

## Supplementary information

### Supplementary Material

#### Construction of *fat-5::gfp* and *fat-7::gfp* transgenic strains

The *fat-5::gfp* and *fat-7::gfp* transgenes were created using methods previously described by Frokjaer-Jensen *et al.* (1), and the transgenic strains were created by microinjection. In brief, an extrachromosomal transgenic strain was made by injection into the EG4322 (*ttTi5605;unc-119(ed3)*). The injection mix consisted of 50 ng/μl pCFJ151 (*ttTi5605\_MCS*) inserted with a target DNA fragment from N2 and the GFP gene from pPD95.75, pJL43.1 (*P<sub>glh-2::MosTase::glh-2utr</sub>*), and 2.5 ng/μl pCFJ90 (*P<sub>myo-2::mCherry</sub>*). DNA mixtures were injected into the gonads of young adult *C. elegans*. The integrated transgenic strains were assigned *kunEx161[unc-119(ed3);fat-5::gfp+unc-119(+)]* and *kunEx162[unc-119(ed3);fat-7::gfp+unc-119(+)]*. The *fat-5::gfp* and *fat-7::gfp* are translational constructs including the promoter regions above 2 kb upstream of the start codon and genomic sequences of *fat-5* and *fat-7*, respectively. The primers used for amplification of *fat-5::gfp* and *fat-7::gfp* were *fat-5F*: GGATATCTGGATCCACGAAAAGTTGCCGGAAATCAAGTG, *fat-5R*: GTCGACCTGCAGGCATGCAATCCCAATTTGTGGAGCATTTTCT, *fat-7F*: GGATATCTGGATCCACGAAGAGCCGAATATGCACAGAAA, *fat-7R*: GTCGACCTGCAGGCATGCAACATGATCGATTTTTTTTCTTGATTCTTC, GFP+unc-54 3'UTR F: TTGCATGCCTGCAGGTCGAC, GFP+unc-54 3'UTR R: CCAGAGCTCACCTAGGTATCTGCCGACTAGTAGGAAACAGT, pCFJ151-F: AGATACCTAGGTGAGCTCTGG, and pCFJ151-R: TTCGTGGATCCAGATATCC.

#### Primer information for *sbp-1* and *sur-7* cDNA

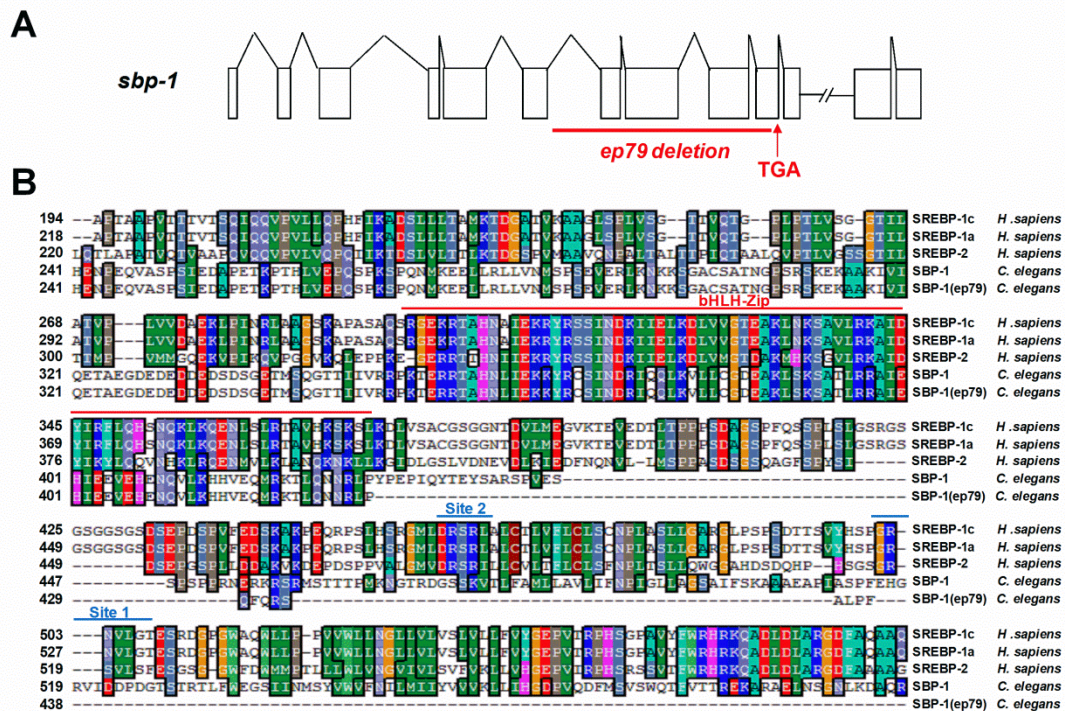
The forward primer sequences for *sbp-1* cDNA were F: CATGGTGGACGCACGTTCTGAC; and the reverse primer was R: TCGGCACGTGAGCTCGTAGAC. The primer sequences for

*sur-7* transcripts in the wild type and three alleles (*kun84*, *tm6523* and *ku119*) were F:  
CAGTTGAAGAGGCTCATGAGC and R: AACGTCTCCAGTACTTTTCGG.

### **Supplementary references**

1. Frokjaer-Jensen, C., M. W. Davis, C. E. Hopkins, B. J. Newman, J. M. Thummel, S. P. Olesen, M. Grunnet, and E. M. Jorgensen. 2008. Single-copy insertion of transgenes in *Caenorhabditis elegans*. *Nat Genet* **40**: 1375-1383.

## Supplementary Figures and Figure Legends

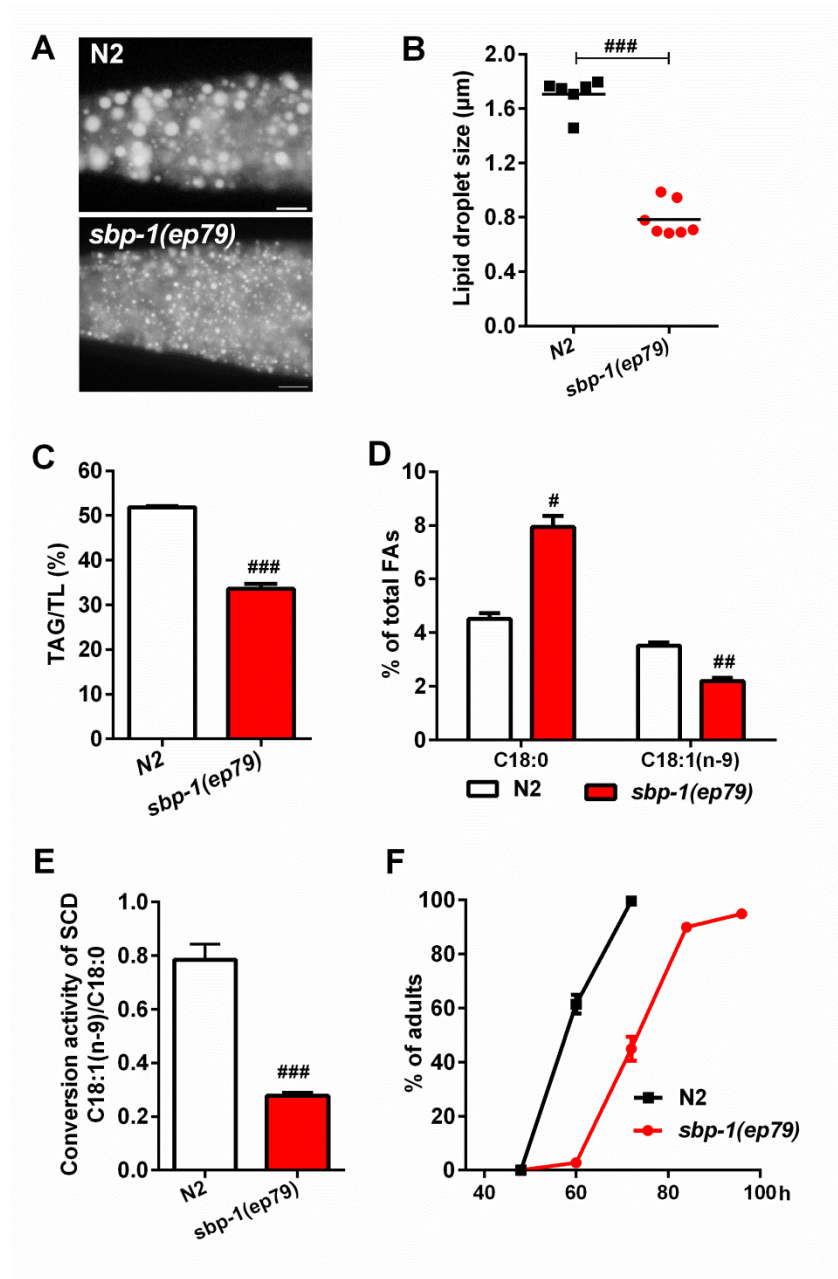


### Supplemental Figure S1. *sbp-1* encodes a homologue of SREBP in *C. elegans*.

(A) The gene structure and *ep79* mutation of *sbp-1*. The *ep79* mutation has a 2171 bp deletion, which leads to early protein maturation by the termination codon of TGA.

(B) Multi-alignment of SREBP from *C. elegans* [SBP-1 and truncated SBP-1(ep79)] and humans. The conserved domain is shown above the sequences. Red bar: The transcriptional domain. Blue bar: The splice site 1 and site 2 by S1P and S2P, respectively. SREBP-1a:

NP\_004167.1, SREBP-1c: NP\_001308025.1, SREBP-2: NP\_004590.2, SBP-1: CAA21042.1.



**Supplemental Figure S2. The phenotypes of *sbp-1(ep79)* mutant.**

(A) Nile Red staining of fixed worms. Representative animals, the anterior is on the right and the posterior is on the left. Scale bar represents 10  $\mu\text{m}$ .

(B) Lipid droplet size in the posterior region of intestines from 6-7 worms of each worm strain. The data are presented as scatter dot plots and means.

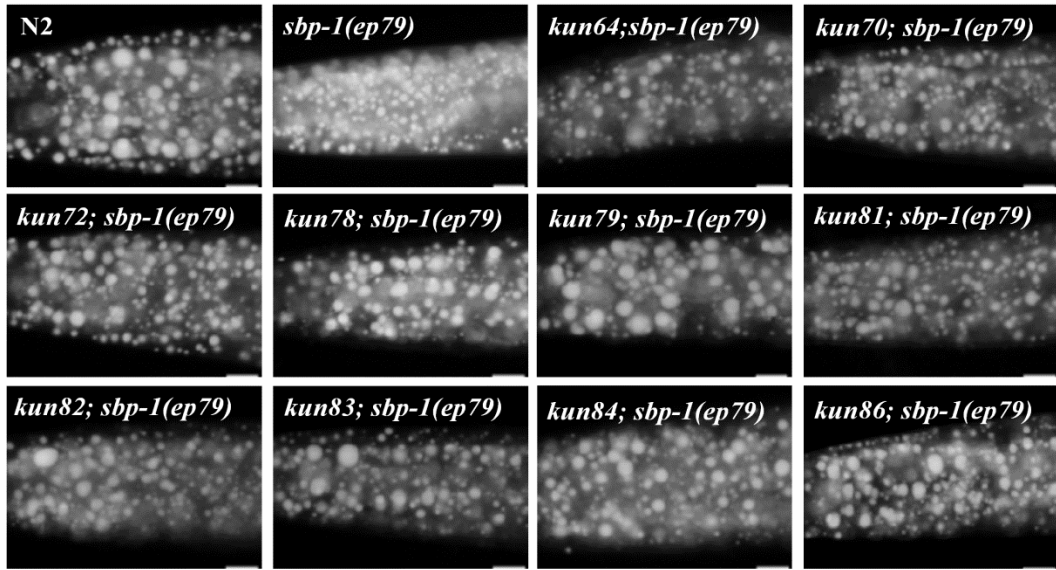
(C) Percentage of triacylglycerol (TAG) in total lipid (TAG + PL). The data are presented as the mean  $\pm$  SEM of 3 biological repeats.

(D) Percentage of C18:0 and C18:1(n-9) in total fatty acids. The data are presented as the mean  $\pm$  SEM of 4 biological repeats.

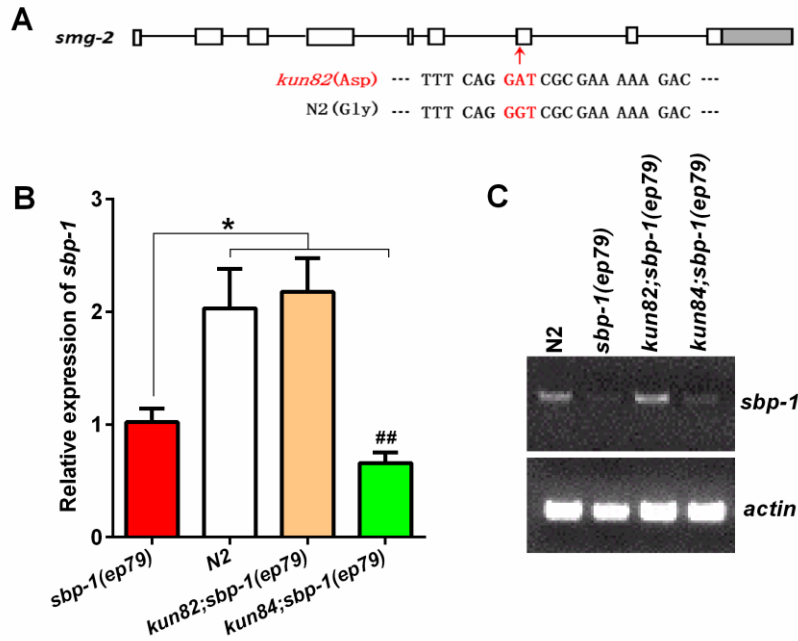
(E) The conversion activity of SCD presented as the ratio of C18:1(n-9)/C18:0. The data are presented as the mean  $\pm$  SEM of 4 biological repeats.

(F) The growth rate of worms. The data are presented as the mean  $\pm$  SEM of 3 biological repeats, of which at least 500 worms were counted for each worm strain.

The statistical analysis was performed using analysis of variance (ANOVA). Significant difference between *sbp-1(ep79)* mutant and wild type (N2), #:  $P < 0.05$ , ##:  $P < 0.01$  ###:  $P < 0.001$ .



**Supplemental Figure S3. Nile Red staining of fixed worms of *sbp-1(ep79)* suppressors.**  
Representative animals, the anterior is on the right and the posterior is on the left. Scale bar represents 10  $\mu$ m.



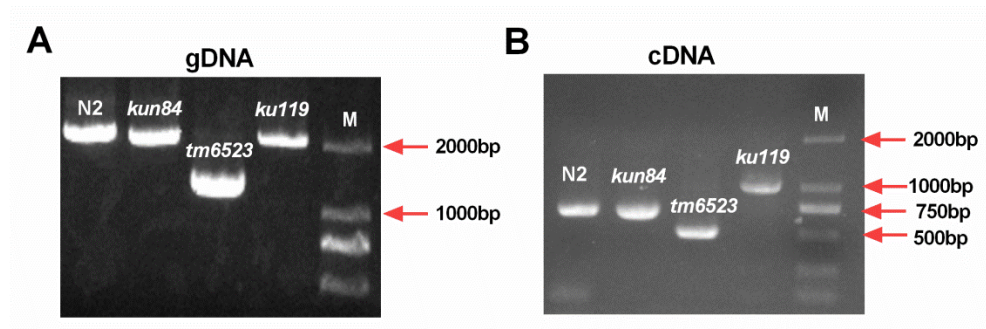
**Supplemental Figure S4. *kun82* mutation of *smg-2*.**

(A) The gene structure and *kun82* mutation of *smg-2*.

(B) Relative mRNA expression of *sbp-1* by QPCR. The data presented are the mean  $\pm$  SEM of 4 biological repeats.

(C) The mRNA expression of *sbp-1* by semi-QPCR.

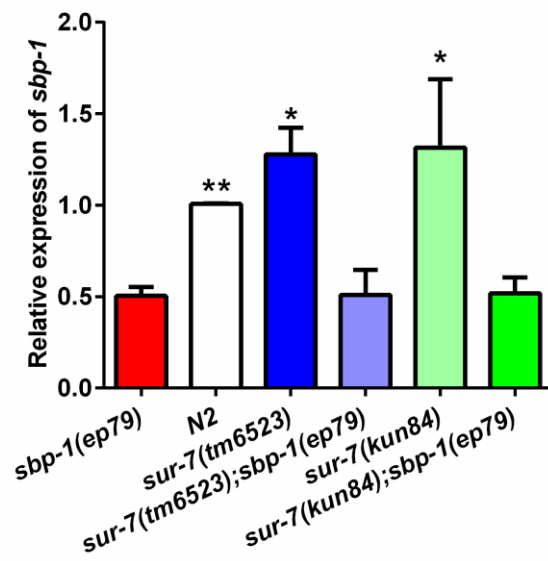
The statistical analysis was performed using analysis of variance (ANOVA). Significant difference between WT (N2) and a specific worm strain, ##:  $P < 0.01$ . Significant difference between *sbp-1(ep79)* mutant and a specific worm strain, \*:  $P < 0.05$ .



**Supplemental Figure S5. The genotypes of *sur-7* alleles.**

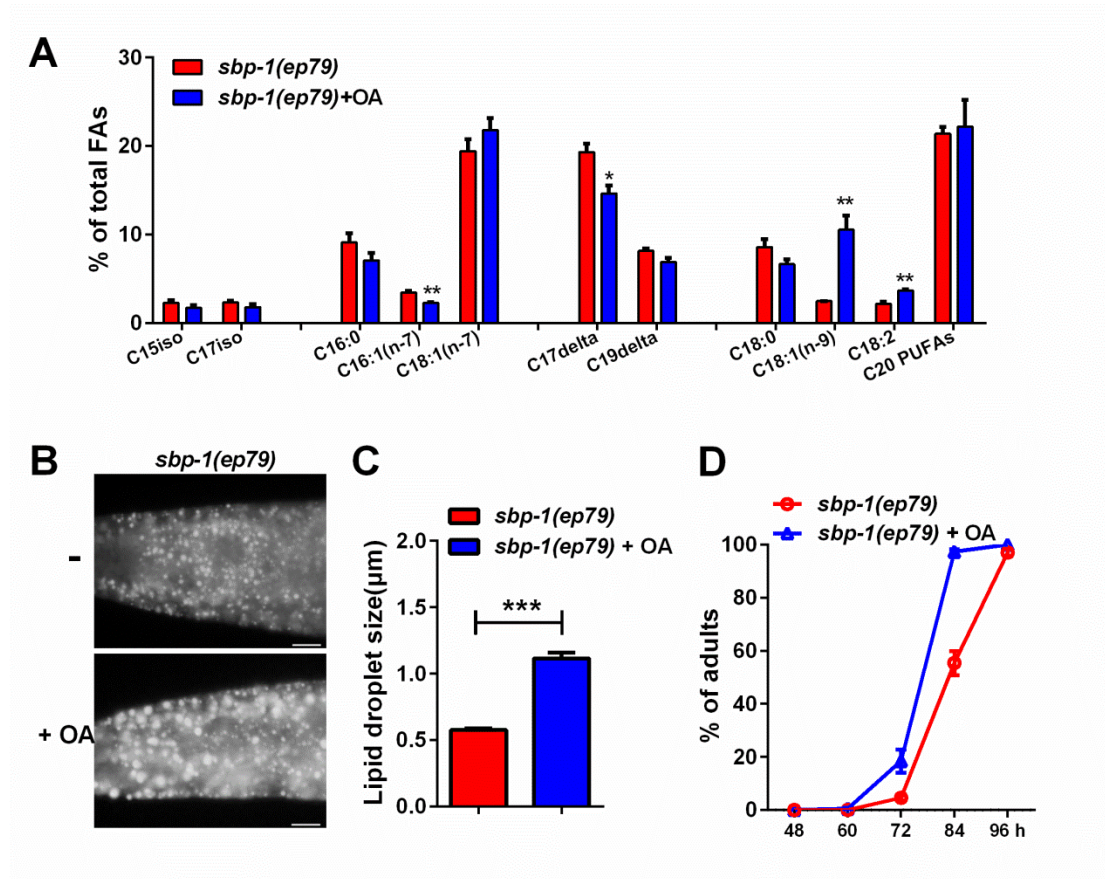
gDNA amplicon (A) and cDNA transcripts (B) of *sur-7* in wild type N2 (686 bp, 1978 bp) and three mutants *kun84* (686 bp, 1978 bp), *tm6523* (490 bp, 1412 bp) and *ku119* (1147 bp, 1978 bp).





**Supplemental Figure S6. Relative mRNA expression of *sbp-1* by QPCR.**

The data presented are the mean  $\pm$  SEM of 3-4 biological repeats. The statistical analysis was performed using analysis of variance (ANOVA). Significant difference between the *sbp-1(ep79)* mutant and a specific worm strain, \*:  $P < 0.05$ , \*\*:  $P < 0.01$ .



**Supplemental Figure S7. Dietary oleic acid (OA, C18:1(n-9)) improves the fat accumulation and growth of *sbp-1(ep79)* mutant.**

(A) Fatty acid profiles. The data presented are the mean  $\pm$ SEM of 4 biological repeats.

(B) Nile Red staining of fixed worms. Representative animals, anterior is on the right and posterior is on the left. Scale bar represents 10  $\mu$ m.

(C) Lipid droplet size in the posterior region of intestines from 6 worms of each worm condition. The data are presented as mean  $\pm$ SEM.

(D) The growth rate of worms. The data are presented as the mean  $\pm$ SEM of 3 biological repeats, of which at least 500 worms were counted for each worm strain.

The statistical analysis was performed using the t-test. Significant difference between with OA and without OA, \*:  $P < 0.05$ , \*\*:  $P < 0.01$ , \*\*\*:  $P < 0.001$ .

## Supplementary Tables

### Supplemental Table S1. Experimental Models: Organisms/Strains

<i>C. elegans</i>	
N2	Caenorhabditis Genetics Center (CGC)
CB4856	CGC
EG4322( <i>ttTi5605;unc-119(ed3)</i> )	CGC
<i>sur-7(ku119)</i>	CGC
XA6900, <i>pha-1(e2123ts)III;qaEx01[Pftn-1::pes-10::GFP-his;pha-1(+)]</i>	CGC
BX115, <i>lin-15(n765)X;waEx16[fat-6::GFP;lin-15(+)]</i>	Lab of Watts, J.L.
KQ377,N2; <i>ftISf[epEx307[unc-119(+);Psbp-1::GFP::SBP-1]</i>	Lab of Watts, J.L.
<i>sbp-1(ep79)</i>	Lab of Watts, J.L.
<i>fat-6(tm331);fat-7(wa37)</i>	Lab of Watts, J.L.
<i>kunEx161[unc-119(ed3);fat-5::gfp+unc-119(+)]</i>	This study
<i>kunEx162[unc-119(ed3); fat-7::gfp+unc-119(+)]</i>	This study
<i>sur-7(tm6523);sbp-1(ep79)</i>	This study
<i>sur-7(ku119);sbp-1(ep79)</i>	This study
<i>sur-7(tm6523)</i>	National Bioresource Project
<i>E. coli</i>	
OP50	CGC
HT115	Ahringer <i>C. elegans</i> RNAi library
Plasmids	
<i>pJL43.1 (Pglh-2::MosTase::glh-2utr)</i>	Lab of Guang, S.H.
<i>pCFJ90 (Pmyo-2::mCherry::unc54utr)</i>	Lab of Guang, S.H.
<i>pCFJ151(ttTi5605_MCS)</i>	Lab of Guang, S.H.
<i>pPD95.75</i>	CGC

**Supplemental Table S2. List of genes that are upregulated or downregulated by zinc reduction (TPEN 2.5  $\mu$ M and 5  $\mu$ M) in the transcriptional profile. The red highlights indicate upregulation and blue indicates downregulation.**

Gene name	log2 (fold-change)		Biological function
	T2.5 vs. CON	T5 vs. CON	
Y53G8AM.5	4.15859	9.1586	Integral to membrane
<i>clec-17</i>	3.48039	7.73523	CUB domain, C-type lectin
R08F11.4	3.37378	8.66818	Methyltransferase type 11
<i>cyp-14A4</i>	3.32968	6.66057	Cytochrome P450 family
Y54G9A.4	3.29906	6.08956	Zinc/iron permease
F44E7.5	3.03179	5.14599	unknown
T02B5.1	3.03005	5.01186	unknown
F56C3.9	2.83383	4.97506	Peptidase, cysteine peptidase active site
<i>cyp-13B1</i>	2.42192	4.082	Cytochrome P450 family
<i>col-176</i>	2.39094	5.47871	Integral to membrane
<i>dpy-6</i>	2.25449	4.51357	Body morphogenesis
Y39G8B.7	2.25276	7.19973	Metridin-like ShK toxin
F40F8.5	2.11251	3.16296	unknown
<i>lip1-4</i>	2.10546	3.30098	LIPase Like
<i>col-128</i>	2.06526	3.05315	COLlagen
<i>cdh-7</i>	2.01772	3.66764	CaDHerin family
<i>dpy-17</i>	1.9434	3.06625	Body morphogenesis
<i>ins-35</i>	1.92629	3.30605	INSulin related
F35F10.1	1.86259	4.08071	PAW domain
<i>wrt-10</i>	1.8498	2.69377	WaRThog (hedgehog-like family)
F59D6.3	1.80578	4.87681	Proteolysis
C15F1.2	1.7826	2.67402	unknown
<i>let-653</i>	1.69241	3.02024	PAN/Apple domain

<i>col-98</i>	1.65801	5.09508	COLlagen
<i>ftn-1</i>	1.64566	3.36036	Ferritin
H19M22.3	1.64332	2.5008	Zinc Metalloprotease
<i>ugt-57</i>	1.63253	3.07465	UDP-glucuronosyl/UDP-glucosyltransferase
F12E12.11	1.57206	4.69358	Oxidation reduction
<i>cdh-10</i>	1.51497	2.75909	CaDHerin family
B0205.13	1.48627	3.93527	unknown
<i>cyp-13A12</i>	1.48343	3.19764	Cytochrome P450 family
<i>clec-70</i>	1.47895	3.59687	Lectin C-type domain
H41C03.1	1.46756	2.92563	CRAL-TRIO lipid binding domain
<i>peb-1</i>	1.45934	2.63915	Pharyngeal Enhancer Binding
R186.1	1.44084	2.67407	Transport and Golgi organization protein 2
C09F9.2	1.38334	2.27175	Cellobiose dehydrogenase
<i>cyp-14A1</i>	1.36872	4.28838	Cytochrome P450 family
H14E04.1	1.34922	2.58371	Cyclopropane-fatty-acyl-phospholipid synthase activity
<i>col-103</i>	1.3482	4.70255	Collagen triple helix repeat
F46F2.3	1.30777	3.06452	Embryo development
<i>ugt-62</i>	1.27479	2.8616	UDP-glucosyltransferase
F19C7.2	1.24814	2.26139	Peptidase activity
Y69H2.14	1.22206	4.67948	Collagen triple helix repeat
F58G6.9	1.19581	2.9528	Ctr copper transporter
<i>ugt-19</i>	1.13324	3.04431	UDP-glucosyltransferase
Y39A1A.9	1.08424	2.32969	Meiosis-specific coiled-coil domain-containing protein MEIOC
<i>cdh-3</i>	1.08326	2.13896	CaDHerin family
<i>nspc-16</i>	1.06096	2.38994	Nematode Specific Peptide family, group C
<i>cdr-4</i>	1.04886	3.88268	CDR-4 protein; Thioredoxin fold
<i>ugt-61</i>	1.02708	2.66684	UDP-glucosyltransferase
DC2.5	1.02045	2.74685	Oxidoreductase activity

F17B5.1	1.01637	2.35435	Thioredoxin-like fold
<i>lec-4</i>	0.985009	2.90739	C-type lectin
<i>cdr-2</i>	0.980387	3.10618	Glutathione S-transferase
F32H5.1	0.776059	2.35806	Peptidase C1A
W05G11.6	0.7679	2.21235	PhosphoenolpyruvateCarboxyKinase
<i>smf-3</i>	-0.412504	-1.4595	NRAMP family
<i>fat-7</i>	-0.434893	-2.98472	Delta-9 fatty acid desaturase
<i>sod-1</i>	-0.663782	-1.71361	Copper/zinc superoxide dismutase
ZK6.11	-0.747434	-3.17365	Transmembrane glycoprotein
F55G11.4	-0.772387	-1.69585	CUB-like domain
<i>ilys-5</i>	-0.805839	-3.40043	Invertebrate lysozyme
T25B6.2	-0.888966	-3.52936	NEPrilysinmetallopeptidase family
<i>fat-5</i>	-0.89422	-1.45747	Delta-9 fatty acid desaturase
C10C5.4	-0.999393	-2.02704	Peptidase M20
<i>col-137</i>	-1.01546	-3.00272	COLlagen
F54D10.8	-1.09477	-4.80133	DUF23
F25B4.8	-1.19106	-3.34302	Carbon-sulfur lyase activity
F15E11.15	-1.19705	-4.45448	Protein Up-regulated in Daf-2(gf)
<i>thn-2</i>	-1.26419	-2.60181	THaumatococcus family
<i>pgp-6</i>	-1.37754	-2.56992	P-Glycoprotein related
Y52E8A.4	-1.57916	-3.22872	Plugged Excretory Pore
<i>gly-1</i>	-1.69268	-4.4856	GLYcosylation related
D1014.6	-1.82807	-8.42167	DUF23
D1014.7	-2.03979	-7.72243	DUF23
Y39B6A.24	-2.06022	-3.95699	ASpartyl Protease
<i>mtl-2</i>	-5.60473	-6.56982	Metallothioneins
C36C5.14	-2.092	-inf	DUF19