The gene responsible for serrate leaflets in *P. sativum* ssp. *abyssinicum* is on linkage group III tightly linked to an STS marker

Lorenzi, V. E.S.I.T.P.A., Rouen, France
Murphy, R.L. and Weeden, N.F. Montana State Univ., Bozeman, MT, USA

The strongly serrate leaflet margins constitute the most obvious morphological character defining *Pisum sativum* ssp. *abyssinicum* (1, 2, 13). The serration is not observed on leaflets of the first two true leaves but is strongly expressed for approximately the next five nodes (see cover). Sutton (11) reported on this phenotype in W808, a line from Palestine that has been confirmed to possess the Abyssinicum genotype (13). He determined that the phenotype was controlled by a dominant allele at a locus he designated *Ser*. Wellensiek (14) suggested that the symbol *Td* be used. Rosen (9) used *td*, believing the gene was recessive or only semi dominant, and Lamprecht (5) used both *Ser* and *Td* to describe serrate phenotypes. Finally, Smirnova (10) suggested that a different locus, *Td'*, might exist in material he examined. At present, W808 is used as the type line for *Td* and is recessive for *ser*, whereas W1414 is the type line for *Ser*, as defined by Lamprecht (5). Thus the literature is very confusing, because the gene responsible for the strongly serrate leaflet margins in line W808 has been referred to as both *Ser* and *Td*. In addition, Lamprecht's (5) drawing of leaflet morphology for the genes *Td*, *Ser* and *Inci* places the phenotype exhibited by W808 squarely in the *Ser* category.

The location of *Td* (as defined by the type lines) on the pea linkage map also has been a subject of discussion in the last 20 years. Marx (7) pointed out inconsistencies between his data and those of Lamprecht. Lamprecht's data suggested linkage between *Td* and *Fa* at a distance of about 30 cM (4), whereas Marx (7) found linkage between *B* and *Td* at an intensity of 14 and 17 cM. Marx reasoned that because *Fa* was on chromosome 4 (Lamprecht's definition) and *B* was on chromosome 3, one of the two linkages must be incorrect. Within the following decade several investigators (3, 8, 12) explored linkage relationships of genes generating serrate leaflet margins. Święcicki (12) observed linkage between *Inci* and *B*, but not between *Td* and *B* or *Ser* and *B*. His experiments did not involve the markers *Le* or *Np* but did test linkage with many standard morphological markers, including *St*. Two papers reported linkage between *Td* and markers on linkage group III. Grajal-Martin and Muehlbauer (3) reported weak linkage (20 to 30 cM) between *Lap1* and *Td*, and concluded that their results indicated that either *Td* was on linkage group III or that there were more than one gene producing the serrate phenotype. In two crosses Polans (8) reported linkage between *Td* and *St* and/or *B*. The two lines he used as sources of the serrate phenotype (82-14n and A778-26-6) were a *P. s.* ssp. *elatius* line selected from PI268480 by N.F. Weeden and a *P. s.* ssp *sativum* line with an introgressed *Td* allele derived from *P.s.* ssp. *abyssinicum* (see source in ref. 3), respectively. The data obtained by Polans placed *Td* between *St* and *B*, a surprising location because Święcicki (12) should have observed linkage between *Td* and *St* in his experiments. As indicated above, most of the reported linkage intensities between the gene generating the serrate leaflet phenotype in *P.s.* ssp. *abyssinicum* and standard marker loci have been greater than 20 cM and therefore are open to question. In this study we attempted to generate more conclusive data for the position of this gene through the use of DNA markers.

A cross was made between *P. s.* ssp. *sativum* cv 'Sparkle' and *P. s.* ssp *abyssinicum* line PI358617. The F_1 plants were tall and displayed a serrate leaflet phenotype matching that in PI358617 (most prominent on leaflets from nodes 5 through 10). The F_1 plants were also partly sterile, indicative of chromosomal rearrangements known to exist between Abyssinicum pea and most *P. sativum* ssp. *sativum* lines (Lamprecht, 6). Thirty-six F_2 plants were scored for *Le*, *Td* and the STS marker, CipPor. Both *Le* and CipPor are known to be on linkage group III of the consensus map, with CipPor mapping approximately 7 cM from *Np* on the opposite side from *Le* (S. Brauner, R. Murphy. J. Przyborowska, and N. Weeden, unpublished). The primer sequences used to amplify the CipPor fragment were 5'-ACTGCTAAGGCTTTGGCTGA and 5'-AGATTTTGTTAGGCTTGGATCACT with standard amplification conditions and an annealing temperature

of 60 C. The resulting 1100 bp fragment was cut with *Hae*III to reveal a polymorphic restriction site (the CipPor fragment from 'Sparkle' possessed a *Hae*III site near the middle of the fragment, whereas the CipPor fragment from PI358617 did not).

Segregation of the individual loci gave monogenic ratios [*Le*:*le* = 27:7, χ^2 =0.37, p<0.01; *Td*:*td* = 20:16 χ^2 =4.6, p<0.05; CipPor (het + PI358617):CipPor ('Sparkle') = 20:16]. Joint segregation analysis is shown in Table 1.

Table 1. Joint segregation analysis of *Le*, *Td* and CipPor in the 'Sparkle x PI358617 F_2

	Number of F_2 plants with phenotype[1]					
Loci	**D/D**	**D/R**	**R/D**	**R/R**	χ^2	**Recombinant Fraction**
Le*:*Td	**15**	**12**	**3**	**4**	**0.36**	**no linkage**
***Le*:CipPor**	**14**	**13**	**3**	**3**	**0.007**	**no linkage**
***Td*:CipPor**	**16**	**2**	**2**	**14**	**19.8**	**9.2+11**

[1]Phenotype designations: D=dominant; R= recessive

The *Td* and CipPor segregation ratios deviate significantly from expectation (3:1), but the deviation may be attributed to linkage with a lethal or some other factor causing the partial sterility. Although neither CipPor nor *Td* display significant linkage with *Le*, if we arrange the loci so that the number of recombinant and double recombinants are both minimized, we obtain a locus order with *Td* in the middle. From the position of CipPor on the consensus map we know the order of the loci is *Lap1—Pepc—CipPor—Np—Le*. Thus our current results would place *Td* near *Np*, although the data do not permit a determination of the distance between *Np* and *Td* or which locus in closer to CipPor. It should be emphasized that we mapped the gene in *P. s.* ssp. *abyssinicum*, and that the gene in the *P. s.* ssp *elatius* accession used by Polans (8) may be different.

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