Supplemental data for:

THE NOVEL HYDROXYLAMINE DERIVATIVE NG-094 SUPPRESSES POLYGLUTAMINE PROTEIN TOXICITY IN *CAENORHABDITIS ELEGANS*

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1) Supplemental Figures: Figure S1 and Figure S2
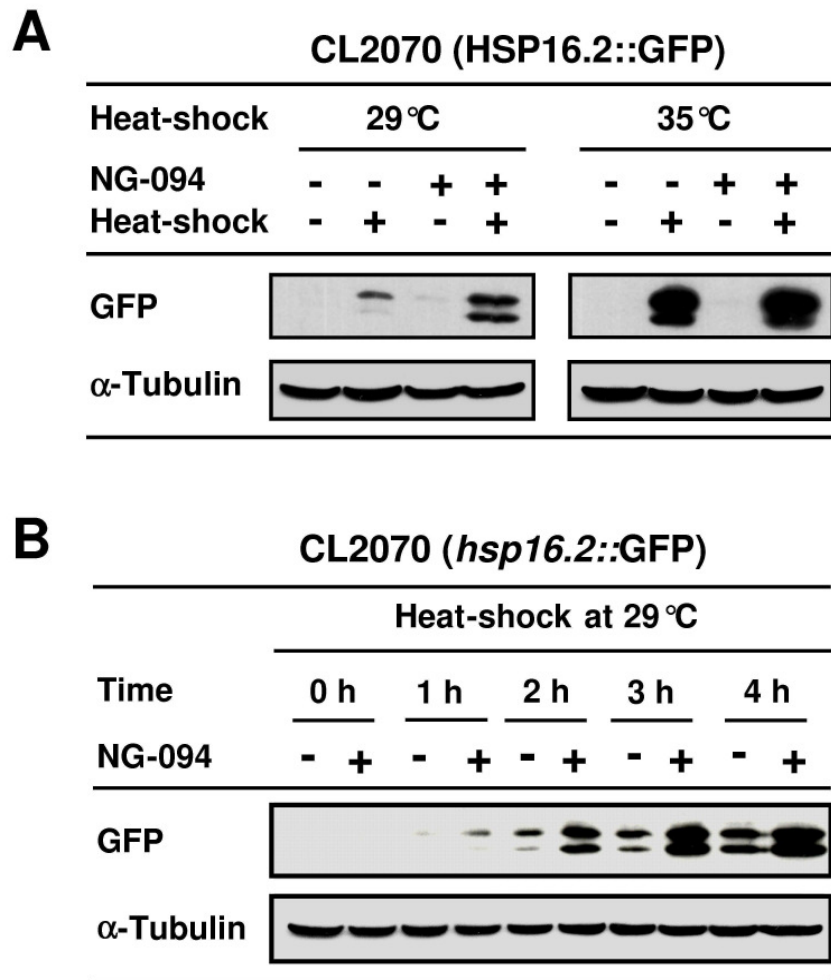**2) Supplemental Table:** Table S1

Figure S1

Fig. S1. (A) Western blot analysis of GFP expression in 3-day-old CL2070 nematodes expressing a *hsp16.2::*GFP transgene. Animals were cultured at 22°C and treated or not with 1 mM NG-094. Analyses were performed for animals that on the third day since hatching were either kept at 22°C or heat-shocked at 29°C or 35°C for 2 h (B) Expression levels of GFP in CL2070 animals that on the third day since hatching were heat-shocked at 29°C for various periods of time. Animals were collected after a 6 h period of recovery at 22° following the heat-shock. α-tubulin was used as a loading control.
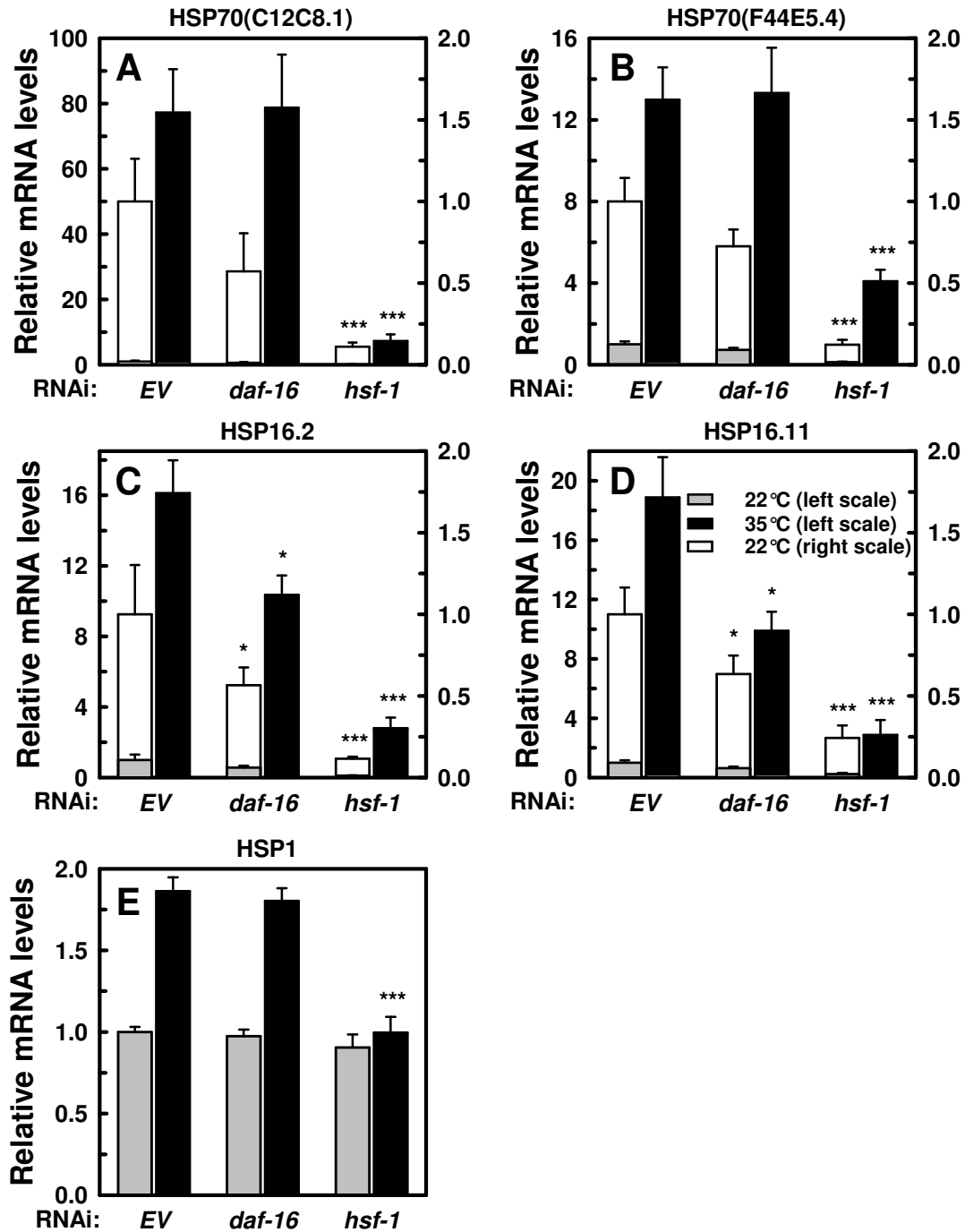
Figure S2

Fig. S2. Relative mRNA levels of (A) HSP70(C12C8.1), (B) HSP70(F44E5.4), (C) HSP16.2, (D) HSP16.11, and (E) HSP1 (HSC70) measured by quantitative RT-PCR for 3-day-old Q35 animals cultured at 22°C on either control bacteria (EV), *daf-16* RNAi bacteria, or *hsf-1* RNAi bacteria. On the third day since hatching, the animals were either kept at 22°C (gray bars - left scale and white bars - right scale) or heat-shocked at 35°C for 90 min (black bars - left scale). Data were normalized to *Act-4* levels. Values were calculated relative to the value of non-heat-shocked animals grown on control bacteria. Data are means ± SE of four independent biological samples. *, *P*<0.05; ***, *P*<0.001, comparing *daf-16* or *hsf-1* RNAi treatment with the corresponding EV treatment.

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Table S1

Table S1. Effect of treatment with NG-094 on the life-span of Q0 and Q35 animals cultured at 22°C.

Strain	NG-094 (1 mM)	Mean ± SE (days)[a]	P	Maximum ± SE (days)[d]	P	75th Percentile (days)[e]	Number of Animals that died/total[f]
Q0	-	17.3 ± 0.3		20.5 ± 0.4		19	75/80
Q0	+	17.9 ± 0.3	0.0915[b]	20.5 ± 0.3	0.9999[b]	20	77/80
Q35	-	14.2 ± 0.4	< 0.0001[b]	17.6 ± 0.3	< 0.0001[b]	16	80/80
Q35	+	15.2 ± 0.3	< 0.0001[b] 0.0972[c]	20.7 ± 0.4	0.6927[b] < 0.0001[c]	17	79/80

[a] Statistical analyses for survival were conducted using the standard chi-squared-based log rank (Mantel cox) test. All animals were cultured in parallel and transferred to fresh plates at the same time.

[b] Compared to Q0 animals that were not treated with NG-094, but analyzed at the same time.

[c] Compared to Q35 animals that were not treated with NG-094, but analysed at the same time.

[d] The maximum life-span was calculated as the mean life span of the eight worms of the population that had the longest life-spans. Statistical analyses were conducted using student's t test.

[e] The 75th percentile is the age when the fraction of animals alive reaches 0.25.

[f] The total number of observations equals the number of animals that died plus the number of censored. Animals that escaped, exploded, or bagged were censored at the time of the event.