QTLs Associated with Resistance in Soybean PI567516C to Synthetic Nematode Population Infecting cv. Hartwig

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Abstract

Worldwide, soybean cyst nematode (SCN, Heterodera glycines Ichinohe) is the most destructive pathogen of soybean [Glycine max (L.) Merr.]. Crop losses are primarily mitigated by the use of resistant cultivars. Nematode populations are variable and have adapted to reproduce on resistant cultivars over time because resistance primarily traces to two soybean accessions, Plant Introduction (PI) 88788 and Peking. Soybean cultivar Hartwig, derived primarily from PI437654, was released for its comprehensive resistance to most SCN populations. A synthetic nematode population (LY1) was recently selected for its reproduction on Hartwig. The LY1 nematode population currently infects known sources of resistance except soybean PI567516C; however, the resistance to LY1 has not been characterized. The objective of this study was to identify quantitative trait loci (QTLs) underlying resistance to the LY1 SCN population in PI567516C, identify diagnostic DNA markers for the LY1 resistance, and confirm their utility for marker-assisted selection (MAS). Resistant soybean line PI567516C was crossed to susceptible cultivar Hartwig to generate 105 recombinant inbred lines (F2-derived F5 families). QTLs were mapped using simple sequence repeats (SSRs) covering 20 Linkage Groups (LGs) and three diagnostic markers, Satt592, Satt331, and Sat_274, were identified on LG O. These markers have a combined efficacy of 90% in identifying resistant lines in a second cross that has been generated by crossing a susceptible cultivar 5601T with resistant PI567516C. F2-derived F4 segregating population was used in MAS to identify resistant lines.

Key words: bioassay, breeding, cyst nematode, Glycine max, Heterodera glycines, mapping, marker assisted selection, resistance, soybean

Introduction

Worldwide, soybean [Glycine max (L.) Merr.], grown for its edible protein and oil, is a very important agronomic crop. Soybean cyst nematode, Heterodera glycines Ichinohe (SCN) reduces yield more than any other soybean pest in the world (Wrather et al. 2001). SCN causes yield reductions by feeding on plant nutrients, retarding root growth, and inhibiting Bradyrhizobium nodulation (Arelli et al. 2009).

Resistant cultivars have been an effective means of control. Soybean breeders have been evaluating the soybean germplasm collection for sources of resistance. Most evaluations for nematode resistance included Races 3, 5, and 14 corresponding to HG Types 0-, 2.5-, 1.3-, respectively (Niblack et al. 2002). A total of 118 resistant PI (Plant Introduction) lines were identified with resistance to one or more of these Races (Arelli et al. 1997). These 118 accessions were screened for resistance to Races 1 and 2 corresponding to HG Types 2.5- and 1.2.5-, respectively (Arelli et al. 1997, 2000). Although, more than 100 SCN resistance sources have been identified, nearly all resistant cultivars that have been developed in the USA have resistance genes from Peking and PI 88788 (Diers and Arelli 1999). Additionally, both soybean lines Peking and PI88788 were reported to have major resistance genes in common (Arelli and Anand 1988). Widespread use of these resistance sources has caused major shifts in nematode populations and nematodes have adapted to reproduce on resistant cultivars over time.

To provide a more comprehensive resistance to most nem-
tode populations, soybean cultivar Hartwig, primarily derived from PI437654, was released (Anand 1992). Only a few isolates of Race 4 population of SCN (corresponding to HG Type 1.2.3.5.6.7) and a synthetic nematode population, LY1 could reproduce on Hartwig. Among known sources of resistance to nematode populations, soybean PI 567516C was the only available resistance source against the LY1 nematode population (Young 1999). Most recently, Chen et al. (2006) using a cluster analysis determined that PI567516C and Hartwig are genetically unrelated.

Understanding the genetic basis of resistance to the LY1 nematode population is important for the development of SCN-resistant cultivars. The identification, map localization, and characterization of genetic factors underlying resistance to SCN are crucial in the development of diagnostic and robust molecular markers for marker-assisted selection (MAS). To date, several sources of resistance have been identified but only PI88788 is the most widely deployed source in more than 95% of all commercial SCN-resistant cultivars produced with Peking as distant second source deployed under 5%. The soybean accession, PI437654, is known to have the broadest spectrum of control against most nematode populations, but is deployed only in a handful of commercial varieties. Recent reports have brought to attention the discovery of highly aggressive populations of SCN that can develop on varieties with resistance derived from PI567516C and Hartwig, and even PI437654.

To protect soybean from the imminent threat of newly-emerging highly aggressive populations of SCN such as LY1, it is imperative to introgress resistance genes from PI567516C into commercial varieties. However, incorporating resistance genes from unadapted germplasm like PI567516C is not without problems. Conventional breeding strategies have to deal with unwanted deleterious linkages often associated with unadapted germplasm. Marker assisted selection could facilitate developing resistant germplasm to the LY1 nematode population with a minimum of linkage drag. Quantitative trait loci (QTLs) have been identified using molecular markers for resistance to several SCN populations in a total of 13 accessions (nine resistance sources). They are located on all linkage groups (LGs) except for D1b, K, and O (Concibido et al. 2004). The QTLs on LGs G and A2 have been studied in detail and molecular markers have been saturated around them. However, resistance in PI567516C for the LY1 nematode population has not been mapped. The objective of this study was to identify QTLs associated with resistance to the LY1 nematode population from PI567516C and assess their potential for marker-assisted selection.

Materials and Methods

Plant materials

Seeds of soybean PI567516C were obtained from Soybean Germplasm Collection, USDA-ARS, Urbana-Champaign, IL (courtesy of Dr. Randall Nelson, Soybean Curator) for making crosses. PI567516C is a maturity group IV soybean, with purple flowers, grey pubescence, greenish yellow seed and having resistance to nematode Races 1, 3, and moderate resistance to Race 2, corresponding to HG Types 2.5.-, 0.-, 1.2.5.-, respectively (Arelli et al. 1997; Young 1999; Niblack et al. 2002). This line was crossed with Hartwig, a soybean cultivar that is susceptible to LY1 in 1999 to generate segregating progenies for genotyping and phenotyping. A total of 105 recombinant inbred lines (F2 families) were used in the evaluations. Soybean PI567516C was also crossed with another susceptible cultivar 5601T (Pantalone et al. 2003) to generate a second population for confirming QTLs associated with resistance to the LY1 nematodes. Several F2 progenies in the second population were advanced to F3 and especially, yellow seeded and agronomically desirable progenies were selected by marker-assisted selection.

Bioassays for reaction to the LY1 nematode population included a modified method of greenhouse evaluation followed an established protocol (Arelli et al. 2000; Arelli and Wang 2008). A set of differentials or indicator lines were included in each evaluation to characterize the nematode population. These indicator lines were PI548402 (Peking), PI88788, PI90763, PI437654, PI209332, PI89772, PI548315 (Cloud), and a susceptible control PI548658 (cv. Lee 74).

Nematode culture

The LY1 nematode population was developed from a mass mating of SCN Race 2 (HG Type 1.2.5-) females with Race 5 (HG Type 2.5-) males (Young 1998). Eggs resulting from the cross were cultured on soybean Hartwig and then on susceptible cultivar Hutcheson (Buss et al. 1988) in alternate generations for 14 cycles, followed by 13 continuous generations of reproduction. A Female Index (FI) was calculated (Golden et al. 1970; Riggs and Schmitt 1988; Schmitt and Shannon 1992; Niblack et al. 2002). After approximately, 26 generations (ca. 30 d each generation) of continuous reproduction and selection of nematode population on Hartwig the female index was 34%, which is a moderately susceptible reaction. The selection process further continued on Hartwig for several more generations until the resulting female index was nearly 100% (susceptible reaction). The resulting synthetic nematode population was called the LY1 and maintained on Hutcheson (Young 1998; Arelli et al. 2009) and was used in this study.

Nematode bioassay

The plants of each recombinant inbred line (F2.5 family) (Hartwig x PI567516C) and indicator lines were grown in 10 cm clay pots (2 - 3 seeds per 7.5-cm-wide pot) filled with silt loam soil and the test was completely randomized. Ten seedlings were grown from each RIL (F2:5 family) except for D1b, K, and O (Concibido et al. 2004). The QTLs on LGs G and A2 have been studied in detail and molecular markers have been saturated around them. However, resistance in PI567516C for the LY1 nematode population has not been mapped. The objective of this study was to identify QTLs associated with resistance to the LY1 nematode population from PI567516C and assess their potential for marker-assisted selection.