Figure S1. **gop-1 encodes an evolutionarily conserved protein.** (A) Sequence alignment of *C. elegans* GOP-1, fly Ema, and human CLEC16A. Identical residues are shaded in black. The mutation sites in gop-1 alleles (tm5384, tm5654, tm5694, and yq79) are indicated. Red box, conserved but uncharacterized FPL domain; green box, putative transmembrane span; blue box, C-type lectin 1 domain in CLEC16A. (B) Gene structure of gop-1. Black boxes show coding segments. The site of the gop-1(yq79) mutation and the regions deleted in gop-1(tm5384), gop-1(tm5654), and gop-1(tm5694) are indicated. (C and D) Time-course analysis of cell corpses during embryonic (C) and germline (D) development in wild type (WT) and gop-1(tm5654), gop-1(tm5694), and gop-1(yq79) mutants. At least 15 animals were scored in each strain at each stage. Data are shown as mean ± SD. Data derived from different genetic backgrounds at multiple developmental stages were compared by two-way ANOVA followed by Bonferroni posttest. *, P < 0.05; **, P < 0.001.
Figure S2. Expression of human CLEC16A rescues the phagosome, endosome, and locomotion defects of gop-1 mutants. (A and B) Germ (A) and somatic (B) cell corpses were quantified in wild-type (WT) and gop-1(tm5384) without or with expression of the indicated transgenes 48 h after L4/adult molt [A, germ cell corpses] or at the 1.5-fold and fourfold stages [B, embryonic cell corpses]. At least 15 animals were scored in each strain. One-way ANOVA with Tukey’s posttest (A) or two-way ANOVA followed by Bonferroni posttest (B) was performed to compare all the other datasets with wild type. Data are shown as mean ± SD. **, P < 0.0001 (A); **, P < 0.001 (B); *, P < 0.05. All other points had P > 0.05. (C–G) DIC images of coelomocytes in WT and gop-1(tm5384) without or with expression of the indicated transgene. White arrowheads, normal endosomes; yellow arrowheads, abnormally enlarged vacuoles. Bars, 5 μm. Quantification is shown in H. At least 30 animals were scored in each strain. (I) gop-1 mutants show locomotion defects. The number of body bends per minute was quantified in the indicated strains. Data are shown as mean ± SD. At least 15 animals were scored in each strain. The WT dataset was compared with all the other datasets by one-way ANOVA with Tukey’s posttest. **, P < 0.0001. All the other points had P > 0.05. (J) Sequence alignment of UNC-108 and human Rab2a. Identical residues are in black. Motifs responsible for binding phosphate and Mg²⁺ (blue boxes) and the guanine base (red boxes) are shown. Switch I and II are domains that undergo dramatic conformational changes during nucleotide exchange. Mutations that arrest Rab2 in the constitutively inactive (S20N), active (Q65L), or nucleotide-free (N119I) forms, or that facilitate nucleotide exchange (K120E), are indicated. The two C-terminal cysteines that undergo prenylation are marked in green.
GOP-1 promotes apoptotic cell degradation

Figure S3. **gop-1 is widely expressed.** (A–I′) DIC and fluorescence images in wild type expressing GFP controlled by the gop-1 promoter. GFP was observed in multiple tissues (arrowheads) throughout embryonic to adult stages. (J–L″) Confocal fluorescence images in wild type coexpressing GFP::GOP-1 and markers of different organelles including Cherry::RAB-5 (J–J″, early endosome), Cherry::RAB-7 (K–K″, late endosome), or NUC-1::Cherry (L–L″, lysosome). White arrowheads, vesicles with overlapping GFP and Cherry fluorescence; pink arrowheads, GFP- and Cherry-positive vesicles that are in close proximity; yellow arrowheads, GFP-positive and Cherry-negative puncta. Quantification is shown at right (mean ± SD). Bars: (A–I′) 10 µm; (J–L″) 5 µm.
Figure S4. **gop-1** mutants show defects in endosome and dense core vesicle maturation. (A and B) Fluorescence images of coelomocytes in wild type (WT), gop-1(tm5384), and unc-108(n3263) expressing RME-8::GFP (A) or LMP-1::GFP (B) and injected with TR-BSA. White and yellow arrowheads indicate the appearance of TR-BSA in GFP-positive and GFP-negative vesicles, respectively. Loss of gop-1 and unc-108 arrests TR-BSA in enlarged hybrids of endosomes and lysosomes. (C–G) Confocal fluorescence images of the dorsal nerve cord in WT, gop-1(tm5384), unc-108(n3263), and unc-108(n3263); gop-1(tm5384) expressing IDA-1::GFP. Quantification is shown in G. (H–L) Confocal fluorescence images of the ventral nerve cord in WT, gop-1(tm5384), unc-108(n3263), and unc-108(n3263); gop-1(tm5384) expressing GLR-1::GFP. Quantification is shown in L. In G and L, at least 15 animals were scored, and data are shown as mean ± SD. One-way ANOVA with Tukey’s posttest was performed to compare mutant datasets with wild type or datasets linked by lines. **, P < 0.0001; N.S., no significance. Bars, 5 µm.
Figure S5. GOP-1 disrupts the UNC-108–GDI-1 complex. (A) The interaction between GOP-1 and RAB-5, RAB-7, and RAB-14 was examined by yeast two-hybrid analysis. GOP-1 did not interact with these Rabs. (B) FLAG-UNC-108-MYC-HIS-GDI-1 complex was purified from yeast cells by two-step affinity purification followed by gel filtration on a S100 column. Protein fractions were collected and analyzed by immunoblotting (left) and Coomassie blue staining (right). (C) Wild-type UNC-108 but not UNC-108 (ΔCC), which lacks the two prenylatable C-terminal cysteines, coprecipitated with GDI-1 in yeast cells. (D) Addition of GOP-1 disrupts the UNC-108–GDI-1 complex. Schematic illustration of the GDI displacement assay is shown at top. At least three independent experiments were performed, and a representative result is shown. (E and F) GOP-1 (E) and UNC-108 (F) were expressed in and purified from *E. coli*, and their binding to PC and PC + PtdIns3P (8%) liposomes was detected by Western blot. No UNC-108 or GOP-1 was recovered in the top fraction in a liposome flotation assay.
Video 1. **RAB-5 transiently associates with apoptotic cell-containing phagosomes.** A somatic cell corpse in a wild-type *C. elegans* embryo expressing GFP::RAB-5 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope (Eclipse Ti-E; Nikon) with an UltraView spinning-disc confocal scanner unit (PerkinElmer). The frames were taken every 1 min for 7 min and are displayed every 1 s. Bar, 2.5 µm. Selected images are shown in Fig. 3 A.

Video 2. **Loss of gop-1 causes prolonged association of RAB-5 with phagosomes.** A somatic cell corpse in a *gop-1(tm5384)* mutant embryo expressing GFP::RAB-5 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 2 min for 28 min and are displayed every 1 s. Bar, 2.5 µm. Selected images are shown in Fig. 3 A.

Video 3. **PtdIns3P transiently accumulates on phagosomes.** A somatic cell corpse in a wild-type *C. elegans* embryo expressing YFP::2xFYVE, the PtdIns3P biosensor, is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 1 min for 13 min and are displayed every 1 s. Bar, 2.5 µm. Selected images are shown in Fig. 3 B.

Video 4. **Loss of gop-1 causes persistent accumulation of PtdIns3P on phagosomes.** A somatic cell corpse in a *gop-1(tm5384)* mutant embryo expressing YFP::2xFYVE, the PtdIns3P biosensor, is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 3 min for 81 min and are displayed every 1 s. Bar, 2.5 µm. Selected images are shown in Fig. 3 B.

Video 5. **UNC-108 transiently associates with apoptotic cell-containing phagosomes.** A somatic cell corpse in a wild-type *C. elegans* embryo expressing both mCherry::MTM-1(C378S) and GFP::UNC-108 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 1 min for 13 min and are displayed every 1 s. Bar, 5 µm. Selected images are shown in Fig. 6 A.

Video 6. **Loss of gop-1 abrogates phagosomal recruitment of UNC-108.** A somatic cell corpse in a *gop-1(tm5384)* mutant embryo expressing both mCherry::MTM-1(C378S) and GFP::UNC-108 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 1 min for 13 min and are displayed every 1 s. Bar, 5 µm. Selected images are shown in Fig. 6 B.
Video 7. **UNC-108 transiently associates with endosomes in coelomocytes.** A coelomocyte in a wild-type *C. elegans* expressing both NUC-1::Cherry and GFP::UNC-108 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 2 min for 14 min and are displayed every 1 s. Bar, 5 µm. Selected images are shown in Fig. 7 A.

Video 8. **Loss of gap-1 abolishes recruitment of UNC-108 to endosomes.** A coelomocyte in a gap-1(tm5384) worm expressing both NUC-1::Cherry and GFP::UNC-108 is followed. Images were analyzed by time-lapse confocal microscopy using an inverted fluorescence microscope with an UltraView spinning-disc confocal scanner unit. The frames were taken every 5 min for 70 min and are displayed every 1 s. Bar, 5 µm. Selected images are shown in Fig. 7 B.

**Table S1. Loss of tbc-2 enhances the somatic cell corpse phenotype of gap-1 and unc-108 mutants**

| Genotype               | No. of somatic cell corpses | Changes | P-value<br>  |
|------------------------|------------------------------|---------|--------------|
| Wild type              | 7.7 ± 2.2                    |         |              |
| tbc-2(qx20)            | 15.7 ± 2.2                   | ↑       | <0.0001      |
| unc-108(n3263)         | 19.7 ± 3.6                   | ↑       | <0.0001      |
| gap-1(tm5384)          | 21.3 ± 2.8                   | ↑       | <0.0001      |
| unc-108, tbc-2         | 35.7 ± 3.4                   | ↑       | <0.0001      |
| tbc-2, gap-1           | 32.7 ± 3.4                   | ↑       | <0.0001      |

*Somatic cell corpses were scored at the twofold embryonic stage. At least 15 animals were scored in each strain and data are shown as mean ± SD.*

Unpaired *t* tests were performed as follows: data derived from the single mutant were compared with wild type. unc-108, tbc-2 was compared with unc-108(n3263), and tbc-2, gap-1 was compared with gap-1(tm5384). ↑, significant change.

**Table S2. Loss of sand-1 enhances the germ cell corpse phenotype of gap-1 and unc-108 mutants**

| Genotype               | No. of germ cell corpses | Changes | P-value<br>  |
|------------------------|----------------------------|---------|--------------|
| Control RNAi           | 5.7 ± 1.2                  |         |              |
| unc-108(RNAi)          | 18.1 ± 1.8                 | ↑       | <0.0001      |
| gap-1(RNAi)            | 12.9 ± 1.9                 | ↑       | <0.0001      |
| sand-1(ak1963); control RNAi | 20.5 ± 4.0                  | ↑       | <0.0001      |
| sand-1(ak1963); unc-108(RNAi) | 60.1 ± 7.7                  | ↑       | <0.0001      |
| sand-1(ak1963); gap-1(RNAi) | 59.0 ± 7.3                  | ↑       | <0.0001      |
| Wild type              | 6.7 ± 2.0                   |         |              |
| tbc-2(qx20)            | 6.9 ± 0.8                   | No      | 0.8119       |
| unc-108(n3263)         | 18.5 ± 6.4                  | ↑       | <0.0001      |
| gap-1(tm5384)          | 17.7 ± 3.9                  | ↑       | <0.0001      |
| unc-108, tbc-2         | 16.9 ± 1.7                  | No      | 0.377        |
| tbc-2, gap-1           | 16.2 ± 1.0                  | No      | 0.1552       |

*Germ cell corpses in one gonad arm were quantified in the indicated strains at 48 h after L4/adult molt. At least 15 animals were scored in each strain, and data are shown as mean ± SD.*

Unpaired *t* tests were performed as follows: data derived from each single mutant or RNAi treatment were compared with wild type. unc-108, tbc-2 was compared with unc-108(n3263), and tbc-2, gap-1 was compared with gap-1(tm5384). ↑, significant change; No, no significant change.