**SUPPLEMENTAL FIGURE LEGENDS**

**Supplemental Figure 1. Schematic of the forward genetic screen for TS-EL morphogenesis-defective mutants and the numbers of alleles at each step.** The boxes represent pools of mutants with the numbers of mutant alleles indicated. The gray triangles represent steps of the forward genetic screen and point to the box(es) that resulted from the step.

**Supplemental Figure 2. Penetrant and variable terminal elongation-defective phenotypes after L4 upshifts for TS-EL mutants. (A-B)** Quantification of 79 penetrant terminal elongation-defective phenotypes for TS-EL mutant (≥70% embryos arrested with similar extents of elongation) and wild-type (N2) embryos after L4 upshifts. 63 TS-EL mutants arrested with 70% or more exhibiting no elongation (A). 16 TS-EL mutants arrested with 70% or more exhibiting similar less extensive elongation arrests (B). **(C)** Quantification of 30 variable terminal elongation-defective phenotypes for TS-EL mutant embryos after L4 upshift (40-70% embryos arrested with a similar extent of elongation). Percent of embryos that differentiated well but arrested with no elongation (purple), or arrested at the 1.25<2-fold stage (light blue), at the 2-fold stage (green), at the 2<3-fold stage (yellow), or with full elongation (≥3-fold), or exhibited differentiation defects (red) were scored using Nomarski optics. Examples of Nomarski images for control embryos at each elongation stage are shown in the Key in Figure 1. TS-EL mutants are shown from left to right by decreasing penetrance for the most penetrant elongation category. If the causal mutation has been identified for a TS-EL mutant, the affected gene is listed next to the TS-EL mutant allele, and the number of embryos scored for each mutant is in brackets.

**Supplemental Figure 3. Quantification of less penetrant elongation defects in TS-EL mutants after late upshifts.** Quantification of terminal elongation-defective phenotypes for TS-EL mutant and wild-type (N2) embryos after late upshifts. **(A)** TS-EL mutants with penetrant elongation defects after L4 upshifts that exhibited weak defects after late upshifts. 50-100% of embryos undergo full elongation and hatch and/or twitch. **(B)** TS-EL mutants with penetrant elongation defects after L4 upshifts that exhibited variable elongation defects after late upshifts. Percent of embryos that differentiated well but arrested with no elongation (purple), or arrested at the 1.25<2-fold stage (light blue), at the 2-fold stage (green), at the 2<3-fold stage (yellow), or with full elongation (≥3-fold), or exhibited differentiation defects (red) were scored using Nomarski optics. Examples of Nomarski images for control embryos at each elongation stage are shown in the Key in Figure 1. TS-EL mutants are shown from left to right by decreasing penetrance for the full elongation category. If the causal mutation has been identified for a TS-EL mutant, the affected gene is listed next to the TS-EL mutant allele, and the number of embryos scored for each mutant is in brackets.

**Supplemental Figure 4. Identification of putative *emb-5(or862*ts) causal mutation. (A)** SNP mapping data for the *or862*ts mutant on linkage group III with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars). Candidate essential genes in which a missense mutation (*emb-5*) or splice site mutation (*mua-3*) were detected are indicated. For complementation test in which *or862*ts failed to complement the known genetic mutation, the gene is dark and bolded; for complementation test in which *or862*ts complemented the known mutation, the gene is gray and bolded. See Table 2 for complementation test results.

**(B)** Sequence alterations in the predicted EMB-5 protein for *or862*ts and the other 4 alleles identified by our screen (*or789*ts, *or975*ts, *or1097*ts, and *or1154*ts). The causal mutation for *hc61*ts, the mutant allele used for the complementation test, has not been curated. EMB-5 is orthologous to human SPT6, a transcription elongation factor (Kwak and Lis, 2013; Nishiwaki et al., 1993). EMB-5 contains an acidic N-terminal domain (brown), a helix-turn-helix DNA binding domain (HTH, purple), a Tex-like domain (light blue), an RNase H-like domain (Yqgf, dark blue), 2 RuvA 2-like domains (pink), an S1 RNA binding domain (green) and an SH2 domain (orange; INTERPRO and Pfam on https://wormbase.org). **(C)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or862*ts missense mutation at the *emb-5* locus. The orange bar in the depth of coverage section indicates the homozygosity of the T to C nucleotide change across the reads. Gray lines indicate all bases matched the reference sequence; blue lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from right to left.

**Supplemental Figure 5. Identification of putative *zim-3*(*or1106*ts and *or1002*ts) causal mutations. (A-B)** SNP mapping data for *or1106*ts (A) and *or1002*ts (B) mutants on linkage group IV with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars). Candidate essential genes in which missense mutations (*or1106*ts: *rpn-1* and *C33D9.13*; *or1002*ts: *zim-3*, *rpn-1*, *let-70*, and *F07C6.4*) or a nonsense mutation (*or1106*ts: *zim-3*) were detected are indicated. For complementation tests in which the TS-EL mutants failed to complement the known genetic mutation, the gene is dark and bolded; for complementation test in which the TS-EL mutant complemented the known mutation, the gene is gray and bolded; for genes in which a complementation test was not performed, genes are not bolded. See Table 2 for complementation test results. **(C)** Sequence alterations in the predicted ZIM-3 protein, isoform a, for *or1106*ts, *or1002*ts, and *tm2303*ts, the known mutant allele used for the complementation test. ZIM-3 is a *Caenorhabditis* specific gene required for homologous pairing of chromosomes during meiosis and contains 2 zinc-finger domains (black box) (Phillips and Dernburg, 2006; Phillips et al., 2009). **(D)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or1106*ts nonsense mutation (LEFT) and the *or1002*ts missense mutation (RIGHT) at the *zim-3* locus. The green bar in the depth of coverage section indicates the homozygosity of the T to A nucleotide change across the reads for *or1106*ts. The orange bar in the depth of coverage section indicates the homozygosity of the T to G nucleotide change across the reads for *or1002*ts. Gray lines indicate all bases matched the reference sequence; blue lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from left to right.

**Supplemental Figure 6. Identification of putative *hlh-1*(*or1312*ts) causal mutation. (A)** SNP mapping data for the *or1312*ts mutant on linkage group II with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars).Candidate essential gene in which a missense mutation was detected is indicated. For complementation test in which *or1312*ts failed to complement the known genetic mutation, the gene is dark and bolded. See Table 2 for complementation test results. **(B)** Sequence alterations in the predicted HLH-1 protein, isoform b, for *or1312*ts and *cc561*ts, the known mutant allele used for the complementation test. HLH-1 is orthologous to human MyoD, a basic helix-loop-helix (bHLH) transcription factor (Krause et al., 1990), and contains a bHLH domain (black box corresponding to amino acids 155-231; INTERPRO, Pfam, smart, and superfamily on https://wormbase.org). **(C)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or1312*ts missense mutation at the *hlh-1* locus. The red bar in the depth of coverage section indicates the homozygosity of the C to T nucleotide change across the reads. Gray lines indicate all bases matched the reference sequence; blue lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). The light yellow read implies the mate pair is on linkage group IV instead of II, and is likely a sequencing artifact. Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from left to right.

**Supplemental Figure 7. Identification of putative *emb-4*(*or1330*ts) causal mutation. (A)** SNP mapping data for the *or1330*ts mutant on linkage group V with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars). Candidate essential genes in which a missense mutation (*Y39B6A.25*) or a 1 bp insertion (*emb-4*) were detected are indicated. For complementation test in which *or1330*ts failed to complement the known genetic mutation, the gene is dark and bolded; for the gene in which a complementation test was not performed, the gene is not bolded. See Table 2 for complementation test results. **(B)** Sequence alterations in the predicted EMB-4 protein, isoform a, for *or1330*ts and *hc60*ts, the known mutant allele used for the complementation test (Checchi and Kelly, 2006). EMB-4 is orthologous to human Aquarius (Katic and Greenwald, 2006) and contains an N-terminus Aquarius intron-binding domain (green) and 2 AAA ATPase DEAD-box helicase domains (blue; INTERPRO and Pfam on https://wormbase.org). **(C)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or1330*ts missense mutation at the *emb-4* locus. The purple “I” indicates an insertion of an A into codon 967. Gray lines indicate all bases matched the reference sequence; blue lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from right to left.

**Supplemental Figure 8. Identification of putative *rib-2*(*or1688*ts) causal mutation. (A)** SNP mapping data for the *or1688*ts mutant on linkage group III with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars). Candidate essential genes in which missense mutations (*rib-2* and *zfp-1*) or splice site mutations (*dcr-1*) were detected are indicated. For complementation test in which *or1688*ts failed to complement the known genetic mutation, the gene is dark and bolded; for complementation test in which *or688*ts complemented the known mutation, the gene is gray and bolded; for the gene in which a complementation test was not performed, the gene is not bolded. See Table 2 for complementation test results. **(B)** Sequence alterations in the predicted RIB-2 protein for *or1688*ts and *gk318*, the known mutant allele used for the complementation test. RIB-2 is orthologous to human exostosin-like glycosyltransferase 3 (EXT3) (Kitagawa et al., 2007) and contains an exostosin-like domain (yellow) and a glycosyltransferase domain (orange; hmmpanther, INTERPRO, Pfam, and superfamily on https://wormbase.org). **(C)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or1688*ts missense mutation at the *rib-2* locus. The green bar in the depth of coverage section indicates the homozygosity of the T to A nucleotide change across the reads. Gray lines indicate all bases matched the reference sequence; blue lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from left to right.

**Supplemental Figure 9. Identification of putative *emb-9* (*or1723*ts). (A)** SNP mapping data for the *or1723*ts mutant on linkage group III with identified causal mutations, showing the frequency of homozygous parental alleles plotted against chromosomal position in bins of either 1 megabase (gray bars) or 0.5 megabase (red bars). Candidate essential genes in which missense mutations were detected are indicated. For complementation test in which *or1723*ts failed to complement the known genetic mutation, the gene is dark and bolded; for complementation test in which *or1723*ts complemented the known mutation, the gene is gray and bolded; for the gene in which a complementation test was not performed, the gene is not bolded. See Table 2 for complementation test results. **(B)** Sequence alterations in the predicted EMB-9 protein, isoform a, for *or1723*ts and *hc70*ts, the known mutant allele used for the complementation test. EMB-9 is orthologous to human type IV collagen α1 subunit (COL4A5) (Guo and Kramer, 1989). EMB-9 contains 10 collagen triple helix repeat domains (green) and 2 c-type lectin fold domains (blue; INTERPRO and Pfam on https://wormbase.org). **(C)** Integrative Genomics Viewer (Broad Institute) screenshot of the sequencing reads at the site of the *or1723*ts missense mutation at the *emb-9* locus. The red bar in the depth of coverage section indicates the homozygosity of the G to A nucleotide change across the reads. Gray lines indicate all bases matched the reference sequence; blue and teal lines imply reads of the opposite strand (https://software.broadinstitute.org/software/igv/interpreting\_pair\_orientations). Single nucleotide changes are indicated on each read (green A, blue C, orange G, and red T). Nucleotide and amino acid sequences are read from right to left.

**Supplemental Figure 10. Additional ventral enclosure defects observe in *or1219*ts mutants after late upshifts.** Maximum projection images of *or1219*ts mutant embryos expressing DLG-1::GFP to mark epidermal cell membranes after late upshifts. Minutes are listed across the top; time 0 corresponds to the start of imaging at the bean stage (one hour at 26°C after the late upshift time point; see Materials and Methods). Embryos were imaged every 20 minutes for 360 minutes. Each row represents a single embryo over time in the lateral orientation (row 1) and ventral orientation (rows 2-4). The top three rows (1-3) are embryos that exhibited incorrect (non-leading) cell contacts during ventral enclosure. The bottom row (4) shows an embryo that made proper leading cell contact at the ventral midline, but the ventral pocket remained.