Cite this poster as:

Labate, J.A., L.D. Robertson, S.M. Sheffer, F.Wu, S.D. Tanksley and A.M. Baldo. 2006. DNA polymorphism estimates within domesticated tomato. PAA/Solanaceae Abstract 366. July 23-27, Madison, WI.

Abstract

Cultivated tomato (*Solanum lycopersicum* L.) is known to have a narrow genetic base. This is in part due to population genetic processes such as founder events, genetic bottlenecks, and natural and artificial selection during domestication. We present empirical estimates of nucleotide polymorphism in 48 genes including COSII, EST-based, and several loci related to fruit quality traits. EST markers included five that were hypothesized to be cryptic wild species alleles (introgressions) within *S. lycopersicum*. All genes were resequenced in a diverse panel of 30 Plant Genetic Resources Unit (PGRU) tomato accessions, line TA496, and *Solanum peruvianum* accession G 32591. The majority of sampled tomato accessions represented the primary center of diversity (Peru, Chile, and Ecuador), and countries contiguous with the primary center. Original collections were made between 1932 and 1976. Within and between-species diversity estimates will be compared for the various marker types. These data will enhance our understanding of single nucleotide polymorphism (SNP) markers and the nature of genetic variation within cultivated tomato.

DNA Polymorphism Estimates within Domesticated Tomato

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PAA/Solanaceae 2006, Madison, WI

❖Within a geographic diversity panel of *S. lycopersicum* (Table 1) we resequenced fragments for 48 loci (Table 2). We verified 150 SNPs in 22.3 kb (about 1 SNP per 150 bp). Three classes of markers gave similar estimates of polymorphism (Table 3).

❖Our panel of 31 *S. lycopersicum* lines was approximately as diverse as one population of *S. pimpinellifolium* (mean $\theta = 1.6 \times 10^{-3}$), the closest wild relative. Nine of the 48 loci in our study were monomorphic, versus 2 of 15 loci in *S. pimpinellifolium* (Roselius et al. 2005, Genetics 171:753-763).

❖One *S. peruvianum* line (Table 1) was sequenced as an outgroup.

Table 1. Tomato diversity panel (31 PGRU accessions and line TA496).

provenance	no. accessions
Primary center of diversity (Chile, Ecuador, Peru), 1932-1974	14
Regions contiguous with primary center (11 countries), 1932-1996	12
Secondary centers of diversity (Afghanistan, China, Cuba, Netherlands), 1932-1960	4
S. peruvianum G32591	1

Table 2. Resequenced fragments from 48 loci.

Locus type	n	attributes
EST-based with predicted SNPs	26	exon and intron
Conserved Ortholog Set II (COSII)	11	mostly intron
Arbitrary genes	11	most related to quality, noncoding regions

Table 3 . Polymorphism of SNP marker types, 31 *S. lycopersicum* DNAs by 48 loci.

marker type	no. loci	mean no. sequences	no. SNPs	nucleotides	mean π*	mean θ*	mean no. haplotypes
EST-based	26	33	86	9,183	1.66	2.07	2.4
COSII	11	34	33	7,587	1.20	1.40	2.6
Arbitrary	11	32	31	5,564	1.20	1.40	2.8

*nucleotide diversity x 1000

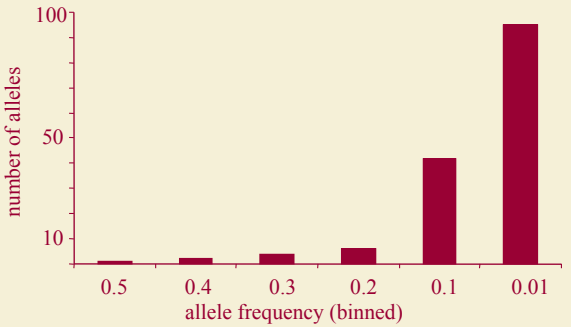
Table 4. Test of the distribution of nucleotide diversity estimates for three classes of *S. lycopersicum* markers.

test of	no. groups	K-W statistic	df	*P*-value
θ	3	3.14	2	0.208
π	3	5.35	2	0.069

❖A Kruskal-Wallis test (one-way nonparametric ANOVA) was used to compare distributions of population parameter estimates for EST-based, COSII, and Arbitrary markers.

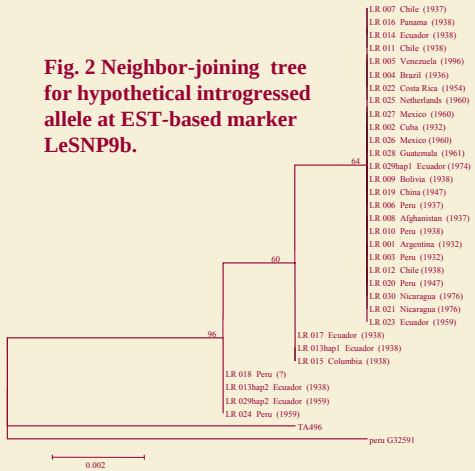
❖The three marker types did not significantly differ from each other in θ and π estimates of nucleotide variation (Table 4).

Fig 1. Frequency of minor allele in *S. lycopersicum*, (39 polymorphic loci, 150 SNPs, 30 to 36 sequences per locus).



❖Allele frequencies in *S. lycopersicum* were highly skewed towards rare SNPs (Fig. 1). The minor allele was at a frequency of less than 10% for 95 SNPs. Only 1 allele was observed at 50% frequency.

Fig. 2 Neighbor-joining tree for hypothetical introgressed allele at EST-based marker LeSNP9b.



❖Several of the 48 loci showed evidence of introgressed alleles from wild species into *S. lycopersicum* (e.g., Fig. 2). This can result from linkage drag during breeding. LeSNP9b maps within 1 cM of a disease resistance allele that was introgressed into TA496 (Labate and Baldo, 2005, Mol. Breeding 16:343-349).

Table 5. HKA neutrality test for 40 loci in *S. lycopersicum* and *S. peruvianum*.

sum of deviations*	df	*P*
90.683	39	0.00001

*observed variation – expected variation

❖**Polymorphism** within a species is correlated with **divergence** between species under neutral evolution, providing the basis for the HKA test (Hudson, Kreitman, and Aguadé, 1987, Genetics 116:153-159).

❖The 40 tested tomato loci highly significantly rejected a neutral equilibrium model (Table 5). This test provides candidate loci for exploring microevolutionary processes such as selection, bottlenecks, and introgression in *S. lycopersicum*.