ITALUS HORTUS
Supplementary Material

Vol. 28 (2021)
doi: 10.26353/j.itahort/2021.1.3243

Pyramiding resistance genes and widening the genetic base of the apple (*Malus* × *domestica* Borkh.) crop

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Received: 09 March 2021; Accepted: 03 May 2021; Published: 13 May 2021

Citation: Testolin, R., Falginella, L., De Carli, A., De Mori, G. and Cipriani, G. (2021). 'Pyramiding resistance genes and widening the genetic base of the apple (*Malus* × *domestica* Borkh.) crop', *Italus Hortus*, 28(1), pp. 32-43. doi: 10.26353/j.itahort/2021.1.3243

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Supplementary Table 1. Number of progeny obtained from each cross combination.

Seed parents / Pollen donors	Ariwa	GK13	GM37	Golden orange	HM100	Total
Annurca rossa	42	0	41	0	106	189
Api Etoilée	16	0	52	13	55	136
Appia	17	27	61	14	82	201
Astrakhan red	100	134	76	102	90	502
Astrakhan white	95	23	22	0	36	176
Calville blanc d'hiver	41	2	64	1	52	160
Calville rouge	72	60	64	141	68	405
Decio	0	0	218	0	67	285
Delicious	72	32	57	29	94	284
Di Corone	91	7	28	28	28	182
Fuji	52	41	64	41	121	319
Gelata	66	60	62	65	92	345
Golden delicious	55	33	181	20	307	596
Granny Smith	17	52	97	66	174	406
Jonagold	40	0	0	6	0	46
Limoncella	24	44	129	62	25	284
M014.05	0	0	0	0	20	20
M015.01	0	0	0	0	36	36
M018.04	47	36	75	79	132	369
McIntosh	50	95	82	89	89	405
Permain dorée	83	80	123	35	89	410
Red chief	84	58	80	79	147	448
Rome Beauty	74	65	106	50	129	424
Rosa mantovana	205	0	172	111	191	679
Royal gala	0	44	68	88	89	289
Stark Splendour	72	89	43	32	44	280
Total	1415	982	1965	1151	2363	7876

Supplementary Table 2. Number of progeny screened with molecular markers associated to the resistant loci.

Seed parents / Pollen donors	Ariwa	GK13	GM37	Golden orange	HM100	Total
Annurca rossa	27	0	13	0	45	85
Api Etoilée	0	0	0	0	38	38
Appia	12	9	29	9	45	104
Astrakhan red	34	1	9	0	7	51
Astrakhan white	21	60	4	35	16	136
Calville blanc d'hiver	8	1	13	0	34	56
Calville rouge	23	18	22	48	30	141
Decio	0	0	50	0	36	86
Delicious	20	15	31	6	47	119
Di Corone	18	7	4	15	23	67
Fuji	12	3	29	5	63	112
Gelata	19	13	4	18	74	128
Golden delicious	14	9	64	6	98	191
Granny Smith	7	37	65	19	58	186
Jonagold	0	0	0	0	5	5
Limoncella	9	6	89	35	14	153
M014.05	0	0	0	0	20	20
M015.01	0	0	0	0	36	36
M018.04	23	9	45	49	55	181
McIntosh	15	25	24	30	13	107
Permain dorée	30	28	36	13	55	162
Red chief	25	27	43	23	96	214
Rome Beauty	19	8	63	16	17	123
Rosa mantovana	78	0	117	33	15	243
Royal gala	0	19	26	39	45	129
Stark Splendour	17	13	16	10	40	96
Total	431	308	796	409	1025	2969

Supplementary Table 3. Observed segregation in the progeny of the R genes Rvi6 and Pl1 from Ariwa.

Seed parent	Rvi6-/ Pl1-	Rvi6+/ Pl1-	Rvi6- Pl1+	Rvi6+ Pl1+	Total R	Segregation model	Expected unit frequency	$\chi 2$	P value
Annurca Rossa	10	6	6	5	17	1:01:01	5.67	0.04	0.98
Appia	3	2	3	4	9	1:01:01	3.00	0.22	0.89
Astrakhan red	6	17	4	7	28	1:01:01	9.33	3.31	0.19
Astrakhan white	11	2	4	4	10	1:01:01	3.33	0.27	0.88
Calville blanc d'hiver	0	5	1	2	8	1:01:01	2.67	1.08	0.58
Calville rouge	4	7	2	10	19	1:01:01	6.33	1.72	0.42
Delicious	1	12	1	6	19	1:01:01	6.33	3.19	0.20
Di Corone	3	10	0	5	15	3:01:03	1.88	3.71	0.16
Fuji	3	4	1	4	9	1:01:01	3.00	0.67	0.72
Gelata	1	5	4	9	18	1:01:01	6.00	0.78	0.68
Golden delicious	5	5	2	2	9	1:01:01	3.00	0.67	0.72
Granny Smith	1	4	0	2	6	1:01:01	2.00	1.33	0.51
Limoncella	1	4	4	0	8	1:01:01	2.67	1.33	0.51
M018.04	8	6	4	5	15	1:01:01	5.00	0.13	0.94
McIntosh	2	6	2	5	13	1:01:01	4.33	0.67	0.72
Permain dorée	10	11	0	9	20	1:01:01	6.67	3.43	0.18
Red chief	7	9	1	8	18	1:01:01	6.00	2.11	0.35
Rome Beauty	4	7	5	3	15	1:01:01	5.00	0.53	0.77
Rosa mantovana	18	35	10	15	60	1:01:01	20.00	5.83	0.05
Stark Splendour	10	3	1	3	7	1:01:01	2.33	0.38	0.83
Total	108	160	55	108	323				

- The analysis of segregation has been carried out only for R genes for which associated markers were available.
- Several seed parents are missing in the cross combinations with the different R parents.
- In all cross combinations, gametes carrying no resistance gene were excluded from the segregation analysis, because the cross populations were phenotypically screened before the molecular screening and susceptible plants were discarded. See Material & Methods for details.
- 'Expected' stays for expected frequency for each gamete, excluded that one that does not carry any R locus. The analysis of segregation was possible because R loci were carried only by R parents that were heterozygous at all R loci, while seed parents did not carry any resistance, except 'Di Corone' that carried the Rvi6 gene and for which the segregation analysis was a little bit different.
- 'Di Corone', was initially included among the susceptible seed parents. During the experiment it was identified as synonymous of 'Prima', a selection from the PRI program and indeed carried the Rvi6 (=Vf) gene. The segregation analysis was carried out accordingly.

Supplementary Table 4. Observed segregation in the progeny of the R genes Rvi2, Rvi4, and Rvi6 from GK13.

Seed parent	Rvi2-/ Rvi4-/ Rvi6-	Rvi2+/ Rvi4-/ Rvi6-	Rvi2-/ Rvi4+/ Rvi6-	Rvi2-/ Rvi4-/ Rvi6+	Rvi2+/ Rvi4+/ Rvi6-	Rvi2+/ Rvi4-/ Rvi6+	Rvi2-/ Rvi4+/ Rvi6+	Rvi2+/ Rvi4+/ Rvi6+	Total R	Segregation model	Expected unit frequency	$\chi 2$	P value
Appia	2	4	0	0	1	0	0	2	7	1:1:1:1:1:1:1:1	0.88	16.13	0.01
Astrakhan red	0	0	0	1	0	0	0	0	1	1:1:1:1:1:1:1:1	0.13	6.88	0.33
Astrakhan white	8	7	9	7	12	6	5	6	52	1:1:1:1:1:1:1:1	6.50	6.12	0.41
Calville blanc d'hiver	0	0	0	0	0	0	1	0	1	1:1:1:1:1:1:1:1	0.13	6.88	0.33
Calville rouge	2	4	0	2	3	3	1	3	16	1:1:1:1:1:1:1:1	2.00	6.00	0.42
Delicious	2	2	1	4	1	1	0	4	13	1:1:1:1:1:1:1:1	1.63	9.38	0.15
Di Corone	1	0	1	1	0	1	2	1	6	1:1:1:2:1:2:2:2	0.38	6.88	0.33
Fuji	2	1	0	0	0	0	0	0	1	1:1:1:1:1:1:1:1	0.13	6.88	0.33
Gelata	1	3	2	1	1	0	2	3	12	1:1:1:1:1:1:1:1	1.50	5.17	0.52
Golden delicious	1	2	1	1	2	1	0	1	8	1:1:1:1:1:1:1:1	1.00	3.00	0.81
Granny Smith	10	5	5	3	7	3	1	3	27	1:1:1:1:1:1:1:1	3.38	7.25	0.30
Limoncella	1	2	1	1	0	0	1	0	5	1:1:1:1:1:1:1:1	0.63	5.58	0.47
M018.04	0	2	0	4	1	1	1	0	9	1:1:1:1:1:1:1:1	1.13	10.32	0.11
McIntosh	6	1	5	3	3	1	4	2	19	1:1:1:1:1:1:1:1	2.38	5.99	0.42
Permain dorée	7	2	2	2	4	5	3	3	21	1:1:1:1:1:1:1:1	2.63	3.42	0.75
Red Chief	2	7	0	9	1	2	2	4	25	1:1:1:1:1:1:1:1	3.13	21.48	0.00
Rome Beauty	3	0	0	1	1	2	0	1	5	1:1:1:1:1:1:1:1	0.63	5.58	0.47
Royal Gala	4	1	0	3	2	1	1	7	15	1:1:1:1:1:1:1:1	1.88	17.79	0.01
Stark Splendour	1	0	2	1	2	1	3	3	12	1:1:1:1:1:1:1:1	1.50	5.17	0.52
Total	53	43	29	44	41	28	27	43	255				

- The analysis of segregation has been carried out only for R genes for which associated markers were available.
- Several seed parents are missing in the cross combinations with the different R parents.
- In all cross combinations, gametes carrying no resistance gene were excluded from the segregation analysis, because the cross populations were phenotypically screened before the molecular screening and susceptible plants were discarded. See Material & Methods for details.
- 'Expected' stays for expected frequency for each gamete, excluded that one that does not carry any R locus. The analysis of segregation was possible because R loci were carried only by R parents that were heterozygous at all R loci, while seed parents did not carry any resistance, except 'Di Corone' that carried the Rvi6 gene and for which the segregation analysis was a little bit different.
- 'Di Corone', was initially included among the susceptible seed parents. During the experiment it was identified as synonymous of 'Prima', a selection from the PRI program and indeed carried the Rvi6 (=Vf) gene. The segregation analysis was carried out accordingly.

Supplementary Table 5. Observed segregation in the progeny of the R genes Rvi5 and Rvi6 from GM37.

Seed parent	Rvi5-/ Rvi6-	Rvi5+/ Rvi6-	Rvi5-/ Rvi6+	Rvi5+/ Rvi6+	Total R	Segregation model	Expected unit frequency	$\chi 2$	P value
Annurca Rossa	4	3	2	4	9	1:01:01	3.00	0.67	0.72
Appia	8	9	9	3	21	1:01:01	7.00	3.43	0.18
Astrakhan red	2	3	1	3	7	1:01:01	2.33	1.14	0.56
Astrakhan white	1	3	0	0	3	1:01:01	1.00	6.00	0.05
Calville blanc d'hiver	1	7	0	5	12	1:01:01	4.00	6.50	0.04
Calville rouge	7	2	6	7	15	1:01:01	5.00	2.80	0.25
Decio	9	13	18	10	41	1:01:01	13.67	2.39	0.30
Delicious	6	10	8	7	25	1:01:01	8.33	0.56	0.76
Di Corone	0	1	2	1	4	1:03:03	0.50	2.17	0.34
Fuji	4	9	9	7	25	1:01:01	8.33	0.32	0.85
Gelata	0	2	0	2	4	1:01:01	1.33	2.00	0.37
Golden delicious	1	23	1	39	63	1:01:01	21.00	34.67	0.00
Granny Smith	14	17	17	17	51	1:01:01	17.00	0.00	1.00
Limoncella	18	20	18	33	71	1:01:01	23.67	5.61	0.06
M018.04	0	25	1	19	45	1:01:01	15.00	20.80	0.00
McIntosh	7	3	6	8	17	1:01:01	5.67	2.24	0.33
Permain Dorée	7	9	11	9	29	1:01:01	9.67	0.28	0.87
Red chief	9	6	13	15	34	1:01:01	11.33	3.94	0.14
Rome Beauty	19	13	13	18	44	1:01:01	14.67	1.14	0.57
Rosa mantovana	19	32	33	33	98	1:01:01	32.67	0.02	0.99
Royal gala	3	13	2	8	23	1:01:01	7.67	7.91	0.02
Stark Splendour	0	10	1	5	16	1:01:01	5.33	7.63	0.02
Total	139	233	171	253	657				

- The analysis of segregation has been carried out only for R genes for which associated markers were available.
- Several seed parents are missing in the cross combinations with the different R parents.
- In all cross combinations, gametes carrying no resistance gene were excluded from the segregation analysis, because the cross populations were phenotypically screened before the molecular screening and susceptible plants were discarded. See Material & Methods for details.
- 'Expected' stays for expected frequency for each gamete, excluded that one that does not carry any R locus. The analysis of segregation was possible because R loci were carried only by R parents that were heterozygous at all R loci, while seed parents did not carry any resistance, except 'Di Corone' that carried the Rvi6 gene and for which the segregation analysis was a little bit different.
- 'Di Corone', was initially included among the susceptible seed parents. During the experiment it was identified as synonymous of 'Prima', a selection from the PRI program and indeed carried the Rvi6 (=Vf) gene. The segregation analysis was carried out accordingly.

Supplementary Table 6. Observed segregation in the progeny of the R gene Rvi6 from Golden orange.

Seed parent	Rvi6-	Rvi6+	Total R
Appia	4	5	5
Astrakhan white	11	24	24
Calville rouge	8	40	40
Delicious	0	6	6
Di Corone	5	10	10
Fuji	1	4	4
Gelata	10	8	8
Golden delicious	5	1	1
Granny Smith	1	18	18
Limoncella	12	23	23
M018.04	16	33	33
McIntosh	3	27	27
Permain dorée	2	11	11
Red chief	4	19	19
Rome Beauty	7	9	9
Rosa mantovana	4	29	29
Royal gala	11	28	28
Stark Splendour	2	8	8
Total	106	303	303

- The analysis of segregation has been carried out only for R genes for which associated markers were available.
- Several seed parents are missing in the cross combinations with the different R parents.
- In all cross combinations, gametes carrying no resistance gene were excluded from the segregation analysis, because the cross populations were phenotypically screened before the molecular screening and susceptible plants were discarded. See Material & Methods for details.
- 'Expected' stays for expected frequency for each gamete, excluded that one that does not carry any R locus. The analysis of segregation was possible because R loci were carried only by R parents that were heterozygous at all R loci, while seed parents did not carry any resistance, except 'Di Corone' that carried the Rvi6 gene and for which the segregation analysis was a little bit different.
- 'Di Corone', was initially included among the susceptible seed parents. During the experiment it was identified as synonymous of 'Prima', a selection from the PRI program and indeed carried the Rvi6 (=Vf) gene. The segregation analysis was carried out accordingly.

Supplementary Table 7. Observed segregation in the progeny of the R genes Rvi5 and Rvi6 from HM100.

Seed parent	Rvi5-/ Rvi6-	Rvi5+/ Rvi6-	Rvi5-/ Rvi6+	Rvi5+/ Rvi6+	Total R	Segregation model	Expected unit frequency	χ^2	P value
Annurca Rossa	13	14	9	9	32	1:01:01	10.67	1.56	0.46
Api Etoilée	13	11	7	7	25	1:01:01	8.33	1.28	0.53
Appia	10	11	14	10	35	1:01:01	11.67	0.74	0.69
Astrakhan red	0	6	0	1	7	1:01:01	2.33	8.86	0.01
Astrakhan white	8	2	3	3	8	1:01:01	2.67	0.25	0.88
Calville blanc d'hiver	2	15	0	17	32	1:01:01	10.67	16.19	0.00
Calville rouge	1	12	3	14	29	1:01:01	9.67	7.10	0.03
Decio	9	3	14	10	27	1:01:01	9.00	6.89	0.03
Delicious	13	5	13	16	34	1:03:03	11.33	5.71	0.06
Di Corone	2	4	10	7	21	1:01:01	7.00	2.57	0.28
Fuji	20	18	17	8	43	1:01:01	14.33	4.23	0.12
Gelata	20	16	13	25	54	1:01:01	18.00	4.33	0.11
Golden delicious	5	46	2	45	93	1:01:01	31.00	40.71	0.00
Granny Smith	6	27	2	23	52	1:01:01	17.33	20.81	0.00
Jonagold	0	2	3	0	5	1:01:01	1.67	2.80	0.25
Limoncella	5	3	2	4	9	1:01:01	3.00	0.67	0.72
M014.05	5	9	0	6	15	1:01:01	5.00	8.40	0.01
M015.01 red	21	8	5	2	15	1:01:01	5.00	3.60	0.17
M018.04	4	27	1	23	51	1:01:01	17.00	23.06	0.00
McIntosh	3	6	2	2	10	1:01:01	3.33	3.20	0.20
Permain dorée	0	29	1	25	55	1:01:01	18.33	25.02	0.00
Red chief	31	17	23	25	65	1:01:01	21.67	1.60	0.45
Rome Beauty	3	6	5	3	14		4.67	1.00	0.61
Rosa mantovana	4	3	4	4	11		3.67	0.18	0.91
Royal gala	18	12	10	5	27		9.00	2.89	0.24
Stark Splendour	11	9	6	14	29		9.67	3.38	0.18
Total	227	321	169	308	798				

- The analysis of segregation has been carried out only for R genes for which associated markers were available.
- Several seed parents are missing in the cross combinations with the different R parents.
- In all cross combinations, gametes carrying no resistance gene were excluded from the segregation analysis, because the cross populations were phenotypically screened before the molecular screening and susceptible plants were discarded. See Material & Methods for details.
- 'Expected' stays for expected frequency for each gamete, excluded that one that does not carry any R locus. The analysis of segregation was possible because R loci were carried only by R parents that were heterozygous at all R loci, while seed parents did not carry any resistance, except 'Di Corone' that carried the Rvi6 gene and for which the segregation analysis was a little bit different.
- 'Di Corone', was initially included among the susceptible seed parents. During the experiment it was identified as synonymous of 'Prima', a selection from the PRI program and indeed carried the Rvi6 (=Vf) gene. The segregation analysis was carried out accordingly.