

Chromosome 4: TG287, 66.00cM

Christopher Martin and Douglas Maxwell
University of Wisconsin – Madison: June 9, 2006

PCR primer pairs were designed for RFLP probe: TG287 (Fig. 1).

Fig. 1: RFLP map of the middle of Chr. 4 (Adapted from Pan et. al., 1999).

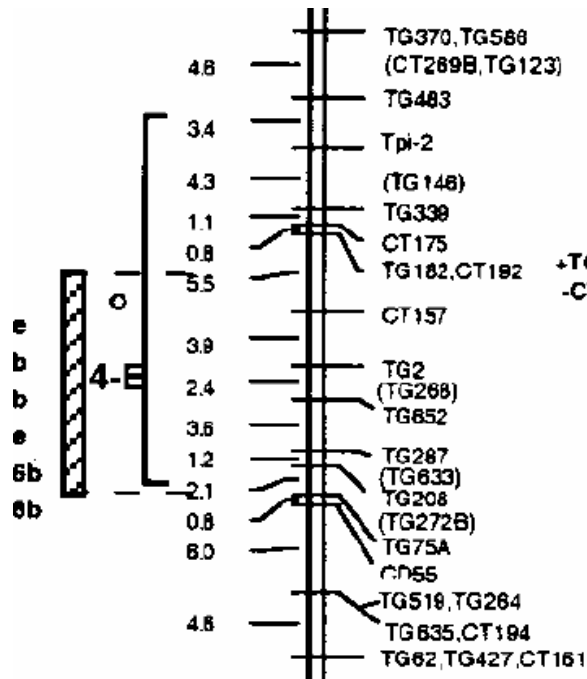


Table 1: Primers from the TG287 probe on Chr. 4.

TG287	Primer Sequence (5' to 3')
PTG287F1	CTCAGAGGTGGGGGCGGAATGG
PTG287F2	CGAACTCAAAGAAGCAACAGAGGGTG
PTG287R1	GGTGTGTTCTCTCTGCTTTATATGC
PTG287R2	CCCCATATATTAACAAAGTGCAAACG

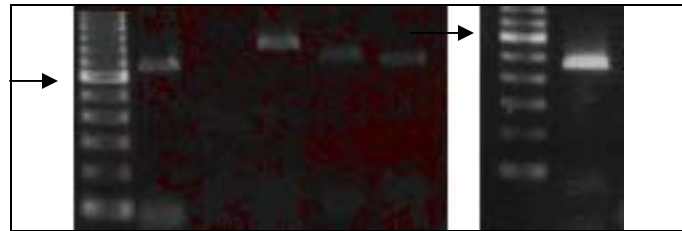


Fig. 2: Agarose gel of the PCR reactions with the four TG287 primer pairs and Heinz 1706 DNA. The PCR reactions were run in two separate gels. In gel one: lane 1, 100-bp ladder; lane 2, control, primer pair PTG301F3/PTG301R2 with Heinz 1706 DNA; lane 3, water; lane 4, PTG287F1/PTG287R1; lane 5, PTG287F1/PTG287R2, lane 6, PTG287F2/PTG287R1. In gel two: lane 1, 100-bp ladder; lane 2, PTG287F2/PTG287R2. The PTG287F2/PTG287R2 primer pair was chosen for sequencing reactions based on the intensity of its band. Arrow marks the 600-bp fragment.

TG287 RFLP Probe: Recent Cornell data (Solanaceae Genomics Network, 2004) shows that this RFLP probe is located roughly 2 cM below the TG208 RFLP probe on Chr. 4. Primers were designed for this RFLP probe in order to determine how far the introgression extended below the TG208 RFLP probe on chromosome four. Four primers were designed from the TG293 RFLP probe: PTG287F1, PTG287F2, PTG287R1, and PTG287R2 (Table 1). All four primer combinations gave single bands between 480 bp and 900 bp with Heinz 1706 DNA (Fig. 2). PTG287F2/PTG287R2 primer pair gave the most intense single band and was thus used with additional genotypes. Heinz 1706, Gc9, Gc173, and Sheriff each gave strong bands at 480 bp. These PCR fragments were sequenced with both the forward and reverse primers. Heinz 1706 (acc. no. DQ222943), Gc173 (acc. no. DQ222942), Gh13, Gc9, and Sheriff each gave sequence with both primers. Just as with the TG208 primers, two patterns were found. Gc173, Gc9, and Gh13 each had the same pattern, while Heinz had a second unique pattern. Sheriff was heterozygous for both patterns (Table 2). Thus, because the TG287 probe has the same correlation between genotypes as the TG208 probe, this would indicate that the introgression extends at least 2 cM below the TG208 locus. Just as with the TG208RFLP probe, the sequence differences do not correlate with begomovirus resistance. Therefore, a molecular marker for begomovirus resistance could not be found at the TG287 RFLP probe. However, the groups of patterns may warrant additional research.

Table 2: Sequence patterns associated with the TG287 RFLP probe. The INDEL is at 51 bp and the SNP is at 122 bp into the sequence.

Pattern (TA)	Pattern (_G)	Pattern (Heterozygous)
Gc173	Heinz 1706	Sheriff
Gc9		
Gh13		

References

- Pan, Q., Liu, Y., Budai-Hadrian, O., Sela, M., Carmel-Goren, L., Zamir, D., and Fluhr, R. 1999. Comparative genetics of nucleotide binding site-leucine rich repeat resistance gene homologues in the genomes of two dicotyledons: tomato and arabidopsis. *Genetics Society of America* 88:309-322.
2004. "Tomato-Arabidopsis Synteny Map." Solanaceae Genomics Network. Cornell University. <http://www.sgn.cornell.edu/maps/tomato_arabidopsis/synteny_map.html> (November 5, 2004).