Registration of the Maize Germplasm CRW3(S1)C6 with Resistance to Western Corn Rootworm

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Maize (*Zea mays* L.) germplasm CRW3(S1)C6 (Reg. No. GP-553, PI 644060) was developed with resistance to western corn rootworm (WCR; *Diabrotica virgifera virgifera* LeConte) by the USDA-ARS Plant Genetics Research Unit in cooperation with the Missouri Agricultural Experiment Station at the University of Missouri-Columbia, and was released in February 2007.

Development of CRW3 began in the 1996–1997 winter nursery when crosses for a diallel of nine parents were made for the purpose of evaluating combining ability for WCR resistance (Hibbard et al., 1999). The parents included a number of inbred lines and populations that showed less root pruning damage by WCR. The diallel crosses and their parents were evaluated for WCR damage in replicated trials in summer 1997. The six most resistant diallel crosses, which included only four of the nine original parents, were crossed as a second diallel in the 1997–1998 winter nursery. Parental materials of the second diallel included NGSDCRW1(S2)C4-15-2S2 × PI340839, TL92A-PAR1779 60-4 × PI340839, TL93A-PAR1774 28-1 × NGSDCRW1(S2)C4-15-2S2, TL92A-PAR1779 60-4 × NGSDCRW1(S2)C4-15-2S2, TL92A-PAR1779 60-4 × TL93A-PAR1774 28-1, and TL93A-PAR1774 28-1 × PI340839. NGSDCRW resulted from interplanting hybrid R802/R109B with open-pollinated SDCRW1SYN population and subsequent selection for large root size and root-pulling resistance. Lines beginning with TL are from the CIMMYT tropical breeding program. PI340839 is a Supergold popcorn from the J.C. Eldridge popcorn collection at Iowa State University. Other pedigree information for these parental materials can be found in Hibbard et al. (1999). A balanced bulk was created and recombined in the summer of 1998 to form CRW3 C0. For the first cycle of selection, approximately 1000 C0 plants were grown and self-pollinated in the 1998–1999 winter nursery. Progeny from the best 300 S_1 ears were evaluated in two replications for WCR damage in the summer of 1999. The 62 most resistant entries (selection intensity of 20%) were recombined from remnant seed in the 1999–2000 winter nursery to form CRW(S1)C1. For cycles 2, 3, and 4, 200 ears were evaluated for WCR resistance in two replications during the summer. Approximately 25 to 30 of the most resistant entries (selection intensity of 12.5–15%) were recombined from remnant seed to form the next cycle. Modifications were made in cycles 5 and 6, where 120 and 220 S_1 lines respectively were screened for WCR reaction. The 25 to 30 most resistant selections were determined before pollen shed and were recombined (bulk pollinated); pollinators were selected within the most resistant rows for lower ear height, flowering synchrony, and absence of excessive disease symptoms. The endemic nursery diseases were Northern corn leaf blight [incited by *Exserohilum turcicum* (Pass.) Leonard & Suggs], common smut [*Ustilago maydis* (DC) Corda], and often Stewart's Wilt (incited by *Pantoea stewartii* Smith).

For each cycle of selection, WCR root damage was evaluated using artificially infested plots in a randomized complete block design, with two replications at a single location. Initially, all plots were hand planted in 1.5-m rows in a randomized complete block design at a planting density of 86,111 plants ha^{-1}, and thinned to a density of 57,408 plants ha^{-1}. Later, a specialized planter was built and plots were lengthened to 1.8 m for a planting density of 53,820 plants ha^{-1} without subsequent thinning. Eggs of the WCR were provided by the USDA-ARS Northern Grain Insects Research Laboratory, Brookings, SD. Eggs were suspended in 0.15% agar solution and were used to mechanically infest plots (Moellenbeck et al., 1994) at the V2 stage of plant development, with approximately 1000 eggs per 30.5 cm of row. Before root excavation, nylon cable ties were used to attach laminated barcodes containing plot information to four roots per plot. When a sufficient number of plants was present, the four plants were not consecutive nor end plants in the plot. Roots were excavated after most WCR feeding was completed (when approximately half of the rootworms from check plots were at the pupal stage) using a specially designed, tractor-mounted implement (Praiswater et al., 1997). Roots were soaked, washed, and rated for damage using the 0 to 3 scale described in Oleson et al. (2005). The average damage rating from four roots per plot was considered one replication.

In 2002 freshly increased seed from random matings within each of CRW3 C0, C1, C2, and C3 was evaluated in replicated

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Table 1. Progress of selection for western corn root worm resistance in CRW3 when averaged across six locations.

Entry	Damage rating (number of nodes pruned)
P34G81† with insecticide treatment	0.31 c‡
CRW3(S1)C3	0.47 bc
CRW3(S1)C2	0.64 b
CRW3(S1)C1	0.70 b
CRW3(S1)C0	0.72 b
P34G81†	1.31 a

†Susceptible Pioneer hybrid check.

‡Means followed by the same letter are not significantly different (LSD, $P < 0.05$).

trials at four Missouri locations and one location each in Illinois, Nebraska, and Iowa, with three replications each. Included in the experiment were two entries of a susceptible commercial Pioneer hybrid, P34G81, with and without the granular insecticide tefluthrin. Evaluations at the Missouri locations were conducted as described above, while evaluations at the other locations were conducted using natural infestations of corn rootworm eggs in trap crop situations. At the Iowa location, damage to P34G81 averaged more than 2.5 nodes of roots pruned. Resistance of CRW3(S1)C6 did not hold up under this level of rootworm pressure. The average damage to P34G81 for the remaining six locations was 1.31 without insecticide and 0.31 with insecticide (Table 1). When excluding the Iowa data, damage to CRW3(C3) was not significantly different ($P > 0.05$) than the insecticide-treated check and was significantly less than both the nontreated check and CRW3(C0).

In 2006 topcrosses of CRW3(S1)C6 with two elite non-stiff stalk (NSS) and two elite stiff stalk (SS) inbred testers from AgReliant were evaluated in South Dakota, Illinois, and two Missouri locations, with three replications each (Table 2). The average damage ratings of the NSS topcrosses were significantly lower ($P < 0.05$) than the average damage to the susceptible hybrid check, B37 × H84. The damage rating for either NSS topcross was not significantly different than the damage rating of either B37 × H84 treated with tefluthrin or a transgenic CRW-resistant Monsanto hybrid. Damage ratings for the SS topcrosses averaged 0.50 and 0.51, both of which were significantly lower than the B37 × H84 check and not significantly different from the check treated with tefluthrin but significantly higher than the transgenic check.

The CRW3 population at cycle 6 is segregating for kernel color with 11.4% white, 16.4% purple, and the remaining 72.2% various shades of yellow. The population is also segregating for endosperm type, including dent, flint, and small-seeded types. In a nonirrigated site in Missouri in 2005, CRW3(S1)C6 flowered 62 d after planting, and the average ear and plant heights were

Table 2. Performance of CRW3(S1)C6 testcrosses with proprietary stiff stalk (SS) and non-stiff stalk (NSS) testers averaged across four locations.

Entry	Damage rating (number of nodes pruned)
transgenic Monsanto hybrid†	0.11 c‡
CRW3(S1)C6 NSS topcross 1§	0.19 bc
CRW3(S1)C6 NSS topcross 2	0.29 bc
B37 × H84¶ with insecticide treatment	0.38 bc
CRW3(S1)C6 SS topcross 1	0.50 b
CRW3(S1)C6 SS topcross 2	0.51 b
B37 × H84¶	1.84 a

† Transgenic rootworm-resistance from the Mon863 event.

‡Means followed by the same letter are not significantly different (LSD, $P < 0.05$).

§Topcrosses made with elite proprietary inbred testers.

¶Susceptible check hybrid.

80 and 175 cm, respectively. Because this is still a segregating population, most other traits do not have consistent values.

Seed from the C6 generation of CRW3 is available in lots of 250 kernels and may be obtained for five years from the Plant Genetics Research Unit, USDA-ARS, 205 Curtis Hall, University of Missouri, Columbia, MO 65211. Seed may be obtained from the North Central Regional Plant Introduction Station thereafter. We ask that appropriate recognition be given when this germplasm contributes to research or to a new cultivar.

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