Sugarbeet Root Maggot Resistance from a Red Globe-shaped Beet (PI 179180)

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ABSTRACT

Sugarbeet root maggot is a major insect pest of sugarbeet in many North American production areas. Chemical insecticides are the primary control method. Host-plant resistance that provides consistent reliable control would provide both an economical and an environmentally favorable alternative to complete dependence on chemical insecticides. This report describes a sugarbeet root maggot-resistant germplasm line, F1043, that is not related to any previously released resistant germplasm lines. F1043 was selected from a cross between a root maggot-susceptible sugarbeet germplasm line and PI 179180, a sugarbeet root maggot-resistant accession with red globe-shaped roots that was originally collected in Turkey. Differences in sugarbeet root maggot damage ratings between F1043 and two previously released resistant lines, F1016 and F1024, were small. The three-year average sucrose concentration of F1043 in the absence of sugarbeet root maggot damage was 12 g kg^{-1} less than the sucrose concentration of a commercial hybrid (136 g kg^{-1}). The average root yield of F1043 was 80% of the root yield of F1016. In all comparisons between F1043 and F1024, F1024 had lower Cercospora leaf spot severity ratings than F1043. There is no indication that F1043 would contribute resistance to rhizomania or curly top when used to introduce sugarbeet root maggot resistance into elite populations or parental lines.

Additional Key Words: *Beta vulgaris*, host-plant resistance, insect resistance, *Tetanops myopaeformis*.

The sugarbeet root maggot (*Tetanops myopaeformis* von Röder), indigenous to North America, is a significant economic pest on two-thirds of the sugarbeet (*Beta vulgaris* L. ssp *vulgaris*) acreage in the United States. Yield reductions sometimes are the result of stand loss early in the season, but more frequently are due to larval feeding on the root surface (Hein, et al., 2009). The primary control is chemical insecticides that reduce larval populations in sugarbeet fields (Campbell et al., 1998; Boetel et al., 2010; 2015). In 42 insecticide trials comparing the absence of insecticide with the most effective control in each trial, root yield reductions due to sugarbeet root maggot (SBRM) feeding ranged from 10% to 84% (Campbell et al., 1998). In these comparisons and a later report by Boetel et al, (2010), the average loss when no insecticide was compared to the most effective insecticide treatment was approximately 42%. No commercially viable alternatives to chemical insecticides are available to sugarbeet growers. SBRM-resistant hybrids would provide an environmentally sustainable option for inclusion in an integrated SBRM management program, and perhaps provide more consistent control than insecticides alone (Campbell, 2005).

Genetic variation for host-plant resistance to SBRM was first demonstrated when Callenbach et al. (1972; 1973) evaluated 340 lines from diverse *Beta* sources and identified 12 lines as potential sources of resistance. Theurer et al. (1982), using divergent selection for low and high damage, demonstrated genetic variation for resistance to SBRM feeding within sugarbeet germplasm. The first publically available sugarbeet germplasm line with SBRM resistance, F1015 (PI 605413), was released in 1996 (Campbell et al., 2000); followed by the release of F1016 (PI 608437) in 1998 (Campbell et al, 2000) and F1024 (PI 658654) in 2009 (Campbell et al., 2011). In a seedling bioassay, a SBRM larva feeding on the roots of susceptible seedlings was more prominent than on resistant seedlings (Smigocki et al., 2006). A serine protease inhibitor with a potential function in the mechanism of resistance was among the more than 150 SBRM-responsive genes identified in F1016 (Puthoff and Smigocki, 2006; Savić and Smigocki, 2012). Serine proteases have a functional role in the gut of SBRM larvae (Wilhite et al., 2000).

In two trials encompassing six environments, four SBRM-resistant pollinators (including F1015 and F1016), and five elite susceptible female (cms) lines, the yield loss attributed to SBRM feeding in hybrids with a resistant pollen parent was substantially less than the corresponding yield loss in commercial susceptible hybrids (Campbell and Niehaus, 2008; Campbell et al., 2008). In the trial that included only hybrids with F1015 as the pollinator, it appeared that the susceptible female (cms) used to produce a SBRM-resistant hybrid may influence the resistance level of the resulting hybrid (Campbell and Niehaus, 2008). Yield differences between testcross hybrids produced by crossing the eight component lines (half-sib families) of F1024 with a common susceptible cms line and susceptible commercial hybrids were small in trials at a site without SBRM, compared to the advantage of SBRM-resistant hybrids at a site with heavy SBRM pressure (Campbell et al., 2011).

This report describes the development and characteristics of a sugarbeet germplasm line, F1043 (PI 676971), with resistance to SBRM. The source of the resistance is a *Beta* accession, PI 179180, with red globe-shaped roots which is not related to previously released SBRM-resistant germplasm lines.

METHODS AND MATERIALS

F1043 was selected primarily for SBRM resistance from a cross between C564 (aa) and PI 179180. PI 179180, an accession with red globe-shaped (elliptic) roots (Fig. 1) that was identified as resistant by Callenbach et al. (1972; 1973), was originally collected near Gemlik, Turkey by Jack Harlan in 1948. C564 (aa) is a SBRM-susceptible, monogerm, self-fertile, O-type line segregating for Mendelian male sterility (PI 610317; Panella et al., 2015). After eight cycles of mass selection for SBRM resistance, 24 full-sib families were formed and subjected to four additional cycles of selection among and within families. F1043 is an increase of one full-sib family.

Selection for SBRM resistance was dependent upon natural infestations of SBRM at sites near St. Thomas, ND. SBRM feeding was assessed in late July or early August. Full-sib families were evaluated for SBRM

Figure 1. Roots of PI 179180, F1043, F1024 and a susceptible commercial hybrid, ACH-817, after infestation with sugarbeet root maggot, St. Thomas, ND.

resistance in replicated trials. The experimental design was a randomized complete block (RCB) with four or five replicates. Experimental units were two rows wide and 9 m long with 56 cm between rows. Five consecutive roots from the center of each of the two rows were hand-dug, washed, and rated on a 0 (no damage) to 9 (more than 75% of the root surface blackened by feeding scars) scale for SBRM damage (Campbell, 2005). Families with relatively low average damage ratings were identified and all remaining roots, except those on the ends of the rows, from all replicates of the selected families were hand-dug and washed. Roots with well-developed single taproots and the least SBRM damage were selected for advancement.

Root yield and sucrose concentration of the selected full-sib families was measured in the absence of SBRM at Fargo, ND in 2014, 2015, and 2016. The experimental design was a RCB with three replicates each year. Experimental units were 10 m long and two rows wide with 56 cm between rows. The trials were planted during the first two weeks of May, managed for optimum yield and quality throughout the growing season, and harvested in late September. Root yield was the fresh weight of all roots from a single plot at harvest expressed as Mg ha^{-1}. Sucrose concentration was determined by polarimetry, based upon a random sample comprised of 10 – 12 roots from each plot, and expressed as grams sucrose per kilogram fresh weight (g kg^{-1} = % sucrose x 10) . Two commercial hybrids, ACH-817 (Crystal Beet Seed, Moorhead, MN) and Beta-1301 (Betaseed, Shakopee, MN), a SBRM-susceptible germplasm line, F1010 (PI 535818; Campbell, 1990), and two SBRM-resistant germplasms, F1016 and F1024 were included for comparisons. The SAS GLM procedure (ver. 9.4, SAS Institute, Inc., Cary, NC) was used for the analysis variance. Fisher's Protected LSD was used to determine when differences among means were significant (P = 0.05).

The family that became F1043 was included in specialized nurseries to obtain an initial assessment of disease development when exposed to *Cercospora beticola* (Cercospora leaf spot), *Fusarium* spp. (Fusarium root rot), *Aphanomyces cochlioides* (Aphanomyces root rot), *Beet necrotic yellow vein virus* (BNYVV; rhizomania) and *Beet severe curly top virus* (curly top). The Cercospora leaf spot nurseries were near East Lansing, MI (USDA-ARS, East Lansing, MI) in 2015 and near Rosemount, MN (Betaseed, Inc, Shakopee, MN) in 2015 and 2016. Aphanomyces root rot and Fusarium root rot were evaluated in nurseries near Shakopee, and Sabin, MN (Betaseed, Inc.), respectively in 2015. Fusarium root rot ratings were based upon the severity of foliar symptoms; however, occasional roots examined exhibited corresponding typical root rot symptoms. The rhizomania and curly top evaluations were conducted by USDA-ARS, Kimberly, ID in 2015 and 2016. These nurseries were located and managed with the objective of providing a reliable indication of the response to a single disease organism with minimal interference from other diseases. Each nursery included entries from other breeding programs and representative resistant and susceptible cultivars selected by the nursery managers. Sugarbeet root aphid (*Pemphigus* sp.) damage was assessed by Betaseed, Inc. in a greenhouse assay in 2015. The root aphid trials were not randomized, so statistical analysis was not appropriate. However, comparisons between lines and with checks provide insight into the relative performance of lines when challenged by sugarbeet root aphid (Panella et al., 2008).

RESULTS

F1043 is a multigerm diploid line. The root surface of F1043 often is slightly darker than that of F1024 and most sugarbeet lines (Fig. 1). In some environments, a light pink or rose color pigment is apparent, often on the upper portion of the root surface below the crown. The pigmentation affects only the root surface. The flesh immediately below any pigmented area is white, similar to F1024 and most other sugarbeet cultivars. Plants with seed stalks (bolters) were observed in all selection-cycles but were never selected as mother roots to produce seed for the next generation. However, approximately 6% and 2% of the plants in the 2015 and 2016 Fargo yield trials produced seed stalks, suggesting that complete elimination of bolters may be difficult. Thirty percent of the PI 179180 plants produced seed stalks in the 2016 Fargo trial; no bolters were observed in F1016 or F1024 in either year. Approximately 20% of the hypocotyls of F1043 seedlings are green, with the remaining 80% red.

Sugarbeet Root Maggot Resistance

Differences in SBRM damage ratings between F1043 and the two previously released SBRM-resistant lines, F1016 and F1024, were small and not significant in any of the three years or when averaged over the three years (Table 1). In all cases, the damage ratings for F1043 were lower than the damage ratings for F1010 and the two susceptible commercial hybrids, ACH-817 and Beta-1301. The average SBRM damage rating of the three susceptible cultivars (ACH-817, Beta-1301, and F1010) was 6.8, compared to a damage rating of 2.9 for F1043; a difference of 3.9 ($CI_{0.90}$ = 3.4 – 4.3). A damage rating of three corresponds to more than ten scattered small scars or up to three large scars. In contrast, a damage rating of seven or above indicates that at least one-fourth of the root surface is blackened by SBRM feeding scars.

Root yield and Sucrose Concentration

The 3-year average sucrose concentration of F1043 was 12 g kg^{-1} less than the sucrose concentration of the commercial hybrid with the highest sucrose concentration, ACH-817 (Table 1). The difference between F1043 and the other commercial hybrid, Beta-1301, was small and not significant. The difference between the 3-year average sucrose concentration of F1043 and the average of the three SBRM-susceptible cultivars was 7.2 g kg^{-1} ($CI_{0.90}$ = 2.2 – 12.2 g kg^{-1}). The small differences in sucrose concentration among the three SBRM-resistant germplasm lines (F1043,

Table 1. Sugarbeet root maggot damage ratings, St. Thomas, ND, and sucrose concentration and root yield of three root maggot resistant germplasm lines (F1043, F1016, and F1024), two susceptible commercial hybrids (ACH-817 and Beta-1301) and a susceptible germplasm line (F1010), Fargo, ND, 2014 – 2016.

	Year			
Cultivar	**2014**	**2015**	**2016**	**Mean**
	-----Sugarbeet root maggot damage (0 – 9)[†] -----			
F1043	2.3 c[‡]	3.8 b	2.6 c	2.9 C
F1016	2.4 c	3.2 b	3.2 c	2.9 C
F1024	2.8 c	3.5 b	2.8 c	3.0 C
ACH-817	6.5 a	8.1 a	8.2 a	7.6 A
Beta-1301	4.5 b	6.9 a	6.7 b	6.0 B
F1010	5.3 ab	7.3 a	7.5 ab	6.7 B
Mean	4.0 B	5.5 A	5.2 A	4.9
	-------------------- Sucrose (g kg^{-1}) ----------------------			
F1043	109 a	123 a-c	140 b	124 BC
F1016	109 a	122 a-c	141 b	124 BC
F1024	116 a	112 c	138 b	122 C
ACH-817	124 a	129 ab	155 a	136 A
Beta-1301	119 a	118 bc	144 ab	127 BC
F1010	119 a	131 a	144 ab	131 AB
Mean	116 C	122 B	144 A	127
	-------------------- Root yield (Mg ha^{-1}) ------------------			
F1043	29.7 bc	22.0 c	21.0 d	23.2 E
F1016	27.7 c	26.2 c	33.3 c	29.1 D
F1024	25.1 c	28.4 c	23.2 d	25.6 DE
ACH-817	46.0 a	59.6 a	53.1 b	52.9 B
Beta-1301	47.4 a	70.2 a	65.9 a	61.2 A
F1010	36.2 b	48.5 b	34.3 c	39.6 C
Mean	35.4 B	42.5 A	38.4 B	38.8

[†]Damage rating: 0, no feeding scars; 1, one to four small scars; 2, five to ten small scars; 3, up to three large scars or numerous small scars; 4, a few large scars or considerable feeding on lateral roots; 5, several large scars and/or extensive feeding on lateral roots; 6, numerous scars, up to 25% of root blackened; 7, 25 – 50% of root blackened with scars; 8, 50 – 75% of root blackened; and 9, > 75% blackened.

[‡]Differences among means within a year followed by the same lower case letter are not significant, according to Fisher's Protected $LSD_{0.05}$; differences among main effect means followed by the same upper case letter are not significant (P = 0.05).

Table 2. Disease and insect damage indices in nurseries to evaluate response of F1043 and F1024 to Aphanomyces root rot, Fusarium root rot, Cercospora leaf spot, Curly Top virus, rhizomania, and sugarbeet root aphid.

Disease/insect	Year	Observation	F1043	F1024	Resistant check	Moderate check	Susceptible check	$LSD_{0.05}$
			(0 - 9)†					
Aphanomyces	2015	Foliar	4.5	5.3	1.7	2.8	5.3	1.4
		Root	4.8	5.5	1.0	4.2	6.3	1.1
		Mean	4.7	5.4	1.3	3.5	5.8	1.1
Fusarium	2015	Early	2.7	2.3	2.2	2.8	3.7	0.7
		Late	5.5	2.8	2.7	5.0	7.8	0.7
		Mean	4.1	2.6	2.4	3.9	5.8	0.5
Cercospora	2015(MN)‡	Last§	7.5	5.3	1.7	8.7	9.0	1.5
		Mean (4)	4.0	2.6	1.3	4.7	8.6	0.7
	2015(MI)	Last	7.0	4.0	3.0	- - - -	7.7	0.8
		Mean (2)	4.6	2.7	2.0	- - - -	5.8	0.7
	2016(MN)	Last	7.0	4.3	1.0	8	9.0	1.3
		Mean (5)	4.5	2.7	1.2	5.1	7.8	0.8
Curly Top	2015	Foliar	6.5	5.9	3.7	- - - -	7.0	0.9
	2016	Foliar	4.0	5.1	2.5	- - - -	6.6	0.6
			(0 - 100)					
Rhizomania	2015	Root	39	33	10	15	36	10
	2016	Root	46	35	18	22	46	6
			(0 - 4)					
Root aphid	2015	Root	3.1	2.6	1.2	- - - -	3.9	- - - -

†Higher indices are indicative of increased severity in all cases.
‡MN indicates evaluations were conducted by Betaseed, Inc. in southern Minnesota; MI refers to trial conducted by USDA-ARS, East Lansing, Michigan.
§The last Cercospora leaf spot rating is almost always the highest (most severe) recorded for the season; the number in parenthesis indicates the number of observation dates included in the mean.

F1016, and F1024) were not significant (P = 0.05).

The 2016 and 3-year average root yields of F1043 were lower than the corresponding root yields of F1016 (Table 1). The difference between F1043 and F1024 was not significant in any of the three years or for the 3-year average root yield. The average root yield of F1043 was 80% of the root yield of F1016 and 60% of F1010. The average root yield of the two commercial hybrids was approximately 2.5 times the root yield of F1043.

Disease Resistance

All Cercospora leaf spot (CLS) severity ratings for F1043 were greater than the ratings for the resistant check (Table 2). In all but the 2015 observations from Michigan, the CLS severity ratings for F1043 were lower than the ratings for the susceptible check (P = 0.05). In all comparisons between F1043 and F1024, the lower CLS rating for F1024 was significant at the P < 0.05 level. Based upon observations from a single trial, it appears that F1043 is susceptible to Fusarium root rot (Table 2). In contrast, the late observation and mean severity ratings for F1024 suggest that F1024 has a useful level of resistance to Fusarium root rot.

There is no indication that F1043, or F1024, would contribute significant resistance to Aphanomyces root rot, rhizomania, or curly top when used to introduce SBRM resistance into elite populations or parental lines (Table 2). Furthermore, the SBRM resistance of F1043 or F1024 does not appear to be accompanied by resistance to sugarbeet root aphid.

DISCUSSION

The SBRM resistance of F1043 is equal to the resistance of F1016 and F1024. F1043 is not related to these previously released SBRM-resistant germplasm lines and as such, may have unique genes for resistance and other traits that could impact combining ability with elite populations and parental lines. The successful transfer of resistance from an accession with red roots to sugarbeet, indicates that the resistance of PI 179180 was not associated with its red pigment and suggests the resistance of PI 181718 (Campbell, 2005), another accession with red roots that has resistance to SBRM, and future accessions with red roots identified as resistant, should be considered as potential sources for production of SBRM-resistant sugarbeet.

F1043 will be maintained by USDA-ARS, Fargo, North Dakota and freely distributed in quantities sufficient for reproduction. Requests for seed should be directed to the USDA-ARS, Sugarbeet and Potato Research Unit, 1605 Albrecht Blvd. N., Fargo, ND 58102. Seed also has been deposited with the USDA National Germplasm System where it will be available for research purposes, including the development of new varieties. Plant Variety Protection will not be pursued for F1043.

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