

File S6

Strains, Plasmids, Transgenes, and Supporting References

C. elegans strains (strain names and complete genotypes; alpha-numerically listed by genotype), DNAs (plasmids and PCR products along with basic construction details, listed alpha-numerically by plasmid name), and transgenes.

C. elegans Strains

Strain name	Genotype
KG1640	<i>cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus] (EDWARDS <i>et al.</i> 2009)
KG2664	<i>cels56; ceEx305</i> [unc-129::unc-116 sas RNAi]
KG2475	<i>cels56; ceEx309</i> [unc-129::dnc-2 gene]
KG1643	<i>cels59</i> [unc-129::YFP-rab-5]
KG1834	<i>cels70</i> [unc-129::aman-2-Venus, RFP-syn-13]
KG2052	<i>cels77</i> [unc-129::unc-16, RFP-syn-13]
KG2126	<i>cels79</i> [unc-129::GFP-pisy-1]
KG2178	<i>cels82</i> [unc-129::GFP-tram-1]
KG2443	<i>cels83</i> [unc-17::CTNS-1-GFP, RFP-RAB-3]
KG2882	<i>cels123</i> [unc-129::GFP]
KG2998	<i>cels134</i> [unc-17b::CTNS-1-mCherry, -GFP]
KG4125	<i>cels195</i> [unc-129::AMAN-2-Venus]
EU828	<i>dhc-1(or195)</i> (HAMILL <i>et al.</i> 2002; KOSHIIKA <i>et al.</i> 2004) (Caenorhabditis Genetics Center)
KG1829	<i>dhc-1(or195); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG2463	<i>dhc-1(or195); unc-16(ce421); cels56</i> [unc-129::CTNS-1-RFP, unc-129::NLP-21-Venus]
JT9887	<i>goa-1(sa734)</i> (ROBATZEK and THOMAS 2000)
KG2164	<i>jnk-1(gk7); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG1746	<i>unc-16(ce421)</i>
KG1788	<i>unc-16(ce421); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG1821	<i>unc-16(ce421); cels59</i> [unc-129::YFP-rab-5]
KG1836	<i>unc-16(ce421); cels70</i> [unc-129::aman-2-Venus, RFP-syn-13]
KG2053	<i>unc-16(ce421); cels77</i> [unc-129::unc-16, RFP-syn-13]
KG2127	<i>unc-16(ce421); cels79</i> [unc-129::GFP-pisy-1]
KG2179	<i>unc-16(ce421); cels82</i> [unc-129::GFP-tram-1]
KG2165	<i>unc-16(ce421); jnk-1(gk7); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG1758	<i>unc-16(ce451)</i>
KG1943	<i>unc-16(ce451); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG2338	<i>unc-16(ce483)</i>
KG2692	<i>unc-16(ce483); ceEx346</i> [unc-129::RFP-SYN-13, unc-129::YFP-RAB-5]
KG2101	<i>unc-16(ce483); cels56</i> [unc-129::CTNS-1-RFP, unc-129::NLP-21-Venus]
KG2444	<i>unc-16(ce483); cels83</i> [unc-17::CTNS-1-GFP, RFP-RAB-3]
KG4109	<i>unc-16(ce483); cels123</i> [unc-129::GFP]
KG3035	<i>unc-16(ce483); cels134</i> [unc-17b::CTNS-1-mCherry, -GFP]
KG4179	<i>unc-16(ce483)/+; cels181</i> [unc-129::CTNS-1A-mCherry]/+; <i>cels192</i> [unc-129::LMP-1-GFP]/+
KG4183	<i>unc-16(ce483)/+; cels185</i> [unc-129::PST-2A-CFP]/+; <i>cels195</i> [unc-129::AMAN-2-Venus]/+
KG4152	<i>unc-16(ce483); cels195</i> [unc-129::AMAN-2-Venus]
FF41	<i>unc-116(e2310)</i> (PATEL <i>et al.</i> 1993) (also known as e2281)
KG1942	<i>unc-116(e2310); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG1944	<i>unc-116(e2310) unc-16(ce421); cels56</i> [unc-129::CTNS-1-RFP, unc-129::nlp-21-Venus]
KG2486	<i>unc-116(e2310); cels56; ceEx319</i> [unc-129::unc-116 cDNA]

Plasmids and PCR Products

DNA Name	Common Name	Description or reference
KG#59	rab-3:: expression vector	(SCHADE <i>et al.</i> 2005)
KG#65	unc-17b:: expression vector	(CHARLIE <i>et al.</i> 2006)
KG#67	ttx-3::GFP	Gift of Oliver Hobert, Columbia University
KG#79	unc-17b::SNB-1-GFP	Used Pfu Ultra polymerase and primers engineered with restriction sites to amplify the 2286 bp snb-1-GFP region from pSB120.65 and cloned into Nhe I/ Apa I cut KG#65
KG#94	unc-17:: expression vector	(Edwards <i>et al.</i> , 2008)
KG#100	unc-17b::SNB-1-GFP	Used Age I/ Apa I to cut out the ~1800 bp GFP/ 3' control region from KG#79, leaving the 3600 bp vector fragment containing the unc-17b promoter. To this vector fragment, we ligated the 1800 bp Age I/ Apa I fragment cut from pPD94.81, which contains the S65C GFP variant.
KG#130	unc-17b::SNT-1	Used StrataScript Reverse Transcriptase and a primer engineered with a restriction site to make the full length snt-1 cDNA minus the stop codon (~1.4 Kb) from <i>C. elegans</i> mRNA. Used Accuprime Pfx and primers engineered with restriction sites to amplify and clone into Nhe I/ Kpn I cut KG#65 (4.2 Kb).
KG#146	unc-17b:: -GFP	(EDWARDS <i>et al.</i> 2009)
KG#148	unc-17b::SNT-1-CFP	Used Kpn I/ Spe I to cut out the ~1000 bp 3' control region from KG#130, leaving the 4600 bp vector fragment containing the unc-17beta promoter hooked to the 1.4 kb snt-1 stop codon-less cDNA. To this vector fragment, we will ligate the 1800 bp Kpn I/ Spe I fragment cut from pPD136.61, which contains CFP.
KG#150	unc-25::GFP	(EDWARDS <i>et al.</i> 2009)
KG#230	unc-129:: expression vector	(EDWARDS <i>et al.</i> 2009)
KG#238	unc-17b::____-RFP	(EDWARDS <i>et al.</i> 2009)
KG#240	unc-129::____-RFP expression vector	(EDWARDS <i>et al.</i> 2009)
KG#244	unc-129:: expression vector with Not I site	(EDWARDS <i>et al.</i> 2009)
KG#253	unc-129::____-CFP	Used Age I/ Apa I to cut out the ~1000 bp unc-54 3' control region from KG#230, leaving the 5.4 kb vector fragment containing the unc-129 promoter. To this vector fragment, we ligated the 1800 bp Age I/ Apa I fragment (containing CFP + the unc-54 3' control region) cut from pPD136.61.
KG#255	ttx-3::RFP	(EDWARDS <i>et al.</i> 2009)
KG#344	acr-2:: expression vector	Used Herculase II and primers engineered with restriction sites to amplify the 3.2 Kb acr-2 promoter from KP#704 and cloned it into Pst I/ Bam HI cut pPD96.52 (6.1 Kb).
KG#353	acr-2::____-GFP	Used Age I/ Spe I to cut out the 1000 bp unc-54 3' UTR from KG#344 leaving the 6.1 Kb vector fragment containing the acr-2 promoter. To this fragment, we ligated the like-digested 1800 bp GFP + unc-54 3' UTR cut from pPD94.81.
KG#354	unc-25::CTNS-1A-GFP	Used StrataScript Reverse Transcriptase and a primer engineered with a restriction site to make the full length ctns-1a cDNA (1.2 Kb) lacking a stop codon from <i>C. elegans</i> mRNA for in-frame fusion to GFP. Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify and clone the cDNA into Nhe I/ Age I cut KG#150 (5.8 Kb).
KG#355	acr-2::CTNS-1a-GFP	Used StrataScript Reverse Transcriptase and a primer engineered with a restriction site to make the full length ctns-1a cDNA (1.2 Kb) lacking a stop codon from <i>C. elegans</i> mRNA for in-frame fusion to GFP. We then used Herculase II polymerase and primers engineered with restriction sites to amplify and clone the cDNA into Nhe I/ Age I cut KG#353 (7.9 Kb).
KG#357	unc-129::YFP-RAB-5	Used Herculase II and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1430 bp YFP-rab-5 insert from pCZ677 unc-25::YFP-rab-5 and clone it into Not I cut KG#244 unc-129:: (6.4 Kb). Analyzed 6 clones to identify those with the correct insert size and use PCR with vector and GFP primers to identify clones with correctly oriented insert.
KG#358	unc-129::CFP-RAB-7	Used Herculase II and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1430 bp CFP-rab-7 insert from pCZGY168 unc-25::CFP-rab-7 and cloned it into Not I cut KG#244 unc-129:: (6.4 Kb). Analyzed 8 clones to identify those with the correct insert size and used PCR

		with vector and GFP primers to identify clones with correctly oriented insert.
KG#359	unc-129::CFP-RAB-11	Used Herculase II and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1430 bp CFP-rab-11 insert from pCZGY171 unc-25::CFP-rab-11 and cloned it into Not I cut KG#244 unc-129:: (6.4 Kb). Analyzed 8 clones to identify those with the correct insert size and used PCR with vector and GFP primers to identify clones with correctly oriented insert.
KG#367	unc-129::____-GFP	Used Kpn I / Spe I to cut out the ~1000 bp unc-54 3' control region from KG#230, leaving the 5.3 kb vector fragment containing the unc-129 promoter. To this vector fragment, we ligated the 1800 bp Kpn I / Spe I fragment (containing GFP + unc-54 3' control region) cut from pPD94.81.
KG#369	unc-129::CTNS-1A-GFP	Used Nhe I / Age I to cut out the 1.2 Kb ctns-1a cDNA from KG#354 and cloned it into the like-digested unc-129::____-GFP vector KG#367.
KG#371	unc-129::CTNS-1A-RFP cDNA	(EDWARDS <i>et al.</i> 2009)
KG#374	unc-129::Venus expression vector	(EDWARDS <i>et al.</i> 2009)
KG#411	unc-129::SYN-13-RFP	Used StrataScript Reverse Transcriptase and a primer engineered with a restriction site to make the full length syn-13 cDNA (0.75 Kb) lacking a stop codon from <i>C. elegans</i> mRNA for in-frame fusion to RFP. We used Herculase II polymerase and primers engineered with restriction sites to amplify and clone the cDNA into Nhe I / Age I cut KG#240 (7.2 Kb).
KG#413	unc-129::SYN-13	Used Herculase II polymerase and primers engineered with restriction sites to amplify the 0.75 Kb syn-13 cDNA (adding a stop codon at the end) from KG#411 and cloned it into Nhe I / Age I cut KG#230 (6.4 Kb).
KG#414	unc-129::RFP-SYN-13	Used Herculase II and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 0.8 Kb RFP fragment (minus stop codon) from KG#238 unc-17 ::RFP and cloned it into Nhe I cut KG#413 unc-129::syn-13 cDNA (7.2 Kb). Used PCR and restriction digests to identify clones with the correct RFP orientation.
KG#429	unc-129::AMAN-2-Venus	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the first 82 codons of the aman-2 gene (301 bp = all of first exon, the 55 bp first intron, and some of second exon, with reading frame adjusted for downstream Venus) from N2 genomic DNA and cloned it into Nhe I / Age I cut KG#374 unc-129::____-Venus (7.2 Kb).
KG#432	unc-17::CTNS-1-GFP	Used Nhe I / Apa I to cut out the ~1000 bp unc-54 3' control region from KG#94, leaving the 5.9 kb vector fragment containing the unc-17 promoter. To this fragment, we ligated the 3.05 Kb Nhe I / Apa I fragment (containing CTNS-1-GFP + unc-54 3' control region) cut from KG#355.
KG#445	unc-129::UNC-16 cDNA	Used AffinityScript Reverse Transcriptase and a primer engineered with a restriction site to make the full length unc-16 cDNA (3471 bp) from <i>C. elegans</i> mRNA. We then used Herculase II polymerase and primers engineered with restriction sites to amplify and clone the cDNA into Nhe I / Kpn I cut KG#230 (6.4 Kb).
KG#468	unc-129::UNC-16 cDNA	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify and clone the 3.5 Kb unc-16 cDNA from KG#445 unc-129::unc-16 cDNA into Nhe I / Kpn I cut KG#230 (6.4 Kb).
KG#469	rab-3::UNC-16 cDNA	Use Nhe I / Kpn I to cut out the 3.5 Kb unc-16 cDNA from KG#468 and clone into the like-digested rab-3:: vector KG#59 (4900 bp).
KG#470	rab-3:: expression vector with Sbf I site	Synthesized 2 complementary oligos containing Nhe I and Age I sticky ends and Sbf I and Kpn I sites in between. After hybridization, we cloned it into Nhe I / Age I cut KG#59.
KG#471	mig-13::expression vector	Used BamH I / Apa I to cut out the ~825 bp mult-cloning site + unc-54 3' control region from KS#1 mig-13:: expression vector, leaving the 6.1 kb vector fragment containing the mig-13:: promoter. To this vector fragment, we ligated the 1000 bp BamH I / Apa I fragment from KG#470.
KG#472	rab-3:: expression vector with Fse I site	Synthesized 2 complementary oligos containing Nhe I and Age I sticky ends and Fse I and Kpn I sites in between. After hybridization, we cloned it into Nhe I / Age I cut KG#59 (rab-3::).
KG#476	mig-13::____-RFP expression vector	Used Kpn I / Apa I to cut out the ~1000 bp unc-54 3' control region from KG#483, leaving the 6.0 kb vector fragment containing the mig-13 promoter. To this vector fragment, we ligate the 1800 bp Kpn I / Apa I fragment (containing RFP + unc-54 3' control region) cut from KG#240.
KG#477	mig-13::GFP-____ expression vector	Used Herculase II polymerase and primers engineered with restriction sites to amplify the 800 bp GFP (S65C version with 3 introns; minus stop codon) from pPD94.81 and cloned it into Asc I / Sbf I cut KG#471 mig-13::____ (7.0 Kb).

KG#483	mig-13:: expression vector	Used BamH I/ Apa I to cut out the ~825 bp multi-cloning site + unc-54 3' control region from KS#1 mig-13, leaving the 6.1 kb vector fragment containing the mig-13:: promoter. To this vector fragment, we ligated the 1000 bp BamH I/ Apa I fragment from KG#472.
KG#484	unc-129::PISY-1 gene	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1042 bp pisy-1 gene coding region from N2 genomic DNA and cloned it into Nhe I/ Age I cut KG#230 (6.4 Kb).
KG#485	unc-129::TRAM-1 gene	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1470 bp tram-1 gene coding region from N2 genomic DNA and cloned it into Nhe I/ Age I cut KG#230 (6.4 Kb).
KG#486	unc-129::GFP-PISY-1 gene	Used Herculase II and primers engineered with restriction sites to amplify the 0.8 Kb GFP fragment (minus stop codon) from KG#146 and cloned it into Nhe I cut KG#484 (7.5 Kb).
KG#488	mig-13::CTNS-1A-RFP	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the stop-codon-less 1215 bp ctns-1 cDNA from KG#371 and cloned it into Fse I/ Kpn I cut KG#476 (7.8 Kb).
KG#490	mig-13::GFP-RAB-5 gene	Used Herculase II polymerase and primers engineered with restriction sites to amplify the 1.3 Kb rab-5 gene coding region from N2 genomic DNA and cloned it into Sbf I/ Kpn I cut KG#477 (7.8 Kb).
KG#491	unc-17::RFPA-RAB-3	Used Herculase II and primers engineered with restriction sites to amplify the 1.5 Kb RFP-rab-3 fragment from KS#3 mig-13::RFP-rab-3 and cloned it into Sbf I cut KG#493 unc-17:: expression vector (6.4 Kb). Used PCR (between RFP and vector) to identify a clone with the correct orientation.
KG#493	unc-17:: expression vector	Synthesized 2 complementary oligos such that hybridization produces a double stranded fragment containing Nhe I and Age I sticky ends and Sbf I and Kpn I sites in between. We then cloned this fragment into Nhe I/ Age I cut KG#94 and tested for the Sbf I site by cutting with Sbf I.
KG#494	unc-129::GFP-TRAM-1 gene	Used Herculase II and primers engineered with restriction sites to amplify the 0.8 Kb GFP fragment (minus stop codon) from KG#146 and cloned it into Nhe I cut KG#485 unc-129::tram-1 gene (7.9 Kb).
KG#546	unc-129::DNC-2 gene	Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify the 1204 bp dnc-2 gene from N2 genomic DNA and clone it into Nhe I/ Age I cut KG#230 (6.4 Kb).
KG#551	unc-129::UNC-116 cDNA	Used AffinityScript Multiple Temperature Reverse Transcriptase and a primer engineered with a restriction site to make the full length unc-116 cDNA (2448 bp) from <i>C. elegans</i> mRNA. Used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify and clone the cDNA into Nhe I/ Age I cut KG#230 (6.4 Kb).
KG#571	unc-129::LMP-1-GFP	Used AffinityScript Multiple Temperature Reverse Transcriptase and a primer engineered with a restriction site to make the full length <i>lmp-1</i> cDNA minus its stop codon (714 bp) from <i>C. elegans</i> mRNA. We then used Herculase II polymerase and primers engineered with restriction sites to amplify and clone the cDNA into Nhe I/ Age I cut KG#367 (7.2 Kb).
KG#611	unc-129::PST-2A-CFP	Used AffinityScript Multiple Temperature Reverse Transcriptase and a primer engineered with a restriction site to make the full length <i>pst-1</i> cDNA minus its stop codon (1.1 Kb; 1095 bp). Then used Herculase II polymerase and primers engineered with restriction sites and the ribosome binding site AAAA to amplify and clone the cDNA into Nhe I/ Age I cut KG#253 unc-129::____-CFP.
KG#645	unc-17b::CTNS-1A-RFP	Used Herculase II polymerase and primers engineered with restriction sites to amplify the 0.5 Kb unc-17b promoter from KG#65. We cut this insert with Hind III/ Bam HI and cloned it into like-digested KG#371 unc-129::CTNS-1-RFP (removing the 2.4 Kb unc-129:: promoter in the process).
KG#657	bacterial tac promoter::MBP-UNC-16[1-462] cDNA	Used Herculase II polymerase and primers engineered with restriction sites to amplify the 1.4 Kb unc-16 [AA 1-462] cDNA from KG#469 (rab-3::unc-16 cDNA) and cloned it into Bam HI/ Sal I cut pMALc2H10T (6.7 Kb).
KGsa7	unc-129::unc-116 sas genomic	Used Expand 20 kb+ to fuse the ~2.6 kb unc-129 promoter with a 2 Kb unc-116 genomic – rich region in forward and reverse orientations to allow sense and anti-sense RNAs to be formed in motor neurons for RNAi experiments. Combined the 2 sas products in equimolar concentrations.
KP#704	acr-2::GFP-SNB-1	Gift of Joshua Kaplan, Massachusetts General Hospital/ Harvard University (Jacob and Kaplan, 2003)
KP#1383	unc-129::NLP-21-Venus	Gift of Joshua Kaplan and Derek Sieburth, Massachusetts General Hospital/ Harvard University (SIEBURTH et al. 2007)
KS#1	mig-13:: expression vector	Gift of Kang Shen, Stanford University (KLASSEN and SHEN 2007)
KS#3	mig-13::RFP-RAB-3	Gift of Kang Shen, Stanford University (KLASSEN and SHEN 2007)
pCZ677	unc-25::YFP-RAB-5	Gift of Yish Jin, University of California San Diego (GRILL et al. 2007)

pCZGY168	unc-25::CFP-RAB-7	Gift of Yish Jin, University of California San Diego (<i>GRILL et al.</i> 2007)
pCZGY171	unc-25::CFP-RAB-11	Gift of Yish Jin, University of California San Diego (<i>GRILL et al.</i> 2007)
pPD94.81	unc-54::GFP	Gift of Andrew Fire, Stanford University
pPD96.52	myo-3:: expression vector	Gift of Andrew Fire, Stanford University
pPD136.61	myo-3::CFP	Gift of Andrew Fire, Stanford University
pMalC2H10	Maltose-binding protein	Gift of John Tesmer, University of Michigan
T	bacterial expression vector	
pRK793	His6-TEV [S219V] protease	Addgene
pSB120.65	snb-1::SNB-1 gene-GFP	Gift of Michael Nonet, Washington University
RM#605p	unc-17b::GFP	Gift of Jim Rand, Oklahoma Medical Research Foundation (<i>CHARLIE et al.</i> 2006)

Transgenic Arrays

Array name	Experimental contents	Relevant organelles labeled and references	Co-transformation marker(s)
<i>ceEx305</i>	KGsa7 [unc-129::UNC-116 sas RNAi fusion PCR product] (80 ng/ ml total; 40 ng/ ml each orientation)	N/A	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>ceEx309</i>	KG#546 [unc-129::DNC-2 gene] (10 ng/ ml)	N/A	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>ceEx319</i>	KG#551 [unc-129::UNC-116 cDNA] (25 ng/ ml)	N/A	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>ceEx346</i>	KG#414 [unc-129::RFP-SYN-13] (5 ng/ ml) KG#357 [unc-129::YFP-RAB-5] (5 ng/ ml)	Early/ recycling endosomes (CHUN <i>et al.</i> 2008; PREKERIS <i>et al.</i> 1998) Early endosomes (GRILL <i>et al.</i> 2007; GROSSHANS <i>et al.</i> 2006; MAXFIELD and McGRAW 2004)	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>cel56</i>	KG#371 [unc-129::CTNS-1a-RFP] (5 ng/ ml)	Lysosomes (KALATZIS <i>et al.</i> 2001; MANGAHAS <i>et al.</i> 2008); this study	KG#255 [ttx-3::RFP] (15 ng/ ml) KP#1383 unc-129::NLP-21-Venus (15 ng/ ml)
<i>cel59</i>	KG#357 [unc-129::YFP-RAB-5] (5 ng/ ml)	Early endosomes (GRILL <i>et al.</i> 2007; GROSSHANS <i>et al.</i> 2006; MAXFIELD and McGRAW 2004)	KG#255 [ttx-3::RFP] (15 ng/ ml)
<i>cel70</i>	KG#429 [unc-129::AMAN-2-Venus] (7 ng/ ml) KG#414 [unc-129::RFP-SYN-13] (5 ng/ ml)	Golgi (ORCI <i>et al.</i> 2000; ROLLS <i>et al.</i> 2002; SUMAKOVIC <i>et al.</i> 2009; VELASCO <i>et al.</i> 1993); this study Early/ recycling endosomes (CHUN <i>et al.</i> 2008; PREKERIS <i>et al.</i> 1998)	KG#255 [ttx-3::RFP] (35 ng/ ml)
<i>cel77</i>	KG#468 [unc-129::UNC-16 cDNA] (35 ng/ ml) KG#414 [unc-129::RFP-SYN-13] (5 ng/ ml)	N/A	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>cel79</i>	KG#486 [unc-129::GFP-PISY-1] (4 ng/ ml) KG#240 [unc-129::RFP] (5 ng/ ml)	Early/ recycling endosomes (CHUN <i>et al.</i> 2008; PREKERIS <i>et al.</i> 1998) Regular/ smooth ER (ROLLS <i>et al.</i> 2002) RFP – soluble cytoplasmic protein	KG#255 [ttx-3::RFP] (25 ng/ ml)
<i>cel82</i>	KG#494 [unc-129::GFP-TRAM-1] (5 ng/ ml) KG#240 [unc-129::RFP] (5 ng/ ml)	Rough ER (ROLLS <i>et al.</i> 2002) RFP – soluble cytoplasmic protein	KG#255 [ttx-3::RFP] (25 ng/ ml)
<i>cel83</i>	KG#432 [unc-17::CTNS-1-GFP] (5 ng/ ml) KG#491 [unc-17::RFPA-RAB-3] (5 ng/ ml)	Lysosomes (KALATZIS <i>et al.</i> 2001; MANGAHAS <i>et al.</i> 2008); this study	KG#255 [ttx-3::RFP] (25 ng/ ml)
<i>cel123</i>	KG#367 [unc-129::GFP] (5 ng/ ml)	N/A	KG#255 [ttx-3::RFP] (25 ng/ ml)
<i>cel134</i>	KG#645 [unc-17b::CTNS-1A-RFP] (3.5 ng/ ml)	Lysosomes (KALATZIS <i>et al.</i> 2001; MANGAHAS <i>et al.</i> 2008); this study	RM#605p [unc-17b::GFP] (10 ng/ ml)
<i>cel181</i>	KG#371 [unc-129::CTNS-1a-RFP] (5 ng/ ml)	Lysosomes (KALATZIS <i>et al.</i> 2001; MANGAHAS <i>et al.</i> 2008); this study	KG#255 [ttx-3::RFP] (25 ng/ ml)
<i>cel185</i>	KG#611 [unc-129::PST-2A-CFP] (5 ng/ ml)	Golgi (DEJIMA <i>et al.</i> 2010); this study	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>cel192</i>	KG#571 [unc-129::LMP-1-GFP] (3 ng/ ml)	Lyosomes (Kostich <i>et al.</i> , 2000); this study	KG#67 [ttx-3::GFP] (25 ng/ ml)
<i>cel195</i>	KG#429 [unc-129::AMAN-2-Venus] (1 ng/ ml)	Golgi (ORCI <i>et al.</i> 2000; ROLLS <i>et al.</i> 2002; SUMAKOVIC <i>et al.</i> 2009; VELASCO <i>et al.</i> 1993); this study	KG#255 [ttx-3::RFP] (25 ng/ ml)

Supporting References

- ARIMOTO, M., S. P. KOUSHIKA, B. C. CHOUDHARY, C. LI, K. MATSUMOTO *et al.*, 2011 The *Caenorhabditis elegans* JIP3 protein UNC-16 functions as an adaptor to link kinesin-1 with cytoplasmic dynein. *J Neurosci* **31**: 2216-2224.
- CHARLIE, N. K., M. A. SCHADE, A. M. THOMURE and K. G. MILLER, 2006 Presynaptic UNC-31 (CAPS) is Required to Activate the G_s Pathway of the Synaptic Signaling Network. *Genetics* **172**: 943-961.
- CHEN, C. C., P. J. SCHWEINSBERG, S. VASHIST, D. P. MARENISS, E. J. LAMBIE *et al.*, 2006 RAB-10 is required for endocytic recycling in the *Caenorhabditis elegans* intestine. *Mol Biol Cell* **17**: 1286-1297.
- CHUN, D. K., J. M. McEWEN, M. BURBEA and J. M. KAPLAN, 2008 UNC-108/Rab2 Regulates Post-endocytic Trafficking in *C. elegans*. *Mol Biol Cell* **19**: 2682-2695.
- DEJIMA, K., D. MURATA, S. MIZUGUCHI, K. H. NOMURA, T. IZUMIKAWA *et al.*, 2010 Two Golgi-resident 3'-Phosphoadenosine 5'-phosphosulfate transporters play distinct roles in heparan sulfate modifications and embryonic and larval development in *Caenorhabditis elegans*. *J Biol Chem* **285**: 24717-24728.
- EDWARDS, S. L., N. K. CHARLIE, J. E. RICHMOND, J. HEGERMANN, S. EIMER *et al.*, 2009 Impaired dense core vesicle maturation in *Caenorhabditis elegans* mutants lacking Rab2. *J Cell Biol* **186**: 881-895.
- GRILL, B., W. V. BIENVENUT, H. M. BROWN, B. D. ACKLEY, M. QUADRONI *et al.*, 2007 *C. elegans* RPM-1 regulates axon termination and synaptogenesis through the Rab GEF GLO-4 and the Rab GTPase GLO-1. *Neuron* **55**: 587-601.
- GROSSHANS, B. L., D. ORTIZ and P. NOVICK, 2006 Rabs and their effectors: achieving specificity in membrane traffic. *Proc Natl Acad Sci U S A* **103**: 11821-11827.
- HAMILL, D. R., A. F. SEVERSON, J. C. CARTER and B. BOWERMAN, 2002 Centrosome maturation and mitotic spindle assembly in *C. elegans* require SPD-5, a protein with multiple coiled-coil domains. *Dev Cell* **3**: 673-684.
- HARRIS, T. W., I. ANTOSHECHKIN, T. BIERI, D. BLASIAR, J. CHAN *et al.*, 2009 WormBase: a comprehensive resource for nematode research. *Nucleic Acids Res.*
- KALATZIS, V., S. CHERQUI, C. ANTIGNAC and B. GASNIER, 2001 Cystinosin, the protein defective in cystinosis, is a H⁺-driven lysosomal cystine transporter. *EMBO J* **20**: 5940-5949.
- KELKAR, N., S. GUPTA, M. DICKENS and R. J. DAVIS, 2000 Interaction of a mitogen-activated protein kinase signaling module with the neuronal protein JIP3. *Mol Cell Biol* **20**: 1030-1043.
- KELKAR, N., C. L. STANDEN and R. J. DAVIS, 2005 Role of the JIP4 scaffold protein in the regulation of mitogen-activated protein kinase signaling pathways. *Mol Cell Biol* **25**: 2733-2743.
- KLASSEN, M. P., and K. SHEN, 2007 Wnt signaling positions neuromuscular connectivity by inhibiting synapse formation in *C. elegans*. *Cell* **130**: 704-716.
- KOSTICH, M., A. FIRE and D. M. FAMBROUGH, 2000 Identification and molecular-genetic characterization of a LAMP/CD68-like protein from *Caenorhabditis elegans*. *J Cell Sci* **113** (Pt 14): 2595-2606.
- KOUSHIKA, S. P., A. M. SCHAEFER, R. VINCENT, J. H. WILLIS, B. BOWERMAN *et al.*, 2004 Mutations in *Caenorhabditis elegans* cytoplasmic dynein components reveal specificity of neuronal retrograde cargo. *J Neurosci* **24**: 3907-3916.
- LUPAS, A., M. VAN DYKE and J. STOCK, 1991 Predicting coiled coils from protein sequences. *Science* **252**: 1162-1164.
- MANGAHAS, P. M., X. YU, K. G. MILLER and Z. ZHOU, 2008 The small GTPase Rab2 functions in the removal of apoptotic cells in *Caenorhabditis elegans*. *J Cell Biol* **180**: 357-373.
- MAXFIELD, F. R., and T. E. McGRAW, 2004 Endocytic recycling. *Nat Rev Mol Cell Biol* **5**: 121-132.
- MONTAGNAC, G., J. B. SIBARITA, S. LOUBERY, L. DAVIET, M. ROMAO *et al.*, 2009 ARF6 Interacts with JIP4 to control a motor switch mechanism regulating endosome traffic in cytokinesis. *Curr Biol* **19**: 184-195.
- NGUYEN, Q., C. M. LEE, A. LE and E. P. REDDY, 2005 JLP associates with kinesin light chain 1 through a novel leucine zipper-like domain. *J Biol Chem* **280**: 30185-30191.
- ORCI, L., M. AMHERDT, M. RAVAZZOLA, A. PERRELET and J. E. ROTHMAN, 2000 Exclusion of golgi residents from transport vesicles budding from Golgi cisternae in intact cells. *J Cell Biol* **150**: 1263-1270.
- PATEL, N., D. THIERRY-MIEG and J. R. MANCILLAS, 1993 Cloning by insertion mutagenesis of a cDNA encoding *Caenorhabditis elegans* kinesin heavy chain. *Proc Natl Acad Sci U S A* **90**: 9181-9185.
- POON, V. Y., M. P. KLASSEN and K. SHEN, 2008 UNC-6/netrin and its receptor UNC-5 locally exclude presynaptic components from dendrites. *Nature* **455**: 669-673.
- PREKERIS, R., J. KLUMPERMAN, Y. A. CHEN and R. H. SCHELLER, 1998 Syntaxin 13 mediates cycling of plasma membrane proteins via tubulovesicular recycling endosomes. *J Cell Biol* **143**: 957-971.
- ROBATZEK, M., and J. H. THOMAS, 2000 Calcium/calmodulin-dependent protein kinase II regulates *Caenorhabditis elegans* locomotion in concert with a G_o/G_q signaling network. *Genetics* **156**: 1069-1082.
- ROLLS, M. M., D. H. HALL, M. VICTOR, E. H. STELZER and T. A. RAPOPORT, 2002 Targeting of rough endoplasmic reticulum membrane proteins and ribosomes in invertebrate neurons. *Mol Biol Cell* **13**: 1778-1791.
- SAKAMOTO, R., D. T. BYRD, H. M. BROWN, N. HISAMOTO, K. MATSUMOTO *et al.*, 2005 The *Caenorhabditis elegans* UNC-14 RUN domain protein binds to the kinesin-1 and UNC-16 complex and regulates synaptic vesicle localization. *Mol Biol Cell* **16**: 483-496.
- SCHADE, M. A., N. K. REYNOLDS, C. M. DOLLINS and K. G. MILLER, 2005 Mutations that Rescue the Paralysis of *C. elegans* ric-8 (Synembryon) Mutants Activate the G_s Pathway and Define a Third Major Branch of the Synaptic Signaling Network. *Genetics* **169**: 631-649.
- SIEBURTH, D., J. M. MADISON and J. M. KAPLAN, 2007 PKC-1 regulates secretion of neuropeptides. *Nat Neurosci* **10**: 49-57.