

***C. elegans* *srf-6* and *nsy-1* mutations result in a similar 2AWCON phenotype and do not complement (*srf-6* is *nsy-1* II)**

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Genotype	% 2AWCON (n)	% 1AWCON (n)	% 0AWCON (n)	Total number scored
Wild-type	1 (1)	99 (97)	0 (0)	98
<i>srf-6(yj13)</i>	92 (108)	5 (6)	3 (3)	117
<i>nsy-1(ok593)</i>	89 (110)	9 (10)	2 (3)	123
<i>srf-6 (yj13) / nsy-1 (ok593)</i>	82 (50)	11 (7)	7 (4)	61
+ / <i>nsy-1 (ok593)</i>	0 (0)	100 (33)	0 (0)	33

Figure 1. Comparison of AWC phenotypes of *srf-6(yj13)* and *nsy-1(ok593)* mutants, and complementation testing of *srf-6(yj13)* and *nsy-1(ok593)* mutants using the 2AWCON^{ON} phenotype. The *nsy-1(ok593)* mutation is a large complex rearrangement that completely deletes the catalytic domain of *nsy-1* (*C. elegans* Deletion Mutant Consortium 2012). Homozygous *srf-6* and *nsy-1* mutants also contained homozygous *unc-4 (e120) II* and *kyls140 I*. Table entries represent percentages of worms with each phenotype, followed by the actual number of worms scored in parentheses. The right-hand column indicates the total number of worms scored per genotype.

Description

C. elegans *srf-6* mutants were isolated using altered surface immunofluorescence as phenotype (Hemmer et al., 1991; Grenache et al., 1996). In Van Sciver et al., 2019, we showed by whole genome sequencing that three different *srf-6* mutants carry mutations in gene *nsy-1*. Well-characterized mutant alleles of *nsy-1* result in hermaphrodites that express the olfactory receptor gene *str-2* in both AWC neurons (2AWCON^{ON}, Troemel et al., 1999), unlike wild type hermaphrodites, which express *str-2* asymmetrically in only one of the two AWC neurons (1AWCON^{ON}, Troemel et al., 1999). Our sequencing results suggested that *srf-6* mutants might have a similar 2AWCON^{ON} phenotype. We therefore examined *srf-6(yj13)* for its AWC phenotype.

In order to introduce an *str-2::GFP* marker into the *srf-6* genotype, first *srf-6(yj13) unc-4(e120) II* and *nsy-1(ok593) unc-4(e120) II* double mutants were constructed as described (Hemmer et al., 1991). Homozygous males carrying the construct *kyls140*, which contains an *str-2::GFP* fusion integrated on chromosome I (Troemel et al., 1999), were mated with *srf-6(yj13) unc-4(e120) II* hermaphrodites, and an Unc F2 hermaphrodite expressing GFP in chemosensory neuron AWC was cloned. An individual hermaphrodite descendant, all of whose offspring expressed GFP, was isolated to establish a strain of genotype *kyls140 I; srf-6(yj13) unc-4(e120) II*. A strain of genotype *kyls140 I; nsy-1(ok593) unc-4(e120) II* was constructed similarly. Adult hermaphrodites from these strains were examined in a fluorescent microscope for their AWC phenotype. Figure 1 shows that *srf-6(yj13)* adults exhibited a 2AWCON^{ON} phenotype similar to that of *nsy-1(ok593)*.

To test whether *srf-6(yj13)* and *nsy-1(ok593)* affect the same or different genetic functions, a complementation test was performed (Figure 1, last two lines). Males of genotype *srf-6(yj13)* were mated with *kyls140 I; nsy-1(ok593) unc-4(e120) II* hermaphrodites, and non-Unc offspring were scored in a fluorescent microscope for the AWC phenotype. The complementation heterozygotes showed a 2AWCON^{ON} phenotype similar to that of *nsy-1(ok593)*. This result included data from two separate crosses. In contrast, when wild type males were mated with *kyls140 I; nsy-1(ok593) unc-4(e120) II* hermaphrodites, the 1AWCON^{ON} phenotype was observed. These results included worms from one cross.

These results indicate that *srf-6(yj13)* and *nsy-1(ok593)* mutations do not complement each other for the 2AWC^{ON} phenotype, and together with the *srf-6* mutant sequencing results (Van Sciver et al., 2019), we conclude that *srf-6* and *nsy-1* are the same gene.

Reagents

C. elegans Strains

N2 *C. elegans* wild type

CX3695 *kyIs140 [str-2::GFP + lin-15(+)] I*

AT18 *srf-6(yj13) II*

AT19 *srf-6(yj13) unc-4(e120) II*

VC390 *nsy-1(ok593) II*

AT28: *srf-6(yj13) unc-4(e120) II; kyIs140 [str-2::GFP + lin-15(+)] I*

AT29: *nsy-1(ok593) unc-4(e120) II*

AT30: *nsy-1(ok593) unc-4(e120) II; kyIs140[str-2::GFP + lin-15(+)] I*

Strains N2, CX3695, and VC390 are available from the CGC. Strains AT28 and AT30 will be submitted to the CGC.

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