

C2_At2g20830, 43 cM, Chromosome 7

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Introduction

The goal for this research was to develop a co-dominant CAPS or SCAR marker for the I3 gene, which conditions 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 C2_At2g20830 was compared with sequences at GenBank. Putative exons were identified by the match with *Vitis vinifera*, AM476255, and forward primer and reverse primers were designed from the putative exon regions to amplify a genomic region, which should include at least one intron.

Table 1. PCR primers on chromosome 7

Primer Name	Primer Sequence (5'-3')
P7-43BF1	CAGTCATTATTAACAAATTCAGGATCG
P7-43BR1	TCTGAGCAATACGTCTAGCAGC

*PCR annealing temperature at 53°C (TGEN53), ~700-bp fragment.

Sequence

M82 (Partial Sequence), (i3/i3)

GENBANK ACCESSION NUMBER: EU926651

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1      CAGTCATTAT TAACAAATTT CAGGATCGTG ATTACAATCG TGTGAACTAC ACTCTGTGAT
61     CATATGTTAT TCATGACAGC ACAGGCTGTC CTATTTATAG TCCCTTGCAT CAAACTGTCTG
121    TAGCCATGGT TGCTGCAGCC TATGACGCGA TAAACCTTGA GCAGCACTCT GGTGCACATC
181    CTCGCCTTGG CGTTGTGGAT GACATTCTCA TTCATCCATT AGCGCGAGCT TCACTGGATG
241    AAGCTTCTTG GCTTGCTAAA AAGATTGCAG CTGATATTGG AAACCGGTTT CAAGGTTAGT
301    AAAAAGTTTG TAACTTTAAA TGAATTCTTC TTTTCTATCA TGTGATGTGC TGTACTGACT
361    TGAAAATGCA AAACGAACCG GTTTTAGAGC TCAAAGTATG TGTATATATG AAAAAACTTT
421    TGCACGCCGT TCTTGACTTT ACATTCTTGA TTCGCCTCTG AATACAGTAC CTGTTTATTT
481    ATATGCTGCT GCTCACCCAA TGGGAAAAGC ACTGGACACC ATAAGGAGGG AGCTAGGTTA
541    TTACCGGCCT AACTTCAGGG GCATCCAATG GGCAGGGTGG GCTCAACCAG AGCAGGTCTC
601    GTCAAACCT GATGAAGTC CAGAGGTGGT GTCTCGTCTG AGAGGTGTCTG TGATGATTGG
661    AGCTCATAAA TGGGTAGCAA TGTACAACAT CCCAATAATG TCCACGGATC TCTCAGCTGC
721    TAGACGTATT GCTCAGA
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NC EBR-8, I3/I3 (Partial Sequence), inbred from R. Gardner

GENBANK ACCESSION NUMBER: EU926652

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1      CAGTCATTAT TAACAAATTT CAGGATCGTG ATTACAATCG TGTGAACTAT ACTCTGTAT
61     CATATGTTAT TCATGACAGC ACAGGCTGTC CTATTTATAG TCCCTTGCAT CAAACTGTCC
121    TAGCCATGGT TACTGCAGCC TATGACGCGA TAAACCTTGA GCAGCACTCT GGTGCACATC
181    CTCGCCTTGG CGTTGTGGAT GACATTCTCA TTCATCCATT AGCGCGAGCT TCACTCGATG
241    AAGCTTCTTG GCTTGCTAAA AAGGTTGCAG CTGATATGG AAACCGATTT CAAGGTTAGT
301    AAAAATTTTA AATGACGACT TCTTTTCTAT CATGTGATAT GATGTACTGA CTTGAAAAATG
361    CATAACGAAC CGGTTTTAGA GCTCAAAGTA TGTGTATATA TGAAAAAAT TTTGCACGCC
421    GTTCTTGACT TTAAATTTCT GATTTCGCCTC TGAATACAGT ACCTGTTTAT TTATATGCTG
481    CTGCTCACCC AATGGGAAAA GCACTGGACA CCATAAGGAG GGAGTTAGGC TATTACCGGC
541    CTAACCTTCAG GGCATTCAA TGGGCAGGGT GGGCTCAACC AGAGCAGGTC CCGGTCAAAC
601    CTGATGAAGG TCCAGAGGTG GTGTCTCGTG CTAGAGGTGT CGTGATGATT GGAGCTCATA
661    AATGGGTAGC AATGTACAAC ATCCAATAA TGTCCACGGA TCTCTCAGCT GCTAGACGTA
721    TTGCTCAGA
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Comparison of M82 (top sequence) with NC EBR-8 (bottom sequence)

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1      CAGTCATTATTAACAAATTT CAGGATCGTGATTACAATCGTGTGAACTA CACTCTTGTAT
      |||
1      CAGTCATTATTAACAAATTT CAGGATCGTGATTACAATCGTGTGAACTA TACTCTTGTAT

61     CATATGTTATTCATGACAGCACAGGCTGTCCTATTTATAGTCCCTTGCATCAAACGTCCG
      |||
61     CATATGTTATTCATGACAGCACAGGCTGTCCTATTTATAGTCCCTTGCATCAAACGTCCG

121    TAGCCATGGTTGCTGCAGCCTATGACGCGATAAACCTTGAGCAGCACTCTGGTGCACATC
      |||
121    TAGCCATGGTTACTGCAGCCTATGACGCGATAAACCTTGAGCAGCACTCTGGTGCACATC

181    CTCGCCTTGGCGTTGTGGATGACATTCTCATTATCCATTAGCGGAGCTTCACTGGATG
      |||
181    CTCGCCTTGGCGTTGTGGATGACATTCTCATTATCCATTAGCGGAGCTTCACTCGATG

241    AAGCTTCTTGGCTTGCTAAAAAGATTGCAGCTGATATTGAAACCGGTTTCAAGGTTAGT
      |||
241    AAGCTTCTTGGCTTGCTAAAAAGGTTGCAGCTGATATTGAAACCGA TTTCAAGGTTAGT

301    AAAAAGTTTGTAACTTTAAATGACTTCTTCTTTTCTATCATGTGATGTGCTGTACTGACT
      |||
301    AAA.....AACTTTAAATGACGACTTCTTTTCTATCATGTGATATGATGTACTGACT

361    TGAAAATGCAAAACGAACCGGTTTTAGAGCTCAAAGTATGTGTATATATGAAAAAATTT
      |||
353    TGAAAATGCATTAACGAACCGGTTTTAGAGCTCAAAGTATGTGTATATATGAAAAAATTT

421    TGCACGCCGTTCTTGACTTTACATTCTTGATTGCCTCTGAATACAGTACCTGTTTATTT
      |||
413    TGCACGCCGTTCTTGACTTTAAATTCTTGATTGCCTCTGAATACAGTACCTGTTTATTT

481    ATATGCTGCTGCTCACCCAATGGGGAAGCACTGGACACCATAAGGAGGGAGCTAGGTTA
      |||
473    ATATGCTGCTGCTCACCCAATGGGAAGCACTGGACACCATAAGGAGGGAGTTAGGCTA

541    TTACCGGCCTAACTTCAGGGGCATCCAATGGGCAGGGTGGGCTCAACCAGAGCAGGTC
      |||
533    TTACCGGCCTAACTTCAGGGGCATCAATGGGCAGGGTGGGCTCAACCAGAGCAGGTC

601    GGTCAAACCTGATGAAGGTCCAGAGGTGGTGTCTCGTGCTAGAGGTGTCGTGATGATTGG
      |||
593    GGTCAAACCTGATGAAGGTCCAGAGGTGGTGTCTCGTGCTAGAGGTGTCGTGATGATTGG

661    AGCTCATAAATGGGTAGCAATGTACAACATCCAATAATGTCCACGGATCTCTCAGCTGC
      |||
653    AGCTCATAAATGGGTAGCAATGTACAACATCCAATAATGTCCACGGATCTCTCAGCTGC
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721 TAGACGTATTGCTCAGA
    |||
713 TAGACGTATTGCTCAGA
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Comments

Partial sequences were obtained for four tomato samples. M82 (susceptible) and NC EBR-8 (resistant) sequences had differences of approximately 16 SNPs and 1 indel. The M82 sequence was the same as the other susceptible line, Purple Russian; and the NC EBR-8 sequence was the same as the other resistant line sequence, NC123S.

The sequence of M82 matched the clone C02SLm0006E22 (110,059 nt) with a 97% nt identity, as did NC EBR-8 with a 93% nt identity, at the GenBank (AC215469.2) site. M82 also matched C07HBa0045O10.1 with 100% nt identity, C02HBa0032J10.3 with a 97% nt identity, and LE_HBa0032J10_T7_140600 with a 95% nt identity at the SGN site. NC EBR-8 also matched these same clones with nt identities of 96%, 93% and 91% respectively.