

INHERITANCE OF RESISTANCE TO *BEEET CURLY TOP VIRUS* IN THE G122 COMMON BEAN LANDRACE

Richard Larsen¹, Phillip N. Miklas¹, and Chet Kurowski²

¹USDA-ARS, Prosser, WA; ²Harris Moran Seed Company, Davis, CA

The curtoviruses *Beet curly top virus* (BCTV), *Beet mild curly top virus*, and *Beet severe curly top virus*, commonly referred to as 'curly top virus' (CTV), are serious virus diseases of common bean in the semiarid regions of the western U.S. The three viruses are vectored in a persistent manner by the beet leafhopper *Circulifer tenellus*. The only effective control of CTV in bean is genetic resistance. We recently reported a SCAR marker (SAS8.1550) directly linked to the dominant *Bct* resistance gene that confers resistance to CTV (Larsen and Miklas, 2004). The marker is located on linkage group B7. During screening procedures to evaluate the robustness of SAS8.1550 for MAS purposes, the landrace Jatu Rong (G122) from India was the only Andean genotype with resistance to CTV that lacked the marker. The objective of this research was to determine whether G122 possesses novel resistance to the curly top viruses.

MATERIALS AND METHODS

Two populations of F_{5:7} Recombinant Inbred Lines (RILs) were derived from separate F₁ seeds from a cross between G122 x Taylor Horticultural (CTV-susceptible). The populations were comprised of 98 RILs in total. Approximately 40 seed per RIL were planted in rows 3 meters in length in a randomized complete block design with three reps over two years at Prosser, WA in 2006 and 2007. Disease incidence based on the number of infected plants within a single-row-plot was used to measure phenotypic response. Because none of the plants expressed intermediate reactions to infection with CTV, individuals were rated either resistant or susceptible. Presence or absence of CTV in select plants was verified by ELISA or polymerase chain reaction (PCR).

The RIL population was also planted in the greenhouse for use in DNA extractions. DNAs bulked from six CTV-resistant and six CTV-susceptible RILs, respectively, were extracted from bean plants at the first trifoliolate stage using FastDNA spin columns (Q-Biogene, Irvine, CA). After adjusting DNA concentrations to 0 ng/μl, random decamer primers (Operon Technologies, Inc. Alameda, CA) were screened for RAPD DNA markers detected as amplified fragments present in one bulk but absent in the other as viewed on agarose gels. RAPDs detected between resistant and susceptible bulk DNAs within populations and verified for cosegregation among individuals comprising the bulks were then assayed across the entire population of 98 RILs. QTL were identified by regression of a marker on disease incidence phenotype using single factor ANOVA (PROC GLM in SAS). A probability level of <0.05 was used as a significance level to declare presence of a QTL. JoinMap 4.0 was used to construct linkage maps.

RESULTS AND DISCUSSION

Infection pressure was uniform across both years as indicated by the high level of disease incidence for the susceptible parent Taylor Horticultural. The mean CTV incidence within 40 plants of the susceptible parent was 8.8 and 11.8 for 2006 and 2007, respectively. Population A (52 RILs) exhibited greater susceptibility (\bar{X} = 4.3 and 4.5 in 2006 and 2007, respectively) than Population B (46 RILs) (\bar{X} = 2.8 and 2.7 in 2006 and 2007, respectively), the reason for which is not immediately clear. G122 landrace may be heterogeneous for minor genes that cause slight differences in reaction

to CTV. We identified three dominant RAPD markers, Q14.925, R15.460, and S11.625, derived from G122 that were completely linked with each other. Genetic analysis of the 98 F_{5:7} RILs revealed that the markers were associated with a major effect QTL that exhibited stable expression across both years and populations (Table 1). The phenotypic variation explained by the QTL in Population A (43.8%) was greater than in Population B (21.9%). Markers Q14.925 and R15.460 also were polymorphic in the BJ core mapping population enabling integration of the QTL to linkage group B6. The *bc-3* gene which conditions resistance to *Bean common mosaic virus* is also located on the B6 linkage group but is not closely linked with the QTL described here. Three additional linked RAPD markers I10.520, S11.580 and K9.925 detected a QTL with minor effect. Expression of this QTL was detected both years, but only in Population A (Table 1). The linkage group associated with this QTL has not yet been identified. Multiple regression analysis indicated these QTL had an additive effect. Additional analysis confirmed that SCAR SAS.1550 for the *Bct* gene located on linkage group B7 was not present in G122. An F₂ from a cross between G122 x Cardinal (which possesses *Bct*) segregated for susceptibility to CTV (data not shown). These findings, combined with the location of the major QTL on B6, indicate that G122 possesses novel resistance to CTV. This is the first report of a QTL for resistance to CTV.

Table 1. Identification (by one-way ANOVA) of two independent QTL conditioning resistance to BCTV as measured by disease incidence across two field trials and two populations, A=52 RILs and B=46 RILs, derived from separate F1 from G122/Taylor Horticulture cross.

| RIL population | Linkage group | RAPD marker | Linkage distance (cM) | % phenotypic variation - explained disease incidence (R^2) | |
|----------------|---------------|-------------|-----------------------|--|------------------------|
| | | | | 2006 | 2007 |
| A | B6 | Q14.950 | 0 | 40.3 ($P<0.0001$) | 47.3 ($P<0.0001$) |
| | | R15.460 | 0 | | |
| | | S11.625 | 0 | | |
| | unknown | I10.520 | 0 | 10.2 ($P<0.022$) | 7.8 ($P<0.047$) |
| | | S11.580 | 1.9 | 14.6 ($P<0.005$) | 11.8 ($P<0.013$) |
| | | K9.925 | 5.2 | 11.3 ($P<0.019$) | 8.2 ($P<0.049$) |
| B | B6 | Q14.950 | 0 | 26.4 ($P<0.0003$) | 17.3 ($P<0.004$) |
| | | R15.460 | 0 | | |
| | | S11.625 | 0 | | |
| | unknown | I10.520 | 0 | 3.9 (ns) | 7.8 (ns) |
| | | S11.580 | 1.9 | 5.8 (ns) | 6.8 (ns) |
| | | K9.925 | 5.2 | ns | ns |

Multiple regression of markers S11.625 + S11.580 was equal to 49% ($P<0.006$) and 32% ($P<0.07$) for populations A and B, respectively, for 2006 and 53% ($P<0.015$) and 24% ($P<0.06$) for populations A and B, respectively, for 2007.

ns - denotes non-significant values.

REFERENCES

Larsen, R.C., and Miklas, P.N. 2004. Generation and molecular mapping of a sequence characterized amplified region marker linked with the *Bct* gene for resistance to *Beet curly top virus* in common bean. *Phytopathology* 94:320-325.