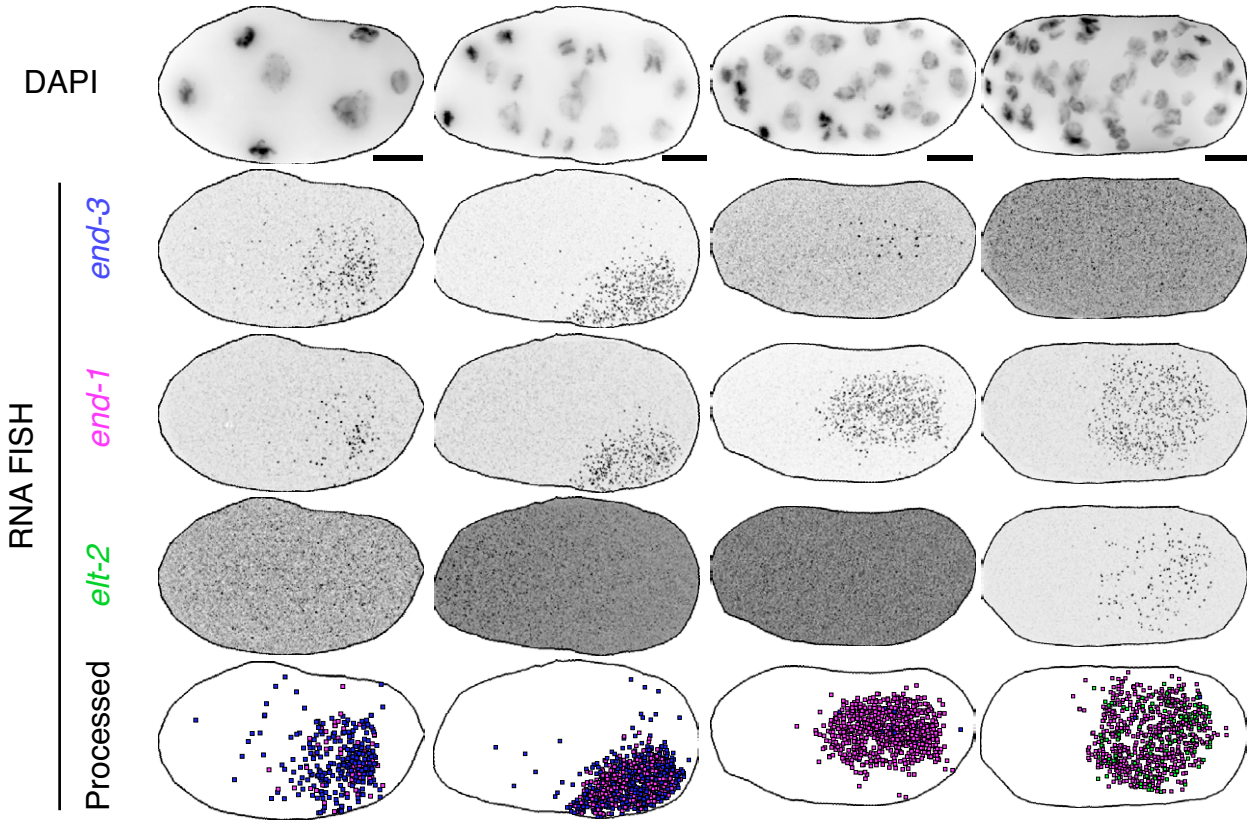

Fig. S1. Embryo images and processed data. Images of the same four fixed embryos as displayed in Fig. 1. First row is a DAPI maximum merge projection. Scale bars: 10 μm. The next three rows are fluorescence images (Alexa594, Tetramethylrhodamine, and Cy5) corresponding to single molecule RNA FISH probes for *end-3*, *end-1* and *elt-2*. The images were filtered through a 2D Laplacian of Gaussian filter, maximum merge projected and auto-contrasted to saturate the top 0.1% of pixels. The last row shows the computationally identified single RNA positions for *end-3*, *end-1* and *elt-2* in blue, magenta and green, respectively.

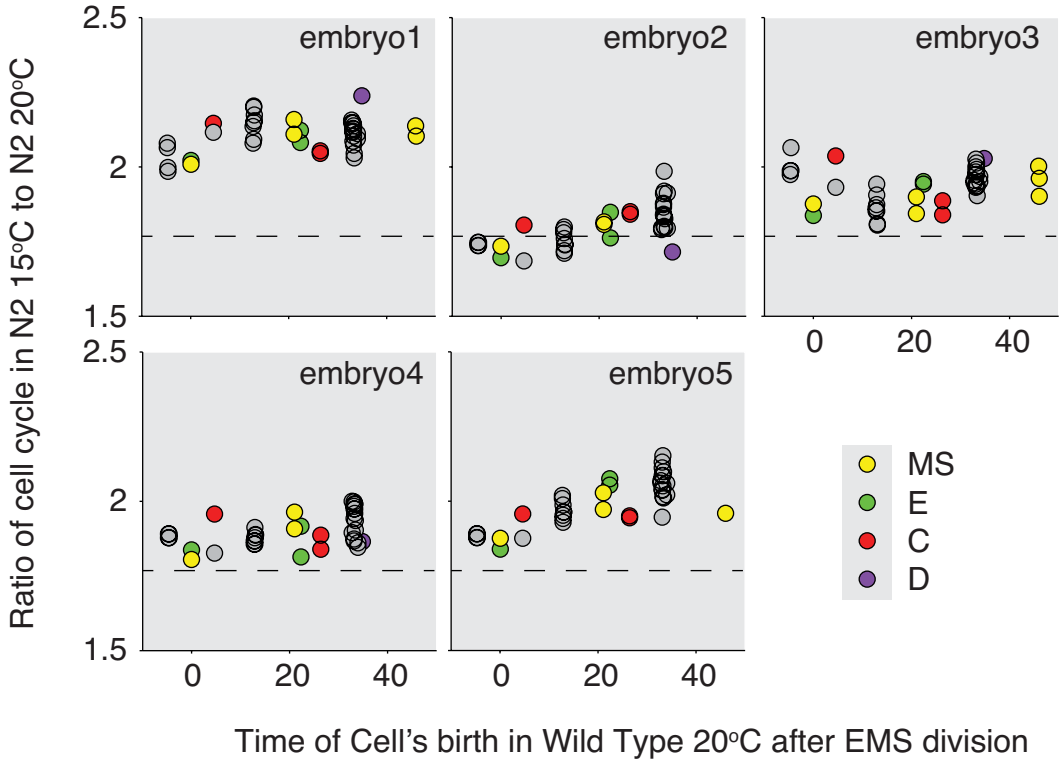

Fig. S2. Cell cycling slowdown at 15°C. Ratio of measured cell cycle times in 5 N2 embryos grown at 15°C to the corresponding cell cycle for N2 embryos grown at 20°C derived from Bao et al. and Sulston et al. (Bao et al., 2008; Sulston et al., 1983). The *x* coordinate is the time that the cell is born in a wild-type 20°C embryo. The dashed line occurs at 1.77, the ratio of overall embryo development time at 15°C to 20°C reported by Wong et al. (Wong et al., 1995). Colored symbols indicate cells of the MS, E, C and D lineages, whereas gray symbols represent all other lineages.

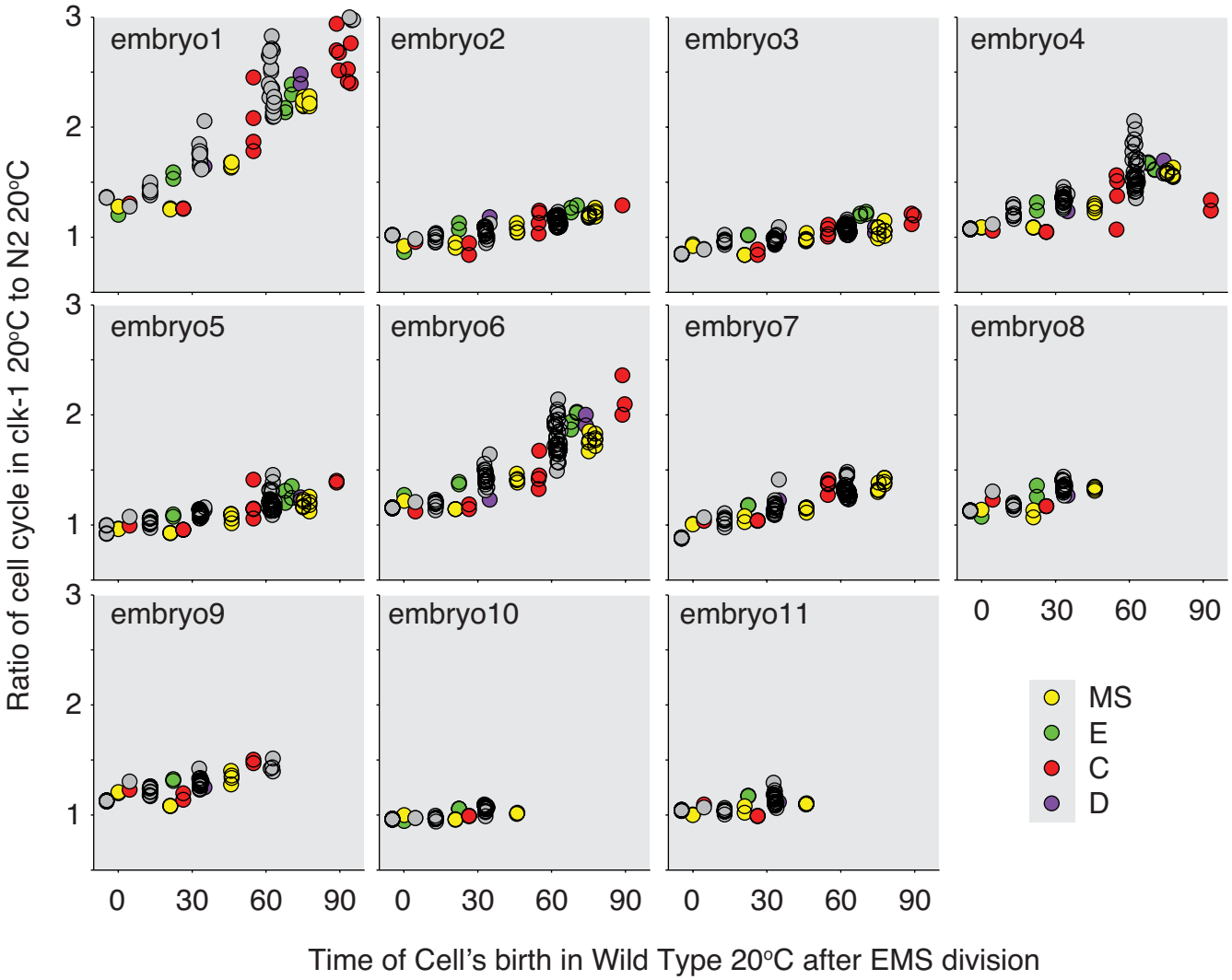

Fig. S3. Cell cycling slowdown in *clk-1(qm30)* mutants. Ratio of measured cell cycle times in 11 *clk-1(qm30)* embryos grown at 20°C to the corresponding cell cycle for N2 embryos grown at 20°C derived from Bao et al. and Sulston et al. (Bao et al., 2008; Sulston et al., 1983). The *x* coordinate is the time that the cell is born in a wild-type 20°C embryo. Colored symbols indicate cells of the MS, E, C and D lineages, whereas gray symbols represent all other lineages.

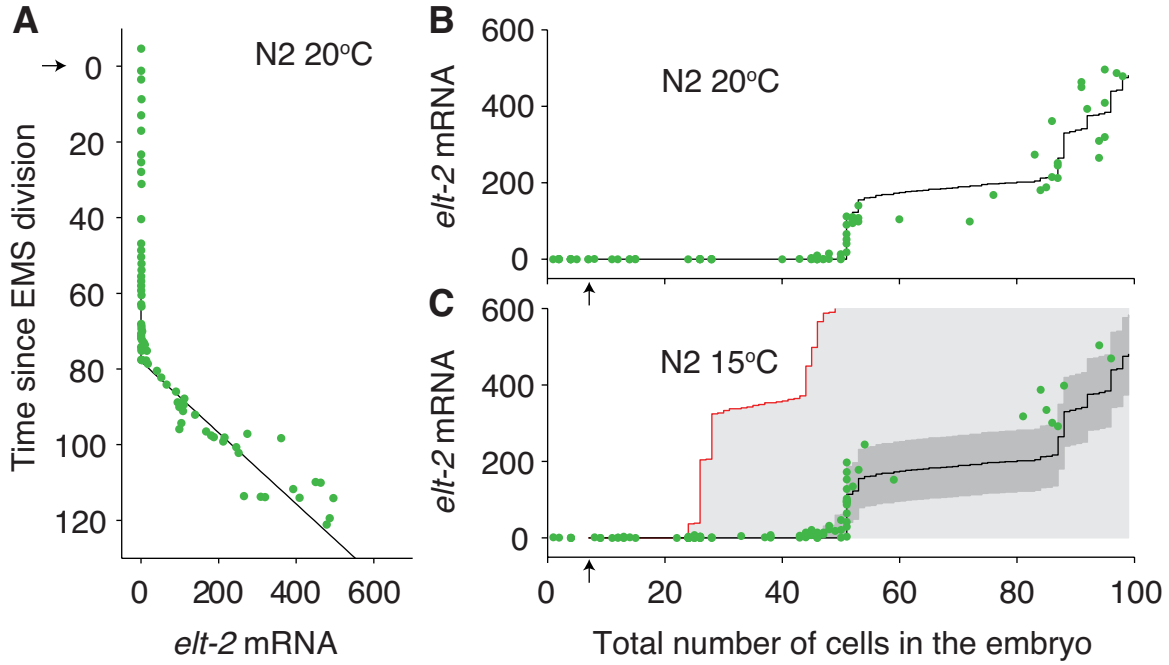

Fig. S4. Comparison of transcription dynamics at 15°C and cell cycle scaling predictions. (**A**) *elt-2* mRNA versus assigned embryo age for N2 embryos grown at 20°C. The black line is a manual fit to the *elt-2* transcription dynamics. The small arrows indicates the time of birth of the E cell. (**B**) As in A, but plotting RNA against the total number of cells in each embryo. The black line is the same as in A after coordinate transformation. (**C**) RNA versus embryo cell count for N2 worms grown at 15°C. Using N2 20°C lineage data scaled by the 1.77 slowdown observed at 15°C (Fig. S2), we overlaid a red line for the expected dynamics if transcription is not slowed down at all ($\alpha = 0$), a black line corresponding to perfect scaling ($\alpha = 1$), a dark-gray region corresponding to a 10% imperfection in scaling ($\alpha \in [0.9, 1.1]$, linear interpolation) and a light-gray region extending from no slowdown to a doubling of the logarithm of slowdown compared with that of cell cycles ($\alpha \in [0, 2]$, geometric interpolation). The small arrows on the *x* axes in B and C indicate an equivalent stage to the arrow in A.

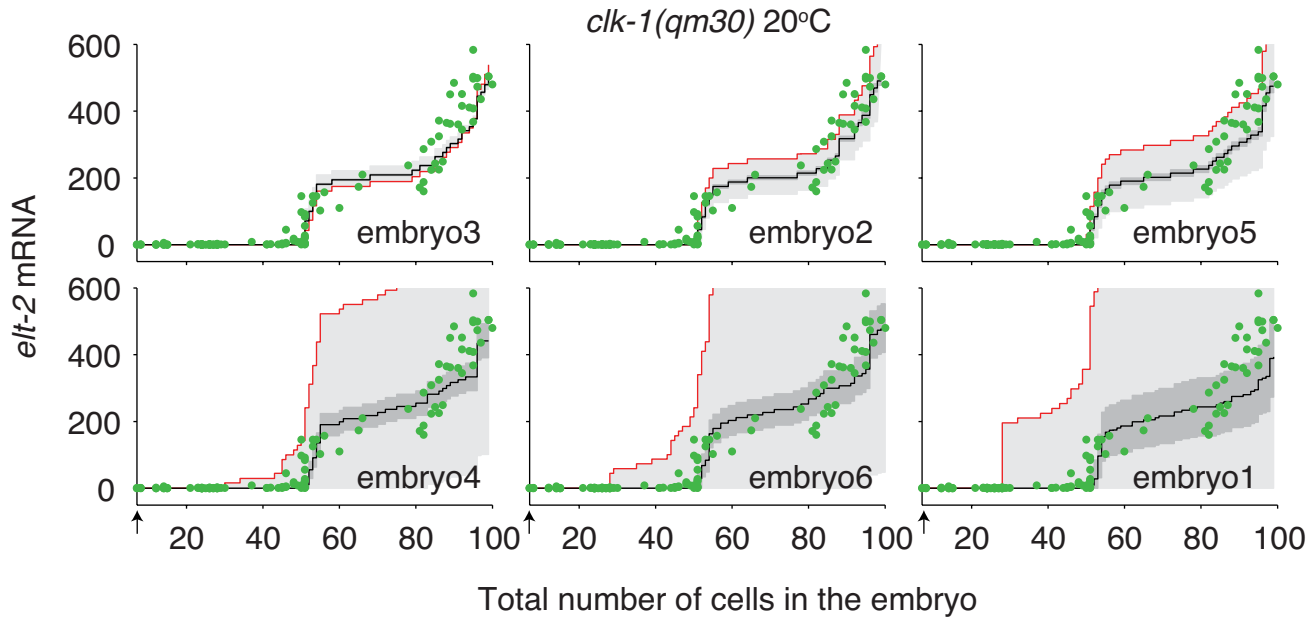

Fig. S5. Comparison of transcription dynamics in *clk-1(qm30)* and cell cycle scaling predictions. The green points in all six panels are the same *elt-2* mRNA versus total embryo cell count data for a sample of *clk-1(qm30)* embryos grown at 20°C. The small arrows on the *x* axes indicate the same cell stage (birth of the E cell) as in Fig. S4B,C. In each panel, we used lineage data from a different *clk-1(qm30)* embryo (numbered consistently with Fig. S3) to calculate scaling prediction lines and regions as in Fig. S4. Only embryos in which at least one of the 4E→8E divisions was observed were used, and the embryos are sorted from least to greatest developmental slowdown.

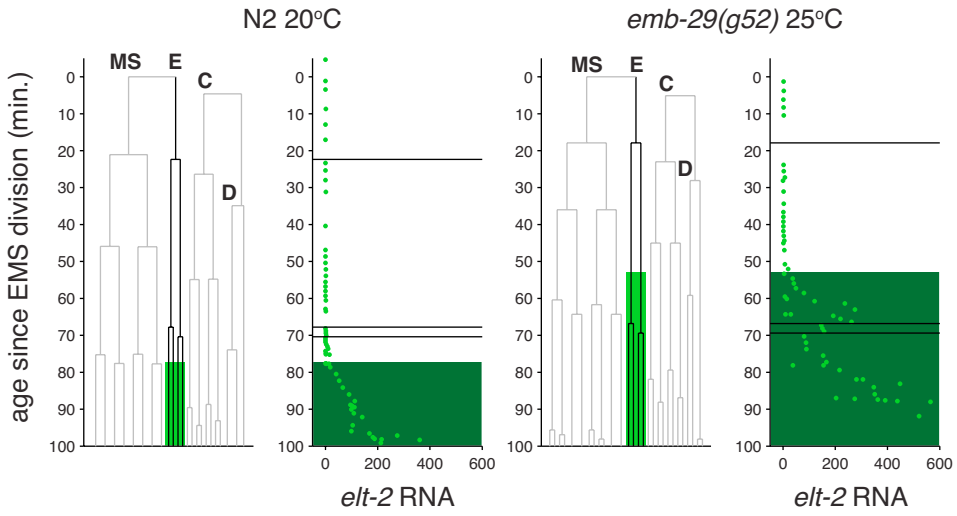

Fig. S6. Temporal transcriptional onset of *elt-2* in N2 and *emb-29(g52)* strains. (Left two panels) Embryonic cell lineage of N2 worms from Bao et al. adjusted to the average cell division rate reported by Sulston et al. at 20°C (Bao et al., 2008; Sulston et al., 1983), and RNA FISH trajectory for *elt-2* as a function of assigned embryo age for a sample of N2 worms grown at 20°C. The E lineage divisions are marked. The dark-green shaded area indicates the period of *elt-2* transcription. (Right two panels) Embryonic cell lineage of an *emb-29(g52)* embryo grown at 25°C and RNA FISH trajectory for a sample of such worms. The *elt-2* onset occurs well before the 2E cell divisions.

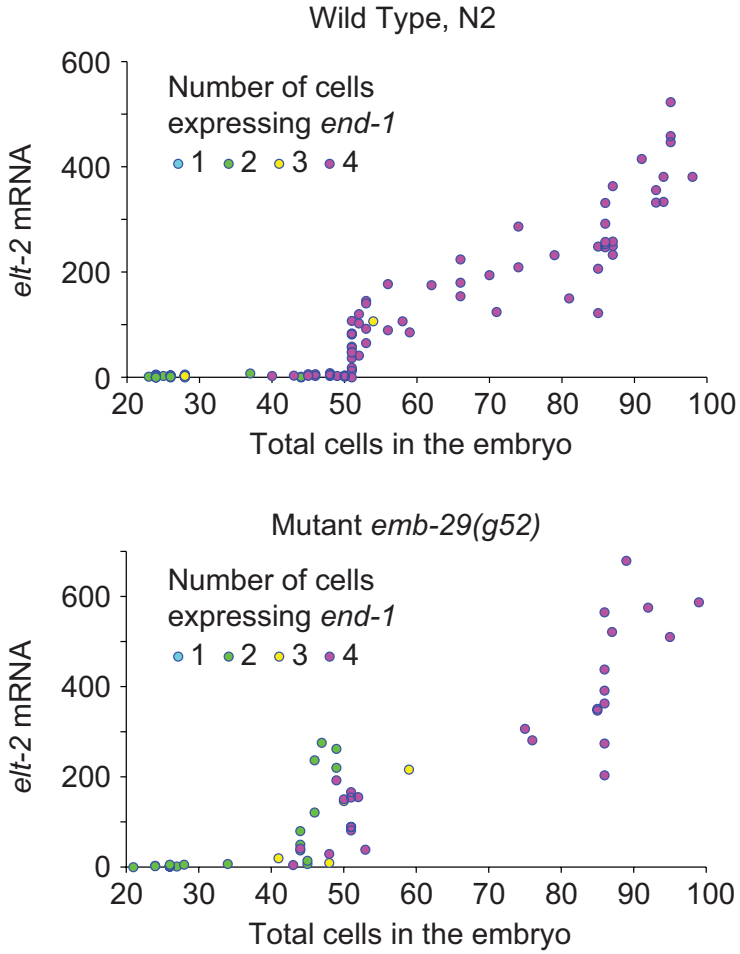

Fig. S7. Number of E cells at the onset of *elt-2* transcription in N2 and *emb-29(g52)* strains. (Top) Wild-type *elt-2* expression. Each data point is color-coded by the number of cells in the embryo expressing *end-1*, a proxy for the number of E cells in the embryo. (Bottom) The same for the mutant strain *emb-29(g52)*. In this case, *elt-2* expression begins even when there are only two E cells.

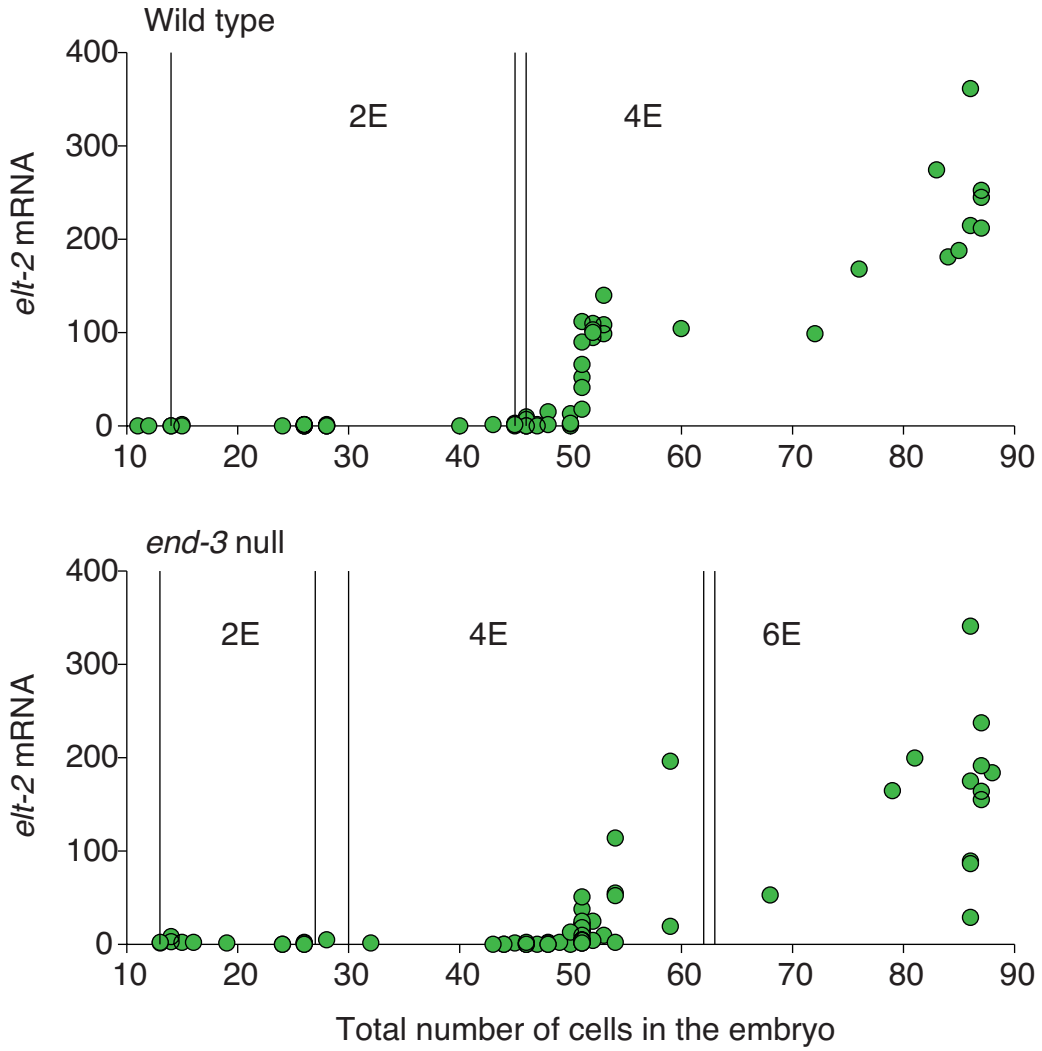

Fig. S8. Effect of deleting *end-3* on *elt-2* transcriptional dynamics. (Top) Wild-type *elt-2* expression. Vertical lines correspond to E lineage division events. (Bottom) *elt-2* expression trajectory in a strain carrying the *end-3(ok1448)* deletion. Timing of the E lineage events shown for this strain were based on the report of Boeck et al. (Boeck et al., 2011). As the timings of all divisions except for the E lineage divisions were found to be the same as in wild type (Boeck et al., 2011), the similar cell stage of *elt-2* onset implies that the onset occurs at the same time as wild type despite the early 2E divisions.

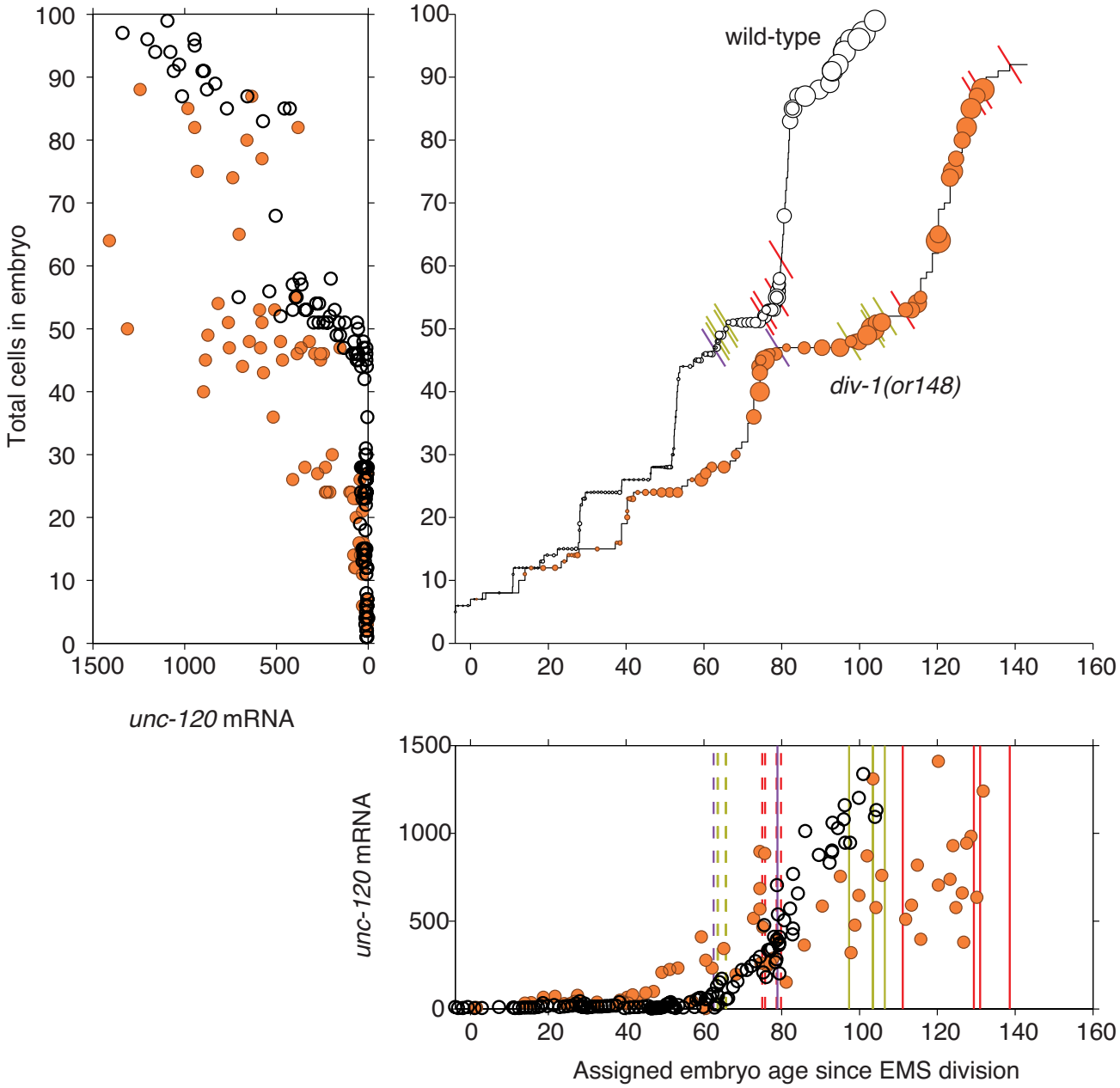

Fig. S9. *unc-120* expression in the *div-1(or148)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Purple, yellow and red solid vertical lines correspond to the divisions $D \rightarrow 2D$, $4C \rightarrow 8C$ and $4MS \rightarrow 8MS$, respectively, in the mutant. Dashed lines correspond to wild type. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks indicate relevant D,MS and C lineage division events.

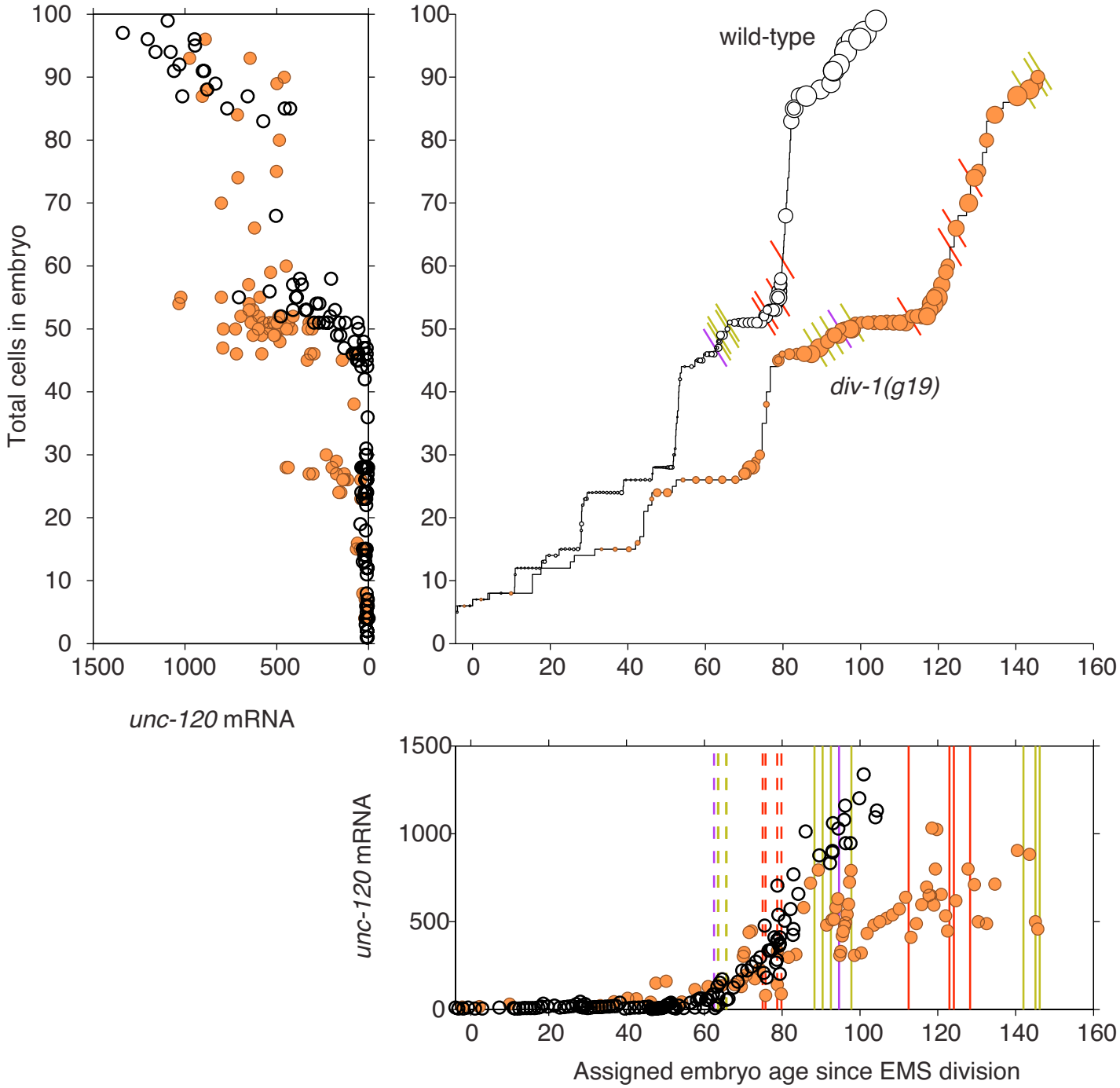

Fig. S10. *unc-120* expression in the *div-1(g19)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Purple, yellow and red solid vertical lines correspond to the divisions $D \rightarrow 2D$, $4C \rightarrow 8C$ and $4MS \rightarrow 8MS$, respectively, in the mutant. Dashed lines correspond to wild type. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks indicate relevant D, MS and C lineage division events.

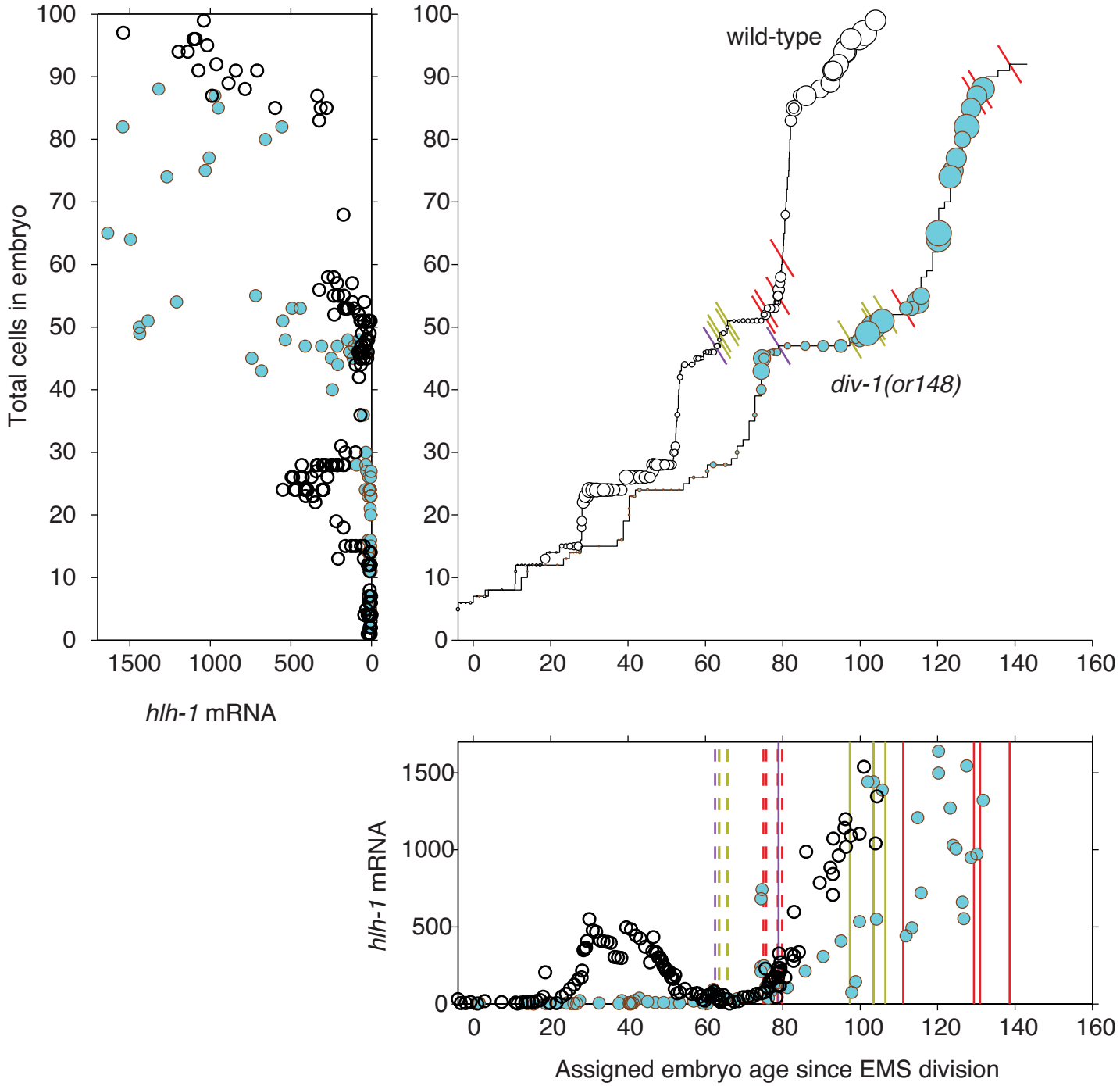

Fig. S11. *hlh-1* expression in the *div-1(or148)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Purple, yellow and red solid vertical lines correspond to the divisions $D \rightarrow 2D$, $4C \rightarrow 8C$ and $4MS \rightarrow 8MS$, respectively, in the mutant. Dashed lines correspond to wild type. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks indicate relevant D, MS and C lineage division events.

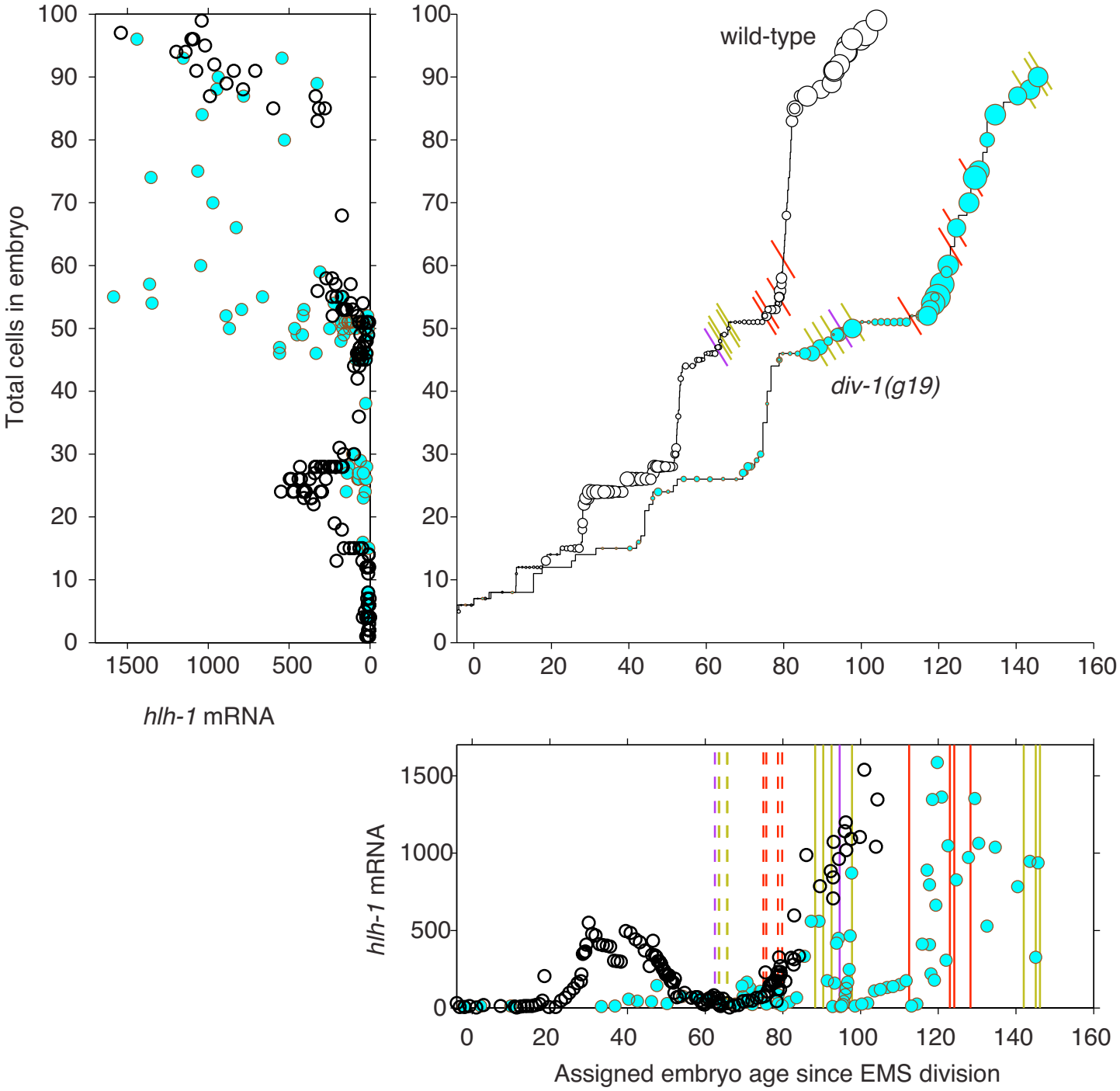

Fig. S12. *hlh-1* expression in the *div-1(g19)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Purple, yellow and red solid vertical lines correspond to the divisions $D \rightarrow 2D$, $4C \rightarrow 8C$ and $4MS \rightarrow 8MS$, respectively, in the mutant. Dashed lines correspond to wild type. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks indicate relevant D, MS and C lineage division events.

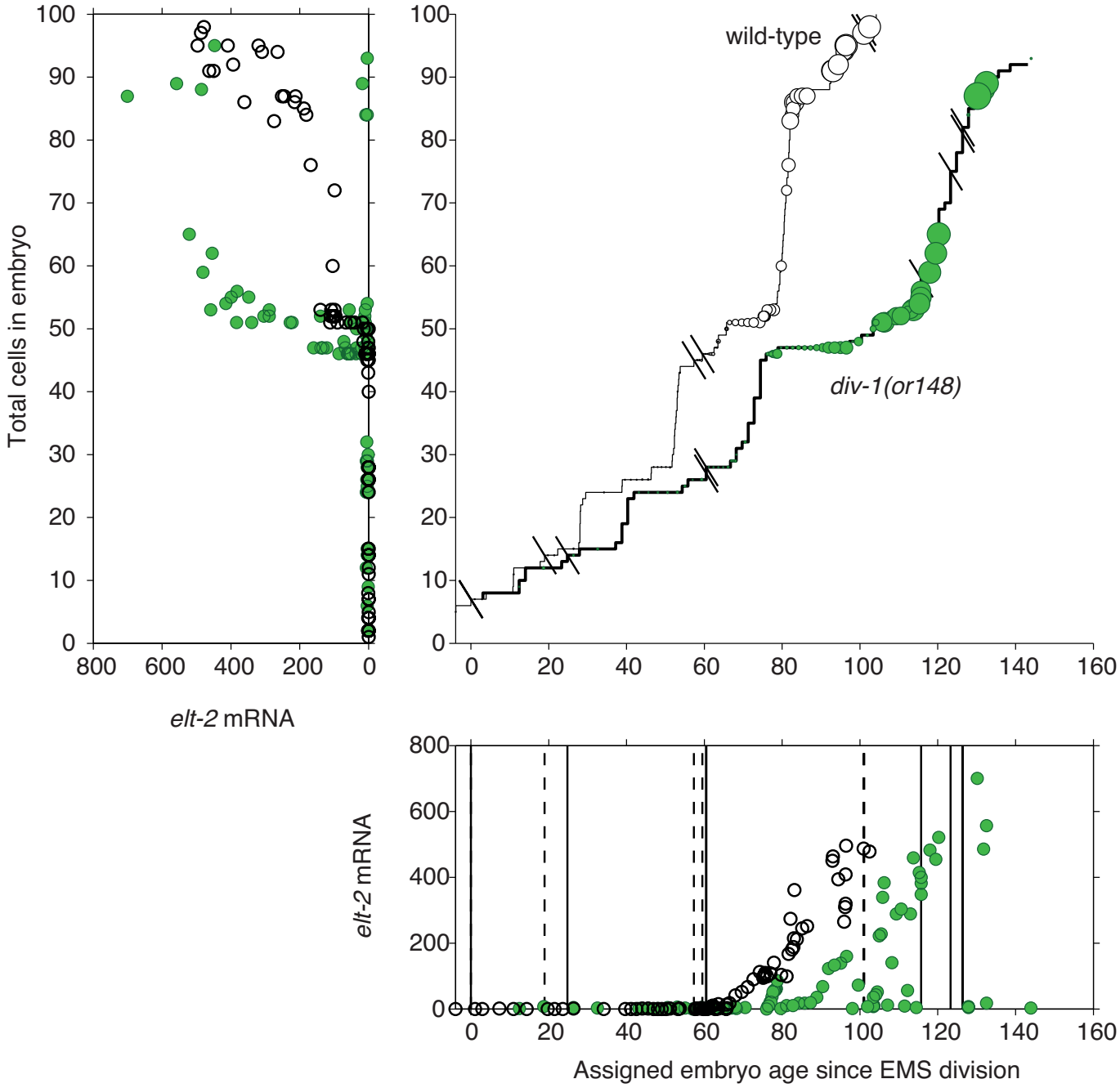

Fig. S13. *elt-2* expression in the *div-1(or148)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Black solid and dashed lines indicate E lineage division events in mutant and wild type, respectively. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks are E lineage division events.

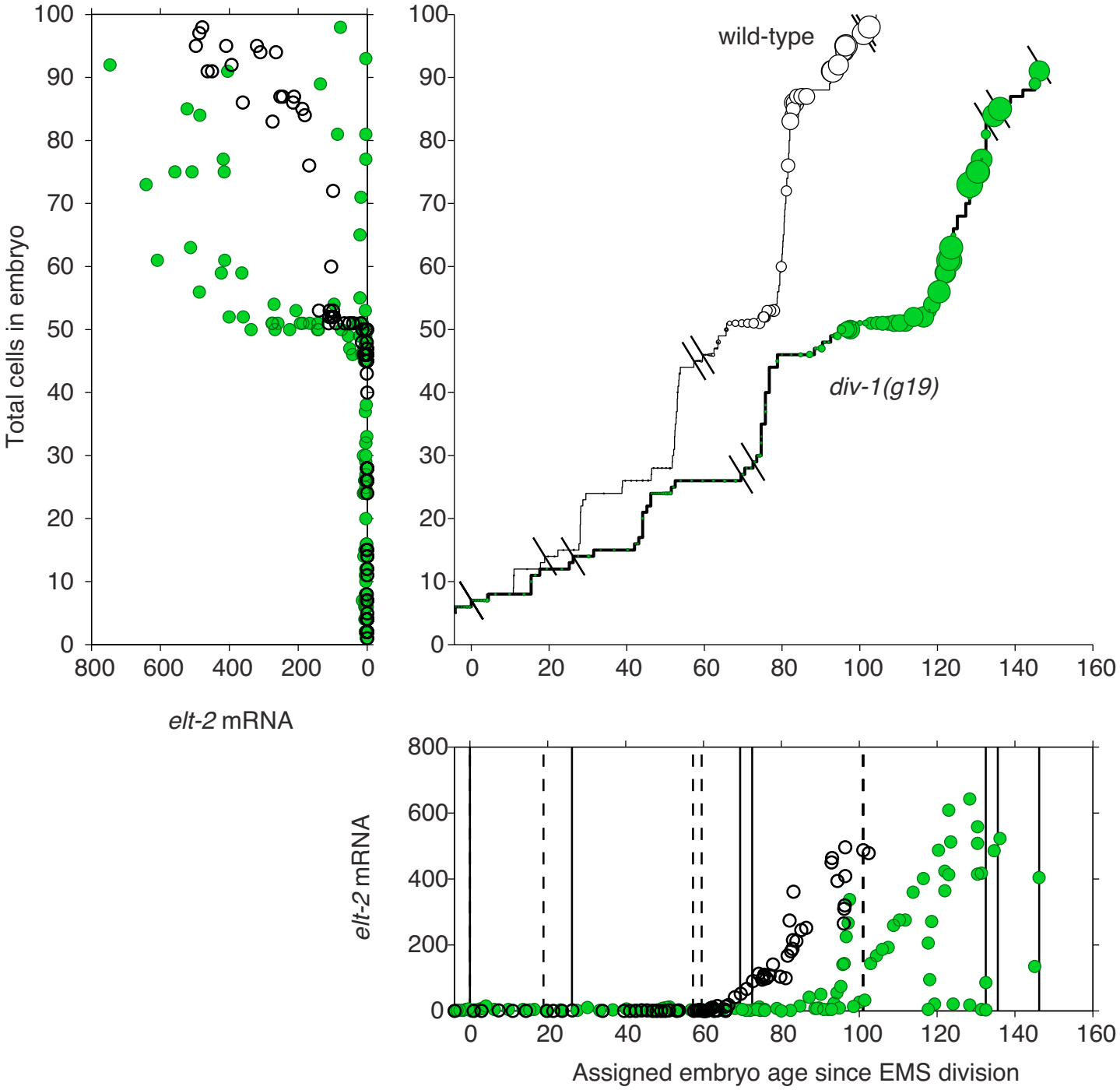

Fig. S14. *elt-2* expression in the *div-1(g19)* strain. In all panels, solid colored symbols correspond to the mutant strain and white or open symbols correspond to data from wild type. (Top left) Number of transcripts versus total number of cells in the embryo. (Bottom right) Number of transcripts versus assigned embryo age (in minutes) based on division times for the mutant at the restrictive temperature 25°C and wild-type division times scaled to the 25°C proliferation rate. Black solid and dashed lines indicate E lineage division events in mutant and wild type, respectively. (Top right) Expression data overlaid on the nuclei versus time trajectory for both strains. Each symbol corresponds to an embryo and the symbol area is proportional to mRNA levels. Solid diagonal marks are E lineage division events.

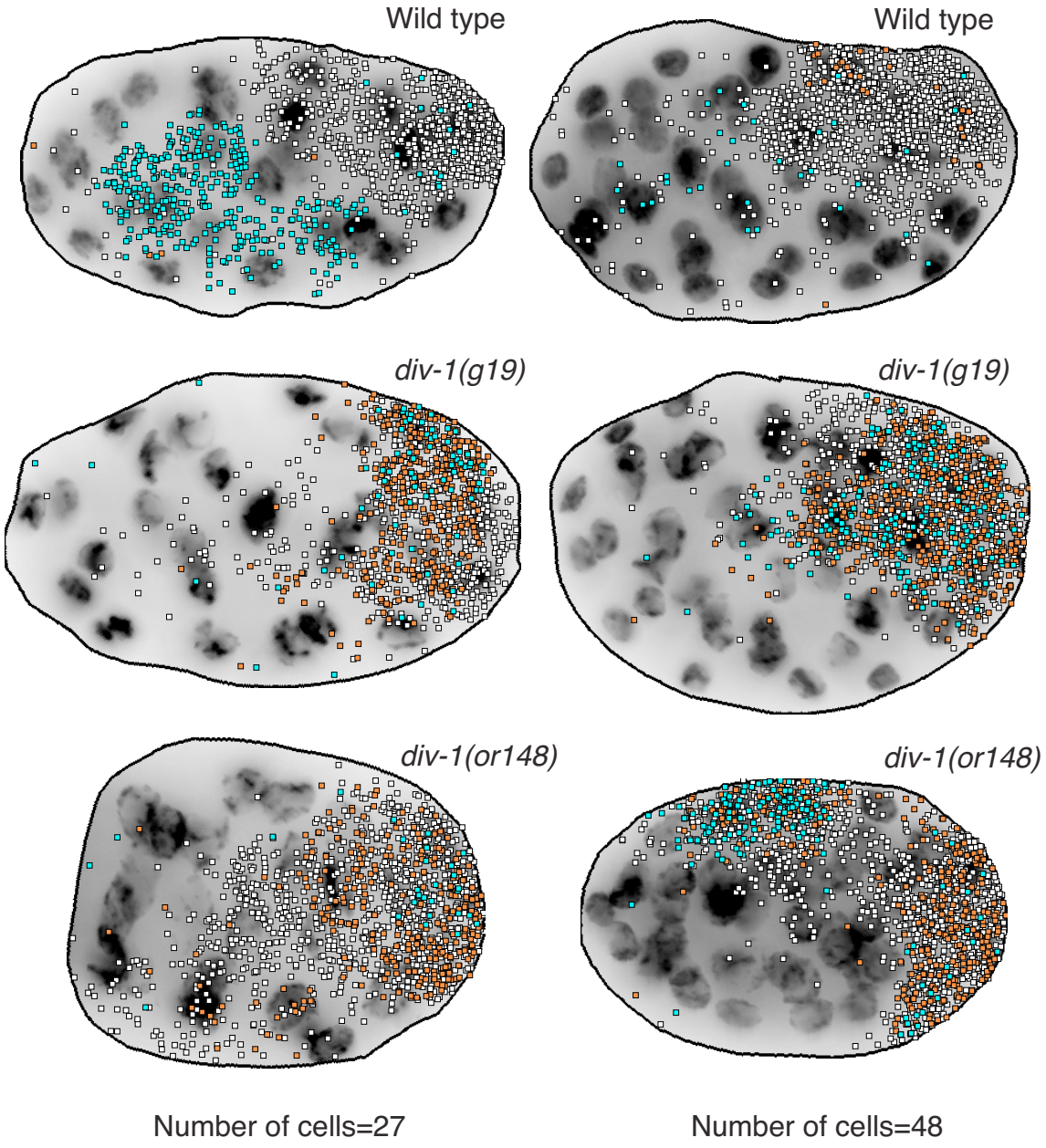

Fig. S15. Spatial pattern of muscle markers in N2 and *div-1* strains. Positions of individual *pal-1*, *unc-120* and *hlh-1* represented as white, orange and cyan squares, respectively, overlaid on a DAPI nuclear stain maximum projection in embryos with the same number of cells form wild-type, *div-1(g19)* and *div-1(or148)* strains. The *hlh-1* expression in early stage wild-type embryos has no known function (Krause, 1995).

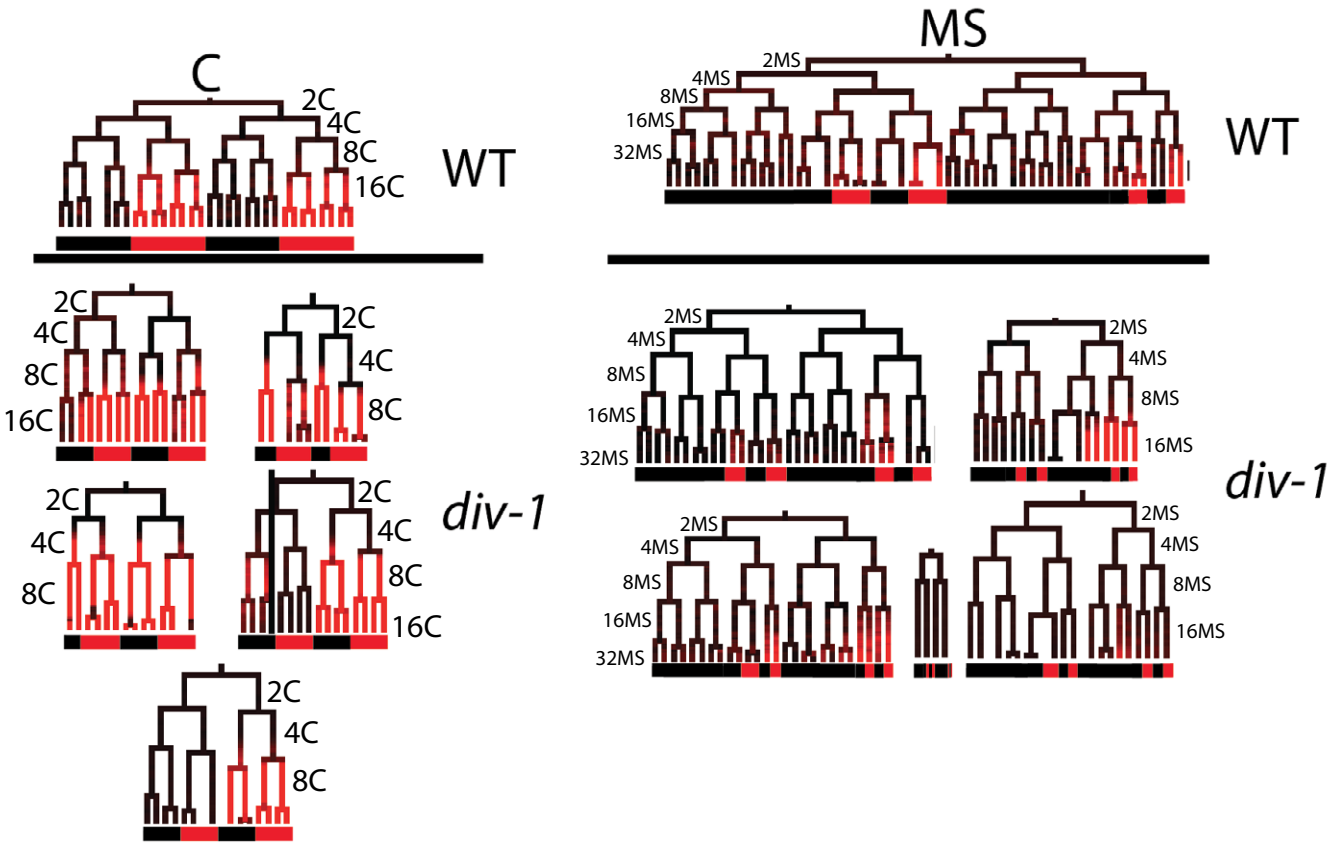

Fig. S16. *hlh-1* reporter expression in live N2 and *div-1(or148)* embryos. Lineages obtained by automated live imaging overlaid with a red color code indicating fluorescence intensity of an RFP *hlh-1* reporter. The horizontal bars below each of the five *div-1* lineages indicate the pattern that is found in wild type.

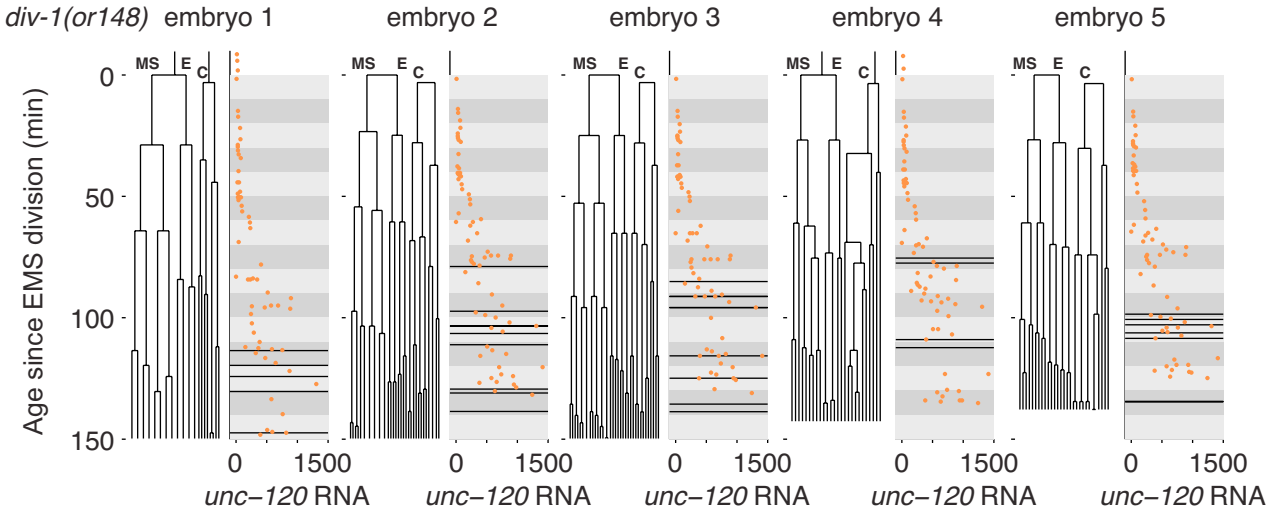

Fig. S17. Lineage variability in *div-1(or148)*. Excerpts of the embryonic lineage determined by automated live embryo imaging for five different *div-1(or148)* embryos at 25°C. The RNA time traces use the same underlying RNA FISH data set from a sample of fixed *div-1(or148)* 25°C embryos, but the assigned embryo ages and locations of the 4*MS*, 4*C*, 1*D*→8*MS*, 8*C*, 2*D* divisions are based on the five different embryo lineages obtained. The fact that *unc-120* onset precedes the divisions in this mutant is independent of which embryo is chosen as the representative lineage.

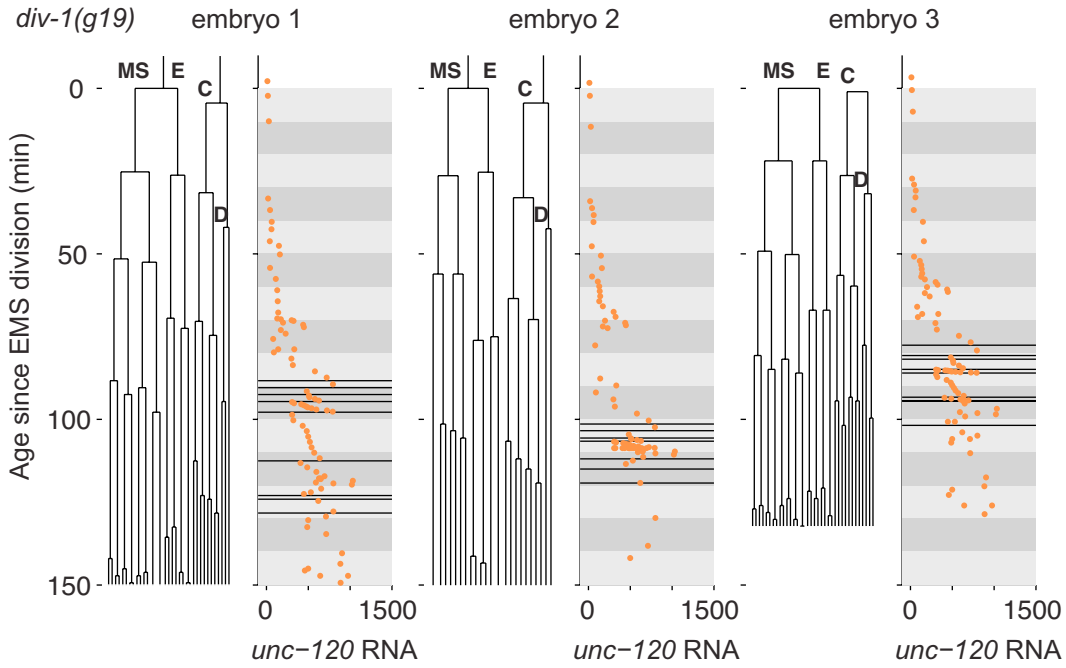

Fig. S18. Lineage variability in *div-1(g19)*. Excerpts of the embryonic lineage determined by automated live embryo imaging for three different *div-1(g19)* embryos at 25°C. The RNA time traces use the same underlying RNA FISH data set from a sample of fixed *div-1(g19)* 25°C embryos, but the assigned embryo ages and locations of the 4*MS*, 4*C*, 1*D*→8*MS*, 8*C*, 2*D* divisions are based on the three different embryo lineages obtained. The fact that *unc-120* onset precedes the divisions in this mutant is independent of which embryo is chosen as the representative lineage.

Table S1. Strains used and purpose in this study

Strain	Genotype	Purpose
MQ130	*clk-1(qm30)*III	Overall metabolic slowdown
GG52	*emb-29(g52)V*	Cell-cycle specific slowdown in E lineage
RB1331	*end-3(ok1448)*V	Gut gene timing
EU548	*div-1(or148)*III	Cell-cycle specific slowdown
GG19	*div-1(g19)*III	Cell-cycle specific slowdown
HH16	*emb-29(b262)unc-60(m35)V*	Whole genome sequencing only
VC271	*end-1(ok558)*V	DNS
GE31	*cib-1(e2300)*I	DNS: *elt-2* RNA is very low
GG39	*emb-23(g39)*II	DNS: *elt-2* RNA is very low
GG32	*emb-22(g32)*V	DNS: excessive gut cells
WM99	*cdk-1(ne2257)*III	DNS: ectopic, low *elt-2* RNA

DNS, data not shown