SUPPLEMENTARY INFORMATION

Supplementary Table 1: Temperature-associated reduced brood size of *daf-2(e1370)* is *skn-1*-independent

Strain	Temperature	# Po	# F1 progeny [mean ± SEM]	P-value vs N2	P-value vs *daf-2*
wild type (N2)	15°C →15°C	5	298.8 ± 29.8		
skn-1(zu135)	15°C →15°C	5	326.4 ± 32.3	n.s.	n.s.
daf-2(e1370)	15°C →15°C	5	333.0 ± 34.2	n.s.	
daf-2(e1370); skn-1(zu135)	15°C →15°C	5	317.6 ± 16.3	n.s.	n.s.
wild type (N2)	**15°C →25°C**	5	223.6 ± 24.1		
skn-1(zu135)	**15°C →25°C**	5	231.0 ± 26.7	n.s.	P < 0.001
daf-2(e1370)	**15°C →25°C**	5	45.2 ± 5.6	P < 0.001	
daf-2(e1370); skn-1(zu135)	**15°C →25°C**	5	24.4 ± 6.5	P < 0.001	n.s.

Temperature-associated reduced motility phenotype of *daf-2(e1370)* is *skn-1*-independent

Strain	Temperature	motility behavior scored 2 days later
wild type (N2)	15°C →15°C	crawling (N=62)
skn-1(zu135)	15°C →15°C	crawling (N=33)
daf-2(e1370)	15°C →15°C	crawling (N=30)
daf-2(e1370); skn-1(zu135)	15°C →15°C	crawling (N=69)
wild type (N2)	**15°C →25°C**	crawling (N=60)
skn-1(zu135)	**15°C →25°C**	crawling (N=31)
daf-2(e1370)	**15°C →25°C**	immobile (N=64) and crawling (N=1)
daf-2(e1370); skn-1(zu135)	**15°C →25°C**	immobile (N=74)

Two dauer-related traits that are characteristic of Class 2 *daf-2* mutants are reduced brood-size and reduced mobility[1]. To assess brood size, animals were maintained and allowed to develop at 15°C. L4 animals were either kept at 15°C, or shifted to 25°C. Each worm was transferred daily onto a fresh NGM OP50 plate and the number of progeny was scored. *P* values were determined with one-way ANOVA post hoc Tukey. n.s. = not significant. For motility assessment, animals were maintained and allowed to develop at 15°C, then one-day adults were either kept at 15°C, or shifted to 25°C. Two days later motility behavior was scored by observing each worm for spontaneous movements over 30 seconds.

Supplementary Table 2. *skn-1*-dependence of *daf-2* lifespan extension under dauer-independent conditions.

Strain	Temp [°C]	Mean lifespan ± S.E.M. [Days]	75th per-centile [Days]	N assayed / Initial N	% mean lifespan change to N2 or control	% mean lifespan change to *skn-1*	P-value (log-rank) vs. N2	P-value (log-rank) vs. *skn-1*	P-value (log-rank) vs. *daf-2*	Figure
Trial at 15°C *										
wild type (N2)	15	26.5 ± 0.6	31	94/115						1a
skn-1(zu67)	15	19.7 ± 0.4	21	84/99	-26		<0.0001			1a
daf-2(e1370)	15	38.0 ± 0.9	45	134/144	+43	+93	<0.0001	<0.0001		1a
daf-2(e1370); skn-1(zu67)	15	18.5 ±0.4	21	95/105		-6	<0.0001	0.0676	<0.0001	1a
Trial at 15°C *										
wild type (N2)	15	22.1 ± 0.4	25	99/114						1a
skn-1(zu67)	15	17.1 ± 0.3	19	102/114	-23		<0.0001			1a
daf-2(e1370)	15	37.4 ± 0.9	46	131/138	+69	+119	<0.0001	<0.0001		1a
daf-2(e1370); skn-1(zu67)	15	17.7 ±0.2	19	113/116		+4	<0.0001	0.3286	<0.0001	1a
Trial at 15°C *										
wild type (N2)	15	21.5 ± 0.5	25	86/103						1a
skn-1(zu67)	15	14.2 ± 0.3	16	92/102	-34		<0.0001			1a
daf-2(e1370)	15	34.3 ± 1.0	42	107/114	+60	+142	<0.0001	<0.0001		1a
daf-2(e1370); skn-1(zu67)	15	15.3 ±0.2	16	100/106		+8	<0.0001	0.0654	<0.0001	1a
Trial at 15°C										
wild type (N2)	15	29.2 ± 0.5	34	105/110						
skn-1(zu129)	15	16.4 ± 0.4	20	107/119	-44		<0.0001			
daf-2(e1370)	15	43.6 ± 1.0	50	100/102	+49	+166	<0.0001	<0.0001		
daf-2(e1370); skn-1(zu129)	15	18.1 ± 0.6	22	113/123		+10	<0.0001	0.0290	<0.0001	
Trial at 15°C										
wild type (N2)	15	24.9 ± 0.4	27	106/125						
skn-1(zu135)	15	18.7 ± 0.3	20	110/124	-25		<0.0001			
daf-2(e1370)	15	33.4 ± 0.7	38	125/133	+35	+79	<0.0001	<0.0001		
daf-2(e1370); skn-1(zu135)	15	20.3 ±0.4	22	91/107		+9	<0.0001	0.0004	<0.0001	
Trial at 15°C										
wild type (N2)	15	26.6 ± 0.3	27	97/109						
skn-1(zu135)	15	16.2 ± 0.2	18	93/103	-39		<0.0001			
daf-2(m596)	15	36.7 ± 0.7	43	112/117	+38	+127	<0.0001	<0.0001		
daf-2(m596); skn-1(zu135)	15	17.5 ± 0.4	18	91/101		+8	<0.0001	0.0062	<0.0001	
Trial at 15°C without FUdR										
wild type (N2) without FUdR	15	26.7 ± 0.7	30	75/112						
skn-1(zu135) without FUdR	15	18.4 ± 0.4	20	91/119	-31		<0.0001			
daf-2(e1370) without FUdR	15	35.6 ± 1.0	44	64/104	+33	+93	<0.0001	<0.0001		
daf-2(e1370); skn-1(zu135) without FUdR	15	18.5 ±0.4	20	94/116		+1	<0.0001	0.9726	<0.0001	
Trial at 15°C and 20°C **										
wild type (N2)	15	22.3 ± 0.4	24	91/112						
skn-1(zu129)[#]	15	19.4 ± 0.7	20	95/122	-13		<0.0001		<0.0001	
skn-1(zu135)[$]	15	17.8 ± 0.2	20	102/121	-20		<0.0001		<0.0001	
daf-2(e1370)	15	35.1 ± 0.7	41	123/129	+57	+80[#], +97[$]	<0.0001			
daf-2(e1370); skn-1(zu129)	15	20.6 ±0.4	21	132/156		+6	0.0052	0.0140	<0.0001	
daf-2(e1370); skn-1(zu135)	15	19.0 ± 0.3	21	108/131		+7	<0.0001	0.0002	<0.0001	
wild type (N2)	**20**	19.8 ± 0.3	21	116/140						
skn-1(zu135)	**20**	14.4 ± 0.5	15	94/111	-38		<0.0001			
daf-2(e1370)	**20**	30.5 ± 0.4	32	141/148	+54	+112	<0.0001	<0.0001		
daf-2(e1370); skn-1(zu135)	**20**	29.4 ±0.7	35	111/134		+104	<0.0001	<0.0001	0.7634	
Trial at 15°C and 20°C										
wild type (N2)	15	25.7 ± 0.3	27	85/92						
skn-1(zu135)	15	17.7 ± 0.3	20	79/85	-31		<0.0001			
daf-2(m596)	15	41.8 ± 0.9	53	139/142	+63	+136	<0.0001	<0.0001		
daf-2(m596); skn-1(zu135)	15	17.1 ± 0.3	18	99/112		-3	<0.0001	0.3016	<0.0001	
wild type (N2)	**20**	23.7 ± 0.3	25	84/92						
skn-1(zu135)	**20**	15.3 ± 0.2	15	90/100	-35		<0.0001			
daf-2(m596)	**20**	38.0 ± 1.1	48	80/83	+60	+148	<0.0001	<0.0001		
daf-2(m596); skn-1(zu135)	**20**	33.8 ± 1.0	41	99/112		+121	<0.0001	<0.0001	0.0130	

Trial at 15°C									
wild type (N2)	15	22.1 ± 0.3	23	80/99					
skn-1(zu67)	15	17.5 ± 0.6	19	54/83	-20		<0.0001		
daf-2(e1368)	15	26.9 ± 0.4	30	136/149	+22	+54	<0.0001	<0.0001	
daf-2(e1368); skn-1(zu67)	15	16.7 ± 0.3	17	82/99		-5	<0.0001	0.1435	<0.0001
Trial downshifted at the first day of adulthood from 20°C to 15°C ***									
wild type (N2)	20->15	27.0 ± 0.4	30	108/118					
skn-1(zu67)	20->15	17.9 ± 0.4	21	94/108	-34		<0.0001		
daf-2(e1370)	20->15	35.8 ± 1.0	44	101/124	+33	+100	<0.0001	<0.0001	
daf-2(e1370); skn-1(zu67)	20->15	21.3 ±0.5	23	95/108		+19	<0.0001	<0.0001	<0.0001
wild type (N2)	**20->20**	23.9 ± 0.6	27	56/59					
skn-1(zu67)	**20->20**	13.9 ± 0.3	14	34/42	-41		<0.0001		
daf-2(e1370)	**20->20**	32.1 ± 0.8	35	54/67	+34	+130	<0.0001	<0.0001	
daf-2(e1370); skn-1(zu67)	**20->20**	30.1 ±1.4	35	39/46		+117	<0.0001	<0.0001	0.8980
Trial at 15°C									
wild type (N2) L4440(RNAi)	15	23.5 ± 0.5	26	113/132					
wild type (N2) *daf-2*(RNAi)	15	38.1 ± 0.9	48	128/140	+62	+120	<0.0001	<0.0001	
skn-1(zu135) L4440(RNAi)	15	17.3 ± 0.6	19	89/108	-26		<0.0001		<0.0001
skn-1(zu135) daf-2(RNAi)	15	18.2 ± 0.5	19	118/144		+5	<0.0001	0.2623	<0.0001
Trial at 20°C									
wild type (N2) L4440(RNAi)	20	25.1 ± 0.4	27	100/115					
wild type (N2) *daf-2*(RNAi)	20	38.4 ± 0.7	43	102/111	+53	+134	<0.0001	<0.0001	
skn-1(zu135) L4440(RNAi)	20	16.5 ± 0.5	17	63/77	-34		<0.0001		<0.0001
skn-1(zu135) daf-2(RNAi)	20	19.7 ± 0.7	25	88/106		+19	<0.0001	0.0002	<0.0001
Trial at 15°C and 20°C									
wild type (N2) *daf-2*(RNAi)	15	42.0 ± 1.7	51	41/56		+79		<0.0001	
skn-1(zu135) daf-2(RNAi)	15	23.5 ± 1.1	28	36/50					
wild type (N2) *daf-2*(RNAi)	**20**	35.5 ± 0.6	38	53/59		+79		<0.0001	
skn-1(zu135) daf-2(RNAi)	**20**	19.8 ± 0.8	23	42/51					
Trial at 15°C and 20°C									
wild type (N2) L4440(RNAi)	15	29.7 ± 0.5	32	107/134					
wild type (N2) *daf-2*(RNAi)	15	50.2 ± 1.0	60	129/137	+69	+87	<0.0001	<0.0001	
skn-1(zu135) L4440(RNAi)	15	26.9 ± 0.5	30	110/130	-9		<0.0001		<0.0001
skn-1(zu135) daf-2(RNAi)	15	28.0 ± 0.7	32	77/91		+4	0.0274	0.0130	<0.0001
wild type (N2) L4440(RNAi)	**20**	24.8 ± 0.2	26	114/129					
wild type (N2) *daf-2*(RNAi)	**20**	45.0 ± 0.8	49	96/104	+81	+117	<0.0001	<0.0001	
skn-1(zu135) L4440(RNAi)	**20**	19.6 ± 0.4	24	100/117	-21		<0.0001		<0.0001
skn-1(zu135) daf-2(RNAi)	**20**	23.4 ± 0.6	26	100/117		+19	0.8297	<0.0001	<0.0001
Trial of crude dauerpheromone at 25°C									
wild type (N2) control	**25**	12.6 ± 0.2	14	28/30					
wild type (N2) crude dauer pheromone	**25**	15.9 ± 0.5	19	30/30	+26		<0.0001		
Trial of crude dauerpheromone at 25°C									
wild type (N2) L4440(RNAi) control	**25**	12.3 ± 0.4	14	27/30					
wild type (N2) L4440(RNAi) crude dauer pheromone	**25**	17.8 ± 0.6	20	28/30	+45	+60	<0.0001	<0.0001	
wild type (N2) *daf-2*(RNAi) control	**25**	20.9 ± 0.7	24	28/30	+70	+88	<0.0001	<0.0001	
wild type (N2) *daf-2*(RNAi) crude dauer pheromone	**25**	23.7 ± 1.0	27	29/30	+92	+114	<0.0001	<0.0001	0.0025
skn-1(zu135) L4440(RNAi) control	**25**	11.1 ± 0.3	13	29/30	-10		0.0016		<0.0001
skn-1(zu135) L4440(RNAi) crude dauer pheromone	**25**	15.3 ± 0.6	18	27/30	+24	+38	<0.0001	<0.0001	<0.0001
skn-1(zu135) daf-2(RNAi) control	**25**	11.2 ± 0.3	13	25/30	-9	+1	0.0163	0.8427	<0.0001
skn-1(zu135) daf-2(RNAi) crude dauer pheromone	**25**	17.1 ± 0.7	20	28/30	+39	+54	<0.0001	<0.0001	0.0003

Analyses that were performed in parallel are grouped together. *e1370* and *m596* are Class 2 *daf-2* alleles, and *e1368* is a Class 1 mutant[1]. (N) = number of animals observed; Lifespans were measured starting from the L4 stage. Animals that left the plates, buried into the agar, bagged, or exploded were censored. *A merger of these three trials is plotted in Fig. 1c. *** animals were maintained at 20°C for several generations, then at the first day of adulthood were either kept at 20°C or downshifted to 15°C. # indicates percent *daf-2(e1370)* lifespan change to *skn-1(zu129)*. $ indicates percent *daf-2(e1370)* lifespan change to *skn-1(zu135).* L4440 empty vector was used as the control in RNAi experiments. 5-Fluoro-2'deoxyuridine (FUdR) was used for all lifespan assays unless otherwise indicated. $ P-value (log-rank) vs. *daf-2(e1368) L4440(RNAi).*

For Supplementary Table 3 (complete list of *daf-2;skn-1*-up- and downregulated gene sets), please see the separate Excel file.

Supplementary Table 4. RNAi knockdown of *daf-2;skn-1*-regulated genes affects adult lifespan

Strain / **RNAi**	Mean lifespan ± S.E.M. [Days]	75th percentile [Days]	N dead/ Initial N	% mean lifespan change to control	P-value (log-rank) vs. control	Figure
Trial of genes from the SKN-1-upregulated *daf-2(-)* set at 20°C						
daf-2(e1368) RNAi **L4440 (control)**	26.5 ± 0.6	31	64/84			Ext.Fig. 2f
daf-2(e1368) RNAi ***col-13***	23.7 ± 0.5	25	65/84	-11	0.0004	Ext.Fig. 2f
daf-2(e1368) RNAi **K04C2.5**	28.5 ± 0.7	34	62/70	+8	0.0253	
daf-2(e1368) RNAi ***col-127***	25.9 ± 0.6	31	58/74	-2	0.5246	
daf-2(e1368) RNAi ***nit-1***	21.6 ± 0.5	24	57/85	-18	<0.0001	Ext.Fig. 2f
daf-2(e1368) RNAi ***pgp-7***	25.9 ± 0.7	31	61/84	-3	0.6322	
daf-2(e1368) RNAi **C06G1.1**	28.4 ± 0.7	31	65/84	+7	0.0322	
Trial of genes from the SKN-1-upregulated *daf-2(-)* set at 20°C						
daf-2(e1368) RNAi **L4440 (control)**	28.0 ± 0.6	30	78/85			Ext.Fig. 2f
daf-2(e1368) RNAi **F55G11.2**	22.1 ± 0.6	25	78/85	-21	<0.0001	Ext.Fig. 2f
daf-2(e1368) RNAi **K10H10.4**	22.9 ± 0.6	27	76/85	-18	<0.0001	Ext.Fig. 2f
daf-2(e1368) RNAi ***dct-5***	23.8 ± 0.6	27	73/85	-15	<0.0001	Ext.Fig. 2f
daf-2(e1368) RNAi **F29G6.1**	26.3 ± 0.6	30	72/85	-6	0.0785	
daf-2(e1368) RNAi ***nspb-7***	29.5 ± 0.6	32	72/85	+5	0.0454	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	16.8 ± 0.2	18	64/87			Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-5***	18.0 ± 0.3	20	56/68	+7	<0.0001	Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-8***	17.7 ± 0.3	19	63/80	+5	0.0038	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	18.5 ± 0.3	20	60/84			Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-12***	18.9 ± 0.3	20	56/84	+2	0.4023	
rrf-3(pk1426) RNAi ***skr-13***	19.4 ± 0.3	21	68/84	+5	0.0169	Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-17***	19.6 ± 0.4	23	56/72	+6	0.0219	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	20.5 ± 0.3	23	73/85			Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-5***	21.8 ± 0.3	23	72/84	+6	0.0057	Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-12***	20.6 ± 0.3	23	79/83	0	0.7817	
rrf-3(pk1426) RNAi ***skr-13***	21.7 ± 0.3	23	59/85	+6	0.0032	Ext.Fig. 2h
rrf-3(pk1426) RNAi ***skr-15***	20.1 ± 0.3	23	81/86	-2	0.5442	
rrf-3(pk1426) RNAi ***skr-17***	19.1 ± 0.4	20	65/82	-7	0.0374	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
N2 RNAi **L4440 (control)**	15.2 ± 0.3	16	45/80			
N2 RNAi **Y43C5A.3**	16.2 ± 0.3	17	46/65	+7%	0.0015	
N2 RNAi **F48C1.8**	15.8 ± 0.3	17	31/60	+4%	0.1308	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	21.0 ± 0.7	26	42/81			
rrf-3(pk1426) RNAi ***scl-5***	23.1 ± 0.5	26	74/109	+15%	0.0015	
rrf-3(pk1426) RNAi **Y43C5A.3**	21.6 ± 0.4	23	75/100	+3%	0.7794	
rrf-3(pk1426) RNAi **F48C1.8**	21.5 ± 0.4	26	77/91	+2%	0.9321	
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	17.8 ± 0.4	21	49/76			
rrf-3(pk1426) RNAi **control (elpc-4)** [$]	23.2 ± 0.5	25	57/109			Ext.Fig. 2g
rrf-3(pk1426) RNAi **T20D4.7**	25.4 ± 0.3	28	74/138	+9%	0.0035	Ext.Fig. 2g
rrf-3(pk1426) RNAi ***cnc-2***	26.2 ± 0.5	29	64/85	+13%	0.0004	Ext.Fig. 2g
rrf-3(pk1426) RNAi ***cnc-7***	27.0 ± 0.5	29	48/78	+16%	<0.0001	Ext.Fig. 2g
rrf-3(pk1426) RNAi **Y50D4B.6**	26.3 ± 0.5	28	47/92	+13%	0.0006	
rrf-3(pk1426) RNAi ***nhr-231/nhr-170***	26.5 ± 0.5	29	66/149	+14%	<0.0001	Ext.Fig. 2g
Trial of genes from the SKN-1-downregulated *daf-2(-)* set at 20°C						
rrf-3(pk1426) RNAi **L4440 (control)**	24.4 ± 0.3	25	59/85			
rrf-3(pk1426) RNAi **GFP**	24.5 ± 0.3	25	57/77	0%	0.9282	
rrf-3(pk1426) RNAi ***nspe-7***	24.9 ± 0.4	27	50/77	+2%	0.2727	
rrf-3(pk1426) RNAi **Y39B6A.24**	25.7 ± 0.4	28	56/78	+5%	0.0161	
rrf-3(pk1426) RNAi ***cnc-2***	26.6 ± 0.5	28	60/81	+9%	0.0002	
rrf-3(pk1426) RNAi ***nhr-147***	24.5 ± 0.2	25	68/90	0%	0.9025	
rrf-3(pk1426) RNAi **F48C1.8**	26.0 ± 0.4	28	69/90	+7%	0.0022	
rrf-3(pk1426) RNAi ***cnc-7***	24.8 ± 0.4	27	69/92	+2%	0.3516	
rrf-3(pk1426) RNAi **Y50D4B.6**	24.0 ± 0.4	25	70/90	-2%	0.3516	
rrf-3(pk1426) RNAi ***cdr-5***	25.4 ± 0.3	27	74/91	+4%	0.0347	

Trials that were performed in parallel are grouped. (N) = number of animals observed; Lifespan was measured from the first day of adulthood. Animals that left the plates, buried into the agar, bagged, or exploded were censored. $ in this trial *elpc-4* RNAi was used as the control. L4440 empty vector was otherwise used as the control. In some cases, knockdown was performed in the RNAi-sensitive strain *rrf-3(pk1426)*[2]. Ext.Fig. stands for Extended Data Figure.

Supplementary Table 5. SKN-1-downregulated *daf-2(-)* genes for which RNAi knockdown was previously found to prolong lifespan

SAM Rank	Gene	Brief description	Reference
2	*pes-2.2*	predicted F box domain	[3]
19	F15D3.8		[4]
42	Y39B6A.24	Aspartyl protease	[4]
53	C31B8.4		[4]
55	Y43C5A.3		[4]
73	Y47H9C.1	Transmembrane glycoprotein	[4]
91	F56D5.5	Tropomysin domain	[5]
159	*pos-1*	CCCH-type Zn-finger protein	[6]
175	F55B11.3		[7]
185	F02D10.6		[8]
208	Y46G5A.20		[4]
249	*aps-3*	AP-3 subunit, intracellular membrane trafficking	[9]
258	ZC196.4		[4]
330	*skr-5*	SCF ubiquitin ligase, Skp1 component	[4]
334	D1054.14	Predicted PRP38-like splicing factor	[6]

Supplementary Table 6. SKN-1 is required for the oxidative stress resistance of *daf-2* mutants

Strain	Mean survival ± S.E.M. [Hours]	75th percentile [Hours]*	N died from stress/ Initial N	% mean change in survival to N2	P-value (log-rank) vs. N2	P-value (log-rank) vs. *daf-2(e1370)*
Trial performed in parallel						
wild type (N2) in 5mM arsenite	11.7 ± 1.2	21	35/36			<0.0001
skn-1(zu67) in 5mM arsenite	6.7 ± 0.5	9	37/37	-42%	<0.0001	<0.0001
daf-2(e1370) in 5mM arsenite	23.8 ± 1.9	31	36/39	+51%	<0.0001	
daf-2(e1370); skn-1(zu67) in 5mM arsenite	8.1 ± 1.0	10	37/39	-31%	0.0263	<0.0001
Trial performed in parallel						
wild type (N2) in 5mM arsenite	18.1 ± 1.8	28	29/35			<0.0001
skn-1(zu67) in 5mM arsenite	8.1 ± 0.8	11	29/30	-55%	<0.0001	<0.0001
daf-2(e1370) in 5mM arsenite	48.1 ± 3.9	72	42/46	+166%	<0.0001	
daf-2(e1370); skn-1(zu67) in 5mM arsenite	9.9 ± 2.5	11	31/32	-45%	<0.0001	<0.0001

Trial with 3 replicates

Strain	Mean survival **after 21** hours ± S.E.M. [%]	N died from stress/ Initial N**	P-value vs. N2	P-value vs. *daf-2(e1370)*
wild type (N2) in 5mM arsenite	22 ± 7.5	31/35		<0.001
skn-1(zu67) in 5mM arsenite	0 ± 0.0	39/39	>0.05	<0.001
daf-2(e1370) in 5mM arsenite	70.3 ± 6.3	40/43	<0.001	
daf-2(e1370); skn-1(zu67) in 5mM arsenite	3.0 ± 3.0	35/39	>0.05	<0.0011

All assays were performed at 20°C. (As) = Arsenite; (N) = number of animals observed. Animals that exploded were censored. *P* values for survival curves were determined by Log-Rank (for survival plots) or one-way ANOVA post hoc Tukey (survival after 21 hours). In parallel control experiments performed without addition of arsenite, the frequency of death was negligible for all genotypes. SKN-1 was also required for the increased resistance of *daf-2* mutants to oxidative stress from tert-butyl hydrogen peroxide[10].

Supplementary Table 7. Overlap between SKN-1-regulated gene sets from _daf-2(-)_, normal, and oxidative stress conditions

Functional Annotation Cluster	Enrichment Score	Genes
9% of SKN-1-upregulated _daf-2(-)_ genes (40 genes) were shared with _skn-1_-upregulated lists identified under normal and oxidative stress (arsenite) conditions[4] (Extended Data Fig. 3g)		
Glutathione S-transferase	4.85	_gst-4_, **_gst-10_**, _gst-12_, **_gst-13_**, _gst-14_, _gst-38_, **F56A4.4**, Y52E8A.3
Proteolysis	1.81	K10C2.1*, K10C2.3, _pcp-2_ ,T16G12.1*
Ageing	1.22	_dod-17_, _dod-24_
others		F55G11.2, _nit-1_, F56D5.3, **C32H11.4**, _pgp-7*_, _nspa-8_, **C32H11.3,** _ifp-1*_, _asm-2*_, F14D7.6*, C33G3.4*, _galt-1*_, _ugt-22*_, _ubc-23*_, F56A4.4, F30F8.5, C46H11.2*, F55E10.6, C06E7.4, C14H10.2, D1086.3*, F09B12.3*, F38B6.4*, Y102A11A.3*, ZK287.1, C26B9.5*
3% of SKN-1-downregulated _daf-2(-)_ genes (13/477) were shared with _skn-1_-downregulated lists identified under normal and oxidative stress conditions[4]		
others		B0348.2, C06E4.8, C31B8.4, _skr-5_, _vang-1_, **Y39B6A.24,** Y43C5A.3, Y46G5A.20, **Y47H9C.1,** Y69A2AR.25, **ZC196.4,** ZK154.5

Genes that were shown to be important for oxidative stress resistance[4] are underlined and shown in bold.

* indicates SKN-1-upregulated _daf-2(-)_ genes that overlap only with _skn-1_-dependent upregulated genes that were identified under normal conditions.

Supplementary Table 8. Overrepresented gene ontology (GO) terms for *skn-1*-dependent up- and down-regulated targets under reduced insulin/IGF-1 signaling (IIS) conditions

GO terms enriched in SKN-1-upregulated *daf-2(-)* genes

48 of 175 collagen genes:
col-7, col-10, col-12, col-13, col-17, col-49, col-58, col-60, col-61, col-65, col-66, col-76, col-81, col-88, col-89, col-91, col-92, col-93, col-97, *col-113, col-115, col-120, col-125, col-127, col-130, col-133, col-136, col-137, col-141, col-144, col-145, col-146, col-150, col-152, col-155, col-156, col-161, col-162, col-165, col-166, col-167, col-168, col-169, col-170, col-174, col-176, col-180, ram-2*

7 of 34 cuticulin precursor genes:
cut-4, cutl-11, cutl-12, cutl-20, cutl-21, cutl-23, ram-5

22 of 37 metallopeptidases genes:
C10C5.3, C10C5.5, F45E1.5, *nas-1, nas-7, nas-8, nas-22, nas-27, nas-29, nas-30, nas-36, nas-38,* R11A5.7, T06A4.3, T16A9.4, T16G12.1, T19D2.1, T21D11.1, *toh-1,* W01A8.6, Y18H1A.9, Y47G6A.19

6 of 44 glutathione S-transferase genes:
gst-4, gst-10, gst-12, gst-13, gst-14, gst-38

10 of 65 UDP-glucuronosyl transferase genes:
F54C1.1, *grl-27, ugt-4, ugt-5, ugt-6, ugt-10, ugt-22, ugt-47, ugt-51, ugt-61*

GO terms enriched in SKN-1-downregulated *daf-2(-)* genes

9 of 21 Skp1-related ubiquitin ligase components:
skr-5, skr-7, skr-8, skr-10, skr-12, skr-13, skr-14, skr-15, skr-17

22 of 390 genes containing a F-box motif:
C47F8.1, F08F3.6, *fbxa-132, fbxa-135, fbxa-153, fbxa-164, fbxa-170, fbxa-215, fbxa-36, fbxa-83, fbxb-59, fbxb-67, fbxc-28, fbxc-32, fbxc-33, fbxc-36, fbxc-42, fbxc-50,* H04D03.4, *pes-2.2*, *skpt-1,* T02G6.5

10 of 21 Broad/complex/Tramtrack/Bric a brac (BTB) domain containing genes:
B0281.5, *btb-6, btb-7, bath-9, btb-11, btb-17, bath-28,* C37C3.9, 10 C17F4.8, *math-10*

5 predicted E3 ligases:
apc-11, B0416.4, F53F8.3, T01C3.3, Y52E8A.4

3 predicted ubiquitin
C16C8.11, F52C6.3, F54D10.7

5 protease
cpr-4, nas-3, T13B5.9, *try-1,* Y39B6A.24

The total number of genes in a given GO term was obtained from wormbase.org WS231.

Supplementary Table 9. Expression of *SKN-1-upregulated daf-2(-)* collagen genes

Gene	SAM#*	SKN-1 site**	SKN-1 ChIP***	Lifespan	Tissues expressioned in adults (wormbase.org)	Expression Cluster (wormbase.org)
col-13	2	0	0	yes	Strong expression in adults	no tissue-specific cluster enrichment
col-127	7	0	0	yes	unknown	L1_muscle enriched
col-66	16	0	0	no	unknown	no tissue-specific cluster enrichment
col-12	17	0	0	yes	Strong expression in adults	L1_muscle enriched
col-146	18	0	0	no	unknown	L1_muscle enriched
col-176	23	1	0	yes	unknown	L1_muscle enriched, muscle genes
col-65	26	1	0	yes	unknown	L1_muscle enriched, muscle genes
col-133	30	1	0	yes	unknown	L1_muscle enriched, muscle genes
col-60	41	0	0	no	unknown	no tissue-specific cluster enrichment
col-120	50	0	0	yes	unknown	no tissue-specific cluster enrichment
col-141	60	1	0	yes	unknown	no tissue-specific cluster enrichment
col-162	63	0	0	no	unknown	no tissue-specific cluster enrichment
col-180	66	0	0	yes	hypodermis	L1_muscle enriched
col-144	68	2	0	yes	hypodermis	L1_muscle enriched
col-115	73	0	0	no	unknown	no tissue-specific cluster enrichment
col-88	76	0	0	no	unknown	L1_muscle enriched
col-7	78	2	0	no	hypodermal, seam cells	L1_muscle enriched
col-136	82	0	0	no	unknown	no tissue-specific cluster enrichment
col-167	92	0	0	yes	unknown	no tissue-specific cluster enrichment
col-168	100	0	0	yes	unknown	no tissue-specific cluster enrichment
col-145	116	1	0	no	unknown	L1_muscle enriched
col-130	127	0	0	no	intestine, vulva (in situ)[11]	L1_muscle enriched
col-150	147	0	0	no	unknown	no tissue-specific cluster enrichment
col-166	153	0	0	no	unknown	no tissue-specific cluster enrichment
col-58	155	0	0	no	unknown	no tissue-specific cluster enrichment
col-156	165	1	0	no	unknown	no tissue-specific cluster enrichment
col-76	167	1	0	no	unknown	muscle enriched (emb)
col-125	171	1	0	no	unknown	L1_muscle enriched
col-89	172	0	0	yes	intestine, vulva, neurons[12]	no tissue-specific cluster enrichment
col-152	183	0	0	no	unknown	no tissue-specific cluster enrichment
col-91	203	1	0	no	unknown	no tissue-specific cluster enrichment
col-174	207	1	0	no	unknown	no tissue-specific cluster enrichment
col-92	212	0	0	no	unknown	L1_muscle enriched
col-170	225	2	0	no	unknown	no tissue-specific cluster enrichment
col-118	239	0	0	no	unknown	L1_muscle enriched
col-93	245	0	0	no	unknown	no tissue-specific cluster enrichment
col-137	265	0	0	no	unknown	no tissue-specific cluster enrichment
col-161	289	0	0	no	unknown	L1_muscle enriched
col-155	290	0	0	no	unknown	L1_muscle enriched
col-113	301	0	0	no	unknown	no tissue-specific cluster enrichment
col-49	304	0	0	no	unknown	no tissue-specific cluster enrichment

col-168	336	1	0	no	unknown	no tissue-specific cluster enrichment
col-61	341	0	0	yes	unknown	intestine enriched[89]
col-10	345	0	0	yes	hypodermis	L1_muscle enriched
col-81	361	1	0	no	unknown	no tissue-specific cluster enrichment
col-165	383	0	0	no	unknown	no tissue-specific cluster enrichment
col-17	388	0	0	no	unknown	L1_muscle enriched
col-97	429	0	0	yes	unknown	L1_muscle enriched

*= SAM rank number of *skn-1*-dependent upregulated genes by *daf-2(-)* (Supplementary Table 3)

**= SKN-1 binding sites: TTDTCATC or GATGAHAA 1500 bp upstream of each of these genes was analyzed for presence of SKN-1 binding sites. *(D=A/G/T; H=T/C/A); This more restrictive version of the SKN-1 consensus motif (WWTRTACT, W=A/T, R=A/G;[13]) should occur randomly every 10923 bp.

***= SKN-1 binding sites identified by genome-wide ChIP-seq using transgenically-expressed SKN-1 at the first larval stage (L1)[14].

L1_muscle enriched and/or muscle genes: mRNA pull-down by P*myo-3*::FLAG::PAB-1[15]

muscle enriched (emb): embryonic P*myo-3*::GFP positive cells selected for microarray[16]

Supplementary Table 10. *C. elegans* collagen genes that decline in expression during ageing, and are upregulated under conditions that increase lifespan

COLLAGENS FOR WHICH EXPRESSION DECLINES WITH AGE

Of 1254 genes that changed expression during ageing, 88 were collagens were downregulated[17]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 37.37; *P*-value 5.6E-66)

shared with SKN-1-upregulated *daf-2(-)* genes
col-7, col-10, col-12, col-13, col-60, col-76, col-81, col-88, col-91, col-92, col-93, col-97, col-118, col-120, col-125, col-127, col-130, col-133, col-141, col-144, col-145, col-146, col-155, col-156, col-161, col-168, col-180

COLLAGENS UPREGULATED BY DRUG TREATMENTS THAT INCREASE *C. ELEGANS* LIFESPAN

8 collagens were among 116 genes induced in the wild type by Resveratrol treatment in young adults[18]

Second highest enrichment score (ES) of all gene ontology clusters: **Collagen** triple helix repeat (ES= 2.21; *P*-value 1.7E-4)

shared with SKN-1-upregulated *daf-2(-) genes*
none

85 collagens were among 1027 genes induced in *daf-16* mutants by Resveratrol treatment in young adults[18]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES=47.21; *P*-value 4.6E-62)

shared with SKN-1-upregulated *daf-2(-) genes*
col-7, col-12, col-13, col-49, col-58, col-60, col-61, col-65, col-76, col-81, col-88, col-89, col-91, col-93, col-97, col-113, col-118, col-120, col-126, col-127, col-130, col-133, col-152, col-155, col-156, col-161, col-174, ram-2

27 collagens were among 740 genes induced by Humic acid treatment in day 11 old adults[19]

Highest enrichment score (ES) of all gene ontology clusters: **Collagen** triple helix repeat (ES= 7.17; *P*-value 6.3E-9)

shared with SKN-1-upregulated *daf-2(-) genes*
col-13, col-92, col-93, col-167, col-133

74 collagens were among 2842 genes induced by Tannic acid treatment in young adults[20]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 14.47; *P*-value 1.1E-20)

shared with SKN-1-upregulated *daf-2(-) genes*
col-10, col-12, col-13, col-49, col-58, col-60, col-65, col-66, col-81, col-88, col-89, col-91, col-97, col-118, col-125, col-130, col-133, col-137, col-141, col-144, col-145, col-146, col-150, col-152, col-155, col-156, col-161, col-166, col-167, col-168, col-174, col-180, ram-2

67 collagens were among 1562 genes induced by Quercetin treatment in young adults[20]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 19.38; *P*-value 4.0E-33)

shared with SKN-1-upregulated *daf-2(-) genes*
col-12, col-13, col-49, col-60, col-81, col-88, col-89, col-91, col-97, col-118, col-130, col-133, col-137, col-150, col-156, col-161, col-174, col-176

64 collagens were among 2380 genes induced by Rotenone treatment in young adults[21]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 13.63; *P*-value 5.6E-25)

shared with SKN-1-upregulated *daf-2(-) genes*
col-10, col-49, col-58, col-60, col-61, col-65, col-88, col-89, col-91, col-97, col-113, col-125, col-130, col-133, col-137, col-141, col-145, col-146, col-150, col-152, col-155, col-156, col-161, col-162, col-170, col-176, ram-2

8 collagens were among 65 genes upregulated by MAHMA (nitric oxide donor) in wild type (N2) L4 worms[22]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 3.25; *P*-value 7.2 E-7)

shared with SKN-1-upregulated *daf-2(-)* genes
col-97

21 collagens were among 99 genes upregulated by MAHMA (nitric oxide donor) in *hsf-1(sy441)* L4 worms[22]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 13.2; *P*-value 2.5 E-23)

shared with SKN-1-upregulated *daf-2(-)* genes
col-97

COLLAGENS UPREGULATED IN GENETIC BACKGROUNDS THAT INCREASE *C. ELEGANS* LIFESPAN

41 collagens were among 2285 genes upregulated in mixed-stage *wdr-23(tm1817)* animals compared to wild type (N2)[23]

Seventh highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 3.91; *P*-value 5.0E-7)

shared with SKN-1-upregulated *daf-2(-) genes* *col-10, col-17, col-76, col-125, col-144, col-145, col-152, col-156, col-166, col-167, col-168, col-169, col-170, col-176, ram-2*

54 collagens were among 791 genes upregulated in young *age-1(mg44)* adults compared to wild type (N2)[24]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 15.01; *P*-value 8.8E-36)

shared with SKN-1-upregulated *daf-2(-) genes*
col-7, col-58, col-91, col-92, col-93, col-113, col-125, col-141, col-152

17 collagens were among 48 genes upregulated by *daf-2(m41)* at day 10 vs wild type at day 6 of adulthood at 25.5°C[25]

Second highest enrichment score (ES) of all gene ontology clusters: **Collagen** triple helix repeat (ES= 9.25; *P*-value 2.1E-17)

shared with SKN-1-upregulated *daf-2(-) genes*
col-141

57 collagens were among 869 genes upregulated by *daf-2(e1370)* at day 5 of adulthood*[26]

Highest enrichment score (ES) of all gene ontology clusters: **Collagen** triple helix repeat (ES= 23.93; *P*-value 9.5E-34)

shared with SKN-1-upregulated *daf-2(-) genes*
col-10, col-12, col-58, col-65, col-66, col-89, col-91, col-97, col-113, col-115, col-125, col-130, col-144, col-145, col-165, col-166, col-167, col-174, ram-2

43 collagens were among 1078 DAF-16-dependent genes expressed in *daf-2(e1370)* in day 1 adults at 20°C**[27]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 12.3; *P*-value 6.2E-20)

shared with SKN-1-upregulated *daf-2(-) genes*
col-60, col-88, col-91, col-92, col-93, col-130, col-137, col-141, col-146, col-150, col-155, col-161, col-165, col-169, col-170, col-176

90 collagens were among 2181 genes upregulated by TGFβ in day 1 adults[27]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 25.56; *P*-value 8.4E-37)

shared with SKN-1-upregulated *daf-2(-) genes*
col-13, col-58, col-61, col-65, col-66, col-76, col-89, col-91, col-92, col-93, col-113, col-115, col-118, col-125, col-126, col-127, col-130, col-141, col-144, col-145, col-146, col-150, col-152, col-155, col-165, col-166, col-167, col-174, col-176, col-180

31 collagens were among 549 genes were upregulated by AMPK and downstream signaling (shared transcriptional output of loss of *crh-1* (CREB) / loss of *tax-6* (calrectulin) / AAK-2 (AMPK) overexpression) in L4 larvae[28]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens**

shared with SKN-1-upregulated *daf-2(-) genes*
col-12, col-13, col-91, col-126, col-127, col-133, col-137, col-141, col-145, col-146, col-150, col-155, col-156, col-167, col-168, col-176, col-180

21 collagens were among 592 genes upregulated by *ash-2(RNAi)* in day 8 adults that lacked a germline[29]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 5.39; *P*-value 1.8E-7)

shared with SKN-1-upregulated *daf-2(-) genes*
col-12, col-118, col-133, col-146

15 collagens were among 709 genes upregulated in young *isp-1* adults[30]

Third highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 2.18; *P*-value 2.6E-3)

shared with SKN-1-upregulated *daf-2(-) genes*
col-88 col-150

51 collagens were among 2459 genes upregulated by *cyc-1(RNAi)* in young adults[30]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 3.89; *P*-value 2.0E-5)

shared with SKN-1-upregulated *daf-2(-)* genes
col-12, col-13, col-89, col-91, col-97, col-113, col-115, col-118, col-120, col-130, col-141, col-144, col-146, col-150, col-165, col-166, col-176, ram-2

13 collagens were among 155 genes upregulated in 2 day old *rsks-1(ok1255)* adults[31]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 7.2; *P*-value 1.1E-8)

shared with SKN-1-upregulated *daf-2(-)* genes *col-92, col-93 col-133*

---

30 collagens were among 213 genes upregulated in *ctbp-1(ok498)* young adults[32]

Highest enrichment score (ES) of all gene ontology clusters: Nematode cuticle **collagens** (ES= 24.51; *P*-value 7.5 E-29)

shared with SKN-1-upregulated *daf-2(-)* genes
col-60, col-65, col-88, col-91, col-97, col-113, col-120, col-125, col-130, col-144, col-145, col-146, col-150, col-166, col-174, col-180

Gene Ontology (GO) enrichment clusters were determined by DAVID using high classification stringency, and *P*-values were determined by DAVID using Fisher Exact test[33-35]. The young adult stage is defined to be 0-13 hours after the last larval molt (L4) into adulthood before eggs appear in the uterus (www.wormbase.org). * Temperature not specified. ***daf-2(e1370)* vs *daf-2(e1370);daf-16(mu86).* A decline in collagen gene expression with age was also observed in[3,36].

Supplementary Table 11. Collagen and extracellular matrix genes are overrepresented in mammalian longevity expression profiles

Comparison / GEO accession	Species	Tissue	Total genes	# collagen	GO Cluster collagen	ES collagen	*P*-value	collagen genes	# ECM	GO cluster ECM	ES ECM	*P*-value	ECM genes	Reference
S6K-/- vs WT	Mouse	muscle	1021	5	60	1.08	8.5 E-3	COL1A2, COL4A5, Col4a1, lox, COL4A4	26	28/270	1.36	2.5 E-3	TIMP3, TRF, ADAMTSL1, LGALS3, COL4A5, COL1A2, LOC100048740, ADAMTS2, Ang, HSD17B12, CRISPLD2, WNT11, MMP11, lox, ANGPTL4, LOC100048740, ADAMTS2, Tgfbr3, TIMP3, Ang, Adamts15, Eln, tgm2, LOC100048740, ADAMTS2, TIMP3, LOC100039744, LOC630776, nid1, GPC3, Col4a1, lox, COL4A4, PTN, Ang, timp4, omd, SPOCK2	[37]
Rapamycin vs control GSE41018	Mouse	heart	1638	12	17/352	1.26	7.1 E-2	Col27a1, COL4A3, COL18A1, Colec12, C1ql4, COL6A2, COL8A2, Col15a1, GLDN, col19a1, COL11A1, COL25A1	31	82/352	0.68	2.5E-1	Adamts3, alb, Mfap1b, Mfap1a, LOC100048751, COL6A2, ADAMTS19, ANGPTL4, COL8A2, Col15a1, Prss34, COL11A1, WNT9A, MATN3, Col27a1, PODNL1, LOC100047082, Fbn2, LAMC2, adam12, LAMA4, MMP10, Rptn, adam28, Wnt16, col19a1, EMILIN3, COL4A3, COL18A1, Gpc5, CRTAC1, wnt8b, LOC677060, kazald1, OC90	[38]
Rapamycin vs control GSE41018	Mouse	brain	1208	9	43/272	0.85	1.4E-1	COL11A1, COL6A2, Col11a2, COL1A1, C1QTNF6, ADIPOQ, COL9A1, LOC100045121, eda, COL9A1, COL4A4	29	11/352	1.73	4.4E-2	COL6A2, ADAMTS19, FREM1, Rptn, MFAP2, Prss34, Adam15, COL11A1, OPTC, CHL1, COL1A1, Adam21, MMP13, COL9A1, MMP11, OGN, ADAM33, adam12, COL9A1, COL4A4, ABI3BP, ADAMTS20, OTOA, Col11a2, ADAMTS8, LOC640499, MMP3, FBLN1, WNT6, Matn2, Ltbp1	[38]
Rapamycin vs control GSE41018	Mouse	kidney	1839	10	173/401	0.44	3.9E-1	COL4A3, Col17a1, COL9A2, Fcna, Fcnb, ADIPOQ, COL25A1, C1QL3, Col4a6, COL9A1	29	320/401	0.15	6.4E-1	NTN4, MATN1, Chad, Prss34, Adam15, GPC6, LOC100045283, Gm4672, lepre1, PODNL1, MMP13, OLFML2A, Col4a6, COL9A1, NEPN, timp4, Col17a1, Gpc5, SPARCL1, Rptn, Wnt16, TRF, fgf1, Adam26a, COL4A3, NTN1, LOC672215, FMOD, COL9A2, WNT11, hapln3, AHSG	[38]
Ames dwarf vs WT GSE1093	Mouse	liver	152	4	15/59	1.34	2.4E-2	COL3A1, COL4A5, Col5a1, Sftpd	7	15/59	1.34	3.4E-2	COL3A1, COL4A5, Col5a1, Ecm1, adam28, Sftpd, Adamts1	[39]
Caloric restriction (40% less Cal.)	Mouse	liver	226	1				COL4A4	10	4/79	2.82	7.9E-4	Sftpd, ZP2, HAPLN1, SPARCL1, EPYC, ACAN, Tff3, Eln, COL4A4, CTGF	[40]

vs ad libitum GSE2431														
pit-1-/- snell dwarf vs WT GSE3129	Mouse	liver	909	6	29/263	1.56	5.1E-2	COL11A1, Col5a1, lox, TNXB, COL7A1, col19a1	27	10/263	2.84	1.6E-3	vwf, Mmp9, FBLN2, ptprz1, MATN3, lox, OGN, LOC100047888, NCAN, Eln, Adamts1, COL11A1, WNT7B, Ang, SPARCL1, WNT2B, ANGPTL4, ENTPD1, TNXB, HAPLN1, mfap4, Col5a1, Lamb1-1, MMP13, AMBN, Tff3, COL7A1, OGN, col19a1	[41]
Caloric restriction (40% less Cal.) vs ad libitum GSE39313	Mouse	liver	228	3	48/84	0.63	2.8E1	col4a2, Col4a1, ADIPOQ	11	17/84	1.61	2.5E-2	col4a2, LGALS1, TIMP3, Col4a1, ANGPTL4, DCN, CTGF, DCN, timp2, SLC1A3, LGALS3	[42]
Caloric restriction (40% less Cal.) vs ad libitum GSE39313	Mouse	epididymal white adipose tissue	389	3	97/148	0.38	5.3E-1	Col15a1, Col18a1, Mbl2	15	62/148	1.21	8.2E-2	Hspg2, ANGPTL4, Ccdc80, PRELP, Col15a1, vtn, TRF, COL18A1, Ang, LOC100046740, SPARC, SMOC1, AHSG, AHSG, Ecm1	[42]
GhRh -/- vs control GSE51108	Mouse	liver	286	4	39/97	0.86	1.1E-1	COL4A5, COL8A1, C1QB, COL3A1	18	13/97	2.16	9.0E-4	Ang, CTGF, COL8A1, tgfbi, LOC100039744, LOC630776, nid1, LGALS3, LOC100048867, ANXA2, Mmp12, COL4A5, LGALS1, Ang, SMOC2, LGALS1, TIMP1, COL3A1	[43]
irs-1 -/- vs control GSE36041	Mouse	murine embryonic fibroblasts	2507	22	22/453	3.28	5.4E-4	Emid1, Adipoq, Col2a1, Col9a1, Col9a3, Col8a2, Col10a1, Col11a2, Col13a1, Col15a1, Col17a1, Col18a1, Col24a1, Col25a1, Col27a1, C1qa, C1ql1, C1ql4, Fcna, Fcnb, Mmp9, Colq	76	4/453	8.09	2.4E-9	WNT3A, MMP9, ENAM, MMP8, MMP23, GPC3, EMID1, FGF1, COL11A2, USH2A, SPN, COL10A1, OPTC, UCMA, ZP3, SPARCL1, EGFL6, CILP, MMP11, PRELP, ADAMTS7, MMP10, ADAMTS9, LAMC3, WNT9A, COL24A1, EPYC, OTOA, ADAMTS18, PODN, ADAMTS14, ADAMTS13, SPOCK2, RBP3, ADAMTSL5, ELN, TIMP4, SPOCK1, COL2A1, DCN, CPZ, COL9A1, SMOC2, COL17A1, LGALS3BP, CRISPLD2, RPTN, SMOC1, COL27A1, TNR, IMPG2, ACAN, TGM2, ADAM33, ENTPD1, TECTB, COL8A2, WNT8A, DPT, ANGPTL4, TECTA, COL18A1, MUC2, HAPLN4, COL15A1, NTN4, PAPLN, OC90, WNT2B, LAMA2, LAMA4, WNT7B, LAMA3, CLEC3B, KAZALD1, DMBT1	[26]

Supplementary Table 12. Collagen and extracellular matrix genes are overrepresented in Nrf2-related expression profiles

Comparison / GEO accession	Species	Tissue	Total genes	# collagen	GO Cluster collagen	ES collagen	*P*-value	collagen genes	# ECM	GO cluster ECM	ES ECM	*P*-value	ECM genes	Ref
Activated NRF2 by Keap1 cKO vs control GSE11287	Mouse	liver	2752	15	127/458	0.78	1.2E-1	Col5a3, COL11A1, COL20A1, SCARA3, COL1A2, col19a1, Pcolce, EMID1, C1qtnf7, COLQ, LOC100046355, ADIPOQ, col19a1, Col9a3, COL4A5, COL4A3	50	24/458	2.19	4.2E-3	mmp16, BCAN, Mmp23, WNT3A, VCAN, FMOD, Wnt8a, loxl1, LAMA3, DMBT1, CHL1, CHL1, TNC, ADAMTS18, col19a1, timp2, Cpz, SLC1A3, TIMP1, NAV2, FMOD, HAPLN1, ADAMTS20, col19a1, MMP3, COL4A3, Col5a3, GPC6, LOC100045283, Gm4672, Ntng1, ADAMTS5, LOC632263, ADAMTSL1, FMOD, COL1A2, CHL1, lox, WNT9A, ADAMTS5, LOC632263, MMP20, CRISPLD2, EMID1, MATN3, Wnt16, Chad, FRAS1, fn1, COL4A5, COL11A1, WNT6, SPOCK2, Hapln4, ADAMTS18, Ltbp1, HAPLN1, FBLN7, OTOG, LOC100039744, LOC630776, nid1, Gpc5, OPTC	[44]
Nrf2-/- vs WT GSE48879	Mouse	kidney	2129	12	250/438	0.39	3.9E-1	COLQ, LOC100046355, COLEC11, C1qtnf7, col14a1, SCARA5, Col16a1, Col6a3, C1qtnf3, EMID1, COL4A5, C1QTNF2	32	395/438	0.08	7.9.E-1	VWA1, CTGF, ANGPTL4, GPC3, NTN4, DCN, OGN, LOC100046740, SPARC, Adamts1, fgf1, MMP13, SPOCK3, BGN, TIMP3, Adam17, EMID1, COL4A5, Ecm1, LOC100039744, LOC630776, nid1, ltbp3, Col16a1, PRELP, Col6a3, Mmp2, FRAS1, Ltbp1, col14a1, Mmp12, LTBP4	[45]
Activated NRF2 by cisplatin comparing Nrf2-/- vs WT GSE48879	Mouse	kidney	2206	24	26/481	2.84	1.7E-5	COL6A2, Col15a1, col14a1, Col27a1, COL18A1, COL1A1, COL4A5, COL4A5, COL4A4, COL4A4, col4a2, COL3A1, Col4a1, Col16a1, Col6a3, EMILIN1, COL18A1, COL6A1, COL18A1, COL1A2, col14a1, Col5a1, EMID1, COL6A1	72	2/481	6.85	4.9E-10	NTN4, TNXB, COL6A2, Mfap1b, Mfap1a, LOC100048751, Col27a1, Adamts1, COL1A1, TIMP3, Ltbp1, COL4A4, Ecm1, ABI3BP, LOC100039744, LOC630776, nid1, ltbp3, fn1, PRELP, LAMA5, COL18A1, lepre1, Col5a1, CTGF, AGRN, loxl1, ADAMTSL2, COL6A1, TIMP1, col14a1, lox, OGN, Podn, LAMC1, MMP14, tgfbi, COL4A5, col4a2, Col4a1, LOC100046740, SPARC, Mmp2, EMILIN1, GPC3, TNC, Mmp12, LOC100048740, ADAMTS2, LAMC2, ADAMTS16, WNT9B, MMP13, ADAMTS4, COL3A1, LOC100048867, ANXA2, Col16a1, fgf1, Tgfbr3, WNT9B, Lama2, NID2, LTBP4, Fbn1, Ccdc80, timp2, Col15a1, DCN, LGALS3BP, BGN, FBLN1, SPARCL1, LAMC3, Col6a3, TRF, tgm2, Lamb1-1, COL1A2, EMID1	[45]

Supplementary Table 13. Adulthood expression of particular ECM genes promotes lifespan extension

Strain / **RNAi**	Mean lifespan ± S.E.M. [Days]	75th percentile [Days]	N dead/ Initial N	% mean lifespan change to control	P-value (log-rank) vs. control	Figure
Trial of adulthood RNAi against collagen genes from SKN-1-upregulated *daf-2(-)* set at 15°C						
wild type (N2) RNAi **L4440 (control)**	28.2 ± 0.7	31	70/81			
wild type (N2) RNAi ***col-10***	28.6 ± 0.5	31	68/83	+1	0.9271	
wild type (N2) RNAi ***col-13***	26.2 ± 0.6	29	61/75	-7	0.0106	
wild type (N2) RNAi ***col-97***	27.6 ± 0.6	30	64/82	-2	0.3896	
wild type (N2) RNAi ***col-144***	29.5 ± 0.6	31	85/100	+5	0.2703	
P-value and % mean lifespan change are relative to wild type (N2) RNAi L4440						
daf-2(e1370) RNAi **L4440 (control)**	32.7 ± 1.0	39	52/59			
daf-2(e1370) RNAi ***col-10***	24.7 ± 0.6	30	88/115	-24	<0.0001	
daf-2(e1370) RNAi ***col-13***	28.2 ± 0.7	33	74/90	-13	0.0001	
daf-2(e1370) RNAi ***col-97***	30.7 ± 0.7	35	81/101	-6	0.0319	
daf-2(e1370) RNAi ***col-144***	26.1 ± 0.5	30	74/94	-20	<0.0001	
P-value relative and % mean lifespan change are relative to *daf-2(e1370)* RNAi L4440						
Trial of adulthood RNAi against collagen genes at 15°C and 20°C in parallel						
rrf-3(pk1426) RNAi **L4440 15°C**	26.2 ± 0.3	28	119/135	-48	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi **L4440 15°C**	50.1 ± 0.6	56	112/128			
daf-2(e1370); rrf-3(pk1426) RNAi ***daf-16* 15°C**	20.5 ± 0.2	21	105/120	-59	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-10* 15°C**	32.8 ± 0.4	37	90/106	-35	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-13* 15°C**	33.1 ± 0.5	35	95/115	-34	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-120* 15°C**	31.9 ± 0.4	33	99/108	-36	<0.0001	
P-value and % mean lifespan change are relative to *daf-2(e1370); rrf-3(pk1426)* RNAi **L4440 15°C**						
rrf-3(pk1426) RNAi **L4440 20°C**	23.0 ± 0.3	23	91/101	-51	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi **L4440 20°C**	47.2 ± 0.6	51	104/117			
daf-2(e1370); rrf-3(pk1426) RNAi ***daf-16* 20°C**	19.4 ± 0.2	21	119/128	-59	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-10* 20°C**	48.1 ± 0.5	51	117/130	+2	0.1452	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-13* 20°C**	47.7 ± 0.6	54	102/118	+1	0.1050	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-120* 20°C**	48.3 ± 0.5	54	101/114	+2	0.0790	
P-value and % mean lifespan change are relative to *daf-2(e1370); rrf-3(pk1426)* RNAi **L4440 20°C**						
Trial of adulthood RNAi against collagen genes at 15°C and 20°C in parallel						
rrf-3(pk1426) RNAi **L4440 15°C**	29.5 ± 0.3	32	121/142	-42	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi **L4440 15°C**	50.5 ± 0.6	57	113/129			
daf-2(e1370); rrf-3(pk1426) RNAi ***daf-16* 15°C**	23.7 ± 0.3	25	118/134	-53	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-10* 15°C**	36.6 ± 0.6	43	109/131	-27	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-13* 15°C**	34.9 ± 0.6	39	107/127	-31	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-120* 15°C**	35.1 ± 0.6	39	113/135	-31	<0.0001	
P-value and % mean lifespan change are relative to *daf-2(e1370); rrf-3(pk1426)* RNAi **L4440 15°C**						
rrf-3(pk1426) RNAi **L4440 20°C**	24.0 ± 0.2	25	121/136	-49	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi **L4440 20°C**	47.2 ± 0.7	50	101/111			
daf-2(e1370); rrf-3(pk1426) RNAi ***daf-16* 20°C**	19.3 ± 0.2	21	116/128	-59	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-10* 20°C**	47.4 ± 0.6	53	123/132	+0.4	0.5363	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-13* 20°C**	46.2 ± 0.4	50	117/124	-2	0.0065	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-120* 20°C**	47.4 ± 0.5	50	103/111	+0.4	0.2526	
P-value and % mean lifespan change are relative to *daf-2(e1370); rrf-3(pk1426)* RNAi **L4440 20°C**						
Trial of adulthood double RNAi against *daf-2* and collagen genes at 20°C						
rrf-3(pk1426) RNAi **L4440/GFP**	22.8 ± 0.3	23	105/116			
rrf-3(pk1426) RNAi ***daf-2/*GFP**	39.1 ± 0.7	45	112/128			
rrf-3(pk1426) RNAi ***daf-2/daf-16***	17.2 ± 0.1	19	123/135	-56	<0.0001	
rrf-3(pk1426) RNAi ***daf-2/skn-1***	24.8 ± 0.5	28	136/150	-37	<0.0001	
rrf-3(pk1426) RNAi ***daf-2/col-10***	30.3 ± 0.7	32	112/125	-23	<0.0001	
rrf-3(pk1426) RNAi ***daf-2/col-13***	33.5 ± 0.6	39	122/132	-15	<0.0001	
rrf-3(pk1426) RNAi ***daf-2/col-120***	33.8 ± 0.6	39	134/144	-14	<0.0001	
P-value and % mean lifespan change are relative to *rrf-3(pk1426)* RNAi ***daf-2/*GFP**						
Trial of adulthood RNAi against collagen genes from the SKN-1-upregulated *daf-2(-)* set and 100μM Rapamycin at 20°C						
wild type (N2) RNAi **L4440 (control)** 0.2% DMSO	22.6 ± 0.2	23	117/125			
wild type (N2) RNAi ***col-10*** 0.2% DMSO	22.6 ± 0.2	23	118/128	0	0.8799	
wild type (N2) RNAi ***col-13*** 0.2% DMSO	22.4 ± 0.3	23	80/87	-1	0.4219	
wild type (N2) RNAi ***col-120*** 0.2% DMSO	22.7 ± 0.2	23	87/94	+1	0.9361	
P-value and % mean lifespan change are relative to wild type (N2) RNAi L4440 (control) 0.2% DMSO						
wild type (N2) RNAi **L4440 (control)** 0.2% DMSO 100μM Rapamycin	27.9 ± 0.5	30	110/121			
wild type (N2) RNAi ***col-10*** 0.2% DMSO 100μM Rapamycin	27.0 ± 0.4	30	112/122	-3	0.1424	
wild type (N2) RNAi ***col-13*** 0.2% DMSO 100μM Rapamycin	26.3 ± 0.4	30	116/123	-6	0.0050	
wild type (N2) RNAi ***col-120*** 0.2% DMSO 100μM Rapamycin	25.2 ± 0.3	28	125/137	-10	<0.0001	
P-value and % mean lifespan change are relative to wild type (N2) RNAi L4440 (control) 0.2% DMSO 100μM Rapamycin						

Trial of adulthood RNAi against collagen genes from the SKN-1-upregulated *daf-2(-)* set and 100μM Rapamycin at 20°C

wild type (N2) RNAi **L4440 (control)** 0.2% DMSO	23.8 ± 0.5	25	48/51			
wild type (N2) RNAi **L4440 (control)** 0.2% DMSO 100μM Rapamycin	28.9 ± 0.7	32	55/58	+21	<0.0001	
wild type (N2) RNAi ***col-13*** 0.2% DMSO 100μM Rapamycin	24.0 ± 0.8	25	43/46	+1	0.7727	
wild type (N2) RNAi ***col-120*** 0.2% DMSO 100μM Rapamycin	23.8 ± 1.3	29	35/37	0	0.5316	

P-value and % mean lifespan change are relative to wild type (N2) RNAi L4440 (control) 0.2% DMSO

Trial of mutated collagen gene (*dpy-1*)* not upregulated in the SKN-1- *daf-2(-)* set at 15°C

wild type (N2)	26.1 ± 0.3	28	72/86			Ext. Fig. 4e
dpy-1(e1) (collagen)	25.3 ± 0.2	28	74/89	-3	0.1032	Ext. Fig. 4e
daf-2(e1370)	37.5 ± 0.6	42	77/85	+43	<0.0001	Ext. Fig. 4e
dpy-1(e1) daf-2(e1370)	25.6 ± 0.3	26	63/75	-2	0.1714	Ext. Fig. 4e

P-value and % mean lifespan change are relative to wild type (N2)

Trial of adulthood RNAi against collagen and protease genes from the SKN-1-upregulated *daf-2(-)* set, cuticle surface barrier and basement membrane genes at 15°C

rrf-3(pk1426) RNAi **L4440 (control)**	26.4 ± 0.3	29	108/120			Ext. Fig. 4f
rrf-3(pk1426) RNAi ***col-61*** **(intestinal expressed collagen)**	27.9 ± 0.3	30	87/94	+6	0.0284	
rrf-3(pk1426) RNAi ***col-89*** **(intestinal expressed collagen)**	27.9 ± 0.3	30	91/96	+6	0.0139	
rrf-3(pk1426) RNAi ***asp-14*** **(Protease)**	28.0 ± 0.3	30	105/116	+6	0.0018	
rrf-3(pk1426) RNAi **C54D10.10 (Protease)**	27.7 ± 0.2	30	104/115	+5	0.0217	
rrf-3(pk1426) RNAi ***nas-1*** **(Protease)**	27.7 ± 0.3	30	81/91	+5	0.0341	
rrf-3(pk1426) RNAi ***nas-36*** **(Protease)**	27.3 ± 0.3	30	98/109	+3	0.1163	
rrf-3(pk1426) RNAi ***suro-1*** **(Protease)**	31.3 ± 0.7	37	118/132	+19	<0.0001	
rrf-3(pk1426) RNAi **T16G12.1 (Protease)**	31.9 ± 0.4	37	104/118	+21	<0.0001	
rrf-3(pk1426) RNAi ***toh-1*** **(Protease)**	28.4 ± 0.3	32	100/117	+8	<0.0001	
rrf-3(pk1426) RNAi **W01A8.6 (Protease)**	27.8 ± 0.2	30	105/117	+5	0.0193	
rrf-3(pk1426) RNAi ***acs-20*** **(cuticle surface barrier)**	26.9 ± 0.4	30	82/90	+2	0.5992	
rrf-3(pk1426) RNAi ***acs-22*** **(cuticle surface barrier)**	24.2 ± 0.3	25	108/122	-8	<0.0001	
rrf-3(pk1426) RNAi ***epi-1*** **(basement membrane)**	27.7 ± 0.3	30	116/120	+5	0.0203	
rrf-3(pk1426) RNAi ***let-2*** **(basement membrane)**	26.9 ± 0.4	28	69/75	+2	0.5942	
rrf-3(pk1426) RNAi ***tsp-15*** **(basement membrane)**	28.1 ± 0.2	30	104/115	+6	0.0024	

P-value and % mean lifespan change are relative to *rrf-3(pk1426)* RNAi L4440 (control)

daf-2(e1370); rrf-3(pk1426) RNAi **L4440 (control)**	47.8 ± 0.5	52	114/130			Ext. Fig. 4f
daf-2(e1370); rrf-3(pk1426) RNAi ***col-61*** **(intestinal expressed collagen)**	41.9 ± 0.8	46	83/88	-12	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***col-89*** **(intestinal expressed collagen)**	46.8 ± 0.9	56	95/100	-2	0.4294	
daf-2(e1370); rrf-3(pk1426) RNAi ***asp-14*** **(Protease)**	36.1 ± 0.6	41	104/114	-24	<0.0001	Ext. Fig. 4f
daf-2(e1370); rrf-3(pk1426) RNAi **C54D10.10 (Protease)**	45.0 ± 0.3	46	101/108	-6	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***nas-1*** **(Protease)**	44.6 ± 0.5	49	112/119	-7	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi ***nas-36*** **(Protease)**	46.2 ± 0.6	49	116/121	-3	0.2931	
daf-2(e1370); rrf-3(pk1426) RNAi ***suro-1*** **(Protease)**	37.6 ± 0.7	46	103/120	-21	<0.0001	Ext. Fig. 4f
daf-2(e1370); rrf-3(pk1426) RNAi **T16G12.1 (Protease)**	45.5 ± 0.5	49	103/115	-5	0.0026	
daf-2(e1370); rrf-3(pk1426) RNAi ***toh-1*** **(Protease)**	45.1 ± 0.4	49	115/128	-6	<0.0001	
daf-2(e1370); rrf-3(pk1426) RNAi **W01A8.6 (Protease)**	43.8 ± 0.6	49	109/123	-8	0.0015	
daf-2(e1370); rrf-3(pk1426) RNAi ***acs-20*** **(cuticle surface barrier)**	32.1 ± 0.5	36	124/138	-33	<0.0001	Ext. Fig. 4f
daf-2(e1370); rrf-3(pk1426) RNAi ***acs-22*** **(cuticle surface barrier)**	29.7 ± 0.5	32	107/122	-38	<0.0001	Ext. Fig. 4f
daf-2(e1370); rrf-3(pk1426) RNAi ***epi-1*** **(basement membrane)**	47.6 ± 0.8	52	79/86	-0.4	0.6226	
daf-2(e1370); rrf-3(pk1426) RNAi ***let-2*** **(basement membrane)**	42.9 ± 0.7	49	118/128	-10	0.0003	
daf-2(e1370); rrf-3(pk1426) RNAi ***tsp-15*** **(basement membrane)**	40.7 ± 0.6	46	106/132	-15	<0.0001	

P-value and % mean lifespan change are relative to *daf-2(e1370); rrf-3(pk1426)* RNAi L4440 (control)

Trial of collagen overexpression at 20°C

wild type (N2)	22.6 ± 0.3	25	65/79	+0.4	0.7880	
IdEx102 [*rol-6(su1006)*] control	22.5 ± 0.3	25	60/71			
IdEx103 [**COL-10**; *rol-6(su1006)*]	24.8 ± 0.4	27	75/85	+10	<0.0001	
IdEx104 [**COL-10**; *rol-6(su1006)*]	24.3 ± 0.4	27	60/70	+8	0.0004	

IdEx105 [**COL-13**; *rol-6(su1006)*]	25.2 ± 0.4	27	67/76	+12	<0.0001	
IdEx106 [**COL-13**; *rol-6(su1006)*]	24.4 ± 0.4	27	82/93	+8	<0.0001	
IdEx107 [**COL-13**; *rol-6(su1006)*]	24.2 ± 0.4	27	55/66	+8	0.0007	
IdEx108 [**COL-13**; *rol-6(su1006)*]	25.5 ± 0.4	27	77/84	+13	<0.0001	
IdEx109 [**COL-120**; *rol-6(su1006)*]	23.3 ± 0.4	25	58/69	+4	0.1570	
IdEx110 [**COL-120**; *rol-6(su1006)*]	23.4 ± 0.3	25	72/84	+4	0.0391	
P-value and % mean lifespan change are relative to *IdEx102* [ROL-16(*su1006*)] control						
Trial of collagen overexpression at 20°C						
wild type (N2)	22.7 ± 0.4	25	55/65	0	0.8246	
IdEx102 [*rol-6(su1006)*] control	22.7 ± 0.3	23	60/67			3f
IdEx103 [**COL-10**; *rol-6(su1006)*]	24.3 ± 0.5	28	69/77	+7	0.0008	3f
IdEx104 [**COL-10**; *rol-6(su1006)*]	23.3 ± 0.5	25	50/59	+3	0.0843	
IdEx106 [**COL-13**; *rol-6(su1006)*]	24.0 ± 0.4	25	80/86	+6	0.0046	
IdEx108 [**COL-13**; *rol-6(su1006)*]	25.5 ± 0.4	28	91/99	+12	<0.0001	3f
IdEx109 [**COL-120**; *rol-6(su1006)*]	24.7 ± 0.5	25	49/60	+9	0.0005	
IdEx110 [**COL-120**; *rol-6(su1006)*]	25.5 ± 0.4	28	79/84	+12	<0.0001	3f
IdEx111 [**COL-10; COL-13; COL-120**; *rol-6(su1006)*]	23.9 ± 0.6	28	46/60	+5	0.0204	3f
P-value and % mean lifespan change are relative to *IdEx102* [ROL-16(*su1006*)] control						
Trial of collagen overexpression at 20°C						
wild type (N2)	21.0 ± 0.4	22	46/53	-0.5	0.9074	
IdEx102 [*rol-6(su1006)*] control	21.1 ± 0.3	22	60/66			
IdEx111 [**COL-10; COL-13; COL-120**; *rol-6(su1006)*]	23.6 ± 0.6	28	50/54	+12	0.0001	
P-value and % mean lifespan change are relative to *IdEx102* [ROL-16(*su1006*)] control						
Trial of overexpression of collagen and extracellular matrix genes not upregulated in the SKN-1- *daf-2(-)* set at 20°C						
wild type (N2)	23.1 ± 0.4	25	52/66			
kaIs12 [COL-19::GFP] (adult specific collagen)	23.6 ± 0.4	25	54/63	+2	0.3474	
kuIs55 [LON-3::GFP] (collagen)	22.1 ± 0.3	23	54/79	-4	0.0185	
chEx1682 [QUA-1::GFP] (peptidase required for molting)	22.6 ± 0.3	23	58/69	-2	0.1328	
upIs1 [MUP-4::GFP] (anchors muscle-hypodermis-cuticle)	22.5 ± 0.2	23	95/115	-3	0.0408	
P-value and % mean lifespan change are relative to wild type (N2)						

Trials that were performed in parallel are grouped. (N) = number of animals observed; Lifespan was measured from the L4 stage. Animals that left the plates, buried into the agar, bagged, or exploded were censored. L4440 empty vector was otherwise used as the control. In some cases, knockdown was performed in the RNAi-sensitive strain *rrf-3(pk1426)*[2]. *col-10*, *col-12,* and *col-97* share >99% protein sequence identity with *col-144*, *col-13*, and *col-109*, respectively. **dpy-1(e1)* affects body length and alae formation[46,47]. For collagen overexpression trials, the extent of overexpression was determined by qRT-PCR. All transgenic lines had significant higher mRNA levels compared to wild type (N2) determined by one sample *t*-test, two-tailed, hypothetical mean of 1 was used for comparison: *IdEx103 col-10* = 6.9±0.6-fold, *IdEx104 col-10* =10.9±0.3-fold, *IdEx106 col-13* = 12.7±0.8-fold, *IdEx108 col-13* = 4.7±0.6-fold, *IdEx109 col-120* = 3.3±0.6-fold, *IdEx110 col-120* = 5.1±0.5-fold relative mRNA levels to wild type (mean ±S.E.M.).

Supplementary Table 14. Potential crossover effects of collagen gene RNAi

Collagen	Sequence name	Coding seq. length (nt)	RNAi clone length (nt)	Paralogues*	Homology region >300nt	Homology region 200-300nt
col-10	B0222.8	885	885	*col-144*	*col-145*	*col-126* *col-127* *col-130*
col-13	F15H10.2	951	951	*col-12*	*col-159* *col-160*	*col-137* *col-136*
col-65	K08C9.4	906	906		*col-107*	none
col-120	Y11D7A.11	942	942		none	none
col-127	F54D1.3	885	885	*col-126* *col-145*		*col-144* *col-10* *col-130*
col-133	F52B11.4	915	915	*col-92* *col-93* *col-94* *col-95* *col-96*		*col-154* *col-155* *col-157* *sqt-3* *col-43*
col-141	T15B7.5	945	945	*col-142*	*col-143*	*col-146* *col-147*
col-167	T10E10.2	906	906	*col-168* *col-169*	*col-171* *col-166* *col-180*	none
col-180	C44C10.1	903	903	*col-170*	*col-166* *col-167* *col-168* *col-169* *col-171*	none

*Paralogues are defined here to share >95% identical amino acid sequence. Homology region is defined as >80% nucleotide(nt) sequence identity with a minimum of 25nt stretch of uninterrupted identical sequence.

Supplementary Table 15. Effects of longevity interventions on 26 proteases upregulated by SKN-1 under rIIs conditions

SAM Rank	Public Name	Seq name	KOG (euKaryotic Orthologous Group) Title	Score(d)	SKN-1 site* TTDTCATC GATGAHAA	*daf-2(-)* vs wild type day 5 adults [26]	*daf-2(-)*; DAF-16-dependent up [27]	*age-1(-)* (PI3K) vs wild type [24]	TGF-β vs wild type adults [27]	Humic acid day 11 adults [19]	Quercitin in young adults [20]	*cyc-1(-)* vs wild type [30]	Resveratrol young adults [18]
4	***asp-14***	**K10C2.3**	**Aspartyl protease**	**4.116**	**0**			**up**	**up**	**up**			
6	*nas-7*	C07D10.4	Meprin A metalloprotease (astacin-like protease)	3.987	1	up			up				
33	*T21D11.1*	T21D11.1	Gelatinase A and related matrix metalloproteases	3.133	1	up							
35	***C54D10.10***	**C54D10.10**	**predicted serine-type endopeptidase inhibitor activity**	**3.096**	**2**		**up**		**up**				
48	***nas-1***	**F45G2.1**	**Meprin A metalloprotease (astacin-like protease)**	**2.867**	**1**	**up**							
54	***W01A8.6***	**W01A8.6**	**metallocarboxypeptidase activity**	**2.802**	**1**	**up**			**up**		**up**		
93	***nas-36***	**C26C6.3**	**Meprin A metalloprotease (astacin-like protease)**	**2.439**	**0**								**up**
98	*nas-27*	T23F4.4	Meprin A metalloprotease (astacin-like protease)	2.411	1								
103	***T16G12.1***	**T16G12.1**	**Puromycin-sensitive aminopeptidase and related aminopeptidases**	**2.404**	**0**				**up**			**up**	
109	*nas-8*	C34D4.9	Meprin A metalloprotease (astacin-like protease)	2.333	0	up							
139	*nas-22*	T11F9.6	Meprin A metalloprotease (astacin-like protease)	2.245	0								
148	***toh-1***	**T24A11.3**	**Meprin A metalloprotease (astacin-like protease)**	**2.183**	**0**		**up**		**up**			**up**	**up**
152	*C10C5.3*	C10C5.3	Aminoacylase ACY1 and related metalloexopeptidases	2.165	3							up	
184	*nas-29*	F58A6.4	astacin-like protease	2.076	0								
185	*C10C5.5*	C10C5.5	Aminoacylase ACY1 and related metalloexopeptidases	2.072	0								
243	*T19D2.1*	T19D2.1	Metallopeptidase	1.926	3	up						up	
259	*nas-38*	F57C12.1	Meprin A metalloprotease (astacin-like protease)	1.904	0	up			up				
272	*F45E1.5*	F45E1.5	carboxypeptidase A	1.886	0	up							
287	*T06A4.3*	T06A4.3	Zinc carboxypeptidase	1.861	0								
294	*nep-21*	T16A9.4	M13 family peptidase (neprilysin)	1.843	0								
299	*nas-30*	Y95B8A.1	Meprin A metalloprotease (astacin-like protease)	1.84	0	up			up			up	
303	*nas-22*	T11F9.6	Meprin A metalloprotease (astacin-like protease)	1.83	0								
349	***suro-1***	**R11A5.7**	**Zinc carboxypeptidase**	**1.743**	**0**	**up**			**up**				
360	*Y47G6A.19*	Y47G6A.19	Zinc carboxypeptidase	1.736	0				up				
410	*K10C2.1*	K10C2.1	Serine carboxypeptidases (lysosomal cathepsin A)	1.675	1								
427	*Y18H1A.9*	Y18H1A.9	Zinc carboxypeptidase	1.659	0	up							

*= SAM rank number of *skn-1*-dependent upregulated genes by *daf-2(-)* (Supplementary Table 3)
**= SKN-1 binding sites: TTDTCATC or GATGAHAA 1500 bp upstream of each of these genes was analyzed for presence of SKN-1 binding sites. *(D=A/G/T; H=T/C/A); This more restrictive version of the SKN-1 consensus motif (WWTRTACT, W=A/T, R=A/G;[13]) should occur randomly every 10923bp.
In bold are the 8 out of the 26 *daf-2(-)*; SKN-1-dependent upregulated protease genes that were tested for their effects on wild type lifespan and *daf-2(e1370)* longevity at 15°C.

Supplementary Table 16. Adulthood knockdown of collagen genes sensitizes to t-BOOH or Arsenite oxidative stress

Strain	Temp. [°C]	Mean survival ± S.E.M. [hours]	75th per-centile [hours]	max. survival [hours]	N died from t-BOOH/ Initial N	% mean lifespan change to L4440 (RNAi)	P-value (log-rank) vs. L4440 (RNAi)	P-value (log-rank) vs. *skn-1* (RNAi)	P-value (log-rank) vs. *daf-2* (RNAi)	Figure
Parallel trial using 15.4 mM t-BOOH at 20°C										
N2 L4440(RNAi)	20	10.5 ± 0.4	12	16	44/61					
N2 *skn-1*(RNAi)	20	5.9 ± 0.2	6		27/42	-44	<0.0001			
N2 *daf-2*(RNAi)*	20	13.6 ± 0.4	16	-	50/76	+30	<0.0001	<0.0001		
N2 *col-120*(RNAi)	20	7.8 ± 0.3	10	11	38/50	-26	<0.0001	<0.0001	<0.0001	
N2 *col-127*(RNAi)	20	8.0 ± 0.4	10	10	24/24	-24	<0.0001	<0.0001	<0.0001	
N2 *col-133*(RNAi)	20	8.1 ± 0.3	10	10	37/40	-23	<0.0001	<0.0001	<0.0001	
N2 *col-144*(RNAi)	20	8.0 ± 0.3	10	11	32/41	-24	<0.0001	<0.0001	<0.0001	
N2 *col-167*(RNAi)	20	8.6 ± 0.3	10	10	28/29	-18	<0.0001	<0.0001	<0.0001	
N2 *col-176*(RNAi)	20	9.5 ± 0.3	11	12	32/34	-10	0.0108	<0.0001	<0.0001	
N2 *col-180*(RNAi)	20	7.8 ± 0.4	10	10	25/34	-26	<0.0001	<0.0001	<0.0001	
Parallel trial using15.4 mM t-BOOH at 20°C										
N2 L4440(RNAi)	20	9.5 ± 0.3	11	14	70/80					
N2 *skn-1*(RNAi)	20	6.6 ± 0.2	8	10	45/50	-30	<0.0001			
N2 *daf-2*(RNAi)*	20	13.0 ± 0.4	16	-	54/74	+37	<0.0001	<0.0001		
N2 *col-120*(RNAi)	20	7.6 ± 0.2	9	9	46/58	-20	<0.0001	0.0008	<0.0001	
N2 *col-127*(RNAi)	20	7.9 ± 0.3	9	10	35/45	-17	<0.0001	0.0003	<0.0001	
N2 *col-133*(RNAi)	20	7.7 ± 0.2	8	9	34/40	-19	<0.0001	0.0035	<0.0001	
N2 *col-144*(RNAi)	20	7.5 ± 0.3	9	11	33/43	-21	<0.0001	0.0086	<0.0001	
N2 *col-167*(RNAi)	20	8.3 ± 0.2	9	9	24/25	-13	0.0007	<0.0001	<0.0001	
N2 *col-176*(RNAi)	20	8.9 ± 0.2	10	11	41/47	-6	0.0049	<0.0001	<0.0001	
N2 *col-180*(RNAi)	20	7.6 ± 0.3	9	10	33/41	-20	<0.0001	0.0052	<0.0001	

(N) = number of animals observed. Animals that crawled off the plates, buried into the agar, or exploded were censored. * scored survival for 16 hours. At 16 hours, several N2 *daf-2(RNAi)* animals were alive. P-values for survival curves were determined by Log-Rank test.

3 combined trials of 5mM Arsenite treatment for 21 hours

Strain	Mean survival **after 21** hours ± S.E.M. [%]	Number of animals	P-value vs. N2 (L4440)	Figure
N2 L4440(RNAi)	52.9 ± 1.8	189		Ext. Fig. 6k
N2 *skn-1*(RNAi)	6.3 ± 4.5	108	<0.001	Ext. Fig. 6k
N2 *daf-2*(RNAi)	87.1 ± 0.2	101	<0.05	
N2 *col-10*(RNAi)	32.7 ± 10.7	171	<0.01	Ext. Fig. 6k
N2 *col-13*(RNAi)	24.0 ± 2.9	155	<0.01	Ext. Fig. 6k
N2 *col-97*(RNAi)	27.3 ± 4.4	86	<0.01	Ext. Fig. 6k
N2 *col-120*(RNAi)	14.2 ± 3.0	132	<0.001	Ext. Fig. 6k
N2 *col-127*(RNAi)	22.3 ± 9.1	70	<0.01	Ext. Fig. 6k
N2 *col-133*(RNAi)	20.1 ± 0.7	65	<0.01	Ext. Fig. 6k
N2 *col-144*(RNAi)	31.5 ± 6.9	181	<0.05	Ext. Fig. 6k
N2 *col-167*(RNAi)	60.5 ± 0.9	124	>0.05	Ext. Fig. 6k
N2 *col-176*(RNAi)	48.0 ± 4.2	96	>0.05	Ext. Fig. 6k
N2 *col-180*(RNAi)	25.7 ± 13.2	76	<0.05	Ext. Fig. 6k
N2 *cut-2*(RNAi)	50.5 ± 1.3	96	>0.05	
N2 F56D5.3(RNAi)	30.7 ± 12.1	80	<0.01	Ext. Fig. 6k
N2 *gst-4*(RNAi)	39.3 ± 3.1	126	<0.01	Ext. Fig. 6k
N2 *nit-1*(RNAi)	32.8 ± 3.8	112	<0.01	Ext. Fig. 6k

3 combined trials of 5mM Arsenite treatment for 30 hours

Strain	Mean survival **after 30** hours ± S.E.M.[%]	Number of animals	P-value vs. *daf-2(e1370)* (L4440)	Figure
daf-2(e1370) L4440(RNAi)	53.8 ± 2.7	125		Ext. Fig. 6l
daf-2(e1370) skn-1(RNAi)	14.8 ± 3.8	149	<0.001	Ext. Fig. 6l
daf-2(e1370) col-10(RNAi)	34.0 ± 7.6	69	<0.05	Ext. Fig. 6l
daf-2(e1370) col-13(RNAi)	32.3 ± 9.2	67	<0.05	Ext. Fig. 6l
daf-2(e1370) col-97(RNAi)	47.0 ± 4.0	63	>0.05	Ext. Fig. 6l
daf-2(e1370) col-120(RNAi)	27.5 ± 3.5	60	<0.05	Ext. Fig. 6l
daf-2(e1370) col-127(RNAi)	52.5 ± 4.5	104	>0.05	Ext. Fig. 6l
daf-2(e1370) col-133(RNAi)	47.5 ± 9.5	116	>0.05	Ext. Fig. 6l
daf-2(e1370) col-144(RNAi)	45.7 ± 4.7	94	>0.05	Ext. Fig. 6l
daf-2(e1370) col-167(RNAi)	28.0 ± 9.0	67	<0.05	Ext. Fig. 6l
daf-2(e1370) col-176(RNAi)	54.5 ± 2.5	92	>0.05	Ext. Fig. 6l
daf-2(e1370) col-180(RNAi)	52.0 ± 1.0	70	>0.05	Ext. Fig. 6l
daf-2(e1370) cut-2(RNAi)	52.7 ± 3.5	62	>0.05	
daf-2(e1370) F56D5.3(RNAi)	35.33 ± 4.3	72	<0.05	Ext. Fig. 6l
daf-2(e1370) gst-4(RNAi)	32.7 ± 5.9	81	<0.05	Ext. Fig. 6l
daf-2(e1370) nit-1(RNAi)	31.3 ± 6.9	125	<0.05	Ext. Fig. 6l

N2= wild type. L4440 = empty vector control. P-values for survival after 21 hours for wild type (N2) or 30 hours for *daf-2(e1370)* mutants were determined by one-way ANOVA post hoc Tukey.

Supplementary Table 17. Primer sequences used for qRT-PCR

Gene	Forward primer (FP) 5'…3	Reverse primer (RP) 5'…3
col-10	GTAACCCTCTCTACCGGAGCTG	ATCTCGGTCCATGCAGAGTCG
col-12/13	GTAGCCTCGGAGTTTCTGGTG	GATCCACCGAATCCTCCGAC
col-65	CGGCTCGTCATGCAACTGTG	TCCGTCGTGACCGTTGTCTC
col-133	TCCAGGACAGAAGGGACCATC	CTCCAGCTTGTCCTGGTTGTC
col-141	GGACAAGCAGGAACTCCAGG	CATTGCAGCAGAACGAGCTGG
col-167	CTCTTGTCGGAGTTGCCGTC	GCGGGTAACCTCTCTCCAGATG
col-176	TACTTGCCCACCAGGACCAC	GGTTGTGGTGCTGTCTTGTCC
daf-16*	TTCCAAGTGATATTGTTGATAGAACTG	TTAATCGGCTTCGACTCCT
dod-17	CTTCGTTGAGAAGCGCGTTGG	CGTCTGGCAGAGAAGGTTGAG
dod-24	CACCGAGCCAGGAGGTTATC	CTGTTGTCCGTCCCGATGTTG
F55G11.2	CGGAACTTTGAACGGAGGGAC	CGAGAGTGAACACGTCTGGAG
gst-38	ACCGATGAGGAGTGGGAGAAG	CTGAGCGAGTGGTTTGCCATC
ins-7*	AGGTCCAGCAGCAGAACCAGAAG	GAAGYCGTCGGTGCATTCTT
nas-7	CGAGCAGCGAAACTATGGCC	TCTCCGGTCTGTCGTGGAAC
nit-1*	AATCCTCCGACTATCCCTTG	AGCGAATCGTTTCTTTTGTG
tubulin _(tba-1)*_	ACTGATCTCTGCTGACAAGG	TTGGCTGGCTCGAAGCAA

Primers for qRT-PCR were designed according to the following criteria: melting temperature (Tm) of each PCR primer between 58-60°C; the Tm of both primers is within 1°C. Amplicon length is 50 to 150. Primers span exon-exon junctions (intron splice-sites) in the target mRNA to prevent amplification of the target from contaminating gDNA. Primers were blasted against the _C. elegans_ genome to ensure unique binding, except that _col-12/13_ binds to both genes.

Primers that have been used in previous publications: *[48] and **[4].

The sequence of the following primers have been published previously:

[49]: _gst-4, skn-1._

[50]: _cdc-42, pmp-3_, Y45F10D.4.

[51]: _col-144_

Supplementary Discussion

Dauer diapause of *C. elegans*

C. elegans develops into a reproductive hermaphrodite adult by passing through four larval stages (L1-L4)[52]. Unfavorable environmental and nutritional conditions (high temperature, crowding, low food availability) induce development into the dauer diapause, a non-reproducing alternative L3 larval stage that is characterized by prolonged periods of immobility, darkening of the body, a decreased body size, a distinct cuticle structure, and shrinkage of the gonad and other structures[1,52,53]. Dauers are resistant to harsh environmental insults and starvation, and can survive for many weeks[53,54]. When environmental conditions become favorable again, dauers reenter the reproductive life cycle by entering the L4 stage[52]. Dauer larvae are thought not to age, because worms that have entered and exited dauer exhibit a normal adult lifespan, even if they previously had remained as dauers for more than 60 days[54]. Development into dauer is dependent upon the activity of DAF-16, which is inhibited directly by IIS through phosphorylation (Extended Data Fig. 1b)[55,56].

Dauer-related *daf-2* Class 2 traits

daf-2 mutations can be categorized as Class 1 or 2[1]. Both mutant classes are predisposed to enter dauer at elevated temperature (e.g. 25°C, Extended Data Fig. 1c, 1i, 1j), but Class 1 mutants appear normal as adults[1]. By contrast, Class 2 mutants are defined by the prevalence of particular traits (decreased movement, pharyngeal pumping, and body size, darkening of the body, gonad shrinkage, reproductive delay, and reduced progeny numbers) in adults at higher temperatures (22.5°C or 25°C)[1]. These Class 2 traits are considered to be dauer-related because of their similarity to the dauer state, and their dependence upon *daf-16*[1]. A striking aspect of

Class 2 traits is their reversibility[1]. For example, when *daf-2(e1370)* adults were downshifted from 25°C to 15°C, mobility and normal morphology were fully restored[1]. Placement of the same animals at 25°C rendered them immobile and morphologically abnormal again within a few days, but this could be reversed by a second downshift to 15°C[1]. These phenotypic shifts could be repeated multiple times in the same adult[1]. Importantly, typical Class 2 mutants such *daf-2*(*e1370*) exhibit a milder version of Class 2 traits at the temperature most frequently used for lifespan and phenotypic analyses (20°C). At 20°C fewer than 1% of *daf-2(e1370)* animals form dauers and adults initially appear normal, but 100% have a prolonged reproductive period and eventually become immobile as adults[1] (not shown; Supplementary Table 1, Video S1 and S2). For simplicity, in the main text we refer to these Class 2 traits as "dauer-related traits" or "dauer traits".

Temperature regulation of dauer entry and adulthood dauer-related traits

Many *daf-2* alleles are frequently referred to as temperature-sensitive (ts) because they induce dauer entry and dauer-like (Class 2) traits in a temperature-dependent manner (see above), and because many of these mutations were identified in screens for temperature-dependent dauer formation[1,52,53]. It is therefore both striking and paradoxical that in the same *daf-2* mutants, other major phenotypes are not dependent upon elevated temperature. Class 2 *daf-2* mutations typically extend adult lifespan comparably at either 15°C, 20°C, or 22.5°C (Fig. 1a, Extended Data Table 1, Supplementary Table 2)[1,57-67], temperatures at which dauer-like Class 2 traits are either absent (15°C), very mild (20°C), or severe (22.5°C)[1]. Moreover, we have shown that in *daf-2* mutants the levels of nuclear DAF-16 and SKN-1, a direct readout of IIS inhibition, are similar at 15°C and 20°C (Extended Data Fig. 1k-o). If these *daf-2* mutations are "ts" in the typical sense of temperature-dependent protein folding, how can the dauer-related phenotypes be strictly dependent upon temperature while other rIIS effects are temperature-independent?

The simplest answer to this question lies in the profound effect of temperature on dauer regulation. Work performed 30 years ago indicated that many if not most mutants with temperature-sensitive effects on dauer are unlikely to be ts with respect to protein folding and gene function, and instead that temperature is a major evolutionarily conserved regulator of dauer entry that operates independently of other variables, so that dauer is essentially a *ts phenotype*[68,69]. This model explains why (1) dauer pheromone-treated wild-type animals and many null mutants readily develop into dauers at higher temperatures, and (2) wild-type animals that are provided with food and have not been exposed to pheromone nevertheless enter dauer at a detectable frequency at 27°C[69,70]. In each case, these animals are "ts" with respect to the dauer phenotype. This model, together with our evidence that IIS regulates an underlying longevity pathway that is dauer-independent, also explains why a Class 2 allele like *daf-2(e1370)* can be temperature-dependent with respect to the phenotypes of dauer entry, dauer traits and *skn-1*-independent lifespan extension, but not with respect to longevity *per se*, or nuclear accumulation of DAF-16 and SKN-1 (see text).

In general, genetic, expression, proteomic, and metabolomic studies of rIIS have involved conditions under which dauer-related traits appear in aging adults, such as analysis of Class 2 *daf-2* mutants at 20°C or higher[3,25,27,71-78]. Given that *daf-2* lifespan extension is *skn-1*-independent in such scenarios (see text), it will be important to determine the extent to which different findings are obtained when such analyses are performed under dauer-independent conditions.

3. Supplementary Videos

For the supplementary videos, *daf-2(e1370)* (Class 2) mutants were grown and maintained at 15°C. At the first day of adulthood, animals were placed on plates containing FUdR and OP50 bacteria, and either kept at 15°C or upshifted to 20°C. At day 14 of adulthood 10 min videos were taken. The videos are shown at 8x speed.

Supplementary References

1 Gems, D. *et al*. Two pleiotropic classes of *daf-2* mutation affect larval arrest, adult behavior, reproduction and longevity in *Caenorhabditis elegans*. *Genetics* **150**, 129-155 (1998).

2 Simmer, F. *et al*. Loss of the putative RNA-directed RNA polymerase RRF-3 makes *C. elegans* hypersensitive to RNAi. *Curr Biol* **12**, 1317-1319, doi:S0960982202010412 [pii] (2002).

3 Murphy, C. T. *et al*. Genes that act downstream of DAF-16 to influence the lifespan of *Caenorhabditis elegans*. *Nature* **424**, 277-283, doi:10.1038/nature01789
nature01789 [pii] (2003).

4 Oliveira, R. P. *et al*. Condition-adapted stress and longevity gene regulation by *Caenorhabditis elegans* SKN-1/Nrf. *Aging Cell* **8**, 524-541, doi:ACE501 [pii]
10.1111/j.1474-9726.2009.00501.x (2009).

5 Kim, Y. & Sun, H. Functional genomic approach to identify novel genes involved in the regulation of oxidative stress resistance and animal lifespan. *Aging Cell* **6**, 489-503, doi:ACE302 [pii]
10.1111/j.1474-9726.2007.00302.x (2007).

6 Curran, S. P. & Ruvkun, G. Lifespan regulation by evolutionarily conserved genes essential for viability. *PLoS Genet* **3**, e56, doi:07-PLGE-RA-0112R1 [pii]
10.1371/journal.pgen.0030056 (2007).

7 Hamilton, B. *et al*. A systematic RNAi screen for longevity genes in *C. elegans*. *Genes Dev* **19**, 1544-1555, doi:19/13/1544 [pii]
10.1101/gad.1308205 (2005).

8 Tacutu, R. *et al*. Prediction of *C. elegans* Longevity Genes by Human and Worm Longevity Networks. *PLoS One* **7**, e48282, doi:10.1371/journal.pone.0048282
PONE-D-12-16927 [pii] (2012).

9 Samuelson, A. V., Klimczak, R. R., Thompson, D. B., Carr, C. E. & Ruvkun, G. Identification of *Caenorhabditis elegans* genes regulating longevity using enhanced RNAi-sensitive strains. *Cold Spring Harb Symp Quant Biol* **72**, 489-497, doi:10.1101/sqb.2007.72.068
10.1101/sqb.2007.72.068 [pii] (2007).

10 Tullet, J. M. *et al*. Direct inhibition of the longevity-promoting factor SKN-1 by insulin-like signaling in *C. elegans*. *Cell* **132**, 1025-1038, doi:S0092-8674(08)00130-X [pii]
10.1016/j.cell.2008.01.030 (2008).

11 Chi, W. & Reinke, V. Promotion of oogenesis and embryogenesis in the *C. elegans* gonad by EFL-1/DPL-1 (E2F) does not require LIN-35 (pRB). *Development* **133**, 3147-3157, doi:dev.02490 [pii]
10.1242/dev.02490 (2006).

12 Hunt-Newbury, R. *et al*. High-throughput in vivo analysis of gene expression in *Caenorhabditis elegans*. *PLoS Biol* **5**, e237, doi:07-PLBI-RA-0103 [pii]
10.1371/journal.pbio.0050237 (2007).

13 Blackwell, T. K., Bowerman, B., Priess, J. R. & Weintraub, H. Formation of a monomeric DNA binding domain by Skn-1 bZIP and homeodomain elements. *Science* **266**, 621-628 (1994).

14 Niu, W. *et al*. Diverse transcription factor binding features revealed by genome-wide ChIP-seq in *C. elegans*. *Genome Res* **21**, 245-254, doi:gr.114587.110 [pii]
10.1101/gr.114587.110 (2011).

15 Roy, P. J., Stuart, J. M., Lund, J. & Kim, S. K. Chromosomal clustering of muscle-expressed genes in *Caenorhabditis elegans*. *Nature* **418**, 975-979, doi:10.1038/nature01012 (2002).

16 Fox, R. M. *et al*. The embryonic muscle transcriptome of *Caenorhabditis elegans*. *Genome Biol* **8**, R188, doi:gb-2007-8-9-r188 [pii]
10.1186/gb-2007-8-9-r188 (2007).

17 Budovskaya, Y. V. *et al*. An elt-3/elt-5/elt-6 GATA transcription circuit guides aging in *C. elegans*. *Cell* **134**, 291-303, doi:S0092-8674(08)00707-1 [pii]
10.1016/j.cell.2008.05.044 (2008).

18 Viswanathan, M., Kim, S. K., Berdichevsky, A. & Guarente, L. A role for SIR-2.1 regulation of ER stress response genes in determining *C. elegans* life span. *Dev Cell* **9**, 605-615, doi:S1534-5807(05)00379-5 [pii]
10.1016/j.devcel.2005.09.017 (2005).

19 Menzel, R. *et al*. The Nematode *Caenorhabditis elegans*, Stress and Aging: Identifying the Complex Interplay of Genetic Pathways Following the Treatment with Humic Substances. *Front Genet* **3**, 50, doi:10.3389/fgene.2012.00050 (2012).

20 Pietsch, K. *et al*. Meta-Analysis of Global Transcriptomics Suggests that Conserved Genetic Pathways are Responsible for Quercetin and Tannic Acid Mediated Longevity in *C. elegans*. *Front Genet* **3**, 48, doi:10.3389/fgene.2012.00048 (2012).

21 Schmeisser, S. *et al*. Neuronal ROS signaling rather than AMPK/sirtuin-mediated energy sensing links dietary restriction to lifespan extension. *Molecular Metabolism* **2**, 1-11 (2013).

22 Gusarov, I. *et al*. Bacterial nitric oxide extends the lifespan of *C. elegans*. *Cell* **152**, 818-830, doi:S0092-8674(13)00014-7 [pii]
10.1016/j.cell.2012.12.043 (2013).

23 Staab, T. A. *et al*. The Conserved SKN-1/Nrf2 Stress Response Pathway Regulates Synaptic Function in *Caenorhabditis elegans*. *PLoS Genet* **9**, e1003354, doi:10.1371/journal.pgen.1003354
PGENETICS-D-12-02463 [pii] (2013).

24 Iser, W. B., Wilson, M. A., Wood, W. H., 3rd, Becker, K. & Wolkow, C. A. Co-regulation of the DAF-16 target gene, *cyp-35B1/dod-13*, by HSF-1 in *C. elegans* dauer larvae and *daf-2* insulin pathway mutants. *PLoS One* **6**, e17369, doi:10.1371/journal.pone.0017369 (2011).

25 Halaschek-Wiener, J. *et al*. Analysis of long-lived *C. elegans daf-2* mutants using serial analysis of gene expression. *Genome Res* **15**, 603-615, doi:gr.3274805 [pii]
10.1101/gr.3274805 (2005).

26 Zarse, K. *et al*. Impaired insulin/IGF1 signaling extends life span by promoting mitochondrial L-proline catabolism to induce a transient ROS signal. *Cell Metab* **15**, 451-465, doi:S1550-4131(12)00094-0 [pii]
10.1016/j.cmet.2012.02.013 (2012).

27 Shaw, W. M., Luo, S., Landis, J., Ashraf, J. & Murphy, C. T. The *C. elegans* TGF-beta Dauer pathway regulates longevity via insulin signaling. *Curr Biol* **17**, 1635-1645, doi:S0960-9822(07)01914-8 [pii]

10.1016/j.cub.2007.08.058 (2007).

28 Mair, W. *et al*. Lifespan extension induced by AMPK and calcineurin is mediated by CRTC-1 and CREB. *Nature* **470**, 404-408, doi:nature09706 [pii]
10.1038/nature09706 (2011).

29 Greer, E. L. *et al*. Members of the H3K4 trimethylation complex regulate lifespan in a germline-dependent manner in *C. elegans*. *Nature* **466**, 383-387, doi:nature09195 [pii]
10.1038/nature09195 (2010).

30 Cristina, D., Cary, M., Lunceford, A., Clarke, C. & Kenyon, C. A regulated response to impaired respiration slows behavioral rates and increases lifespan in *Caenorhabditis elegans*. *PLoS Genet* **5**, e1000450, doi:10.1371/journal.pgen.1000450 (2009).

31 Chen, D. *et al*. Germline signaling mediates the synergistically prolonged longevity produced by double mutations in *daf-2* and *rsks-1* in *C. elegans*. *Cell reports* **5**, 1600-1610, doi:10.1016/j.celrep.2013.11.018 (2013).

32 Chen, S. *et al*. The conserved NAD(H)-dependent corepressor CTBP-1 regulates *Caenorhabditis elegans* life span. *Proc Natl Acad Sci U S A* **106**, 1496-1501, doi:0802674106 [pii]
10.1073/pnas.0802674106 (2009).

33 Huang da, W., Sherman, B. T. & Lempicki, R. A. Bioinformatics enrichment tools: paths toward the comprehensive functional analysis of large gene lists. *Nucleic Acids Res* **37**, 1-13, doi:gkn923 [pii]
10.1093/nar/gkn923 (2009).

34 Huang, D. W., Sherman, B. T. & Lempicki, R. A. Systematic and integrative analysis of large gene lists using DAVID Bioinformatics Resources. *Nature Protoc.* **4**, 44-57 (2009).

35 Dennis, G., Jr. *et al*. DAVID: Database for Annotation, Visualization, and Integrated Discovery. *Genome Biol* **4**, P3 (2003).

36 Golden, T. R., Hubbard, A., Dando, C., Herren, M. A. & Melov, S. Age-related behaviors have distinct transcriptional profiles in *Caenorhabditis elegans*. *Aging Cell* **7**, 850-865, doi:ACE433 [pii]
10.1111/j.1474-9726.2008.00433.x (2008).

37 Selman, C. *et al*. Ribosomal protein S6 kinase 1 signaling regulates mammalian life span. *Science* **326**, 140-144, doi:10.1126/science.1177221 (2009).

38 Neff, F. *et al*. Rapamycin extends murine lifespan but has limited effects on aging. *J Clin Invest* **123**, 3272-3291, doi:10.1172/JCI67674 (2013).

39 Tsuchiya, T. *et al*. Additive regulation of hepatic gene expression by dwarfism and caloric restriction. *Physiological genomics* **17**, 307-315, doi:10.1152/physiolgenomics.00039.2004 (2004).

40 Dhahbi, J. M., Mote, P. L., Fahy, G. M. & Spindler, S. R. Identification of potential caloric restriction mimetics by microarray profiling. *Physiological genomics* **23**, 343-350, doi:10.1152/physiolgenomics.00069.2005 (2005).

41 Boylston, W. H., DeFord, J. H. & Papaconstantinou, J. Identification of longevity-associated genes in long-lived Snell and Ames dwarf mice. *Age (Dordr)* **28**, 125-144, doi:10.1007/s11357-006-9008-6 (2006).

42 Zhang, Y. *et al*. The starvation hormone, fibroblast growth factor-21, extends lifespan in mice. *eLife* **1**, e00065, doi:10.7554/eLife.00065 (2012).

43 Sun, L. Y. *et al*. Growth hormone-releasing hormone disruption extends lifespan and regulates response to caloric restriction in mice. *eLife* **2**, e01098, doi:10.7554/eLife.01098 (2013).

44 Osburn, W. O. *et al*. Genetic or pharmacologic amplification of nrf2 signaling inhibits acute inflammatory liver injury in mice. *Toxicol Sci* **104**, 218-227, doi:10.1093/toxsci/kfn079 (2008).

45 Joo, M. S., Lee, C. G., Koo, J. H. & Kim, S. G. miR-125b transcriptionally increased by Nrf2 inhibits AhR repressor, which protects kidney from cisplatin-induced injury. *Cell death & disease* **4**, e899, doi:10.1038/cddis.2013.427 (2013).

46 Brenner, S. The genetics of *Caenorhabditis elegans*. *Genetics* **77**, 71-94 (1974).

47 Hedgecock, E. M. & Herman, R. K. The *ncl-1* gene and genetic mosaics of *Caenorhabditis elegans*. *Genetics* **141**, 989-1006 (1995).

48 Robida-Stubbs, S. *et al*. TOR Signaling and Rapamycin Influence Longevity by Regulating SKN-1/Nrf and DAF-16/FoxO. *Cell Metab* **15**, 713-724, doi:S1550-4131(12)00147-7 [pii]
10.1016/j.cmet.2012.04.007 (2012).

49 Li, X. *et al*. Specific SKN-1/Nrf Stress Responses to Perturbations in Translation Elongation and Proteasome Activity. *PLoS Genet* **7**, e1002119, doi:10.1371/journal.pgen.1002119 PGENETICS-D-11-00172 [pii] (2011).

50 Hoogewijs, D., Houthoofd, K., Matthijssens, F., Vandesompele, J. & Vanfleteren, J. R. Selection and validation of a set of reliable reference genes for quantitative sod gene expression analysis in *C. elegans*. *BMC Mol Biol* **9**, 9, doi:1471-2199-9-9 [pii]
10.1186/1471-2199-9-9 (2008).

51 Liby, P. *et al*. BIR-1, the homologue of human Survivin, regulates expression of developmentally active collagen genes in *C. elegans*. *Folia Biol (Praha)* **52**, 101-108, doi:FB2006A0013 [pii] (2006).

52 Riddle, D. L., Blumenthal, T., Meyer, B. J. & Priess, J. R. in *C. elegans II* (eds D. L. Riddle, T. Blumenthal, B. J. Meyer, & J. R. Priess) (1997).

53 Riddle, D. L., Swanson, M. M. & Albert, P. S. Interacting genes in nematode dauer larva formation. *Nature* **290**, 668-671 (1981).

54 Klass, M. & Hirsh, D. Non-ageing developmental variant of *Caenorhabditis elegans*. *Nature* **260**, 523-525 (1976).

55 Kenyon, C. J. The genetics of ageing. *Nature* **464**, 504-512, doi:nature08980 [pii]
10.1038/nature08980 (2010).

56 Shore, D. E. & Ruvkun, G. A cytoprotective perspective on longevity regulation. *Trends in cell biology* **23**, 409-420, doi:10.1016/j.tcb.2013.04.007 (2013).

57 Dorman, J. B., Albinder, B., Shroyer, T. & Kenyon, C. The *age-1* and *daf-2* genes function in a common pathway to control the lifespan of *Caenorhabditis elegans*. *Genetics* **141**, 1399-1406 (1995).

58 Qi, W., Huang, X., Neumann-Haefelin, E., Schulze, E. & Baumeister, R. Cell-nonautonomous signaling of FOXO/DAF-16 to the stem cells of *Caenorhabditis elegans*. *PLoS Genet* **8**, e1002836, doi:10.1371/journal.pgen.1002836
PGENETICS-D-12-00777 [pii] (2012).

59 Narasimhan, S. D. *et al*. PDP-1 links the TGF-beta and IIS pathways to regulate longevity, development, and metabolism. *PLoS Genet* **7**, e1001377, doi:10.1371/journal.pgen.1001377 (2011).

60 Padmanabhan, S. *et al*. A PP2A regulatory subunit regulates *C. elegans* insulin/IGF-1 signaling by modulating AKT-1 phosphorylation. *Cell* **136**, 939-951, doi:S0092-8674(09)00073-7 [pii]
10.1016/j.cell.2009.01.025 (2009).
61 Boehm, M. & Slack, F. A developmental timing microRNA and its target regulate life span in *C. elegans*. *Science* **310**, 1954-1957, doi:310/5756/1954 [pii]
10.1126/science.1115596 (2005).
62 Melendez, A. *et al*. Autophagy genes are essential for dauer development and life-span extension in *C. elegans*. *Science* **301**, 1387-1391, doi:10.1126/science.1087782
301/5638/1387 [pii] (2003).
63 McCulloch, D. & Gems, D. Body size, insulin/IGF signaling and aging in the nematode *Caenorhabditis elegans*. *Exp Gerontol* **38**, 129-136, doi:S053155650200147X [pii] (2003).
64 Gerisch, B., Weitzel, C., Kober-Eisermann, C., Rottiers, V. & Antebi, A. A hormonal signaling pathway influencing *C. elegans* metabolism, reproductive development, and life span. *Dev Cell* **1**, 841-851, doi:S1534-5807(01)00085-5 [pii] (2001).
65 Larsen, P. L., Albert, P. S. & Riddle, D. L. Genes that regulate both development and longevity in *Caenorhabditis elegans*. *Genetics* **139**, 1567-1583 (1995).
66 Tissenbaum, H. A. & Ruvkun, G. An insulin-like signaling pathway affects both longevity and reproduction in *Caenorhabditis elegans*. *Genetics* **148**, 703-717 (1998).
67 Hars, E. S. *et al*. Autophagy regulates ageing in *C. elegans*. *Autophagy* **3**, 93-95, doi:3636 [pii] (2007).
68 Golden, J. W. & Riddle, D. L. The *Caenorhabditis elegans* dauer larva: developmental effects of pheromone, food, and temperature. *Dev Biol* **102**, 368-378, doi:0012-1606(84)90201-X [pii] (1984).
69 Golden, J. W. & Riddle, D. L. A pheromone-induced developmental switch in *Caenorhabditis elegans*: Temperature-sensitive mutants reveal a wild-type temperature-dependent process. *Proc Natl Acad Sci U S A* **81**, 819-823 (1984).
70 Ailion, M. & Thomas, J. H. Dauer formation induced by high temperatures in *Caenorhabditis elegans*. *Genetics* **156**, 1047-1067 (2000).
71 McElwee, J., Bubb, K. & Thomas, J. H. Transcriptional outputs of the *Caenorhabditis elegans* forkhead protein DAF-16. *Aging Cell* **2**, 111-121 (2003).
72 Lee, S. S., Kennedy, S., Tolonen, A. C. & Ruvkun, G. DAF-16 target genes that control C. elegans life-span and metabolism. *Science* **300**, 644-647, doi:10.1126/science.1083614 (2003).
73 Dong, M. Q. *et al*. Quantitative mass spectrometry identifies insulin signaling targets in C. elegans. *Science* **317**, 660-663, doi:10.1126/science.1139952 (2007).
74 Stout, G. J. *et al*. Insulin/IGF-1-mediated longevity is marked by reduced protein metabolism. *Mol Syst Biol* **9**, 679, doi:10.1038/msb.2013.35 (2013).
75 Samuelson, A. V., Carr, C. E. & Ruvkun, G. Gene activities that mediate increased life span of *C. elegans* insulin-like signaling mutants. *Genes Dev* **21**, 2976-2994, doi:21/22/2976 [pii]
10.1101/gad.1588907 (2007).
76 Curran, S. P., Wu, X., Riedel, C. G. & Ruvkun, G. A soma-to-germline transformation in long-lived *Caenorhabditis elegans* mutants. *Nature* **459**, 1079-1084, doi:nature08106 [pii]

10.1038/nature08106 (2009).

77 Castro, C. *et al*. A study of *Caenorhabditis elegans* DAF-2 mutants by metabolomics and differential correlation networks. *Molecular bioSystems* **9**, 1632-1642, doi:10.1039/c3mb25539e (2013).

78 Depuydt, G. *et al*. LC-MS proteomics analysis of the insulin/IGF-1-deficient *Caenorhabditis elegans daf-2(e1370)* mutant reveals extensive restructuring of intermediary metabolism. *Journal of proteome research* **13**, 1938-1956, doi:10.1021/pr401081b (2014).