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daf-2 modulates regeneration of mechanosensory neurons II

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Description:

The *daf-2* gene encodes an insulin-like growth factor/IGF-1 receptor that regulates *C. elegans* embryonic and larval development. It has previously been shown that DAF-2 inhibits neurite regeneration of the GABAergic motor neurons and PVD sensory neurons in an age-dependent fashion [1, 2]. Following injury, the posterior lateral microtubule (PLM) neurons are capable of regenerating through axonal fusion, a highly efficient regrowth mechanism in which separated fragments fuse back together [3-6]. We previously established that a critical event for axonal fusion to occur is the exposure of injury-induced phosphatidylserine (PS) 'save-me' signals [5]. The level of PS exposed increases with advancing age [3]. To determine if *daf-2* is involved in this age-dependent modulation of PS exposure, we visualised and quantified the level of PS exposed after PLM axotomy using a secreted, tagged version of Annexin V [5, 7]. Mutation of *daf-2* had no effect on PS exposure 1 h post-axotomy, with no significant differences observed on either the distal or proximal axon segments (Table 1).

Table 1. Quantification of the relative level of PS exposed 1 h post-axotomy.

Genotype	PS exposed on distal axon (relative to pre-axotomy)	*n*	PS exposed on proximal axon (relative to pre-axotomy)	*n*
wild-type	1.53 ± 0.105	28	1.44 ± 0.0855	28
daf-2(e1370)	1.51 ± 0.167	26	1.57 ± 0.166	26

Reagents

One-day-old adult hermaphrodites were used for all experiments, and were grown under standard conditions at 20°C. The BXN301 [*daf-2(e1370); smIs95(Phsp-16.2::sAnxV::mRFP); zdIs5(Pmec-4::GFP)*] strain was used along with the CU4204 [*smIs95(Phsp-16.2::sAnxV::mRFP); zdIs5(Pmec-4::GFP)*] control strain. The *daf-2(e1370)* allele has been considered temperature sensitive for the dauer phenotype, but not for the long-lived phenotype. At 20°C, *daf-2(e1370)* animals display a greater than 2-fold increase in lifespan compared to the wild-type [8]. Laser axotomy, microscopy and quantification of data was performed as previously described [3].

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