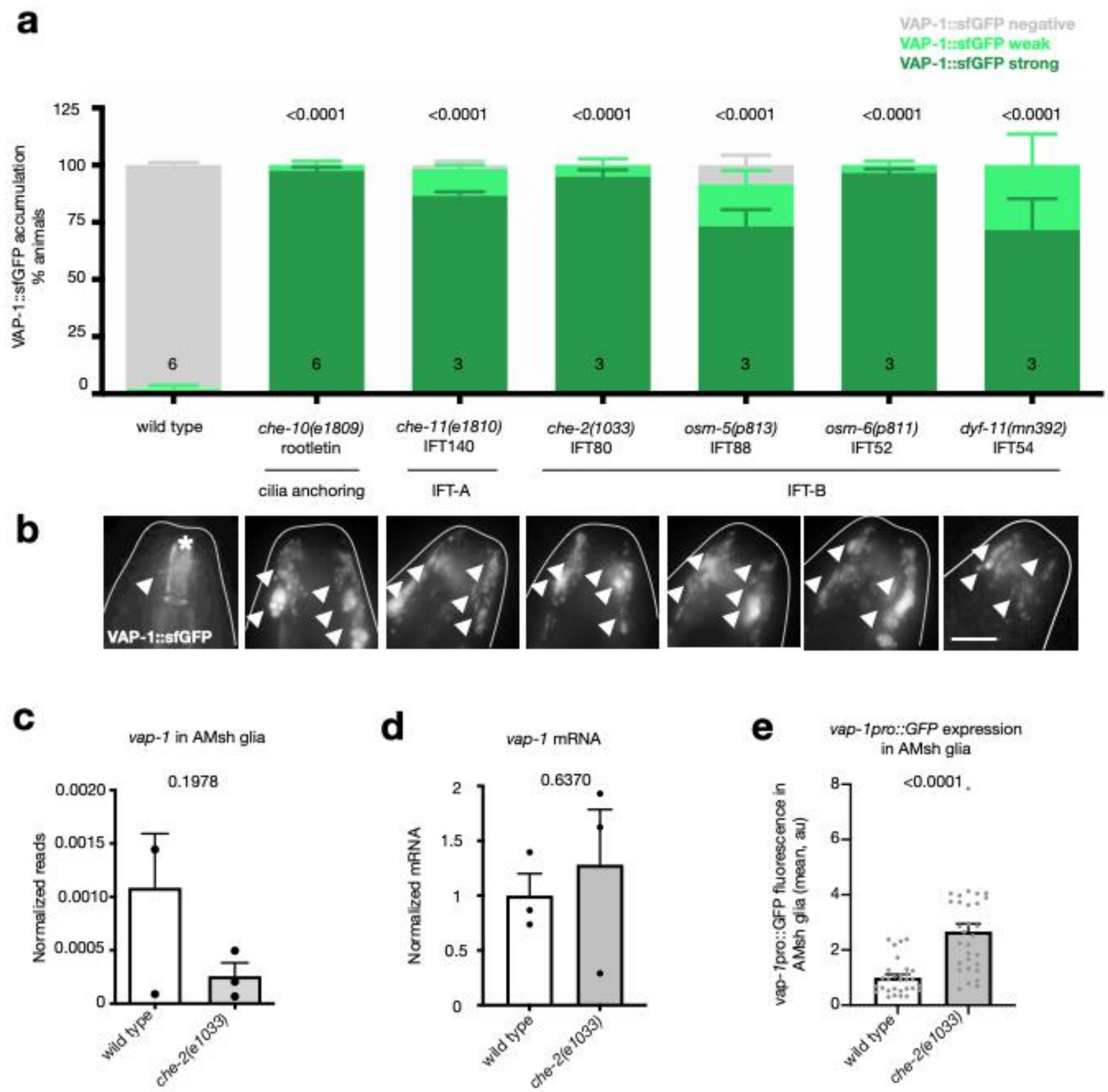
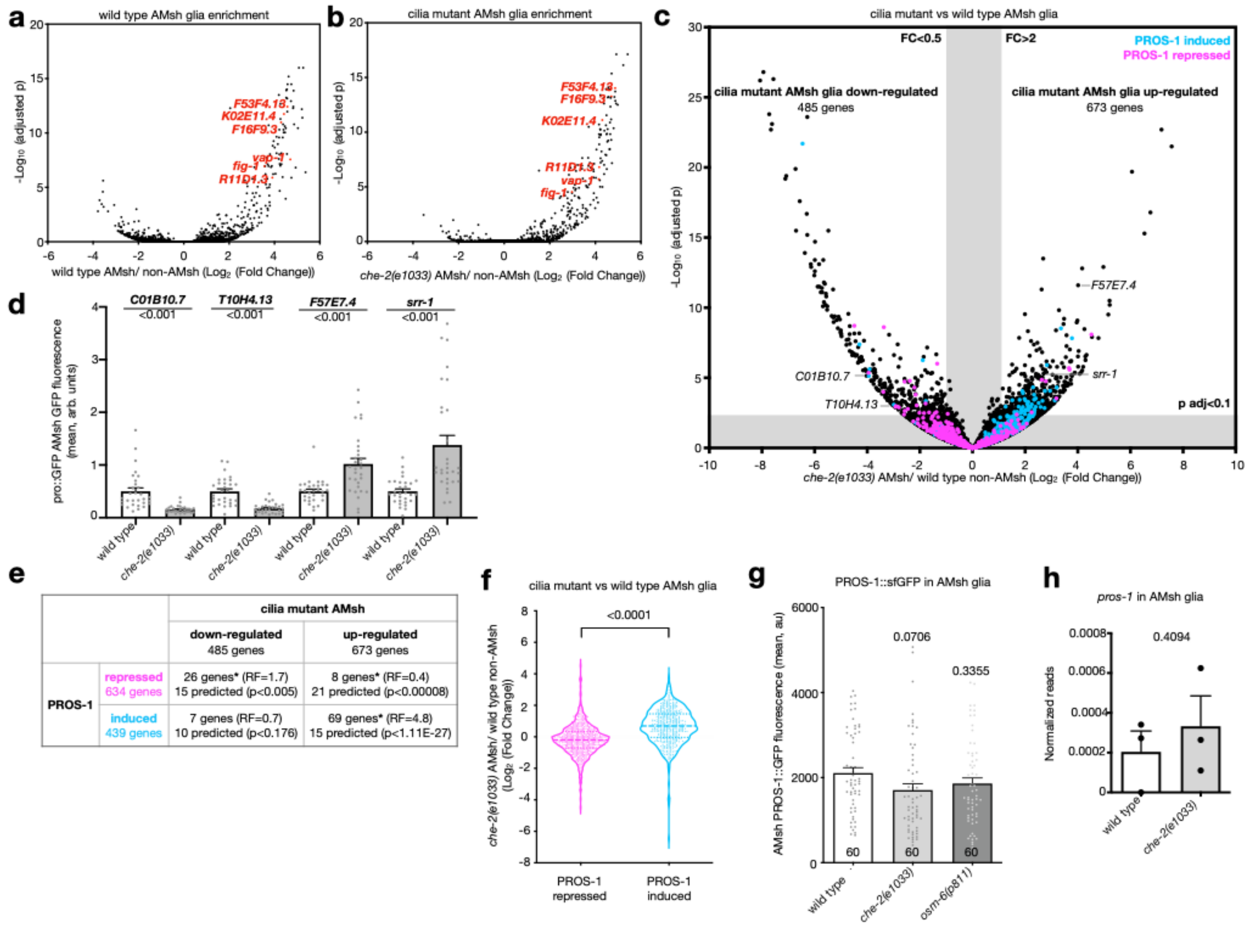


Supplementary Figure 1. AMsh glia ensheathing dendrites with mutations in cilia genes respond by accumulating secreted matrix and increasing expression of *vap-1*.

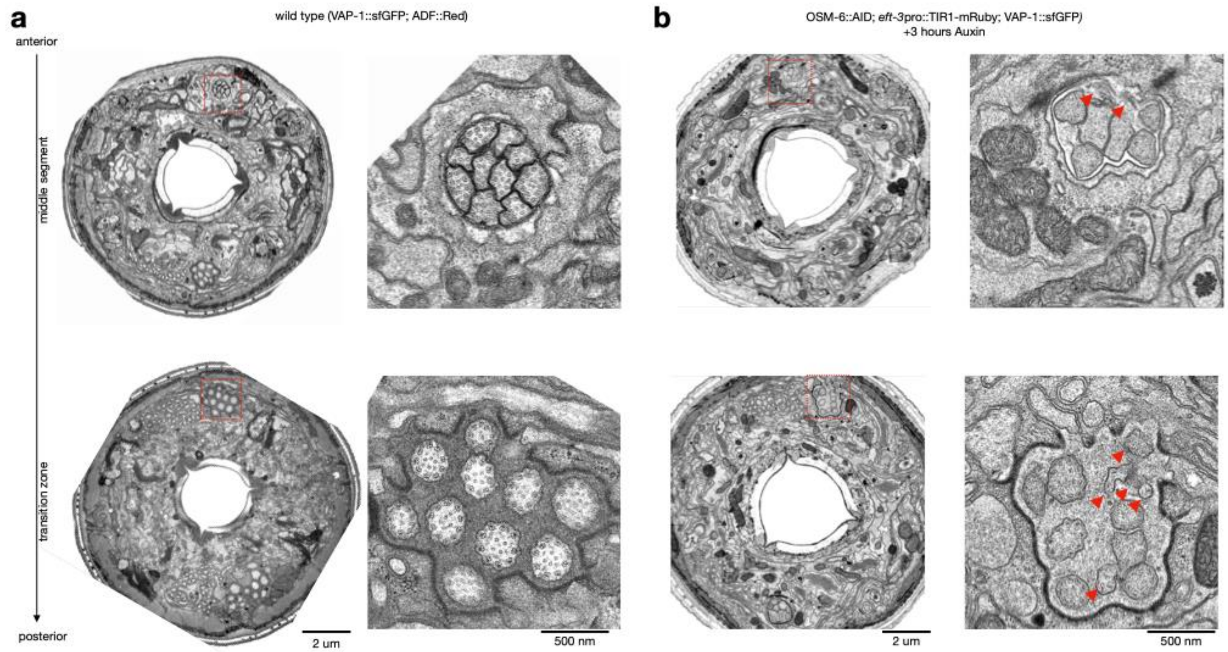
VAP-1::sfGFP accumulation scoring (**a**) and representative brightness and contrast matched images of VAP-1::sfGFP (**b**) of wild type and indicated cilia mutant animals. Data are represented as mean+SEM. *n* (number of trials, 20 animals scored / trial) indicated inside bars. *P* values calculated using calculated using Dunnett's multiple comparison test following one-way ANOVA. Arrowheads, VAP-1::sfGFP puncta. Asterisk, anterior buccal cavity auto-fluorescence. Images are maximum intensity projections of widefield z-stacks. Scalebar= 10μm. (**c**) *vap-1* expression (normalized RNA-seq reads) in AMsh glia FACS-purified from wild type and cilia mutant [*che-2(e1033)*] animals. *n*=3 independent experiments. (**d**) Normalized *vap-1* mRNA levels determined by qPCR of whole worm lysates from wild type and *che-2(e1033)* animals. *n*=3 independent experiments. (**e**) Quantification of *vap-1*pro::GFP fluorescence in AMsh glia in wild type and *che-2(e1033)* mutants. *n*=30 AMsh glia from 3 independent experiments. *P*-values calculated using unpaired, two-tailed t test assuming Gaussian distribution in panels c, d, and e. Data represented as mean+SEM. Source data are provided as a Source Data file.



		cilia mutant AMsh	
		down-regulated 485 genes	**up-regulated** 673 genes
PROS-1	repressed 634 genes	26 genes* (RF=1.7) 15 predicted (p<0.005)	8 genes* (RF=0.4) 21 predicted (p<0.00008)
	induced 439 genes	7 genes (RF=0.7) 10 predicted (p<0.176)	69 genes* (RF=4.8) 15 predicted (p<1.11E-27)

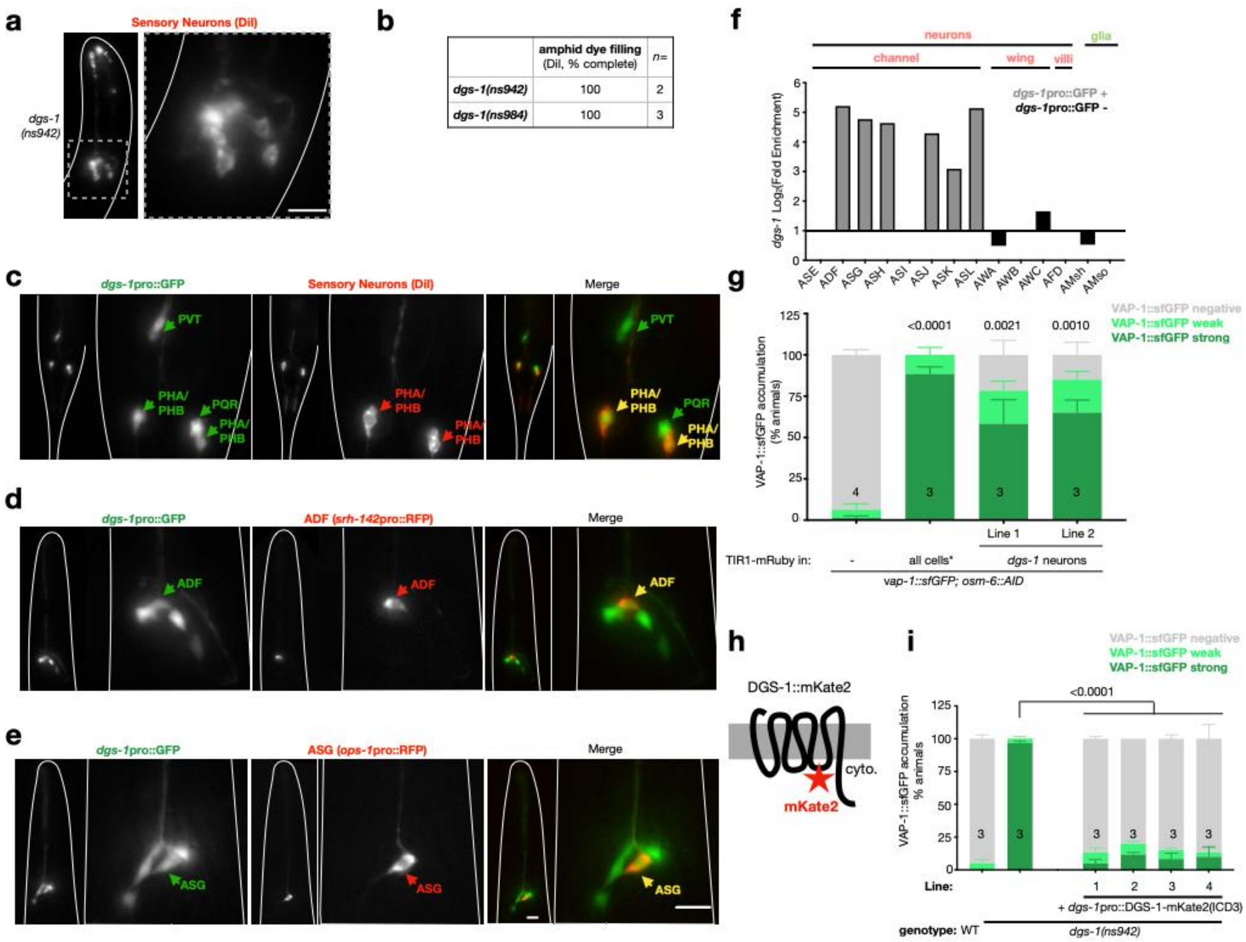
Supplementary Figure 2. AMsh glia transcriptionally respond to defects in ensheathed dendrite cilia, correlating with increased PROS-1 activity.

Volcano plots of AMsh enriched genes (reads in purified dsRed + AMsh glia compared to dsRed- cells) from wild type (**a**) and cilia mutant [*che-2(e1033)*] (**b**) animals expressing AMsh glia-specific reporter *F16F9.3*pro::dsRed. Known AMsh glia-enriched genes are highlighted in red. (**c**) Volcano plot of gene expression changes between purified wild type and cilia mutant [*che-2(e1033)*] AMsh glia. PROS-1 induced and repressed genes are labeled in cyan and magenta, respectively.[23] Genes used for transcriptional reporter validation in panel d are labeled. Values calculated from 3 independent experiments. (**d**) Quantification of *C01B10.7, T10H4.13, F57E7.4,* and *srr-1* transcriptional reporter expression in wild type versus *che-2(e1033)* mutant AMsh glia. n=30 AMsh glia quantified from 3 independent experiments. P values calculated using unpaired, two-tailed t tests. (**e**) Table comparing overlap of genes down- and up-regulated in cilia mutant versus wild type AMsh glia and genes regulated by PROS-1. Total number of genes and % of total protein-coding genes in the *C. elegans* genome is listed for each group. RF= representation factor (# overlapping genes/ expected # of overlapping genes). *P*-values calculated using normal approximation to the hypergeometric probability. *, $P \leq 0.005$. (**f**) Violin plot of PROS-1 induced and repressed gene enrichment in cilia mutant [*che-2(e1033)*] versus wild type AMsh glia. n= 634 PROS-1 repressed genes, 439 PROS-1 induced genes. *P*-value calculated using unpaired, two-tailed t test assuming Gaussian distribution. (**g**) Quantification of PROS-1::GFP fluorescence in AMsh glia of wild type and two cilia mutants: *che-2(e1033)* and *osm-6(p811)*. n=60 AMsh glia quantified from 3 independent experiments. *P*-values calculated using Dunnett's multiple comparison test following ordinary one-way ANOVA and shown in relation to wild type. (**h**) *pros-1* expression (normalized RNA-seq reads) in AMsh glia FACS-purified from wild type and cilia mutant [*che-2(e1033)*] animals. n=3. *P*-values calculated using paired, two-tailed t test assuming Gaussian distribution. Data in bar graphs are represented as mean+SEM. Source data are provided as a Source Data file.



Supplementary Figure 3. Inducible OSM-6 degradation causes acute cilia disruption.

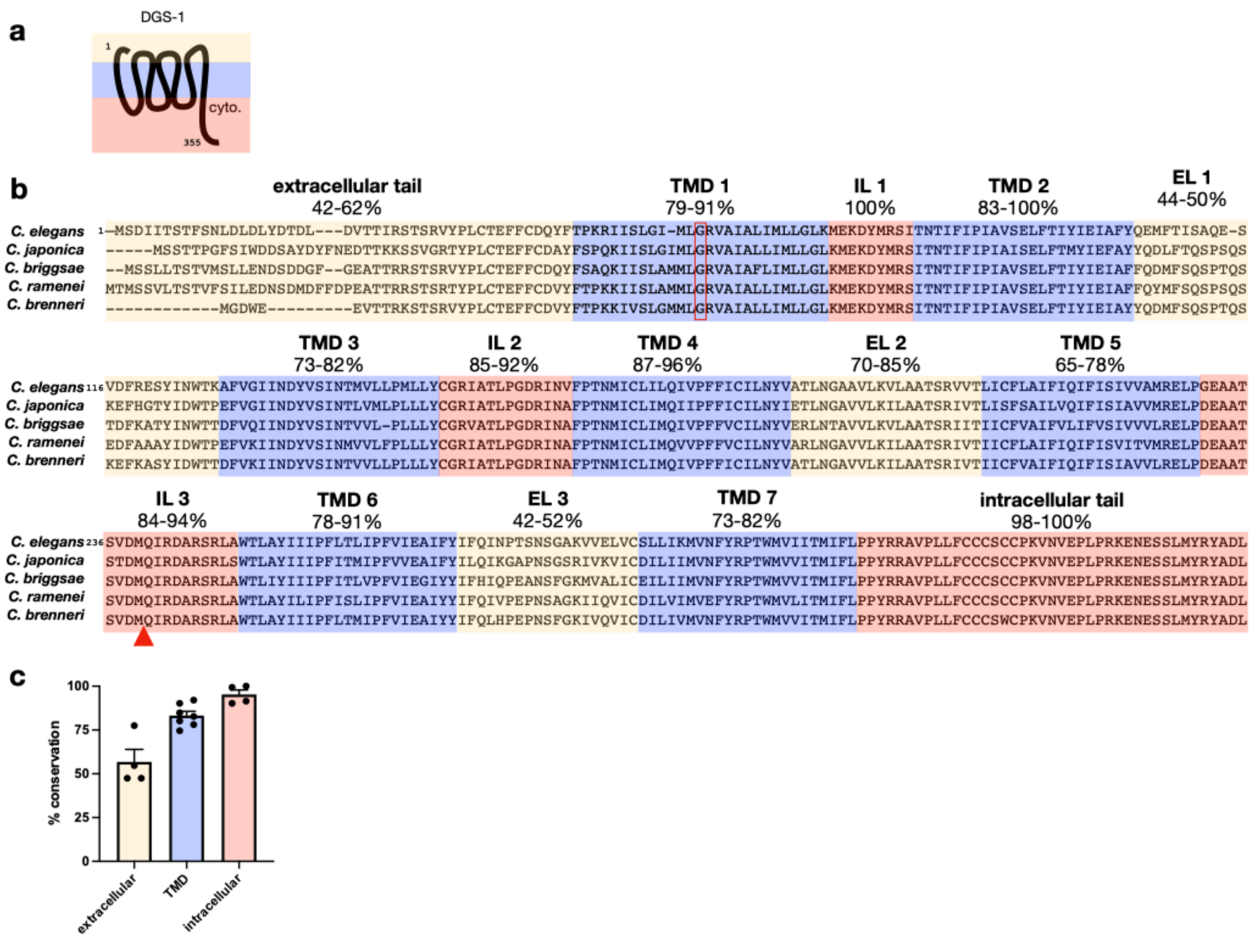
TEM cross sections of through (**a**) an untreated wild-type animal (VAP-1::sfGFP; (*srh-142pro::dsRed*)) and, (**b**) a dye filling defective OSM-6::AID; *eft-3*pro::TIR1-mRuby; *vap-1(ns831)* animal following 3 hours of auxin treatment. Whole animal cross sections are shown on the left with enlarged insets of channel cilia at higher resolution shown on the right. Both the middle segment (anterior) and transition zone (posterior) regions of the cilia are shown for each animal. Arrows, abnormal cilia morphology.



Supplementary Figure 4. Additional data related to *dgs-1*.

(**a**) Image and inset of a *dgs-1(ns942)*; *vap-1(ns831)* animal labeled with DiI. Scale bar=10µm. (**b**) Neuronal DiI uptake in *dgs-1* mutants. *n* indicated in table, 20 animals/trial. Representative images of animals expressing *dgs-1*pro::GFP co-labeled (**c**) with DiI (subset of sensory neurons), (**d**) an ADF neuron reporter (*srh-142*pro::RFP), or (**e**) an ASG neuron reporter (*ops-1*pro::RFP). Scale bars=10µm. (**f**) RNA-seq expression of *dgs-1* in the amphid. Cells with no RNA-seq reads have no bars. All data is from the CeNGEN project[33] except AMsh glia, which is from our RNA-seq experiments (Supplementary Fig. 2). (**g**) VAP-1::sfGFP scoring in *osm-6::AID* animals expressing TIR1-mRuby in all cells or only from the *dgs-1* promoter. *n* (number of trials scored, 20 animals/

trial) indicated inside bars. *P*-values calculated using Tukey's multiple comparison test following ordinary one-way ANOVA from % animals with strong VAP-1::sfGFP accumulation.*,TIR1-mRuby expressed in all cells (*eft-3pro::TIR1-mRuby*) is expressed from a genetically integrated transgene, while *dgs-1* neuron-specific TIR1-mRuby is expressed from an extrachromosomal array. (**h**) DGS-1::mKate2 predicted topology. (**i**) VAP-1::sfGFP accumulation in wild type, *dgs-1(ns942)* mutant, and *dgs-1(ns942)* mutants expressing DGS-1 fused to mKate2 in the 3rd intracellular domain. *n*=3 trials/ condition, 20 animals scored/trial. *P*-values calculated using Dunnett's multiple comparison test following ordinary one-way ANOVA from % animals with strong VAP-1::sfGFP accumulation. Data represented as mean+SEM. Source data are provided as a Source Data file.



Supplementary Figure 5. Comparison of DGS-1 sequences in *Caenorhabditis* species.

(**a**) DGS-1 topology with highlighting corresponding to panel b. (**b**) Amino acid sequence alignment of DGS-1 and its homologs in *Caenorhabditis* species. Extracellular, transmembrane, and intracellular regions are highlighted in yellow, blue, and pink, respectively, as in schematic. Range of percent conservation between *C. elegans* and other species listed below each domain. Red box indicates conserved Glycine (G58) in *C. elegans*, which is mutated in *ns942*. Red arrowhead indicates insertion site for Linker-mKate2-Linker sequence in DGS-1::mKate2. (**c**)

Comparison of percent conservation between topological domains of DGS-1. Source data are provided as a Source Data file.

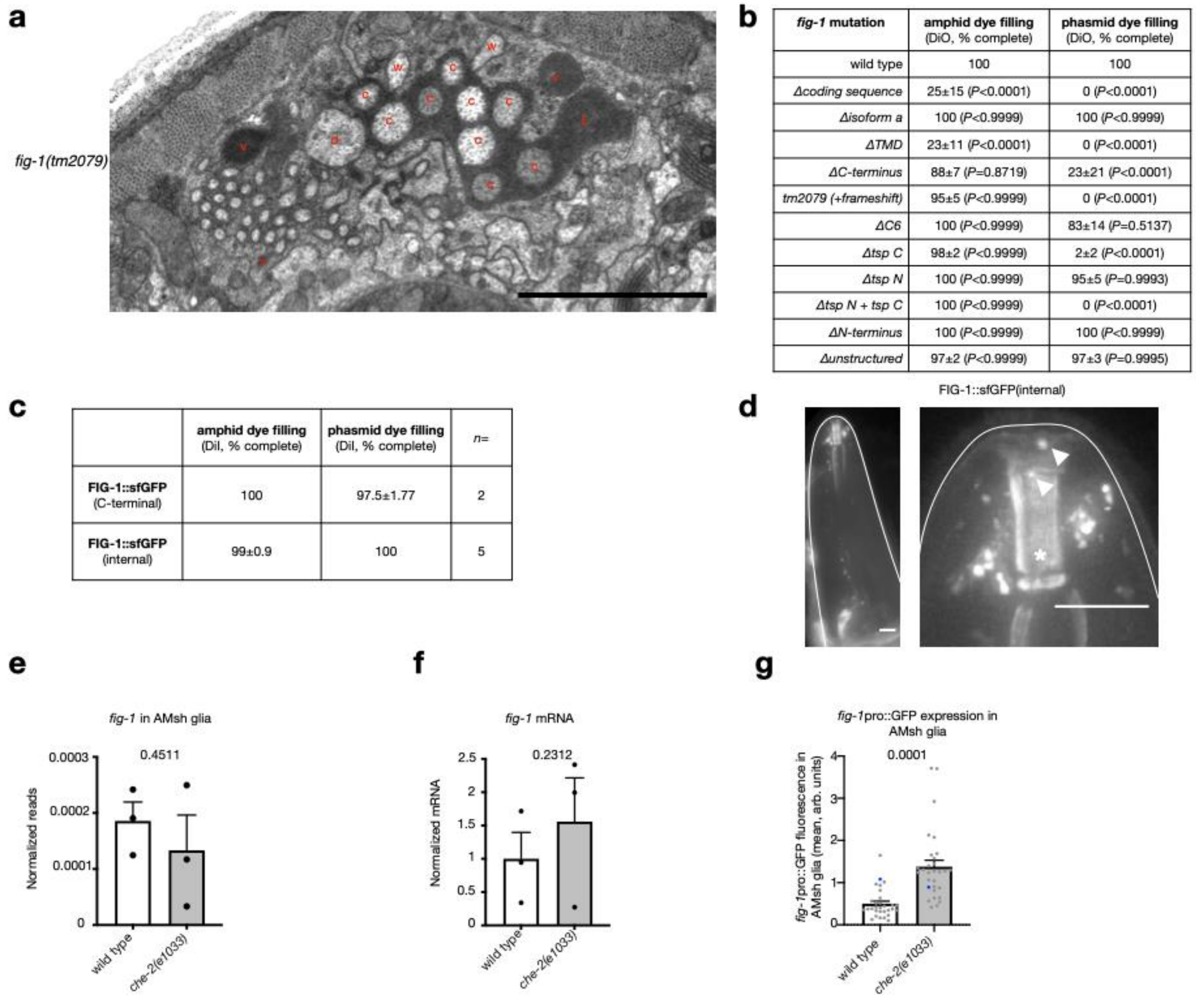
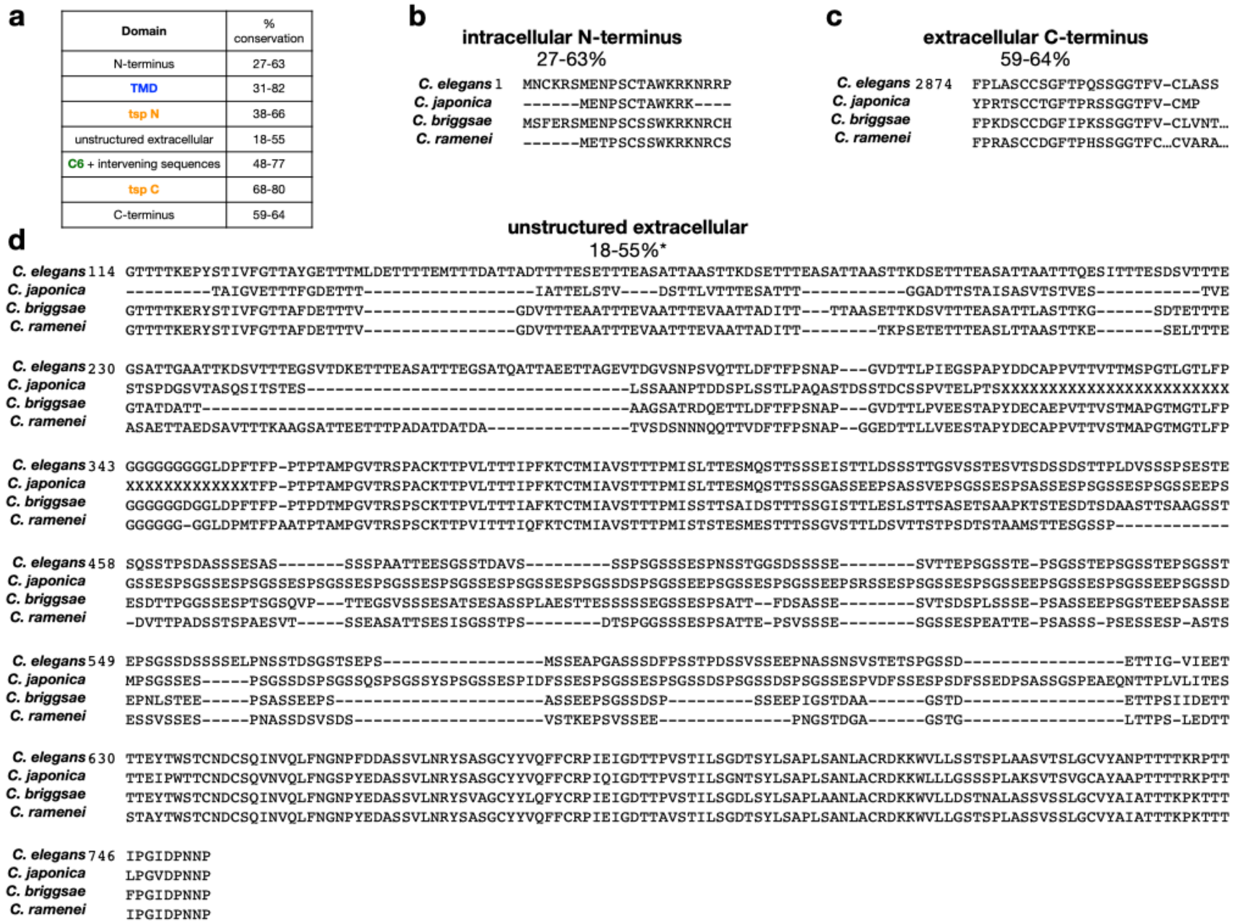


fig-1 mutation	amphid dye filling (DiO, % complete)	phasmid dye filling (DiO, % complete)
wild type	100	100
Δcoding sequence	25±15 (*P*<0.0001)	0 (*P*<0.0001)
Δisoform a	100 (*P*<0.9999)	100 (*P*<0.9999)
ΔTMD	23±11 (*P*<0.0001)	0 (*P*<0.0001)
ΔC-terminus	88±7 (*P*=0.8719)	23±21 (*P*<0.0001)
tm2079 (+frameshift)	95±5 (*P*<0.9999)	0 (*P*<0.0001)
ΔC6	100 (*P*<0.9999)	83±14 (*P*=0.5137)
Δtsp C	98±2 (*P*<0.9999)	2±2 (*P*<0.0001)
Δtsp N	100 (*P*<0.9999)	95±5 (*P*=0.9993)
Δtsp N + tsp C	100 (*P*<0.9999)	0 (*P*<0.0001)
ΔN-terminus	100 (*P*<0.9999)	100 (*P*<0.9999)
Δunstructured	97±2 (*P*<0.9999)	97±3 (*P*=0.9995)

	amphid dye filling (DiI, % complete)	phasmid dye filling (DiI, % complete)	*n*=
FIG-1::sfGFP (C-terminal)	100	97.5±1.77	2
FIG-1::sfGFP (internal)	99±0.9	100	5

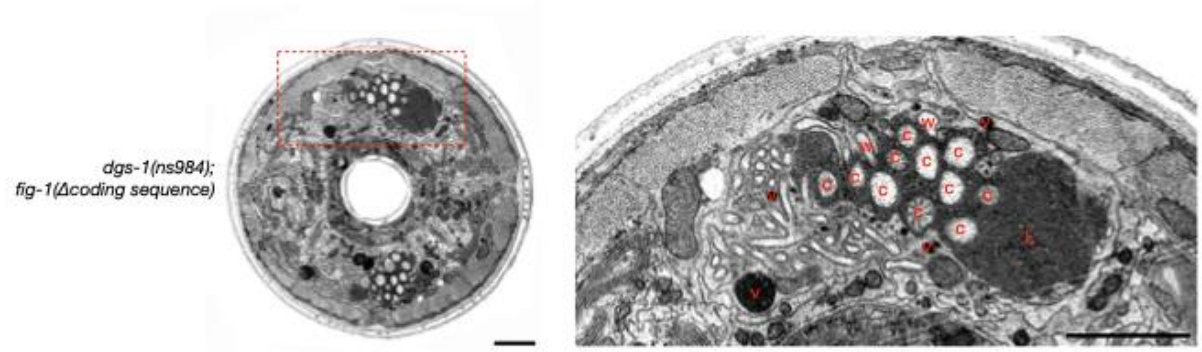
Supplementary Figure 6. Additional data related to *fig-1*.

(**a**) Electron micrograph cross sections through *fig-1(tm2079)* mutant animals, with enlarged inset of the right amphid shown in red dotted lines. L, amphid channel lobes. v, matrix filled AMsh vesicle. c, channel cilia. d, dendrite. w, wing cilia. Scale bars=2μm. Samples originally prepared for previous publication.[29] (**b**) Neuronal DiO dye uptake in indicated *fig-1* mutants. *n*=3 trials, 20 animals per trial. Adjusted *P* values, in relation wild type, calculated using Dunnett's multiple comparison following one-way ANOVA. (**c**) Dye filling of FIG-1::sfGFP strains. *n*= numbers of trials20 animals per trial. (**d**) Widefield z-stack of FIG-1::sfGFP(internal) animal. Arrowheads, anterior FIG-1::sfGFP puncta, Asterisk, posterior buccal cavity auto-fluorescence. Scale bars=10μm. (**e**) *fig-1* expression (normalized RNA-seq reads) in AMsh glia FACS-purified from wild type and cilia mutant [*che-2(e1033)*] animals. *n*=3 independent experiments. (**f**) Normalized *fig-1* mRNA levels determined by qPCR of whole worm lysates from wild type and *che-2(e1033)* animals. *n*=3 independent experiments. (**g**) Quantification of *fig-1*pro::GFP fluorescence in AMsh glia in wild type and *che-2(e1033)* mutants. *n*=30 AMsh glia from 3 independent experiments. *P*-values calculated using unpaired, two-tailed t test assuming Gaussian distribution in panels e, f, and g. Data represented as mean+SEM. Source data are provided as a Source Data file.



Supplementary Figure 7. Comparison of FIG-1 sequences in *Caenorhabditis* species.

(**a**) Percent amino acid conservation of FIG-1 domain sequences, listed as a range comparing *C. elegans* to *C. japonica*, *C. briggsae*, and *C. ramenei.* Note that no FIG-1b homolog is annotated in *C. brenneri*. Amino Acid sequence alignment of FIG-1 domains required for matrix secretion: the intracellular N-terminus (**b**), extracellular C-terminus (**c**), and unstructured extracellular domain (**d**). Range of percent conservation between *C. elegans* and indicated *Caenorhabditis* species listed below each domain.



Supplementary Figure 8. ***dgs-1; fig-1*** **double mutants do not accumulate additional glia-secreted matrix.**

Full TEM cross section through *dgs-1(ns984)*; *fig-1(Δcoding sequence)* mutant (left), and inset (right). c, channel cilia. w, wing cilia. L, amphid channel lobes. v, matrix-filled AMsh vesicle. Scale bars=2μm.

Additional information on FACS gating strategy:

Cells were sorted by size (FSC-A>~30,000) and granularity (<~200,000) to exclude cell debris and aggregates. Cells with DAPI staining (>~10^2 measured with 405nm laser) were excluded as dead cells, as were cells without nuclear Draq5 staining (<~10^3 -10^3 measured with 635nm laser) to obtain a population of live, nucleated cells. Finally, populations of AMsh glia labeled with *F16F9.3*pro::dsRed (>~10^2 measured with 561 laser) were sorted from all other cells (<~10^2 measured with 561 laser) for subsequent RNA-seq.

Table S1. Strains and recombinant DNA used in this study.

Experimental models: Organisms/strains		
REAGENT or RESOURCE	**SOURCE**	**IDENTIFIER**
C. elegans wild type strain N2	*Caenorhabditis* Genetics Center (CGC)	N2 (RRID:WB-STRAIN:WBStrain00000001)
Hawaiian	*Caenorhabditis* Genetics Center (CGC)	CB4856
unc-119(ed3)III	*Caenorhabditis* Genetics Center (CGC)	DP38
*vap-1(ns831 [vap-1::sfGFP])*X; nsEx6547 [*F16F9.3*pro::myr-mKate2 + *unc-122*pro::RFP]	This paper	OS13459
oyIs51 [*srh-142*pro::RFP] V; *vap-1(ns831 [vap-1::*sfGFP*])* X;	This paper	OS12214
*vap-1(ns831 [vap-1::sfGFP])*X	This paper	OS11927
*osm-6(p811)*V; *vap-1(ns831 [vap-1::sfGFP])*X	This paper	OS11948
*vap-1(ns831[vap-1::sfGFP]) che-2(e1033)*X	This paper	OS12195
*che-10(e1809)*II; *vap-1(ns831 [vap-1::sfGFP])*X	This paper	OS12462
*che-11(e1810)*V; *vap-1(ns831 [vap-1::sfGFP])*X	This paper	OS12463
*nsIs143[F16F9.3*pro::dsRed*]*X	Procko et al.[61]	OS4079
*nsIs143[F16F9.3*pro::dsRed*] che-2(e1033)* X	This paper	OS11549
nsIs698 [mir-228pro::nslRFP + coel::mCherry]	Wallace et al.[23]	OS11313
nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6094 [PC01B10.7::GFP + Punc-122::GFP]	This paper	OS12053
che-2(e1033); nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6094 [C01B10.7pro::GFP + Punc-122::GFP]	This paper	OS12116
nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6116 [T10H4.13pro::GFP + Punc-122::GFP]	This paper	OS12114
che-2(e1033)X; nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6116 [T10H4.13pro::GFP + Punc-122::GFP]	This paper	OS12124
nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6095 [srr-1pro::GFP + Punc-122::GFP]	This paper	OS12054
che-2(e1033)X; nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6095 [srr-1pro::GFP + Punc-122::GFP]	This paper	OS12115
nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6113 [F57E7.4pro::GFP + Punc-122::GFP]	This paper	OS12111

che-2(e1033)X; nsIs698 [mir-228pro::nslRFP + coel::mCherry]; nsEx6113 [F57E7.4pro::GFP + Punc-122::GFP]	This paper	OS12127
wgIs500 [PROS-1/ CEH-26::TY1::EGFP::3xFLAG]	Nui et al.[62]	OP500
che-2(e1033)X; wgIs500 [PROS-1/ CEH-26::TY1::EGFP::3xFLAG]	This paper	OS12932
osm-6(p811)V; wgIs500[PROS-1/ CEH-26::TY1::EGFP::3xFLAG]	This paper	OS12933
ieSi57 [eft-3p::TIR1::mRuby::unc-54 3'UTR + Cbr-unc-119(+)] II; osm-6(syb2906 [osm-6::linker-GFP-AID]) V	This paper	OS13059
ieSi57[eft-3p::TIR1::mRuby::unc-54 3'UTR + Cbr-unc-119(+)]II, osm-6(syb2906 syb4401 [osm-6::linker-AID])V, vap-1(ns831[vap-1::sfGFP])X	This paper	PHX4401
osm-6(syb2906 syb4401 [osm-6::linker-AID])V, vap-1(ns831[vap-1::sfGFP])X	This paper	OS13359
ieSi57[eft-3p::TIR1::mRuby::unc-54 3'UTR + Cbr-unc-119(+)]II; vap-1(ns831[vap-1::sfGFP])X	This paper	OS13360
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6577 [dyf-11pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13494
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6579 [dyf-11pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13496
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6580 [dyf-11pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13497
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6581 [F16F9.3pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13498
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6622 [F16F9.3pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13566
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6623 [F16F9.3pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13567
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6625 [srh-142pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13569
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6626 [srh-142pr::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13570
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6629 [srh-142pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13573

osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6627 [srg-47pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13571
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6628 [srg-47pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13572
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6631 [srg-47pro::TIR1-mRuby + unc-122pro::GFP]	This paper	OS13575
oyIs51[srh-142pro::RFP]V; vap-1(ns831 [vap-1::sfGFP])X	This paper	OS12214
oyIs51[srh-142pro::RFP]V; che-2(e1033) vap-1(ns831 [vap-1::sfGFP])X	This paper	OS12215
oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; dgs-1(ns942)IV	This paper	OS12566
dgs-1(ns984)IV; oyIs51[srh-142pro::RFP]; vap-1(ns831[vap-1::sfGFP])X	This paper	OS12876
dgs-1(ns942 syb3113[e58>G])IV; oyIs51[srh-142pro::RFP]; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX3113
oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; ns942; nsEx6306 [dgs-1pro::dgs-1 cDNA + mir-228pro::nlsRFP]	This paper	OS12903
oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; ns942; nsEx6312 [dgs-1pro::dgs-1 cDNA + mir-228pro::nlsRFP]	This paper	OS12915
oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; ns942; nsEx6307 [dgs-1pro::dgs-1 cDNA + mir-228pro::nlsRFP]	This paper	OS12904
nsIs972[dgs-1pro::GFP + unc-122pro::GFP]	This paper	OS14591
dgs-1(ns942)IV; oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; nsEx6324 [dyf-11pro::dgs-1 cDNA + unc-122pro::GFP]	This paper	OS12935
dgs-1(ns942)IV; oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; nsEx6326 [dyf-11pro::dgs-1 cDNA + unc-122pro::GFP]	This paper	OS12937
dgs-1(ns942)IV; oyIs51[srh-142pro::RFP]V; vap-1(ns831)X; nsEx6325 [dyf-11pro::dgs-1 cDNA + unc-122pro::GFP]	This paper	OS12936
oyIs51[srh-142pro::RFP]V; nsEx6300 [dgs-1pro::GFP + unc-122pro::GFP]	This paper	OS13236
oyIs47[ops-1pro::RFP] II; nsEx6300 [dgs-1pro::GFP + unc-122pro::GFP]	This paper	OS13254
nsIs971[DGS-1::mKate2 + coel::GFP] / oyIs26 [ops-1:: GFP + lin-15(+)] lin-15(n765ts) X	This paper	OS14036

osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6632 [dgs-1pro::TIR1-mRuby + coel::GFP]	This paper	OS13576
osm-6(syb2906 syb4401 [osm-6::linker-AID])V; vap-1(ns831[vap-1::sfGFP])X; nsEx6633 [dgs-1pro::TIR1-mRuby + coel::GFP]	This paper	OS13577
dgs-1(ns942) IV; vap-1(ns831[vap-1::sfGFP]) X; nsEx6556[dgs-1pro::DGS-1-mKate2(between TMD5 and TMD6)]	This paper	OS13470
dgs-1(ns942) IV; vap-1(ns831[vap-1::sfGFP]) X; nsEx6557[dgs-1pro::DGS-1-mKate2(between TMD5 and TMD6)]	This paper	OS13471
dgs-1(ns942) IV; vap-1(ns831[vap-1::sfGFP]) X; nsEx6558[dgs-1pro::DGS-1-mKate2(between TMD5 and TMD6)]	This paper	OS13472
dgs-1(ns942) IV; vap-1(ns831[vap-1::sfGFP]) X; nsEx6559[dgs-1pro::DGS-1-mKate2(between TMD5 and TMD6)]	This paper	OS13473
oyIs51 fig-1(syb6983[Δcoding sequence])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX6983
oyIs51 fig-1(syb5898[ΔSS/ TMD])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX5898
fig-1(tm2079) V; vap-1(ns831[vap-1::sfGFP])X	This paper	OS13602
oyIs51 fig-1(syb7619[ΔC-terminus])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX7619
oyIs51 fig-1(syb7600[ΔN-terminus])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX7600
oyIs51 fig-1(syb7606[Δunstructured])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX7606
oyIs51 fig-1(syb6326[ΔtspC])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX6326
oyIs51 fig-1(syb6326 syb6968[ΔtspN + tspC])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX6968
oyIs51 fig-1(syb7051[Δisoform a])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX7051
oyIs51 fig-1(syb5954[ΔtspN])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX5954
oyIs51 fig-1(syb6039[ΔC6])V; vap-1(ns831[vap-1::sfGFP])X	This paper	PHX6039
fig-1(syb6983) oyIs51 V; vap-1(ns831) X; nsEx7266[fig-1pro::FIG-1ΔC6cDNA + coel::GFP]	This paper	OS14709
fig-1(syb6983) oyIs51 V; vap-1(ns831) X; nsEx7214 [F16F9.3pro::FIG-1ΔC6cDNA + coel::GFP]	This paper	OS14590

fig-1(syb6983) oyIs51 V; vap-1(ns831) X; nsEx7161 [F16F9.3pro::FIG-1ΔC6cDNA + coel::GFP]	This paper	OS14508
fig-1(syb6983) oyIs51 V; vap-1(ns831) X; nsEx7173 [dyf-11pro::FIG-1ΔC6cDNA + coel::GFP]	This paper	OS14548
fig-1(syb6983) oyIs51 V; vap-1(ns831) X; nsEx7163 [dyf-11pro::FIG-1ΔC6cDNA + coel::GFP]	This paper	OS14510
fig-1(syb7231 [FIG-1::sfGFP (Cterm)] V; nsEx6547 [F16F9.3pro::myr-mKate2 + coel::RFP]	This paper	OS14481
fig-1(tm2079)	Gift from S. Mitani lab	FX2079
dgs-1(ns984) IV; oyIs51 V; vap-1(ns831) che-2(e1033)X	This paper	OS13361
fig-1(tm2079) oyIs51 V; vap-1(ns831) che-2(e1033) X	This paper	OS13941
dgs-1(ns984) IV; fig-1(tm2079) V; vap-1(ns831)X	This paper	OS13603
dgs-1(ns984)IV; oyIs51 fig-1(syb6983)V; vap-1(ns831)X	This paper	OS14619
che-2(e1033)X; nsIs971	This paper	OS13703
fig-1(tm2079) V; nsIs971	This paper	OS13705
fig-1(syb7231[fig-1::sfGFP(C-terminal)]) V	This paper	PHX7231
fig-1(syb7231 [FIG-1::sfGFP Cterm] V; che-2(e1033)X; nsIs971	This paper	OS14469
dgs-1(ns984)IV; fig-1(syb7231)V	This paper	OS14470
fig-1(syb7231) V; nsIs971	This paper	OS14468
dgs-1(ns984) IV; vap-1(ns831) X	This paper	OS14306
ieSi57 II; C09B9.1(ns984) IV; osm-6(syb2906 syb4401) V; vap-1(ns831)X	This paper	OS14498
ieSi57 II; *dgs-1(ns942)* IV; *osm-6(syb2906 syb4401)* V; *vap-1(ns831)* X	This paper	OS14477
ieSi57 II; *dgs-1(ns942)* IV; *osm-6(syb2906 syb4401)* V; *vap-1(ns831)* X; nsEx6306 *[dgs-1pro::dgs-1 cDNA + mir-228pro::nlsRFP]*	This paper	OS14778
ieSi57 II*; osm-6(syb2906 syb4401) fig-1(syb8275)*V*; vap-1(ns831)*X	This paper	PHX8275
Schneider's *Drosophila* Line 2 (S2 cells)	ATCC	CRL-1963
Recombinant DNA		
P*eft-3*::Cas9 + Empty sgRNA	AddGene	pDD162
P*eft-3*::Cre	AddGene	pDD104
P*eft-3*::TIR1-linker-mRuby	AddGene	pLZ31
P*actin*(*Drosophila melanogaster*)::sax-7s::gfp	Dong et al[63]	pXD49
vap-1 sgRNA guide + Cas9 plasmid	This paper	pKV2

vap-1::sfGFP homologous recombination template	This paper	pKV3
*dgs-1*pro::DGS-1 mKate2(TMD5/6)	This paper	pKV12
*dyf-11*pro::TIR1-mRuby	This paper	pKV13
*F16F9.3*pro::TIR1-mRuby	This paper	pKV14
*dgs-1*pro::TIR1-mRuby	This paper	pKV15
srh-142 pro(ADF)::TIR1-mRuby	This paper	pKV16
*srg-47*pro(ASI)::TIR1-mRuby	This paper	pKV17
*fig-1*pro::FIG-1ΔC6repeats cDNA	This paper	pKV34
*F16F9.3*pro::FIG-1ΔC6 cDNA	This paper	pKV35
*dyf-11*pro::FIG-1ΔC6 cDNA	This paper	pKV36
*actin(Drosophila melanogaster)*pro::sfGFP	This paper	pKV40
*actin(Drosophila melanogaster)*pro::mKate2	This paper	pKV41
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP	This paper	pKV42
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2	This paper	pKV42

Table S2. Oligonucleotides used in this study.

Oligonucleotides		
REAGENT or RESOURCE	**SOURCE**	**IDENTIFIER**
vap-1 sgRNA FWD primer gttgcatagaaaatttaCTAGTTTTAGAGCTAGAAATAGCAAGT	This paper	oKV25
DD universal REV primer Caagacatctcgcaatagg	Dickinson et al.[51]	DD universal REV
*F16F9.3*pro::myr-mKate2 primer A ACAGCTATGACCATGATTACGC	This paper	oKV327
*F16F9.3*pro::myr-mKate2 primer B agcttttccaatacatgatccCATTTTTGTTTCTTACTGTCTTGGG	This paper	oKV416
*F16F9.3*pro::myr-mKate2 primer C ATGggatcatgtattggaaaag	This paper	oKV417
Primer D AAGGGCCCGTACGGCCGACTA	Hobert et al.[53]	oKV113/ primer D
*F16F9.3*pro::myr-mKate2 primer A* GCTTATGAAATGCGGAAC	This paper	oKV430
Primer D* GGAAACAGTTATGTTTGGTATA	Hobert et al.[53]	oKV114/ primer D*
*C01B10.7*pro::GFP primer A ttcgtatggttgattattaattgc	This paper	oKV122
*C01B10.7*pro::GFP primer A* attgcacaatttcgtttgttatct	This paper	oKV123
*C01B10.7*pro::GFP primer B AGTCGACCTGCAGGCATGCAAGCTgatggccgcacaagtgaa	This paper	oKV124
*T10H4.13*pro::GFP primer A ggagaattgcacgtaattattg	This paper	oKV134
*T10H4.13*pro::GFP primer A* agtgtcaaacattgaaacaaataag	This paper	oKV135
*T10H4.13*pro::GFP primer B AGTCGACCTGCAGGCATGCAAGCTgaagtcctgcctctgaag	This paper	oKV136
F57E7.4::GFP primer A gacgagagcgcaaaaggc	This paper	oKV124
F57E7.4::GFP primer A* tttgagggttggaagtatatggaatt	This paper	oKV125
F57E7.4::GFP primer B AGTCGACCTGCAGGCATGCAAGCTgatgagggacaattttctgtcttc	This paper	oKV126
*srr-1*pro::GFP primer A gcctaattatgtgaaaggtttaca	This paper	oKV119
*srr-1*pro::GFP primer A*	This paper	oKV120

catatgtactctaggtgttttctc		
*srr-1*pro::GFP primer B AGTCGACCTGCAGGCATGCAAGCTttcaacgttgac agcaattg	This paper	oKV121
*dgs-1*pro primer A gggcattgacctgaaaaccag	This paper	oKV301
*dgs-1*pro::GFP primer B AGTCGACCTGCAGGCATGCAAGCTatacctgaaact aaaaataataaaagtggc	This paper	oKV303
Primer C AAGGGCCCGTACGGCCGACTA	Hobert et al.[53]	oKV112/ Primer C
*dgs-1*pro primer A* ctaattgtcagtttcaaccag	This paper	oKV302
dgs-1pro::dgs-1 cDNA primer B ttagttatatacctgaaactaaaaataataaaagtggctgaaatagtc	This paper	oKV312
dgs-1 cDNA primer C ttttagtttcaggtatataactaaatATG	This paper	oKV313
dgs-1 cDNA primer D/ D* attttttcaaccaatttattcaattc	This paper	oKV314
*dyf-11*pro::*dgs-1* cDNA primer A ACAGCTATGACCATGATTACGC	This paper	oKV327
*dyf-11*pro::*dgs-1* cDNA primer B CATatttagttatatacctgaaactaaaaGCACTTTTACTGCA AAAATCTC	This paper	oKV329
*dyf-11*pro::*dgs-1* cDNA primer A* *GTGGCTGCTCATTAGGAAAC*	This paper	oKV328
*dyf-11*pro::TIR1-mRuby Frag FWD GATTTTTGCAGTAAAAGTGCATGCAAAAGAG AATCGCCTTGT	This paper	oKV450
TIR1-mRuby Frag REV CCGTACGGCCGACTAGTAGGAAACAGTTATG TTTGGTATATTGGGAATGTATTCTGTCATTTA AGGC	This paper	oKV451
TIR1-mRuby Vect FWD TATACCAAACATAACTGTTTCCTACTAGTCGG C	This paper	oKV448
*dyf-11*pro::TIR1-mRuby Vect REV AAGGCGATTCTCTTTTGCATGCACTTTTACTG CAAAAATCTCATTACAAGTTGC	This paper	oKV449
*F16F9.3*pro::TIR-1mRuby Frag FWD CAAGACAGTAAGAAACAAAAATGCAAAAGA GAATCGCCTTGT	This paper	oKV453
*F16F9.3*pro::TIR-1mRuby vect REV AAGGCGATTCTCTTTTGCATTTTTGTTTCTTAC TGTCTTGGGTATTTTTAGAGGAAT	This paper	oKV452
*srh-142*pro::TIR-1mRuby Frag FWD	This paper	oKV459

tcttttttctttttgccaatATGCAAAAGAGAATCGCCTTG TCG		
*srh-142*pro::TIR-1mRuby Vect REV AAGGCGATTCTCTTTTGCATattggcaaaaagaaaaaag aggtgca	This paper	oKV458
*srg-47*pro::TIR-1mRuby Frag FWD atcttttcttcgaattaaaaATGCAAAAGAGAATCGCCTT GT	This paper	oKV461
*srg-47*pro::TIR-1mRuby Vect REV AAGGCGATTCTCTTTTGCATttttaattcgaagaaaagatt atcaaaaaatcaaatgtgttgaatg	This paper	oKV460
*C09B9.1*pro::TIR1mRuby Frag FWD atttcagccacttttattatATGCAAAAGAGAATCGCCTT GTCG	This paper	oKV456
*C09B9.1*pro::TIR1mRuby Frag REV AAAGGGCCCGTACGGCCGGGAAACAGTTATG TTTGGTATATTGGGAATGTATTCTG	This paper	oKV457
*C09B9.1*pro::TIR1mRuby Vect FWD TATACCAAACATAACTGTTTCCCGGCCGTACG	This paper	oKV454
*C09B9.1*pro::TIR1mRuby Vect REV AAGGCGATTCTCTTTTGCATataataaaagtggctgaaat agtctgaaaaacaaatgg	This paper	oKV455
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Vect FWD ATGAGCTCTACAAAGGATCCTGACGTAAGCT AGCAGGATC	This paper	oKV680
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Vect REV ATtgatCTTTTACAATTCATatcaacTTTTTTGTACA AACTTGT	This paper	oKV681
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Frag 1 FWD GTTTGTACAAAAAAgttgatATGAATTGTAAAAG atcaATGGAAAATCCT	This paper	oKV682
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Frag 1 REV GGCCAATCCCGGggatctGGAGAAGACGCGAGA CAAAC	This paper	oKV683
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Frag 2 FWD TTGTTTGTCTCGCGTCTTCTCCagatccCCGG	This paper	oKV684
*actin(Drosophila melanogaster)*pro::*fig-1(ΔC6)*::sfGFP Frag 2 REV GATCCTGCTAGCTTACGTCAGGATCCTTTGTA GAGCTCA	This paper	oKV685
mKate2 exon 2+3 PCR Fusion A*	This paper	oKV686

GTACGGATCCAAGgtaag		
mKate2 exon 2+3 PCR Fusion B GGATGGGAAGTTGACTCCACGGATCTTGACG TTGTAGATGAGGCATCC	This paper	oKV687
mKate2 exon 2+3 PCR Fusion C ATCCGTGGAGTCAACTTCCC	This paper	oKV688
mKate2 exon 2+3 PCR Fusion D* CTTGATACGCTCAAGACG	This paper	oKV689
mKate2 exon 2+3 PCR Fusion A AGGGAACCGTCAACAACCAC	This paper	oKV690
mKate2 exon 2+3 PCR Fusion D AGCTTGGATGGGAGGTCG	This paper	oKV691
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Vect FWD TGTATCGATATGCCGATTTGTGACGTAAGCTA GCAGGA	This paper	oKV692
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Vect REV GATGTGATTATATCCGACATATCTGGATCCGG GGT	This paper	oKV693
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 1 FWD CAGAGACCCCGGATCCAGATATGTCGGATAT AATCACATCGAC	This paper	oKV694
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 1 REV TGGGTGTGGTTGATGAAGGTCTTGGATCCGT ACATGAAGG	This paper	oKV695
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 2 FWD CCTTCATGTACGGATCCAAGACCTTCATCAAC CACAC	This paper	oKV696
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 2 REV TTCTTGGAACGGTAGGTGGTCTTGAGGTTGCA GATGAGGT	This paper	oKV697
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 3 FWD ACCTCATCTGCAACCTCAAGACCACCTACCGT TCC	This paper	oKV698
*actin(Drosophila melanogaster)*pro::*dgs-1*::mKate2 Frag 3 REV GATCCTGCTAGCTTACGTCACAAATCGGCAT ATCGATACATAAG	This paper	oKV699
*actin(Drosophila melanogaster)*pro::sfGFP Vect REV	This paper	oKV700

AGTTCTTCTCCTTTGCTCATATCTGGATCCGG GGT		
*actin(Drosophila melanogaster)*pro::sfGFP Frag FWD CAGAGACCCCGGATCCAGATATGAGCAAAGG AGAAGAACTTTTCACTG	This paper	oKV701
*actin(Drosophila melanogaster)*pro::mKate2 Vect FWD CATCCAAGCTCGGACACCGTTGACGTAAGCT AGCAGGATCTTTGTGAAG	This paper	oKV702
*actin(Drosophila melanogaster)*pro::mKate2 Vect REV TTAATGAGCTCGGAGACCATATCTGGATCCG GGGT	This paper	oKV703
*actin(Drosophila melanogaster)*pro::mKate2 Frag 1 FWD CAGAGACCCCGGATCCAGATATGGTCTCCGA GCTCATTAAcGAA	This paper	oKV704
*actin(Drosophila melanogaster)*pro::mKate2 Frag 3 REV GATCCTGCTAGCTTACGTCAACGGTGTCCGA GC	This paper	oKV705
vap-1 qPCR FWD AGAGCGGTTGCACAGACGTT	This paper	oKV731
vap-1 qPCR REV GCTCAAGCCCTTTAGCCATGC	This paper	oKV732
cdc-42 qPCR FWD TTCGACAATTACGCCGTCACA	This paper	oKV739
cdc-42 qPCR REV ACGTCGGTCTGTGGATACGA	This paper	oKV740
fig-1 qPCR FWD ATGGGGTGATTGGGGTGCAT	This paper	oKV727
fig-1 qPCR REV TCAGACATTTCACCTTCGCACG	This paper	oKV728