Figure S1

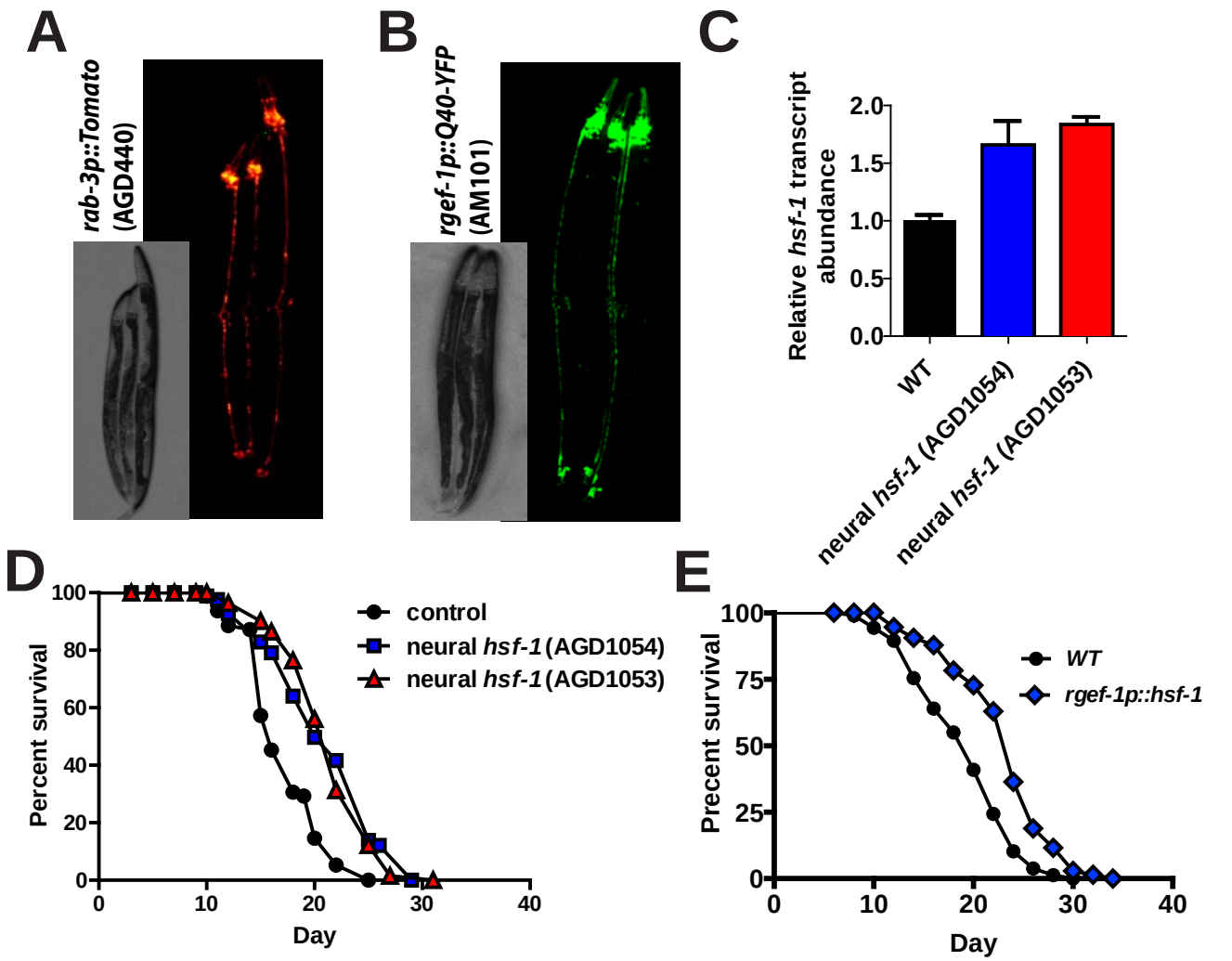

Figure S1. Ectopic overexpression of *hsf-1* in the nervous system extends lifespan of *C. elegans*, Related to Figure 1. (A-B) Characterization of neural specific promoters in which worms express transgenic copies of the fluorescent proteins, Tomato or Q40-YFP, under control of the *rab-3* (AGD440) and *gref-1* (AM101) promoter elements, respectively. L4 and day 1 adult animals were cultured at 20°C. **(C)** Quantitative RT-PCR of *hsf-1* mRNA from wild type (N2) and *rab-3p::hsf-1* transgenic worms (AGD1053 and AGD1054). **(D)** Lifespan survival curves of animals used in (C) were performed at 20°C. **(E)** Lifespan survival curves of wild type and *rgef-1p::hsf-1* (AGD1441). Lifespan statistics are found in Table 1.

Figure S2

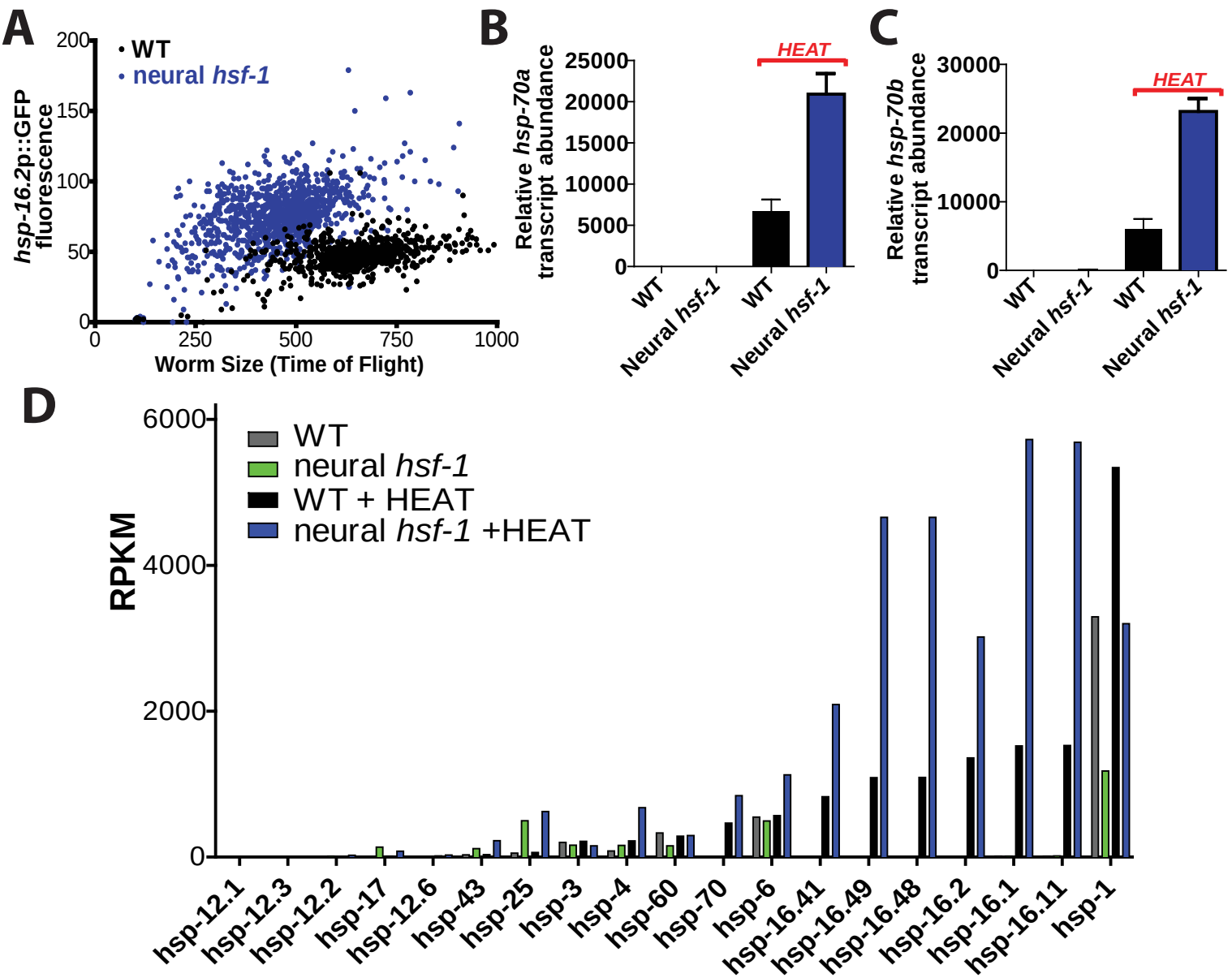

Figure S2. Neural overexpression of *hsf-1* increases the induction of the heat shock network, Related to Figure 2. **(A)** Flow cytometry shows the distribution of heat inducible GFP flourescence from the *hsp-16.2p::GFP* reporter in a worm population of wild type (CL2070) and *rab-3p::hsf-1* transgenic worms (AGD1448). **(B-C)** Quantitative RT-PCR of *hsp-70a(C12C8.1)* and *hsp-70b(F44E5.4)* mRNAs in wild type (N2) and *rab-3p::hsf-1* transgenic worms (AGD1289). **(D)** Quantification of all annotated heat shock transcripts (hsp) under heat stress (34°C) and at permissive temperatures (20°C) in wild type (N2) and neural *hsf-1* overexpressing worms (AGD1289). Transcript abundance was normalized by read per kilobase of transcript per million sequencing reads (RPKM) for each condition or genotype.

Figure S3

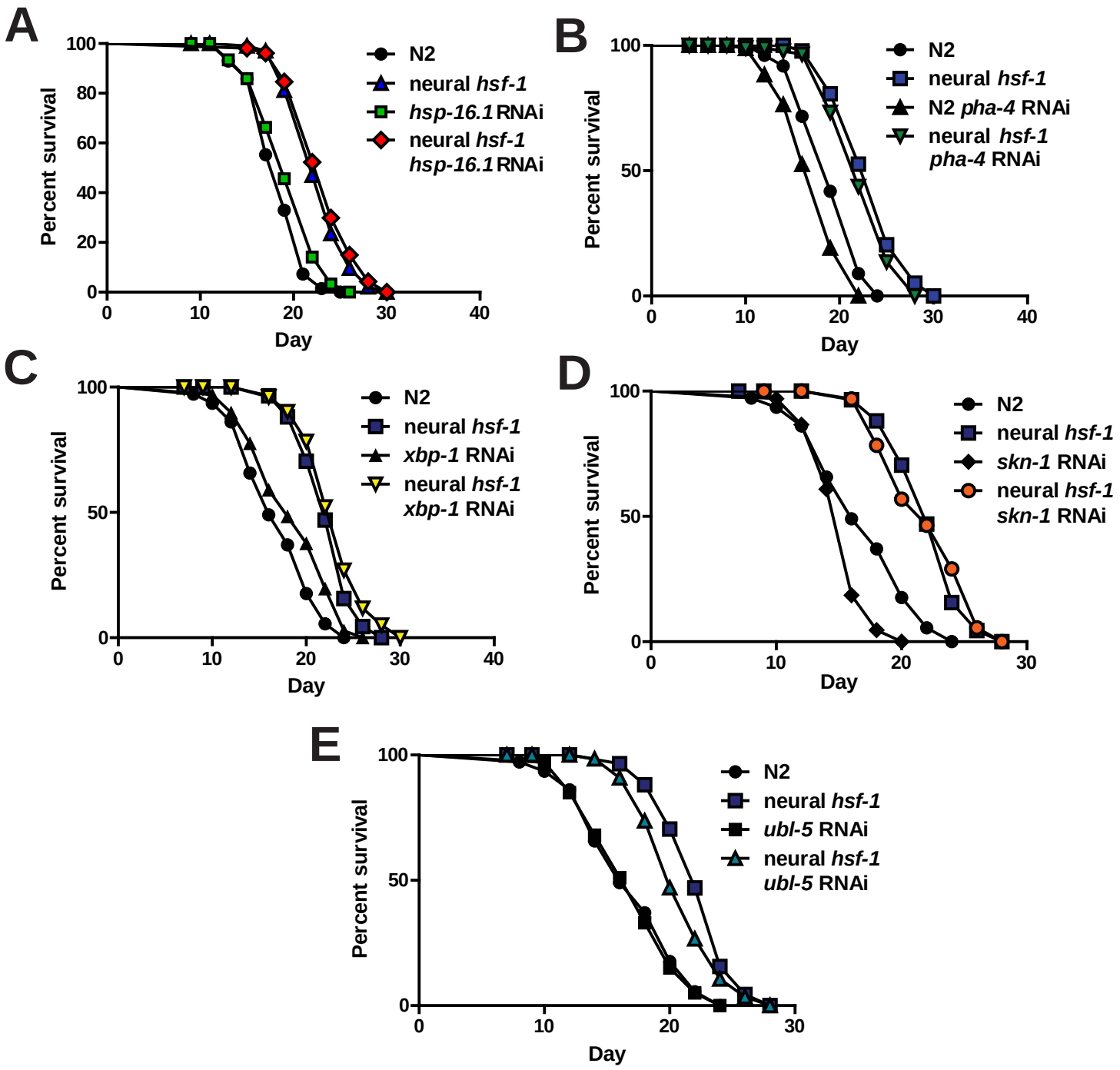

Figure S3. Lifespan extension by neural *hsf-1* overexpression is not dependent on *hsp-16* or several regulatory transcription factors, Related to Figure 3. (A-E) Lifespan survival curves of wild type (N2) and *rab-3p::hsf-1* transgenic worms (AGD1289) cultured at 20°C on HT115 *E. coli* harboring interferring RNAs for (A) *hsp-16*, (B) *pha-4*, (C) *xbp-1*, (D) *skn-1* and (E) *ubl-5*. Lifespan statistics are found in Table 1.

Figure S4

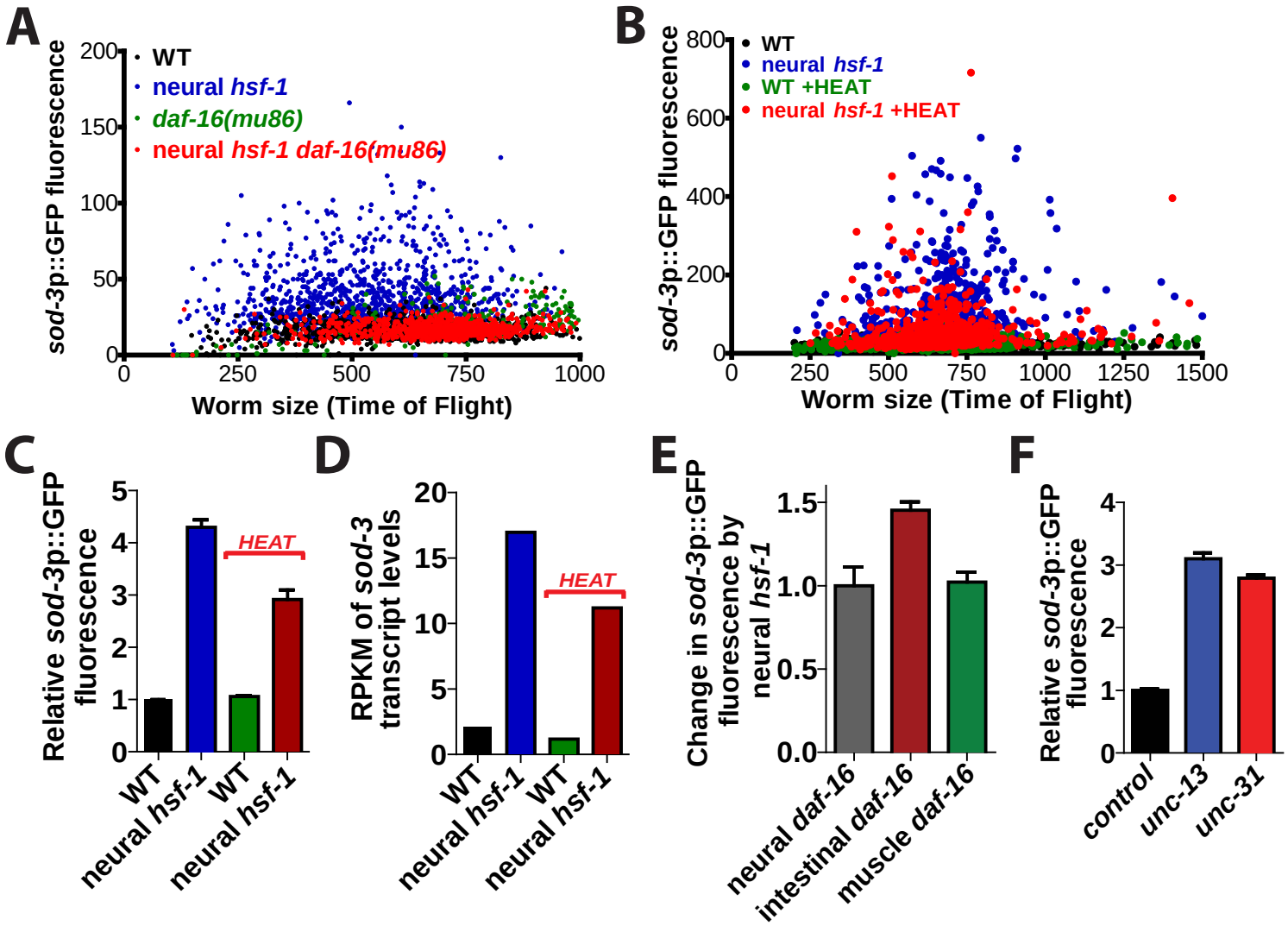

Figure S4. Dynamics for the activation of the DAF-16 transcriptional target, *sod-3*, Related to Figure 4. (A) DAF-16 dependent induction of *sod-3* by neural *hsf-1* overexpression. Flow cytometry shows the distribution of GFP fluorescence in individual *sod-3p::GFP* reporting animals (CF1553). The *daf-16(mu86)* allele was used to assess dependence of the *daf-16* on neural *hsf-1* induced *sod-3* expression in day 1 adult nematodes. Fluorescent expression of the *sod-3* reporter in the presnce of *rab-3p::hsf-1* (AGD1198) was compared to the same worms in the *daf-16(mu86)* null background (AGD1457). **(B-D)** Influence of heat shock (34°C) on *sod-3* activation. **(B)** Flow cytometry shows the distribution of GFP flouorescence in individual *sod-3p::GFP* reporting animals with (AGD1189) and without (CF1553) *rab-3p::hsf-1* under heat stress (34°C) and at permissive temperatures (20°C). **(C)** Quantification of GFP fluorescence from flow cytometry in (B). **(D)** Through RNA-sequencing analysis, transcript abundance of *sod-3* was normalized by reads per kilobase of transcript per million sequencing reads (RPKM) in wild type and *rab-3p::hsf-1* transgenic worms (AGD1289). **(E)** Neural *hsf-1* overexpression requires *daf-16* in the intestine to activate *sod-3.* Flow cytometry was used to quantify the distribution of GFP fluorescence in individual *sod-3p::GFP* reporting worms. Cultured at 20°C on OP50 *E. coli*, all worms were in a *daf-16(mu86)* null background and extopic expression of *daf-16* was rescued in indivdual tissues including the nervous system (AGD1313), intestine (AGD1315) and body-wall muscle (AGD1316). Shown is a normalized ratio of quantified GFP fluorescence (GFP signal from neural *hsf-1* overexpression/GFP signal from the respective control strain). **(F)** Abolishing dense-core vesicle and small-clear vesicle release from neurons induces expression of *sod-3*. Flow cytometry was used to quantify the distribution of GFP fluorescence in individual *sod-3p::GFP* reporting animals in *unc-13* (AGD1475) and *unc-31* (AGD1576) mutant backgrounds. Show in the quantification of GFP fluorescence.

Table S1

Element	Sequence	Times Present	e-value
DAE	CTTATCA	60	1.20E-03
HSE	TTCTAGAA	25	N.D.

seq	CTTATCA	TTCTAGAA
B0336.2.1	2	0
C04F12.1	1	0
C04F6.1.3	1	0
NP_495640.1	1	0
C06G8.1	2	0
C27H6.4b	0	0
C31B8.4	0	0
C32F10.8b.1	0	1
C34F11.3b	0	1
ADF1_CAEEL	0	1
C49G7.5	0	0
EEED8.4	0	0
F01F1.6.1	0	1
F08H9.4	0	1
F09E5.8	0	0
F11A6.1b	0	3
F11D11.3	1	0
F11G11.2	1	0
F15B9.1	1	0
F19B6.2b	0	0
F22A3.6a	1	1
F22B7.9	0	0
F28D1.5	1	0
NP_491282.1	0	0
F35C11.3	0	1
F35C8.6	0	1
NP_505928.1	1	0
F41C3.5.1	1	0
F45E1.4	1	1
F45F2.12	0	0
F46E10.10c.2	1	0
F47H4.2	1	0
F52E1.7a	1	0
F54C1.7.4	0	0
F55H2.2.1	0	0
F56B6.4c	1	0
F57B10.3b.2	0	0
F57F4.3	0	0
F58B3.1	0	0
H15N14.1f	0	0
ASP3_CAEEL	0	1
H23L24.5	0	0
K07E3.4b	1	0
K08E3.5a.1	2	1
K08F4.9	0	2
K10G4.3	0	0
K12C11.1.1	1	0
K12C11.4	0	1
M03F4.7b	1	0
M05D6.4	1	0
R05F9.10	1	0
R05F9.6	3	0
R05H10.5	0	0
R07E3.3	0	0
R107.7.2	0	1
R11H6.1.2	0	0
R12E2.11	0	0
T04H1.9	0	0
T05H4.6.3	0	0
T07C4.9b.2	1	0
T12A2.2.2	0	0
T14F9.1.2	0	1
T14G11.3	0	0
T19D12.4b	2	1
T20G5.1	1	0
T21B4.9	3	1
T25C8.2.1	4	1
W01F3.2	2	1
W03D2.6	2	0
W03G1.7b	2	0
Y105C5B.15	1	0
Y105E8B.1e.2	0	1
Y113G7B.2	0	0
Y47G7B.2	0	0
Y49A3A.2.2	1	0
Y50D7A.7.1	3	0
Y54E2A.11b.2	0	0
Y55B1AR.1.1	4	0
Y69H2.3d.2	0	0
Y87G2A.5	0	0
ZK1037.5	3	1
ZK666.6	2	0
total	60	25

Table S1. The *daf-16* associated element (DAE) was detected de novo as an overrepresented sequence on promoters of genes that are upregulated by *hsf-1* overexpression and correlate with heat protection, Related to Figure 3. The canonical *C. elegans* heat shock element (HSE) was not detected (N.D.) as an overrepresented sequence on promoters of genes upregulated by elevated *hsf-1*. The CTTATCA and TTCTAGAA nucleotide sequences correspond to the DAE and HSE respectively.