Supplementary material

Table S1 List of 57 genes that show larval arrest after RNAi inactivation

RNAi clone	Gene	Brief description	Functional group
I-5I22	B0511.8	Mitochondrial 28S ribosomal protein S30	Mitochondria
I-6F08	C01A2.3	Cytochrome oxidase biogenesis protein	Mitochondria
II-3D02	C01F1.2	Putative cytochrome C oxidase assembly protein	Mitochondria
II-7L05	C09H10.3	NADH:ubiquinone oxidoreductase, NDUFV1/51kDa subunit	Mitochondria
II-7K16	C14A4.14	Mitochondrial 28S ribosomal protein S22	Mitochondria
I-3O24	F22D6.4	NADH:ubiquinone oxidoreductase, NDUFS6/13 kDa subunit	Mitochondria
I-4J22	F26E4.4	Cell death regulator Aven	Mitochondria
I-2O16	F27C1.7	Mitochondrial F1F0-ATP synthase, subunit OSCP/ATP5	Mitochondria
II-7A04	F54B3.3	AAA+-type ATPase	Mitochondria
I-2D03	F59A3.3	Mitochondrial/chloroplast ribosomal protein L24	Mitochondria
I-5C22	F59C6.5	NADH-ubiquinone oxidoreductase, subunit NDUFB10/PDSW	Mitochondria
II-6G09	K01C8.6	Mitochondrial ribosomal protein L10	Mitochondria
X-6J17	R03E1.2	ATPase membrane sector associated protein	Mitochondria
II-7G09	R53.4	Mitochondrial F1F0-ATP synthase, subunit f	Mitochondria
II-1K06	T02H6.11	Ubiquinol cytochrome c reductase, subunit QCR7	Mitochondria
I-2P08	T08B2.8	Predicted mitochondrial ribosomal protein L23	Mitochondria
II-5H24	T09A5.5	Putative NADH dehydrogenase	Mitochondria
X-1D12	T14F9.1	Vacuolar H+-ATPase V1 sector, subunit H	Mitochondria
II-7F06	VW02B12L.1	Vacuolar H+-ATPase V0 sector, subunit a	Mitochondria
I-3O12	W01A8.4	NADH-ubiquinone oxidoreductase B15 subunit	Mitochondria
I-7B02	Y47G6A.10	AAA+-type ATPase containing the peptidase M41 domain	Mitochondria
I-7G09	Y63D3A.7	NADH:ubiquinone oxidoreductase NDUFA2/B8 subunit	Mitochondria
I-1L24	ZK973.10	NADH:ubiquinone oxidoreductase, NDUFS4/18 kDa subunit	Mitochondria
I-2B19	C09D4.5	60s ribosomal protein L19	Translation
II-7P06	C47D12.6	Threonyl-tRNA synthetase	Translation
I-2C07	D1007.12	60s ribosomal protein L24	Translation
II-6N03	D2013.7	Translation initiation factor 3, subunit f (eIF-3f)	Translation
II-6E20	D2085.3	Translation initiation factor 2B, epsilon subunit (eIF-2Bepsilon/GCD6)	Translation
I-3O22	F22D6.3	Asparaginyl-tRNA synthetase	Translation
I-3D16	F26A3.2	Nuclear cap-binding protein complex	Translation
I-2H21	F55F8.3	WD40-repeat-containing subunit of the 18S rRNA processing complex	Translation
I-5C20	F59C6.4	Exosomal 3'-5' exoribonuclease complex subunit Rrp40	Translation
I-1O18	W01B11.3	Ribosome biogenesis protein - Nop58p/Nop5p	Translation
I-7J09	Y39G10AR.8	Translation initiation factor 2, gamma subunit (eIF-2gamma; GTPase)	Translation
I-7D04	Y48G1A.4	Nucleolar protein involved in 40S ribosome biogenesis	Translation
I-4L13	C17E4.5	Splicing factor RNPS1, SR protein superfamily	Transcription
X-5I11	C33D3.1	GATA-4/5/6 transcription factors	Transcription
I-2P20	C48E7.2	RNA polymerase III (C) subunit	Transcription
I-4H13	F14B4.3	RNA polymerase I, second largest subunit	Transcription
I-2H23	F55F8.4	CBF1-interacting corepressor CIR and related proteins	Transcription
I-2G04	F56A3.2	GIY-YIG type nuclease	Transcription
I-3M11	F57B10.1	CREB/ATF family transcription factor	Transcription
II-5E09	C56E6.1	Transporter, ABC superfamily	Signaling
II-6L23	D2013.5	Dynamin-like GTP binding protein	Signaling
I-1B06	F28H1.2	Calponin	Signaling
I-3I14	T21G5.4	PDZ domain	Signaling
I-5G21	C25A1.5	Cytochrome b5 ::Sphingolipid fatty acid hydroxylase	Metabolism
I-2I23	F54D7.2	sn-1,2-diacylglycerol ethanolamine- and cholinephosphotranferases	Metabolism
I-3G22	K02F2.2	S-adenosylhomocysteine hydrolase	Metabolism
I-2P06	T08B2.7	Hydroxyacyl-CoA dehydrogenase/enoyl-CoA hydratase	Metabolism
I-2J01	F55F8.5	Microtubule binding protein YTM1 (contains WD40 repeats)	Cell structure
I-1F10	R12E2.3	26S proteasome regulatory complex	Protein degradation
II-7D06	B0491.5	Uncharacterized protein	Unknown
I-3L20	F20G4.1	Uncharacterized conserved protein	Unknown
II-6N11	F42A8.1	Uncharacterized protein	Unknown
I-7I06	K11B4.1	Uncharacterized conserved protein	Unknown
I-7J11	Y39G10AR.7	Unnamed protein	Unknown

RNAi inactivation of 57 genes caused larval arrest phenotypes based on the previous genome-wide screen performed by Kamath *et al.* (2003). 23 genes (40%) are involved in mitochondrial functions; 12 genes are involved in mRNA translation (21%); 7 genes (12%) are involved transcription; 4 genes are involved in signal transduction (7%); 4 genes (7%) are involved in metabolism; 1 gene (2%) is involved in cellular function; 1 gene (2%) is involved in protein turnover; and 5 genes (9%) are uncharacterized with unknown functions.

Table S2 An RNAi screen identified 23 novel genes with antagonistic pleiotropy functions in *C. elegans* development and aging

RNAi clone	Gene	Brief description	Functional group	Blast E-value		
				Fly	Mouse	Human
I-7J09	Y39G10AR.8	eIF-2γ	Translation	6.4e-183	5.3e-188	3e-103
II-6E20	D2085.3	eIF-2Bε	Translation	3.3e-32	6e-36	6.7e-32
II-6N03	D2013.7	eIF-3f	Translation	2.1e-37	1.9e-43	3.1e-43
I-2B19	C09D4.5	60s ribosomal protein L19	Translation	2.5e-68	1e-46	8.9e-68
I-1O18	W01B11.3	Ribosome biogenesis protein Nop58p/Nop5p	Translation	3.5e-137	4e-126	2.3e-140
I-5C20	F59C6.4	3'-5' exoribonuclease complex subunit involved in rRNA processing	Translation	7.6e-36	7.2e-37	2.3e-38
I-7D04	Y48G1A.4	Nucleolar protein involved in 40S ribosome biogenesis	Translation	8.8e-55	1.4e-39	2.9e-46
II-7P06	C47D12.6	Threonyl-tRNA synthetase	Translation	4.4e-257	1.1e-248	3.2e-247
I-3O22	F22D6.3	Asparaginyl-tRNA synthetase	Translation	6.3e-173	1.2e-164	4.4e-174
I-2P20	C48E7.2	RNA polymerase III subunit	Transcription	1.6e-36	2.1e-52	2.5e-45
I-4H13	F14B4.3	RNA polymerase I subunit	Transcription	1.1e-243	2.4e-269	2.2e-266
II-7G09	R53.4	Mitochondrial F1F0-ATP synthase subunit f	Mitochondria	1.1e-11	5.2e-08	6.3e-05
I-5C22	F59C6.5	NADH:ubiquinone oxidoreductase subunit NDUFB10/PDSW	Mitochondria	4.3e-34	1.3e-15	6.5e-18
I-1L24	ZK973.10	NADH:ubiquinone oxidoreductase subunit NDUFS4	Mitochondria	9.2e-31	5.4e-36	4.3e-35
II-3D02	C01F1.2	Cytochrome C oxidase assembly protein	Mitochondria	2.8e-55	5.3e-54	3.8e-54
II-6G09	K01C8.6	Mitochondrial ribosomal protein L10	Mitochondria	1e-08	2.9e-08	3.5e-05
I-2D03	F59A3.3	Mitochondrial ribosomal protein L24	Mitochondria	2.1e-36	1.5e-25	1.5e-27
I-5I22	B0511.8	Mitochondrial 28S ribosomal protein S30	Mitochondria	0.0016	1.2e-07	1.3e-06
I-7B02	Y47G6A.10	AAA+-type ATPase	Mitochondria	5e-234	2e-232	3.3e-232
I-4J22	F26E4.4	Cell death regulator Aven	Mitochondria	1.3e-08	0.0032	0.00028
II-5E09	C56E6.1	ABC superfamily transporter	Signaling	1.8e-30	7.2e-41	7.5e-43
I-7I06	K11B4.1	Uncharacterized conserved protein	Unkown	5.7e-17	1.1e-05	1.1e-05
II-7D06	B0491.5	Uncharacterized protein	Unkown	0.0014	3.6	9.2

RNAi inactivation of 23 genes during adult extends lifespan. 9 genes (39%) are involved in mRNA translation; 9 genes (39%) are involved in mitochondrial functions; 2 genes (9%) are involved transcription; 1 genes are involved in signal transduction (4%); and 2 genes (9%) are uncharacterized with unknown functions. Blast-P analysis showed that most of the 23 genes have homologs/orthologs in other organisms with high sequence similarity.

Table S3 Lifespan extension in different genetic backgrounds

Gene	N2 lifespan					*daf-2(e1370)* lifespan					*daf-16(mu86); daf-2(e1370)* lifespan					*eat-2(ad465)* lifespan				
	Mean	**Max**	**Ext.**[a]	**n**[b]	***p***[c]	**Mean**	**Max**	**Ext.**[a]	**n**[b]	***p***[c]	**Mean**	**Max**	**Ext.**[a]	**n**[b]	***p***[c]	**Mean**	**Max**	**Ext.**[a]	**n**[b]	***p***[c]
Control RNAi	20.3	28	/	114	/	33.9	58	/	85	/	11.9	15	/	66	/	22.9	34	/	184	/
Y39G10AR.8	25.2	38	24%	86	<0.0001	47.8	68	41%	107	<0.0001	13.0	15	9%	85	0.0007	30.2	40	32%	109	<0.0001
D2085.3	24.0	32	18%	90	<0.0001	49.2	74	45%	101	<0.0001	13.3	17	12%	66	<0.0001	28.1	38	23%	95	<0.0001
D2013.7	25.9	42	28%	87	<0.0001	44.5	70	31%	113	<0.0001	13.5	19	13%	82	<0.0001	32.3	42	41%	83	<0.0001
C09D4.5	22.5	32	11%	115	<0.0001	50.9	72	50%	98	<0.0001	13.9	17	17%	59	<0.0001	28.3	38	24%	106	<0.0001
W01B11.3	23.4	32	15%	106	<0.0001	51.1	72	51%	68	<0.0001	13.3	16	12%	75	<0.0001	30.8	42	35%	94	<0.0001
F59C6.4	22.7	32	12%	84	<0.0001	45.5	68	34%	90	<0.0001	13.6	18	14%	63	<0.0001	29.2	40	28%	64	<0.0001
Y48G1A.4	24.5	40	21%	99	<0.0001	47.2	66	39%	101	<0.0001	13.1	15	10%	51	0.0010	27.3	36	19%	71	<0.0001
C47D12.6	22.8	30	12%	65	<0.0001	40.3	58	19%	72	0.0026	12.9	16	8%	73	0.0019	25.3	34	11%	66	0.0060
F22D6.3	22.5	36	11%	108	<0.0001	43.9	64	30%	96	<0.0001	13.0	16	9%	86	0.0005	29.2	40	28%	83	<0.0001
C48E7.2	25.5	40	26%	105	<0.0001	43.6	66	29%	109	<0.0001	13.6	19	14%	70	<0.0001	30.2	42	32%	57	<0.0001
F14B4.3	23.4	36	15%	96	<0.0001	44.4	64	31%	113	<0.0001	13.0	17	9%	59	0.0039	29.1	40	27%	76	<0.0001
R53.4	24.3	32	20%	111	<0.0001	53.3	74	57%	112	<0.0001	13.0	19	9%	82	0.0012	32.8	44	43%	76	<0.0001
F59C6.5	23.6	36	16%	102	<0.0001	50.4	70	49%	101	<0.0001	13.1	15	10%	60	0.0010	28.2	38	23%	92	<0.0001
ZK973.10	23.7	36	17%	107	<0.0001	44.5	64	31%	103	<0.0001	13.2	16	11%	54	0.0001	29.8	42	30%	55	<0.0001
C01F1.2	25.0	38	23%	89	<0.0001	42.1	66	24%	106	<0.0001	13.7	17	15%	59	<0.0001	26.7	42	17%	107	<0.0001
K01C8.6	23.7	38	17%	107	<0.0001	46.6	70	38%	108	<0.0001	13.4	16	13%	50	<0.0001	28.9	42	26%	67	<0.0001
F59A3.3	23.1	36	14%	97	<0.0001	43.3	68	28%	108	<0.0001	13.5	16	12%	75	<0.0001	29.4	40	28%	75	<0.0001
B0511.8	24.5	36	21%	97	<0.0001	48.7	74	44%	109	<0.0001	13.9	19	17%	55	<0.0001	28.7	38	25%	56	<0.0001
Y47G6A.10	25.2	40	24%	109	<0.0001	48.6	68	43%	105	<0.0001	13.4	18	13%	89	<0.0001	28.6	38	25%	68	<0.0001
F26E4.4	23.8	38	17%	91	<0.0001	43.0	62	27%	105	<0.0001	13.0	15	9%	65	0.0008	27.4	34	20%	62	<0.0001
C56E6.1	23.5	36	16%	87	<0.0001	46.9	68	38%	97	<0.0001	13.6	16	14%	71	<0.0001	29.1	40	27%	39	<0.0001
K11B4.1	23.6	36	16%	83	<0.0001	43.8	68	29%	71	<0.0001	13.2	17	11%	59	0.0001	29.4	44	28%	87	<0.0001
B0491.5	23.6	36	16%	95	<0.0001	48.3	66	43%	105	<0.0001	13.0	15	9%	66	<0.0001	30.8	42	35%	67	<0.0001

RNAi inactivation of the 23 identified genes during adult cause lifespan extension in the wild-type N2, *daf-2*, *daf-16; daf-2* and *eat-2* backgrounds. All lifespan experiments were performed at 20°C. Lifespan of animals treated with different RNAi were compared with those treated with the control RNAi using the log-rank method.

[a] Mean lifespan extension compared to animals treated with the control RNAi.

[b] Total numbers of animals scored.

[c] *p*-values were calculated using the log-rank method (Graphic Prism 4).

Table S4 Heat stress, oxidative stress and fecundity assays

Gene	Brief description	Functional group	Heat stress[a]			Oxidative stress[e]			Fecundity[f]		
			Extension[b]	n[c]	*p*[d]	Extension[b]	n[c]	*p*[d]	Brood size[g]	n[c]	*p*[h]
/	Control RNAi	/	/	167	/	/	33	/	330.2 ± 6.4	36	/
Y39G10AR.8	eIF-2γ	Translation	11%	37	0.0021	51%	43	<0.0001	29.4 ± 1.9	10	<0.0001
D2085.3	eIF-2Bε	Translation	-3%	35	0.8083	10%	34	0.1069	94.7 ± 7.7	11	<0.0001
D2013.7	eIF-3f	Translation	16%	40	<0.0001	46%	33	<0.0001	52.5 ± 2.6	12	<0.0001
C09D4.5	rpl-19	Translation	10%	39	0.0084	54%	45	<0.0001	49.8 ± 5.4	10	<0.0001
W01B11.3	Ribosome biogenesis protein Nop58p/Nop5p	Translation	17%	47	<0.0001	0%	36	0.8704	192.0 ± 15.6	10	<0.0001
F59C6.4	3'-5' exoribonuclease involved in rRNA processing	Translation	11%	62	0.0005	8%	44	0.3722	264.4 ± 10.3	11	<0.0001
Y48G1A.4	Nucleolar protein involved in 40S ribosome biogenesis	Translation	8%	31	0.0422	67%	32	<0.0001	233.2 ± 13.9	13	<0.0001
C47D12.6	Threonyl-tRNA synthetase	Translation	11%	43	0.0032	0%	11	0.8757	68.2 ± 9.4	11	<0.0001
F22D6.3	Asparaginyl-tRNA synthetase	Translation	15%	51	<0.0001	-10%	42	0.0880	67.1 ± 7.2	12	<0.0001
C48E7.2	RNA polymerase III subunit	Transcription	12%	48	<0.0001	54%	23	<0.0001	307.9 ± 12.5	12	0.0999
F14B4.3	RNA polymerase I subunit	Transcription	7%	35	0.0991	-8%	43	0.1575	264.1 ± 13.7	13	<0.0001
R53.4	Mitochondrial F1F0-ATP synthase subunit f	Mitochondria	19%	50	<0.0001	44%	38	<0.0001	73.4 ± 8.5	11	<0.0001
F59C6.5	NADH:ubiquinone oxidoreductase subunit NDUFB10/PDSW	Mitochondria	17%	48	<0.0001	41%	35	<0.0001	174.4 ± 12.8	13	<0.0001
ZK973.10	NADH:ubiquinone oxidoreductase subunit NDUFS4	Mitochondria	16%	45	<0.0001	3%	35	0.7561	252.3 ± 20.1	9	<0.0001
C01F1.2	Cytochrome C oxidase assembly protein	Mitochondria	10%	32	0.0051	15%	42	0.0242	237.0 ± 19.3	12	<0.0001
K01C8.6	Mitochondrial rpl-10	Mitochondria	8%	42	0.0221	28%	33	0.0043	186.4 ± 15.3	10	<0.0001
F59A3.3	Mitochondrial rpl-24	Mitochondria	5%	40	0.1515	-5%	43	0.5427	284.5 ± 10.8	13	0.0007
B0511.8	Mitochondrial rps-30	Mitochondria	21%	67	<0.0001	49%	45	<0.0001	202.5 ± 13.6	11	<0.0001
Y47G6A.10	AAA+-type ATPase	Mitochondria	6%	37	0.0298	28%	27	0.0049	66.0 ± 5.3	13	<0.0001
F26E4.4	Cell death regulator Aven	Mitochondria	7%	57	0.0076	13%	36	0.0666	270.0 ± 9.1	10	<0.0001
C56E6.1	ABC superfamily transporter	Signaling	3%	41	0.4018	0%	45	0.8226	238.0 ± 12.5	11	<0.0001
K11B4.1	Uncharacterized protein	Unkown	11%	29	0.0082	-8%	47	0.1101	242.4 ± 6.2	11	<0.0001
B0491.5	Uncharacterized protein	Unkown	10%	40	0.0097	41%	34	<0.0001	172.8 ± 15.5	11	<0.0001

RNAi inactivation of some identified genes during adult results in increased heat stress resistance, oxidative stress resistance, and reduced fecundity.

[a] Heat stress assays were performed at 35°C. Survival of animals treated with different RNAi were compared with those treated with the control RNAi using the log-rank method. Significant increases in heat stress resistance ($p < 0.05$) were highlighted.

[b] Mean survival extension compared to animals treated with the control RNAi.

[c] Total numbers of animals scored.

[d] *p*-values were calculated using the log-rank method (Graphic Prism 4).

[e] Oxidative stress assays were performed by exposing animals to juglone (360 mM). Survival of animals treated with different RNAi were compared with those treated with the control RNAi using the log-rank method. Significant increases in oxidative stress resistance ($p < 0.05$) were highlighted.

[f] Brood sizes of animals treated with different RNAi were compared with those treated with the control RNAi using the *t*-test. Significant reductions in fecundity ($p < 0.05$) were highlighted.

[g] Mean number of progeny produced by one hermaphrodite animals ± standard error of mean.

[h] *p*-values were calculated using the *t*-test method (Graphic Prism 4).