

## *Coiling shoot (csh)*, a new mutation affecting shoot gravitropism

Gorel, F.L. and Berdnikov, V.A.

Inst. of Cytology and Genetics, Russian Academy of Sciences  
Novosibirsk, Russia

In 1997 we developed a pea line, Delta, that permitted the maintenance of a heterozygous segment of linkage group V between loci *det* and *curl* (Fig. 1) (1). This construct is based on a sporophytic lethal  $tl^x$ , which is probably a small deletion of a chromosome fragment carrying the *Tl* gene. As a result, recombination between loci *r* and *tl* is significantly suppressed. For this reason, all round seeds of the line Delta have genotype  $R\ tl^x / r\ Tl$ . Allelic composition of the *Sca* locus coding for a seed protein is easily determined by electrophoresis of proteins in a small quantity of cotyledon flour. Thus, to maintain this line, round seeds heterozygous for *Sca* should be picked out and the resulting plant self-pollinated. The presence of the *det* and *curl* markers is verified by sowing of wrinkled seeds. In the line Delta meiotic recombination is decreased about three times as compared to the standard map, for this reason the majority of seeds retain the flanking markers (1).

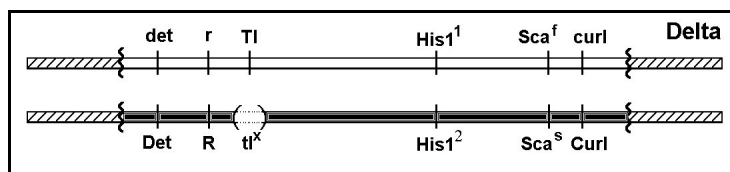


Fig. 1. A scheme showing a segment of Linkage group V maintained heterozygous in the line Delta.

The line Delta can be used to obtain recessive mutations in a chromosome segment between *R* and *Sca*. These mutations can be identified by simply comparing phenotypes of plants grown from round and wrinkled seeds. One hundred and fifty seeds of the line Delta were treated with 0.1% EMS and planted in a hydroponic bed. Seed was collected from the 100 most fertile plants. Two seeds from each of these plants were germinated and grown to maturity. Round and wrinkled seeds from  $M_2$  plants were sown in the field. In one family (E-65) seedlings from only the round seeds emerged, the seedlings from the wrinkled seeds often staying buried because shoots apparently had lost negative gravitropism (Fig.2). The direction of growth of the shoot apex in the mutants seemed to be randomly determined and changed when the shoot tip contacts with any mechanical obstacle. While under ground, etiolated shoots grow by epicotyl extension. However, once the shoots breaks through the soil surface, the plant starts growing upward, presumably responding by positive phototropism. Hence, the traits of the adult plant can be registered. We called this mutation affecting normal negative gravitropism of shoots *coiling shoot (csh)*.

The plants grown from round seeds of the E-65 family were crossed with the line DAG (*Det, a, R, Tl, His1^f, SCA^s, Curl*), isolated from the accession VIR-5538 (Daghestan). Four  $F_1$  seeds heterozygous for *SCA* produced plants hetero-zygous for *His1*, *tl* and *r*. Segregation of phenotypes in the  $F_2$  is shown in Table 1.

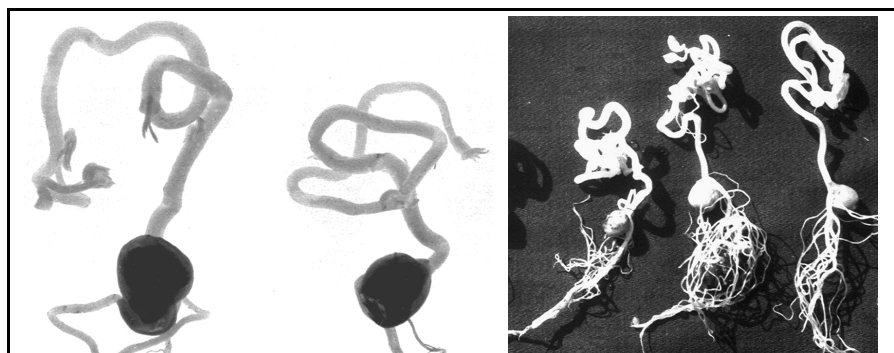


Fig. 2. Pea seedlings exhibiting the phenotype *csh*. left, 3-days old seedlings; right, 7-day old seedlings.

**Table 1. Joint segregation data for the  $F_2$  progeny of the cross E-65 (*r, tl, csh, His1^s, SCA^f, curl*) X DAG (*R, Tl, Csh, His1^f, SCA^s, Curl*).**

Gene A Gene B	Number of plants with designated phenotype <sup>1</sup>	Rec.	St.	Joint
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		A/B	A/h	A/b	h/B	h/h	h/b	a/B	a/h	a/b	fract %	Error	seg. $\chi^2$
<i>r</i>	<i>Sca</i>	45	79	15				3	18	30	21.41	3.30	50.1 <sup>2</sup>
<i>r</i>	<i>csH</i>	133		6				14		37	11.19	2.45	99.2 <sup>2</sup>
<i>r</i>	<i>tl</i>	55	84	0				0	5	46	2.71	1.19	166.0 <sup>2</sup>
<i>r</i>	<i>His1</i>	39	85	15				2	17	32	19.34	3.14	56.2 <sup>2</sup>
<i>csH</i>	<i>SCA</i>	47	88	12				1	9	33	12.89	2.58	87.5 <sup>2</sup>
<i>Sca</i>	<i>tl</i>	32	16	0	23	58	16	0	15	30	20.28	2.36	92.1 <sup>2</sup>
<i>Sca</i>	<i>His1</i>	41	7	0	0	93	4	0	2	43	3.49	0.96	307.2 <sup>2</sup>
<i>csH</i>	<i>tl</i>	54	83	10				1	6	36	10.23	2.30	107.7 <sup>2</sup>
<i>csH</i>	<i>His1</i>	41	95	11				0	7	36	9.72	2.24	104.7 <sup>2</sup>
<i>tl</i>	<i>His1</i>	32	14	0	15	65	9	0	23	32	17.53	2.19	115.0 <sup>2</sup>

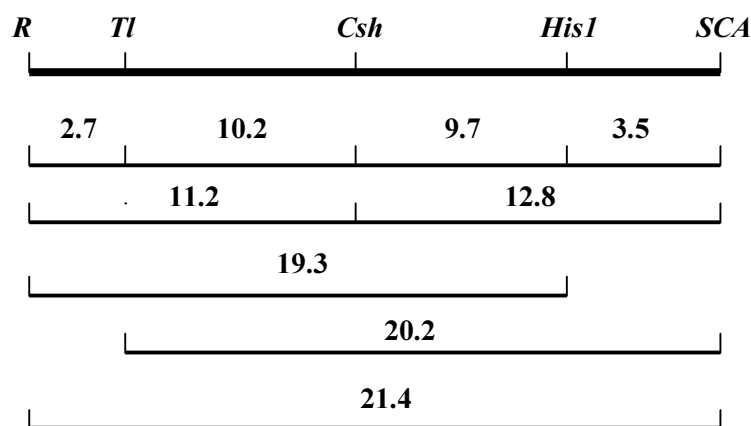
<sup>1</sup> 'h' stands for heterozygotes. Total number of plants analyzed is 190.

<sup>2</sup> P<0.0001.

All dominant genes are in coupling phase.

All calculations were made with the program 'Cros' using the maximum likelihood method.

Recombination relationships of the new locus are given in the following diagram.



Thus, the new locus *Csh* is situated in linkage group V between *tl* and *His1* loci at approximately equal distances.

In higher plants, the shoots exhibit negative gravitropism, which is one of the most important phenomena in plant growth and development. In pea, the *ageotropic* mutation is known (2), which affects root growth and to a less extent decreases negative gravitropism of the shoot. It has been mapped to linkage group IV (3). The new mutation *csH* does not effect gravitropism of the roots and together with *age* can be used to study one of the most important features of the plant.

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