağan, Halilbey, Ece, Kıral, Neğiş, Krasnodarsky-424, Edirne, Şumnu, and

Gönen in Trakya Agricultural Research Institute. In 2007. Thirteen cross combinations were conducted (Table-1). Seven of them were also used for backcross breeding in 2008 (Table-2). The commercial varieties were used as recurrent parent in backcross combinations.

Table 1. The cross combination conducted in 2007.

No	Pedigree	Cross Combination
1	2007020-TR2500	Osmancık-97 x IMI Variety
2	2007041-TR2521	IMI Variety x Durağan
3	2007043-TR2523	IMI Variety x Halilbey
4	2007044-TR2524	IMI Variety x Ece
5	2007045-TR2525	IMI Variety x Kıral
6	2007046-TR2526	IMI Variety x Neğiş
7	2007047-TR2527	IMI Variety x Osmancık-92
8	2007048-TR2528	IMI Variety x Krasnodarky-424
9	2007049-TR2529	IMI Variety x Kızıltan
10	2007050-TR2530	IMI Variety x Edirne
11	2007051-TR2531	IMI Variety x Şumnu
12	2007074-TR2554	Halilbey x IMI Variety
13	2007134-TR2614	Gönen x IMI Variety

Table.2 The backcross combination started in 2008.

No	Pedigree	Cross Combination
1	2007020-TR2500	Osmancık-97 x IMI Variety
2	2007043-TR2523	IMI Variety x Halilbey
3	2007044-TR2524	IMI Variety x Ece
4	2007046-TR2526	IMI Variety x Neğiş
5	2007047-TR2527	IMI Variety x Osmancık-92
6	2007049-TR2529	IMI Variety x Kızıltan
7	2007074-TR2554	Halilbey x IMI Variety

Method

The single cross was conducted to create segregating population. The modified bulk selection method was practiced for selection. For this, F_2 was planted and harvested as bulk and F_3 was planted as bulk and harvested as single plant selection. Afterward, the pedigree selection method was practiced. The selection continued until obtaining the pure lines to F_6-F_7 generation.

After selecting the pure lines, they were tested in observation nursery, preliminary yield and yield trial, respectively.

Observation nurseries were seeded in dry conditions as row planting. Each plot was 5 m long and consisted of four rows, spaced 25 cm apart.

The yield trails were conducted with randomised complete block experiment design with three replications at Trakya Agricultural Research Institute. The seed rate was 450 seeds m^{-2} and fertilize dose was $N_{180}P_{60}$ kg ha^{-2} for the experiments. The plot size was 3x5= 15 m^{-2} at planting and it was 2,5x4,5= 11,25 m^{-2} for preliminary yield trial, and it was 4x5= 20 m^{-2} at planting and 3,5x4,5= 15,75 m^{-2} at harvesting for yield trials, respectively. The planting was done into standing water broadcasting by hand.

The observations were recorded for flowering and maturity days, plant height and panicle length, the number of panicle per squarmetter, spikelet sterility, grain shattering, lodging, paddy yield, 1000 grain weight of rice and milled grains, head and total milled rice yield.

Backcross Breeding

As mentioned above a backcross program started in 2008. The commercial varieties used as recurrent parent in backcross combination and the backcrossing continued until BC_6

Herbicide Application

Imidazolinone group herbicides, imazamox was used in this study, 40 g imazamox ha^{-2} was applied at 3-4 leaf stages as first postmergence application and 80 g imazamox ha^{-2} herbicide applied as second application after 40-45 days of planting to avoid the late emerging red rice and weeds. After imazamox application the plants did not have resistant gene died, and then selection and backcrossing practiced on remaining alive plants.

Results and Discussion

Conducting an herbicide resistant rice breeding program between 2007 and 2013, some herbicide resistant promising lines obtained and they tested in the yield trials in 2012 and 2013. Also, some of them were evaluated in the observation nurseries . However, some data related to yield trials and backcross studies will be given in this paper.

The analysis of variance revealed high significant difference among the genotypes for rice yield in the experiments conducted in 2012 and 2013 (Table-3 and table-4). Most of IMI lines had higher rice yield than conventional varieties, Osmancık-97 and Edirne. Also, some of them have higher yield potential than the IMI variety, the source of resistant gene used in this breeding program.

Table .3 The paddy yields of preliminary yield trial conducted in 2013.

Entry No.	Variety Name	Rice Yield (ton/ha)
17	2007051-TR2531-1-1-1	9,94 a
15	2007047-TR2527-2-3-1	9,91 ab
12	2007134-TR2614-3-1-2	9,57 abc
16	2007047-TR2527-2-3-3-2	9, 53abcd
9	2007050-TR2530-4-1-1	9,48 abcd
11	2007050-TR2530-4-1-2	9,23 abcd
3	2007041-TR2521-5-3-1	9,01 abcde
4	2007043-TR2523-3-1-1	8,91 abcde
2	2007020-TR2500-6-1-1	8,87 abcde
18	2007074-TR2554-3-3-1	8,73 bcde
6	2007044-TR2524-4-1-1	8,67 cde
1	**Osmancık-97 (Check)**	**8,61 cde**
5	2007043-TR2523-6-1-1	8,54 cdef
13	2007044-TR2524-1-2-1	8,48 cdef
8	2007046-TR2526-7-1-1	8,43 cdef
7	2007044-TR2524-4-4-1	8,36 cdef
14	2007044-TR2524-3-3-2-1	8,35 def
10	**IMI Variety (Check)**	**8,32 def**
19	2007134-TR2614-3-1-1	7,85 ef
20	**Edirne (Check)**	**7,37 f**
	CV (%) = 8,30	LSD (%5)= 1,21

The some agronomic and technological traits of IMI lines and check varieties were given in table-5 and 6. As it is seen in the tables, the characteristics of IMI lines are similar to the traits of conventional popular rice varieties, Osmancık-97 and Edirne at the moment in Turkey. Thus, their agronomic and technological traits can be accepted by the Turkish rice farmers and consumers.

Backcross Breeding

Backcross breeding to develop IMI resistant variety started in 2008 and it continued until BC_6 generation in 2013. This work finished in 2013 and the selection will be carried out in BC_6F_1 populations in 2014. The backcross combination reached to BC_6 generation given in table-7.

At the same time, some single plants were selected in BC_5F_1 generations in 2013 (see table-8) and they were planted in the experiment field as BC_5F_2 in 2014 rice growing season. The selection will be continued in these populations as well.

Introducing the IMI varieties to the Turkish rice farmers will take time, because, it is a new technology for the farmers, the Turkish rice farmers are very conservative in terms of changing technology. Similar situation occurred in the United States, the IMI resistant varieties were first used on a limited areas in 2002 in this country, however, this area steadily increased through the years and IMI rice varieties were grown on more than 60% of the rice area in the southern United States.

The promising IMI lines may be registered as commercial varieties in a few years, after then, the farmers will be able to use them to control red rice. Using this technology, they do not control only red rice but also they can have chance to control some other weeds emerging in the rice fields such as *Echinochloa Spp*, Leptochloa Spp., and *Cyperus Spp*. etc. Similarly Meins et al., (2003) excellently controlled red rice using imazamox. On the other hand, Chin et al., (2007) controlled *Echinochloa cruss-galli*, *Leptochloa chinensis*, *Cyperus iria*, *Cyperus difformis*, *Ludwigia octovalvis*, and weedy rice with imidazolinone herbicide in Vietnam. Also similar results reported by Levy (2004).

Table .4 The paddy yields of yield trials conducted in 2012 and 2013.

Entry No.	Variety Name	Rice Yield (ton/ha)			Yield Range
		2013	2012	Average	
13	2007051-TR2531-1-3	8,77 a	7,53	8,15	2
11	2007074-TR2554-2-2-1	8,46 ab	8,01	8,24	1
9	2007047-TR2527-2-1-2	8,31 abc	7,11	7,71	4
8	2007046-TR2526-2-1-1	8,03 abcd	6,14	7,08	12
3	2007041-TR2521-1-2-1	8,01 abcd	7,33	7,67	5
7	IMI Variety (Check)	7,95 abcde	7,26	7,61	6
2	2007020-TR2500-1-2-1	7,94 abcde	7,53	7,73	3
12	2007046-TR2526-2-2	7,91 abcde	6,64	7,28	8
4	2007043-TR2523-1-3-1	7,85 abcdef	6,75	7,30	7
1	Osmancık-97 (Check)	7,70 bcdef	6,64	7,17	11
5	2007043-TR2523-3-1-1	7,41 cdef	7,14	7,27	9
6	2007044-TR2524-1-2-2	7,38 def	7,04	7,21	10
10	2007051-TR2531-3-1-1	7,03 ef	5,48	6,25	14
14	Edirne (Check)	6,93 f	5,71	6,32	13
	CV (%)= 7,08	LSD (%)= 0,93			

Table.5 The some agronomic characteristics of lines or varieties tested in the yield trial in 2013.

Entry No	Variety Name	Days to Flower	Days to Maturity	Plant Height (cm)	Panicle Length (cm)	Panicle Number M^{-2}	Grain Shattering (%)	Spikelet Sterility (%)	Lodging (1-9)
1-	**Osmancık-97 (Check)**	88	126	98,7	13,7	289	1	5,2	5
2-	2007020-TR2500-1-2-1	90	129	101,0	16,1	357	1	8,0	1
3-	2007041-TR2521-1-2-1	91	137	94,7	14,1	326	2	15,9	1
4-	2007043-TR2523-1-3-1	90	130	108,1	16,8	371	8	17,1	3
5-	2007043-TR2523-3-1-1	93	136	87,3	17,9	477	8	17,8	1
6-	2007044-TR2524-1-2-2	89	126	81,1	15,7	437	1	11,2	1
7-	**IMI Variety (Check)**	102	144	83,1	19,6	513	4	21,9	1
8-	2007046-TR2526-2-1-1	96	140	107,5	18,1	330	1	11,4	1
9-	2007047-TR2527-2-1-2	87	126	96,7	16,4	399	1	10,8	3
10-	2007051-TR2531-3-1-1	92	126	93,3	16,7	403	1	9,1	1
11-	2007074-TR2554-2-2-1	93	137	99,5	15,5	361	2	13,9	1
12-	2007046-TR2526-2-2	97	140	110,4	19,3	369	1	5,7	3
13-	2007051-TR2531-1-3	95	140	110,7	15,4	381	2	12,4	3
14-	**Edirne (Check)**	89	126	105,9	16,8	340	1	7,2	3

Table.6 The some technological characteristics of lines or varieties tested in the yield trial in 2013 .

				Rice Grain		Milled Grain		Milled Yield		
Entry No	Variety Name	Rice Grain 1000 Grain Weight (g)	Milled Grain 1000 Grain Weight (g)	Length (mm)	Width (mm)	Lenth (mm)	Width (mm)	Total Milled	Head Rice	Appearance of Milled Grain
1	**Osmancık-97 (Check)**	32,4	24,4	8,6	3,2	6,1	2,9	73,9	64,3	Translucent
2	2007020-TR2500-1-2-1	31,7	23,3	9,5	3,0	6,7	2,5	72,7	66,5	Translucent
3	2007041-TR2521-1-2-1	26,0	19,7	7,6	2,9	5,6	2,6	72,8	64,5	Translucent
4	2007043-TR2523-1-3-1	25,7	19,3	9,0	2,5	6,7	2,3	73,9	63,9	Translucent
5	2007043-TR2523-3-1-1	26,8	20,2	8,8	2,5	7,0	2,4	73,5	57,8	Translucent
6	2007044-TR2524-1-2-2	30,6	22,1	9,0	2,9	6,4	2,6	72,7	61,3	Translucent
7	**Clierfield (Check)**	23,8	18,1	9,0	2,5	6,7	2,2	73,4	65,3	Translucent
8	2007046-TR2526-2-1-1	37,0	27,7	9,4	3,2	7,2	2,8	70,2	48,7	Translucent with white belly
9	2007047-TR2527-2-1-2	28,2	20,8	8,9	2,9	6,2	2,5	74,5	55,9	Translucent
10	2007051-TR2531-3-1-1	22,8	16,8	8,7	3,1	2,7	2,5	71,7	62,2	Translucent
11	2007074-TR2554-2-2-1	28,1	21,1	8,7	3,2	6,3	2,6	72,2	64,3	Translucent
12	2007046-TR2526-2-2	33,0	24,7	9,1	3,3	6,5	2,7	72,7	62,8	Translucent with white centre
13	2007051-TR2531-1-3	24,4	18,9	8,3	2,9	6,0	2,6	72,4	63,9	Translucent
14	**Edirne (Check)**	40,0	30,0	9,3	3,6	7,1	3,1	73,3	55,9	Translucent

Table. 7 The backcross generation in the end of 2013 crop season.

Pedigree	Cross Combination	Backcross level reached in 2013.
2011147-TR3086	Osmancık-97x IMI Variety	BC_6
2011149-TR3088	IMI Variety x Halilbey	BC_6
2011150-TR3089	IMI Variety x Ece	BC_6
2011151-TR3090	IMI Variety x Neğiş	BC_6
2011152-TR3091	MI Variety x Osmancık-97	BC_6
2011153-TR3092	IMI Variety x Kızıltan	BC_6
2011154-TR3093	Halilbey x IMI Variety	BC_6

Table. 8 The backcross combination in BC_5F_1 generation and selected plants in 2013.

No	Pedigree	Backcross Combination	Generation	The number of selected plants
1	2011147-TR3086	Osmancık-97 x IMI Variety	BC_5F_1	6
2	2011149-TR3088	IMI Variety x Halilbey	BC_5F_1	10
3	2011150-TR3089	IMI Variety x Ece	BC_5F_1	4
4	2011151-TR3090	IMI Variety x Neğiş	BC_5F_1	9
5	2011152-TR3091	IMI Variety x Osmancık-97	BC_5F_1	7
6	2011153-TR3092	IMI Variety x Kızıltan	BC_5F_1	12
7	2011154-TR3093	Halilbey x IMI Variety	BC_5F_1	11
Total				**65**

Conclusion

The herbicide resistant rice breeding program started in Trakya Agricultural Resaerch Institute in Turkey in 2008. Since then, the breeding works have been continued. At the moment, some herbicide resistant promising lines were developed. Also, some IMI resistant backcross population developed. Using these material, IMI herbicide resistant varieties may be registered in the near future, and then, they can be used to control red rice and some weeds in the rice fields in Turkey. Thus, an alternative tool to control the weeds will be given to the rice farmers.

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