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Two New Necrotic Root Mutants

In soybean [*Glycine max* (L.) Merr.], five necrotic root mutants have been identified. Three were recovered among germinal revertant progeny of the w_4-mutable line (Palmer et al., 1989) and they are allelic (Kosslak et al., 1996). Twenty three necrotic root mutants were identified in a second gene tagging study with the w_4-mutable line. One has been tested and was allelic to the three from w_4-mutable (Andersen and Palmer, 1997). The fifth characterized necrotic root mutant was identified in family EMS-95 which was descended from EMS treated cultivar AgriPro 1776 (Palmer and Wubben, 1998). This EMS mutant necrotic root line was allelic to the previous four characterized necrotic root mutant lines. Necrotic root is a programmed cell death mutant (Kosslak et al., 1997).

Our objective was to determine the inheritance and the allelism of two additional necrotic root mutants, NR-4 and NR-5, which were identified (spontaneous mutations) in our genetic studies.

Materials and Methods

Necrotic root mutant NR-4 was identified in genetic line A97-356, which was a cross of Clark-k_2 X T323 [y_{20} (Ames 2) *Mdh1-n* (Ames 2)]. T323 originated as a germinal revertant in T322 (w_4-*m*). In the F_2 generation of the Clark-k_2 X T323 cross, a chlorophyll leaf chimera plant was observed. Among the self-pollinated progeny of this chimera, normal root and necrotic root plants were evident.

NR-5 was identified in a classical linkage group chromosome interchange cross of genotype w_1 w_1 adh_1 adh_1 ms_1 ms_1 X KS172-11-3 (*W1 W1 Adh1 Adh1 Ms1 Ms1*) . The F_2 segregated normal root and necrotic root plants.

Allelism crosses were T328H [Rn_1 rn_1 (Ames 1)] X NR-4, NR-5 X T329H [Rn_1 rn_1 (Ames 2)], and reciprocal crosses between NR-4 X NR-5.

Only F_1 seed that was the result of cross-pollinations between heterozygous female X heterozygous male parents were used for the allelism test. The parent plants were numbered and the heterozygous genotypes identified by progeny testing of self-pollinated plants. The normal root F_1 plants were advanced to the F_2 generation.

The F_1 and F_2 seed from all cross-pollinations were placed on germination paper. The seedlings were scored for root necrosis 10 days after planting.

Results and Discussion

We believe that NR-4 and NR-5 necrotic root mutants are spontaneous mutations, not contamination from known necrotic root mutant lines. NR-4 and NR-5 only showed segregation of the genetic traits expected from the original genetic crosses.

The three allelism crosses each indicated that the unknown necrotic root mutations were allelic to known necrotic mutant lines (Table 1). Furthermore, the two unknown necrotic root mutant lines were allelic to each other (Table 1). Both F_1 and F_2 data support the conclusion that NR-4 and NR-5 are allelic to the five characterized necrotic root mutations in soybean. In addition, the NR-4 and NR-5 heterozygous necrotic root lines showed 3 normal: 1 necrotic root plant segregation upon self-pollination.

Four of the six necrotic root mutants have a direct common origin. Three are germinal revertants from *w_4-m*. The NR-4 mutant line traces to the *w_4-m* line. NR-4 descended from a cross involving *w_4-m*.

EMS-95 is an EMS mutagen-derived mutant from commercial cultivar AgriPro 1776. The NR-5 mutant line is from a linkage group 8 chromosome interchange mapping study. KS172-11-3 has one interchanged chromosome with linkage group 8 (Mahama and Palmer, 2003). The other interchanged chromosome has not been associated with any linkage group. Perhaps the other interchanged chromosome involves the same chromosome that has the *Rn_1* locus. The *Rn_1* locus has not been tested for linkage with KS172-11-3.

It seems that the necrotic root locus is a ‘hot spot’ for mutations, either from a putative transposable element (*w_4-m*), a chemical mutagen (EMS), or spontaneous mutation.

References

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Table 1. Allelism tests of unknown necrotic root mutants NR-4 and NR-5 with known necrotic root mutants T328 and T329; F_1 and F_2 data.

	No. F_1 plants				No. F_2 plants			
Cross	Normal	Necrotic	χ^2(1:1)	P	Normal	Necrotic	χ^2(3:1)	P
T328H X NR-4 het.	72	54	2.57	0.11	1876	592	1.35	0.25
NR-5 het. X T329H	13	15	0.14	0.71	404	146	0.70	0.40
NR-4 het. X NR-5 het.	20	20	0	1	673	254	2.85	0.09

*Data from reciprocal cross-pollinations were combined since the statistical test indicated that the data were homogeneous.