

T1329, 43 cM, Chromosome 7

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July 11, 2008

Introduction

The goal for this research was to develop a co-dominant CAPS or SCAR marker for the *I3* gene, which corresponds to resistance to *Fusarium oxysporum* f. sp. *lycopersici* race 3. PCR primers for markers in the chromosomal region between the molecular markers TG183 (42 cM) and TG639 (43.3 cM) (Hemming et al., 2004) were evaluated on homozygous susceptible and homozygous resistant tomato inbred lines as well as heterozygous F1 hybrids obtained from J. W. Scott, University of Florida, R. Gardner, North Carolina State University or commercial hybrids.

Reference:

Hemming, M. N., S. Basuki, D. J. McGrath, B. J. Carroll, and D. A. Jones. 2004. Fine mapping of the tomato *I-3* gene for Fusarium wilt resistance and elimination of a co-segregating resistance gene analogue as a candidate for *I-3*. *Theor. Appl. Gen.* 109:409-418.

Primers

The sequence of T1329 (3,498 nt for SGN-U312610) was compared with sequences at GenBank. Putative exons were identified by the match with *Vitis vinifera*, AM439610, a forward and a reverse primers were designed from the putative exon regions to amplify a genomic region, which should include at least one intron. The primer pair listed below gave the strongest single PCR fragment and the best sequence.

Table 1. PCR primers on chromosome 7

Primer Name	Primer Sequence (5'-3')
P7-43AF2	GGTACTTTCGCCAATATCCGCCTTGT
P7-43AR2	GTCAATCCAAGTGTATCTGCATCCTC

*PCR annealing temperature at 53°C (TGEN53), ~510-bp fragment.

Sequences

Purple Russian, i3/i3 (Partial Sequence); Purple Russian is a heritage tomato (OP) from Seed Savers Exchange, Decorah, Iowa

GENBANK ACCESSION NUMBER: EU920052

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1      GGTACTTTCG CCAATATCCG CCTTGTAAC AAGCTGTAA ACGGGGAGGT TGGTCCAAAG
61     ACGGTCCACA TTCCCTCTGG AGAAAAGCTA TCTGTGTTG ATGCTGCTAT GGTACTTTCC
121    TTTCTCTGTT TCTTAATGGT TCAAATCCTG TTTTCATTAA AGATAAGAGT TTGCATTGCG
181    CATTTCACTT TTGCTGTTTT ATGTTGACAC TTGTTAAACA CGTGATGCCT TCAGAAATAC
241    AAATCCGCTG GACAAAAGCAC TATTATCTTG GCTGGAGCTG AATATGGAAG TGAAGACTCC
301    CGAGATTGGG CTGCCAAGGG ACCAATGTTG TTGGTGGGTT TTCTTGTGCA TTTATTCTTA
361    ATTTGGTTGA TCCTGTCCTT TTCTTTTTTA CCAGAGGATT CCCTAACTAC TAAAGAACAA
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421 TTTAACAGGG AGTTAAAGCT GTAATTGCTA AGAGCTTCGA GAGGATTCAT CGCAGTAACT
 481 TGGTAGGAAT GGGTATTGTG CCTCTATGTT TTAAGGCTGG GGAGGATGCA GATACACTTG
 541 GATTGAC

NC EBR-8, I3/I3 (Partial Sequence), inbred from R. Gardner

GENBANK ACCESSION NUMBER: EU926650

1 GACTTCAACT CATATGGTAG CCGTCGTGGT AATGATGAGA TTATGGCTAG GGGTACTTTC
 61 GCCAATATCC GCCTTGTAAC CAAGCTGTTA AACGGGGAGG TTGGTCCAAA GACGGTCCAC
 121 ATTCCTTCTG GAGAAAAGCT ATCTGTGTTT GATGCTGCTA TGGTACTTTC CTTTCTCTGT
 181 TTCTTAATGG TTCAAATCCT GTTTTCATTA AAGATAAGAG TTTGCATTGC GCATTTCACT
 241 TTTGCTGTTT TATGTTGACA CTTTTTAAAC ACGTGATGCC TACAGAAATA CAAATCTGCT
 301 GGACAAAGCA CTATTATCTT GGCCTGGAGCT GAATATGGAA GTGGAAGCTC CCGAGATTGG
 361 GCTGCCAAGG GGCCAATGTT GTTGGTGGGT TTTCTTGTC ATTTATTCTT AATTTGGTTG
 421 ATCCTGTCTT TTTCTTTTTT ACCAGAGGAT TCCCTAATA CTACAGAACA ATTTAACAGG
 481 GAGTAAAAGC TGTAATTGCT AAGAGCTTCG AGAGGATTCA TCGCAGTAAC TTGGTAGGAA
 541 TGGGTATTGT GCCTCTATGT TTTAAGGCTG GGGAGGATGC AGATACACTT GGATTGACAG
 601 GTCAAGAGCG CTATCCATTG ACTTGG

Comparison of Purple Russian (top sequence) with NC EBR-8 (bottom sequence)

Identity= 98.72%(540/547)

1GGTACTTTC
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 1 GACTTCAACTCATATGGTAGCCGTCGTGGTAATGATGAGATTATGGCTAGGGGTACTTTC
 10 GCCAATATCCGCCTTGTAACAAGCTGTTAAACGGGGAGGTTGGTCCAAAGACGGTCCAC
 |||||
 61 GCCAATATCCGCCTTGTAACAAGCTGTTAAACGGGGAGGTTGGTCCAAAGACGGTCCAC
 70 ATTCCTTCTGGAGAAAAGCTATCTGTGTTTGATGCTGCTATGGTACTTTCCTTTCTCTGT
 |||||
 121 ATTCCTTCTGGAGAAAAGCTATCTGTGTTTGATGCTGCTATGGTACTTTCCTTTCTCTGT
 130 TTCTTAATGGTTCAAATCCTGTTTTTCATTAAGATAAGAGTTTGCATTGCGCATTTCACT
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 181 TTCTTAATGGTTCAAATCCTGTTTTTCATTAAGATAAGAGTTTGCATTGCGCATTTCACT
 190 TTTGCTGTTTATGTTGACACTTGTAAACACGTGATGCCTCAGAAATACAAATCCGCT
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 241 TTTGCTGTTTATGTTGACACTTTTAAACACGTGATGCCTACAGAAATACAAATCTGCT
 250 GGACAAAGCACTATTATCTTGGCTGGAGCTGAATATGGAAGTGAAGCTCCCGAGATTGG
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 301 GGACAAAGCACTATTATCTTGGCTGGAGCTGAATATGGAAGTGAAGCTCCCGAGATTGG
 310 GCTGCCAAGGGACCAATGTTGTTGGTGGGTTTTCTTGTCATTTATTCTTAATTTGGTTG
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 361 GCTGCCAAGGGCCAATGTTGTTGGTGGGTTTTCTTGTCATTTATTCTTAATTTGGTTG
 370 ATCCTGTCTTTTTCTTTTTTACCAGAGGATTCCTAATACTAAGAACAATTTAACAGG
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 421 ATCCTGTCTTTTTCTTTTTTACCAGAGGATTCCTAATACTACAGACAATTTAACAGG
 430 GAGTTAAAGCTGTAATTGCTAAGAGCTTCGAGAGGATTCATCGCAGTAACTTGGTAGGAA
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 481 GAGTAAAAGCTGTAATTGCTAAGAGCTTCGAGAGGATTCATCGCAGTAACTTGGTAGGAA

490 TGGGTATTGTCCTCTATGTTTTAAGGCTGGGGAGGATGCAGATACACTTGGATTGAC
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541 TGGGTATTGTCCTCTATGTTTTAAGGCTGGGGAGGATGCAGATACACTTGGATTGACAG

Comments

Partial sequence was obtained for six tested samples. Purple Russian and NC EBR-8 sequences had 7 SNPs. The sequence for Purple Russian was identical to that for L40, a F2 from Llanero (i3/i3). The sequence for NC EBR-8 was identical to Fla7547 and NC-123S, both reported to be homozygous for I3. NC07196 was heterozygous as reported.

For the sequence of Purple Russian, there was 100% nt identity with BAC clone C07HBa0073N22 (28,687 nt) at GenBank (AC212624) and SGN sites. NC EBR-8 matched the same BAC clone with a 98% nt identity. The exons of the P7-43A sequence corresponded to the mRNA for *Solanum pennellii* aconitase (AY250115) and a putative aconitase from *Capsicum chinense* (AB372268) and *Solanum tuberosum* (X97012).