Li et al., http://www.jcb.org/cgi/content/full/jcb.201111053/DC1

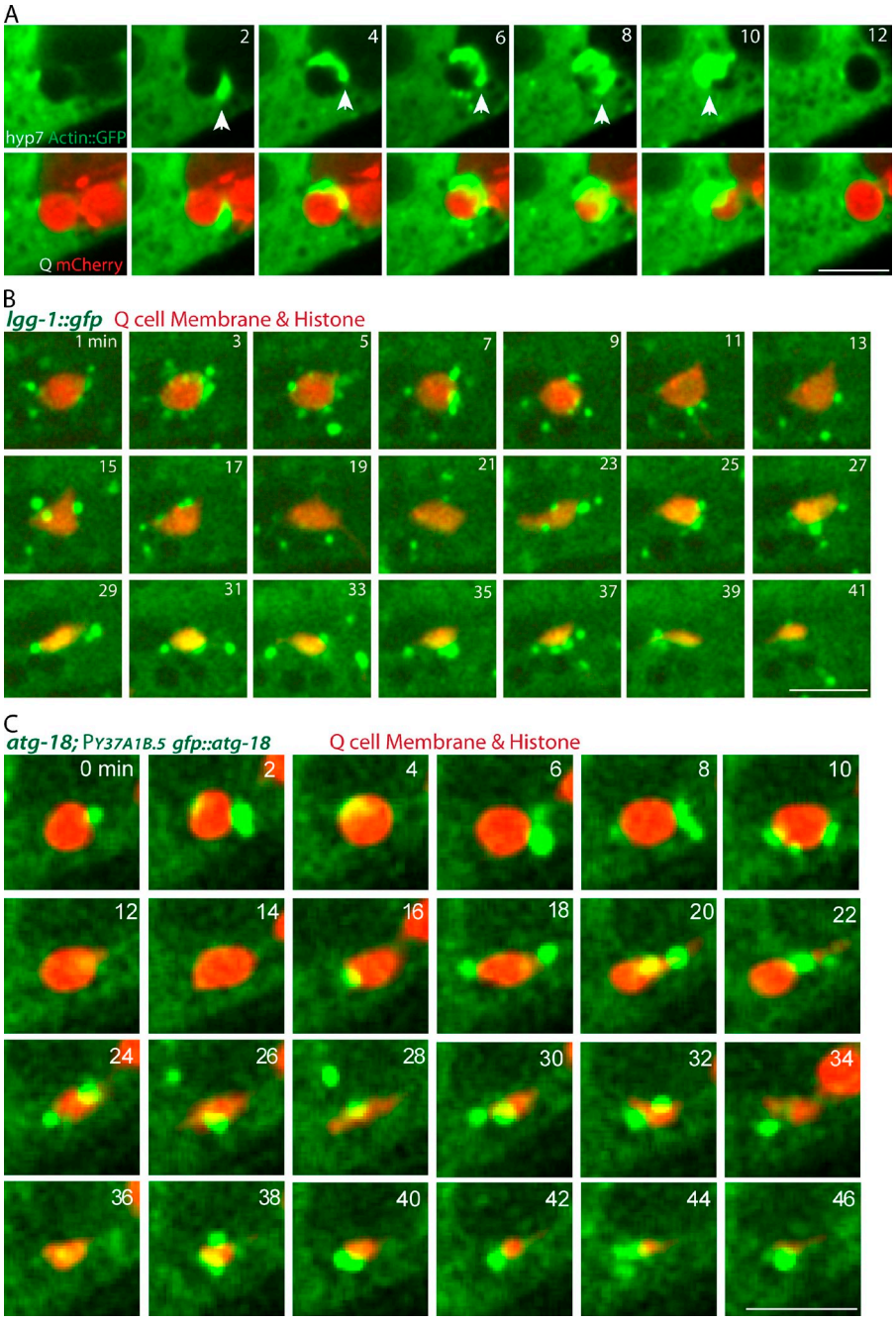


Figure S1. **Actin halo formation and LGG-1 and ATG-18 recruitment of the Q cell corpse.** (A) Still images of actin::GFP in an epithelial cell, hyp7, show actin halo on an apoptotic Q cell (labeled by cytosolic mCherry). Time on the right top is in minutes. The arrows show the formation of actin halo on the Q cell corpse. (B and C) Still images of autophagy markers LGG-1/LC3 (green in B) or ATG-18 (green in C) recruitment onto the outer surface of the Q cell corpse. GFP-tagged LGG-1 and ATG-18 proteins were expressed under either the endogenous promoter for *lgg-1* or hyp7 cell–specific promoter for *atg-18*. The Q cell plasma membrane (mCherry with a myristoylation signal) and histone (his-24::mCherry) were specifically labeled by the *egl-17* promoter. Bars, 5 µm.

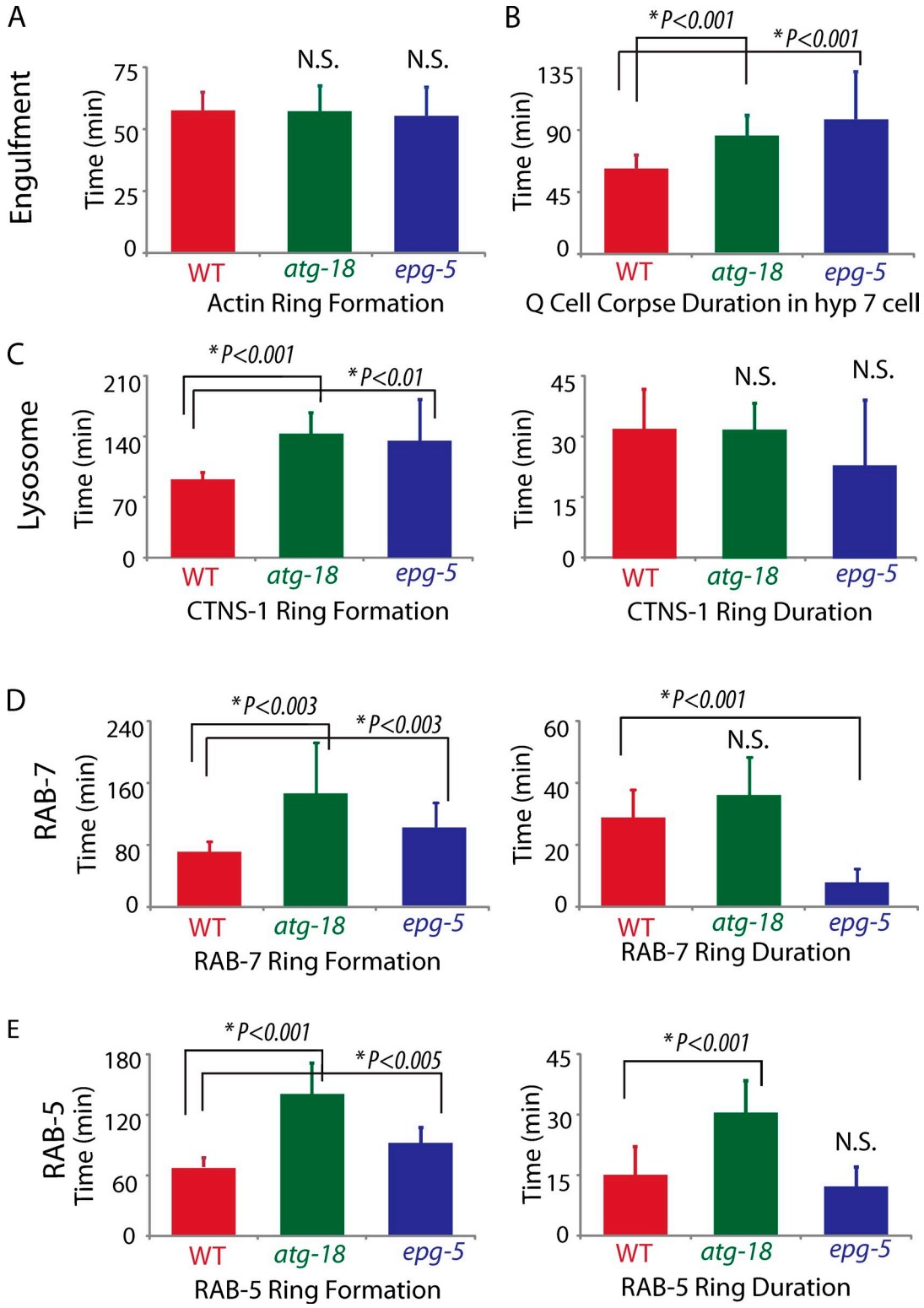


Figure S2. **Statistical analysis of Q cell corpse degradation in WT and autophagy mutants.** (A) Q cell corpse engulfment. (B) Q cell corpse duration in the hyp7 cell. (C) Lysosome/CTNS-1::GFP recruitment and duration. (D) GFP::RAB-7 recruitment and duration. (E) GFP::RAB-5 recruitment and duration. Data in A and B and C–E are the same as data in Fig. 2 C and Fig. 3 (A–C), respectively. Data shown are the means ± SD; *n* = 10–29 per group in a single experiment.

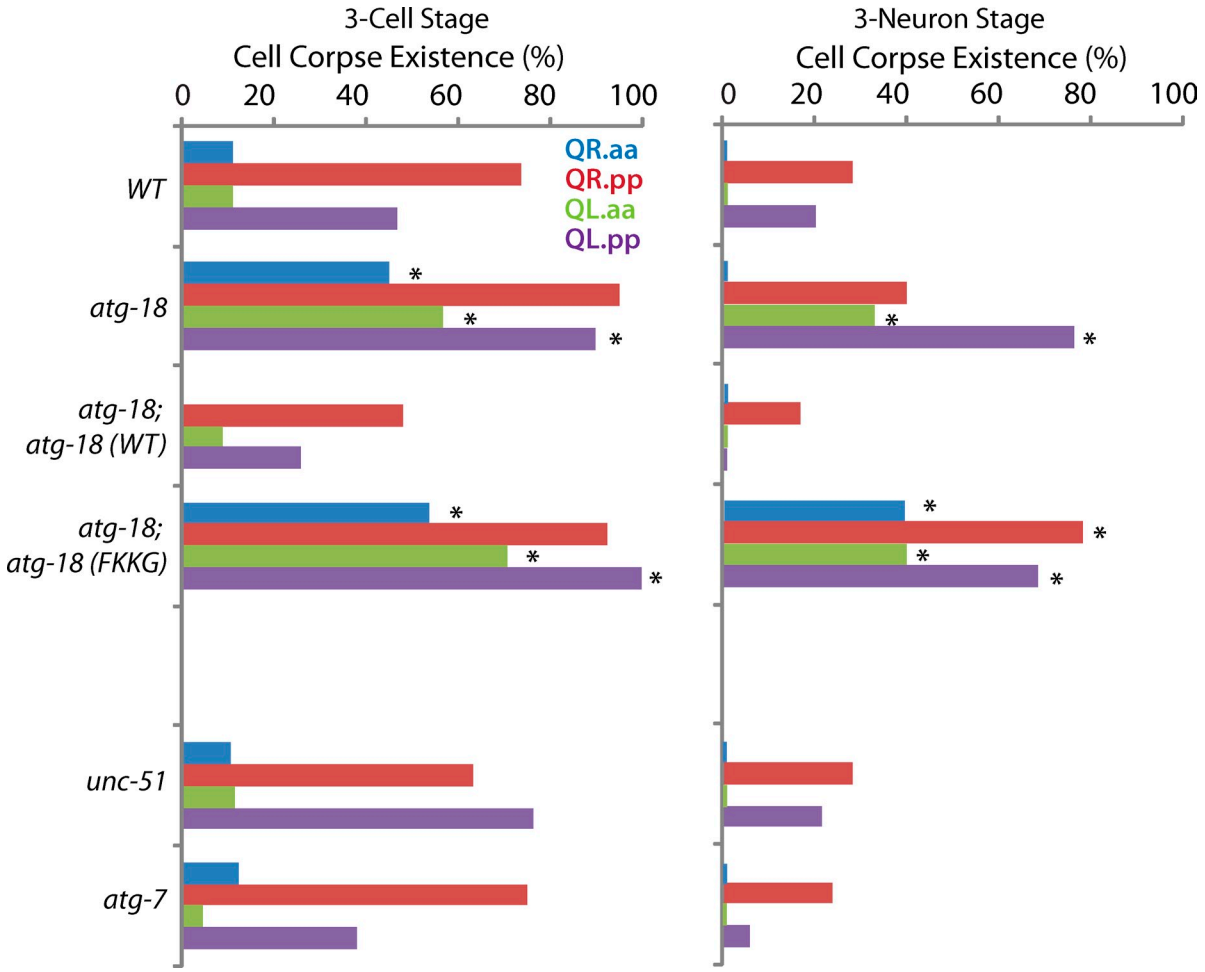
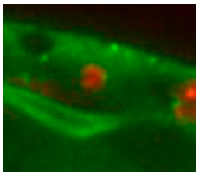
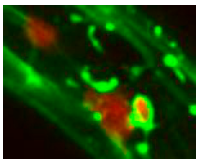


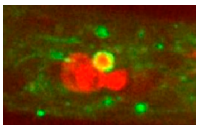
Figure S3. **Quantifications of Q cell corpse degradation in WT and autophagy mutants at three-cell and three-neuron developmental stages.** *, $P < 0.01$, $\chi 2$ test (mutant paired with WT). For each data point, $n = 15–22$ from a single experiment.



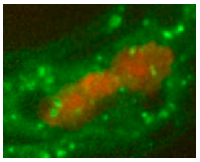
Video 1. **The birth, engulfment, and degradation of Q cell corpse.** Transgenic *C. elegans* strain (XW6462) expressing GFP-tagged actin cytoskeleton (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 124 min. The display rate is 15 frames per second.



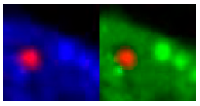
Video 2. **The recruitment of CTNS-1 (lysosome) onto Q cell corpse.** Transgenic *C. elegans* strain (XW7289) expressing GFP-tagged CTNS-1 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 116 min. The display rate is 15 frames per second.



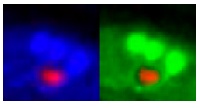
Video 3. **The recruitment of RAB-7 onto Q cell corpse.** Transgenic *C. elegans* strain (XW6195) expressing GFP-tagged RAB-7 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 137 min. The display rate is 15 frames per second.



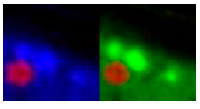
Video 4. **The recruitment of RAB-5 onto Q cell corpse.** Transgenic *C. elegans* strain (XW6193) expressing GFP-tagged RAB-5 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 141 min. The display rate is 15 frames per second.



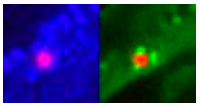
Video 5. **The recruitment of RAB-5 and ATG-18 onto Q cell corpse.** Transgenic *C. elegans* strain (Ex525) expressing BFP-tagged RAB-5 (blue) and GFP-tagged ATG-18 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 16 min. The display rate is seven frames per second.



Video 6. **The recruitment of actin and EPG-5 onto Q cell corpse.** Transgenic *C. elegans* strain (Ex489) expressing BFP-tagged actin (blue) and GFP-tagged EPG-5 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 43 min. The display rate is seven frames per second.



Video 7. **The recruitment of RAB-7 and LGG-1 onto Q cell corpse.** Transgenic *C. elegans* strain (Ex507) expressing BFP-tagged RAB-7 (blue) and GFP-tagged LGG-1 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 28 min. The display rate is seven frames per second.



Video 8. **The recruitment of CTNS-1 and LGG-1 onto Q cell corpse.** Transgenic *C. elegans* strain (Ex522) expressing BFP-tagged CTNS-1 (blue) and GFP-tagged LGG-1 (green) in the hyp7 cell and mCherry in Q cells (red). Images were taken by a time-lapse fluorescence microscope (Axio Observer.Z1) attached to a spinning-disk confocal scan head (CSU-X1 Spinning Disk Unit). Frames were taken every minute for 31 min. The display rate is seven frames per second.

Table S1. ***C. elegans* strains used in this study**

Strain name	Genetic background	Description	Method	Resource
GOU344	*N2*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{lgg-1} *lgg-1::gfp*	Cross with *rdvls1; adls2122*	CGC and this study
XW6462	*N2*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *act-5::gfp*	Cross with *rdvls1; qxls289*	This study
XW7289	*N2*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *ctns-1::gfp*	Cross with *rdvls1; qxls281*	This study
XW6195	*N2*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-7*	Cross with *rdvls1; qxls317*	This study
XW6193	*N2*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-5*	Cross with *rdvls1; qxls318*	This study
Ex473	*unc-76*	P_{egl-17}*myri-mCherry;* P_{egl-17}*mCherry::his-24;* P_{ced-1} *epg-5::gfp*	Microinjection	This study
Ex489	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *epg-5::gfp;* P_{hyp7} *bfp::act-1*	Microinjection	This study
Ex507	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *gfp::lgg-1;* P_{hyp7} *bfp::rab-7*	Microinjection	This study
Ex510	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *epg-5::gfp;* P_{hyp7} *bfp-TEV-S::rab-5*	Microinjection	This study
Ex522	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *gfp::lgg-1;* P_{hyp7} *ctns-1::bfp::unc-54 3′*	Microinjection	This study
Ex525	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::atg-18;* P_{hyp7} *bfp:rab-5*	Microinjection	This study
Ex568	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::atg-5b*	Microinjection	This study
Ex570	*unc-76*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::atg-7*	Microinjection	This study
Ex513	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::atg-18*	Microinjection	CGC and this study
Ex505	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *act-5::gfp*	Microinjection	CGC and this study
XW8030	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24*	Cross with *rdvls1*	CGC and this study
XW8031	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *act-5::gfp*	Cross with *rdvls1; qxls289*	CGC and this study
Ex527	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *ctns-1::gfp*	Microinjection	CGC and this study
Ex538	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *gfp::rab-7*	Microinjection	CGC and this study
Ex504	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-5*	Microinjection	CGC and this study
Ex545	*atg-18 (gk378)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp:: atg-18* (FRRG → FKKG)	Microinjection	CGC and this study
GOU345	*epg-5 (tm3425)*	*Pegl-17::myri-mCherry; Pegl-17::mCherry::his-24*	Cross with *rdvls1*	S. Mitani's[a] laboratory and this study
Ex488	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *epg-5::gfp*	Microinjection	S. Mitani's laboratory and this study
XW7535	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *act-5::gfp*	Cross with *rdvls1; qxls289*	S. Mitani's laboratory and this study
XW8821	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{ced-1} *ctns-1::gfp*	Cross with *rdvls1; qxls281*	S. Mitani's laboratory and this study
XW7534	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-7*	Cross with *rdvls1; qxls317*	S. Mitani's laboratory and this study
XW7538	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-5*	Cross with *rdvls1; qxls318*	S. Mitani's laboratory and this study
XW7538	*epg-5 (tm3425)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24;* P_{hyp7} *gfp::rab-5*	Cross with *rdvls1; qxls318*	S. Mitani's laboratory and this study
GOU104	*unc-51 (e1189)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24*	Cross with *rdvls1*	CGC and this study
XW8524	*atg-7 (bp422)*	P_{egl-17} *myri-mCherry;* P_{egl-17} *mCherry::his-24*	Cross with *rdvls1*	H. Zhang's[b] laboratory and this study

CGC, Caenorhabditis Genetics Center.
[a]Tokyo Women's Medical University School of Medicine, Tokyo, Japan.
[b]National Institute of Biological Sciences, Beijing, China.

Table S2. **PCR products for *C. elegans* transgenesis**

PCR product	Primer 5′	Primer 3′	Template
egl-17 promoter	CAGATGGATGTTTACTGCCAACTGG	AGCTCACATTTCGGGCACCTGAA	N2 genomic DNA
myr-mCherry	CAGGTGCCCGAAATGTGAGCTATGGGTTCC TGTATTGGAAAAGTCTC	CTAAAGGGAACAAAAGCTGGAGC	pJWZ50.3
P_{egl-17} *myr-mCherry*	CTTCCGTTCTATGGAACACTC	GAATCATCGTTCACTTTTCACGG	*egl-17* promoter + Myristoylation-mCherry
P_{egl-17} *mCherry::his-24*	CTTCCGTTCTATGGAACACTC	GAAGACGTTGAACGTCAAATTATC	*egl-17* promoter + mCherry + his-24
hyp7 promoter	GCGGATCCAAACTTTATTAGACGTCGCAATTT	TTTGGTTTTTGGGATTTTTGATC	N2 genomic DNA
gfp::rab-5	GATCAAAAATCCCAAAAACCAAAATGAGTA AAGGAGAAGAAC	GAAACGCGCGAGACGAAAGGGCCCGT	Plasmid pPD49.26-P_{ced-1} *gfp::rab-5*
P_{hyp7} *gfp::rab-5*	TTTATTAGACGTCGCAATTTAAT	AAGGGCCCGTACGGCCGACTAGTAGG	hyp7 promoter + *gfp::rab-5*
gfp::rab-7	GATCAAAAATCCCAAAAACCAAAATGAGTA AAGGAGAAGAAC	GAAACGCGCGAGACGAAAGGGCCCGT	Plasmid pPD49.26-P_{ced-1} *gfp::rab-7*
P_{hyp7} *gfp::rab-7*	TTTATTAGACGTCGCAATTTAAT	AAGGGCCCGTACGGCCGACTAGTAGG	hyp7 promoter + *gfp::rab-7*
bfp-tev-s	AGACCCAAGCTTGGTACCATGATGTCAGAG CTTATTAAGGAG	AGTACCTCCACCTCCGCTGTCCATGT	Plasmid pDONR P4-P1R-*bfp-tev-s*
act-1::unc-54 3′ utr	ACATGGACAGCGGAGGTGGAGGTACTATG TGTGACGACGAGGTTGC	AAGGGCCCGTACGGCCGACTAGTAGG	Plasmid pPD49.26-*act-1::unc-54 3′ utr*
P_{hyp7} *bfp-tev-s:: act-1::unc-54 3′ utr*	GACGTCGCAATTTAATTTATACAATGACAC	GGAAACAGTTATGTTTGGTATATTGGG	P_{hyp7} + *bfp-tev-s+act-1 unc-54 3′ utr*

Table S3. **Plasmids constructed for *C. elegans* transgenesis**

Plasmid name	Primer 5′	Primer 3′	Notes
pCFJ151-MCS-FLAG-6×HIS-UNC-54 3′UTR	GCACTAGTGCTAGCTCGCGA-ACGCGTGACTACAAGGATGA-CGATGACAAGCACCACCACC-ACCACCACTAACGCATCGGC-CGCTGTCATCAGATC	CGACTAGTAGGAAACAGT-TATGTTTGG	MCS (NheI–NruI–MluI)-FLAG-6×HIS-UNC-54 3′UTR was amplified by PCR and then digested by SpeI. It was inserted into pCFJ151 via its SpeI site.
pCFJ151-P_{hyp7}-MCS-FLAG-6×HIS-UNC-54 3′UTR	GCGGATCCAAACTTTATTAGAC-GTCGCAATTT	GCGGATCCTTTGGTTTTTGG-GATTTTTGATC	P_{hyp7} was amplified by PCR. N2 genomic DNA was used as template. Then, it was digested by BamHI and inserted into vector via the BglII site.
pCFJ151-P_{hyp7} *act-5::gfp*-FLAG-6×HIS-UNC-54 3′UTR	CGACGCGTATGGAAGAAGAA-ATCGCCGCCCTC	GCACGCGTCTATTTGTATAGT-TCATCCATGCC	act-5::GFP was amplified by PCR from plasmid *95.77*-P_{ced-1}::*act-5::gfp*. Then, it was digested by MluI and inserted into vector via the MluI site.
pPD49.26-*gfp*	GCGCTAGCATGAGTAAAGG-AGAAGAAC	GCGAGCTCTATTTGTATAGTTC-ATCCATGCC	*gfp* was amplified by PCR and then digested with NheI and SacI. It was inserted into vector via NheI–SacI sites.
pPD49.26-*epg-5::gfp*	GCCCCGGGATGGCGGAATT-GGTTCGTCC	GCGCTAGCTTGCTTACCTAAC-AATTGCAA	*epg-5* was amplified by PCR from N2 genomic DNA and digested by SmaI and NheI. Then, it was inserted into vector via SmaI–NheI sites.
pPD49.26-P_{ced-1}::*epg-5::gfp*	na	na	*ced-1* promoter (5 kb) was digested by BamHI from plasmid and inserted into vector via the BamHI site.
pPD49.26-*gfp*	GCGCTAGCATGAGTAAAGGA-GAAGAAC	GCGGTACCTTTGTATAGTTCAT-CCATGCC	*gfp* was amplified by PCR and then digested with NheI and KpnI. It was inserted into vector via NheI–KpnI sites.
pPD49.26-P_{ced-1} *gfp*	na	na	*ced-1* promoter (5 kb) was digested by BamHI from plasmid and inserted into vector via BamHI site.
pPD49.26-P_{ced-1} *gfp::lgg-1*	GCGGTACCATGAAGTGGGC-TTACAAGGAGGAG	CGGGTACCTTATTCCTTCTTTTC-GACCTC	*lgg-1* was amplified by PCR from N2 genomic DNA and digested by KpnI. Then, it was inserted into vector via KpnI site.
pPD49.26-P_{hyp7}	GCGGATCCAAACTTTATTAGA-CGTCGCAATTT	GCGGATCCTTTGGTTTTTGGGA-TTTTTGATC	P_{hyp7} was amplified by PCR. N2 genomic DNA was used as template. Then, it was digested by BamHI and inserted into vector via the BamHI site.
pPD49.26-P_{hyp7} *bfp*	GCGCTAGCATGTCAGAGCTT-ATTAAGGAG	GCGGTACCATTAAGCTTGTGAC-CCAGTTTG	*bfp* was amplified by PCR and digested by NheI and KpnI. Then it was inserted into vector via NheI–KpnI sites.
pDONRP4-P1 R P_{hyp7}	AGTGACCTGTTCGTTGTTGC-AGAAAAATATTTCACTGTTTCAC	CATTTCGGGCACCTGGGGATTT-TTGATCTGCAAATATTGAC	P_{hyp7} was amplified from N2 genomic DNA and inserted into pDONRP4-P1 R via In-Fusion Advantage PCR Cloning kit (Takara Bio Inc.).
pDONRP4-P1 R P_{hyp7} *bfp-tev-s*	CCCGAAATGTGAGCTATGT-CAGAGCTTATTAAGGAG	AGGTCACTAATACCAAGTACCTC-CACCTCCGCTGTCCATGT	*bfp-tev-s* sequence was amplified from plasmid and inserted into the pDONRP4-P1 R P_{hyp7} plasmid via In-Fusion Advantage PCR Cloning kit.
pDONRP4-P1 R P_{hyp7} *gfp-tev-s*	CCCGAAATGTGAGCTATG-AGTAAAGGAGAAGAACTTTTCAC	AGGTCACTAATACCAAGTACCTC-CACCTCCGCTGTCCATGT	*gfp-tev-s* sequence was amplified from plasmid and inserted into the pDONRP4-P1 R P_{hyp7} plasmid via In-Fusion Advantage PCR Cloning kit.

Table S3. **Plasmids constructed for *C. elegans* transgenesis** *(Continued)*

Plasmid name	Primer 5′	Primer 3′	Notes
pDONRP4-P1 R P_{hyp7} *bfp::rab-5*	GGAGGTGGAGGTACTATG-GCCGCCCGAAACGCAGGA	AGGTCACTAATACCAGAG-TTTCATCTGATGGTATTGC	*rab-5* was amplified from N2 genomic DNA and inserted into pDONRP4-P1 R P_{hyp7}::BFP-TEV-S plasmid via In-Fusion Advantage PCR Cloning kit.
pDONRP4-P1 R P_{hyp7} *gfp::atg-7*	GGAGGTGGAGGTACTATG-GCCACGTTTGTTCCCTTTGTT	AGGTCACTAATACCAGTAC-ATGAATAATTTCTGACATTAAG	*atg-7* was amplified from N2 and inserted into the pDONRP4-P1 R P_{hyp7}::BFP-TEV-S plasmid via In-Fusion Advantage PCR Cloning kit.
pDONRP4-P1 R P_{hyp7} *gfp::atg-18*	GGAGGTGGAGGTACTATG-TCGGCTACAACATCAGAA	AGGTCACTAATACCATTTACT-CGAATGAGAATGCCA	*atg-18* was amplified from N2 genomic DNA and inserted into pDONRP4-P1 R P_{hyp7}::GFP-TEV-S::atg-18 3′UTR plasmid via In-Fusion Advantage PCR Cloning kit.
pDONRP4-P1 R P_{hyp7} *gfp::atg-18* (FRRG → FKKG)	CCAAATGGACATCGGCTC-TTTGAATTCAAAAAGGGCGT-AACTCGCTGTGTCAATATC	GATATTGACACAGCGAGTTA-CGCCCTTTTTGAATTCAAAGA-GCCGATGTCCATTTGG	*atg-18* FKKG was amplified from the pDONRP4-P1 R P_{hyp7} *gfp::atg-18* plasmid and then inserted into the P_{hyp7}::*gfp-tev-s* plasmid via In-Fusion Advantage PCR Cloning kit.
pPD95.77-*bfp*	GTACCGGTAGAAAAAATGT-CAGAGCTTATTAAGGAGAATATG	ATTCTACGAATGCTAATTAAGC-TTGTGACCCAGTTTGCTCGG	The *gfp* sequence of pPD95.77 was replaced with *bfp* by In-Fusion Advantage PCR Cloning kit.
pPD95.77-P_{hyp7} *bfp*	GTACCGGTAGAAAAAGTT-GCAGAAAAATATTTCACTGTTTCAC	ACCAAGCTTGGGTCTGGGATT-TTTGATCTGCAAATATTGAC	P_{hyp7} was amplified from N2 genomic DNA and inserted into pPD95.77-*bfp*::unc-54 *3′utr* plasmid via In-Fusion Advantage PCR Cloning kit.
pPD95.77-P_{hyp7} *ctns-1:: bfp*	CAGATCAAAAATCCCATG-AGTTTCCCGGTGGCATTTTTG	GGTACCAAGCTTGGGTCTGTCA-TGTACAATAATAGGTTCCG	*ctns-1* was amplified from N2 genomic DNA and inserted into the pPD95.77-P_{hyp7}::*bfp* plasmid via In-Fusion Advantage PCR cloning kit.
pPD49.26-P_{hyp7} *bfp:: rab-7*	na	na	rab-7 genomic DNA was digested from pPD49.26-P_{ced-1}::*gfp::rab-7* with KpnI. Then, it was inserted into vector via the KpnI site.

MCS, multiple cloning site; na, not available.