SUPPLEMENTARY INFORMATION

Both Nuclear Size and DNA Amount Contribute to Midblastula

Transition Timing in *Xenopus laevis*

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SUPPLEMENTARY FIGURE LEGENDS

**Supplementary Figure 1. GS17 in situ hybridization in embryos with altered
nuclear size and ploidy**

See Figure 2 legend for experimental approach.

(a) GS17 staining intensity was quantified in both halves of 7 hpf diploid and haploid
control embryos, and normalized to the diploid control. N = number of embryos. Error
bars represent SD. * p < 0.05.

(b) GS17 staining intensity was quantified in pre-MBT diploid and haploid embryo
halves with increased nuclear size, and normalized to the diploid with increased nuclear
size. N = number of embryos. Error bars represent SD. * p < 0.05.

(c) Haploid embryos were microinjected with mRNA to increase nuclear size. Cells in
4.5 hpf embryos that received the microinjected mRNA were identified by rhodamine
dextran fluorescence, and nuclear size was quantified for GS17-positive and GS17-
negative cells. The bottom panels show representative images of two cells that received
the microinjected mRNA and showed differential GS17 staining and nuclear sizes. N =
number of cells. Error bars represent SD. *** p <0.001. Scale bar, 20 μm.

(d) Diploid and haploid embryos were microinjected with mRNA to increase nuclear
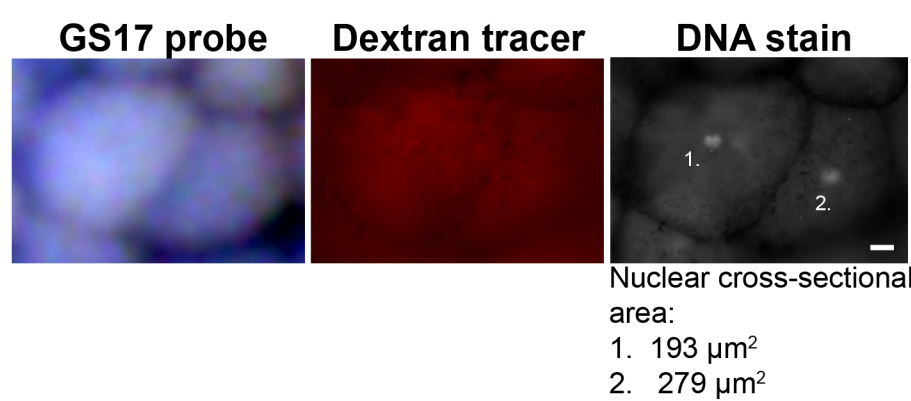size. Cells in 4.5 hpf embryos that were positive for GS17 staining and exhibited similar
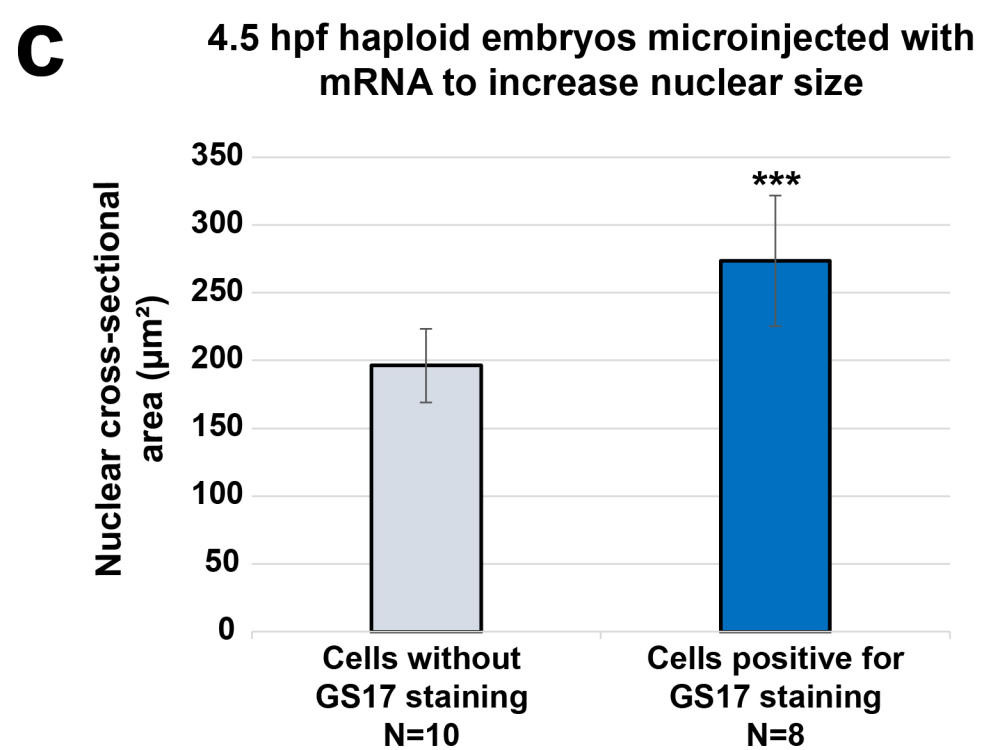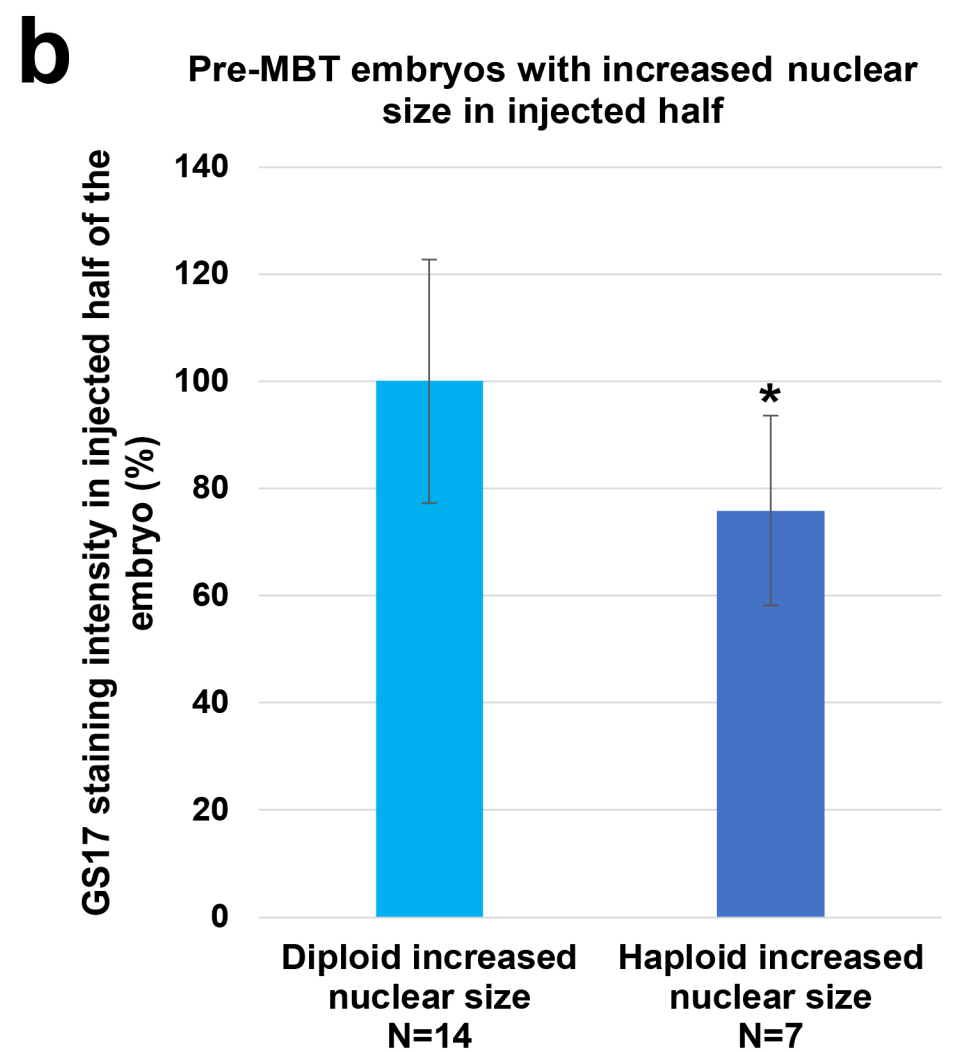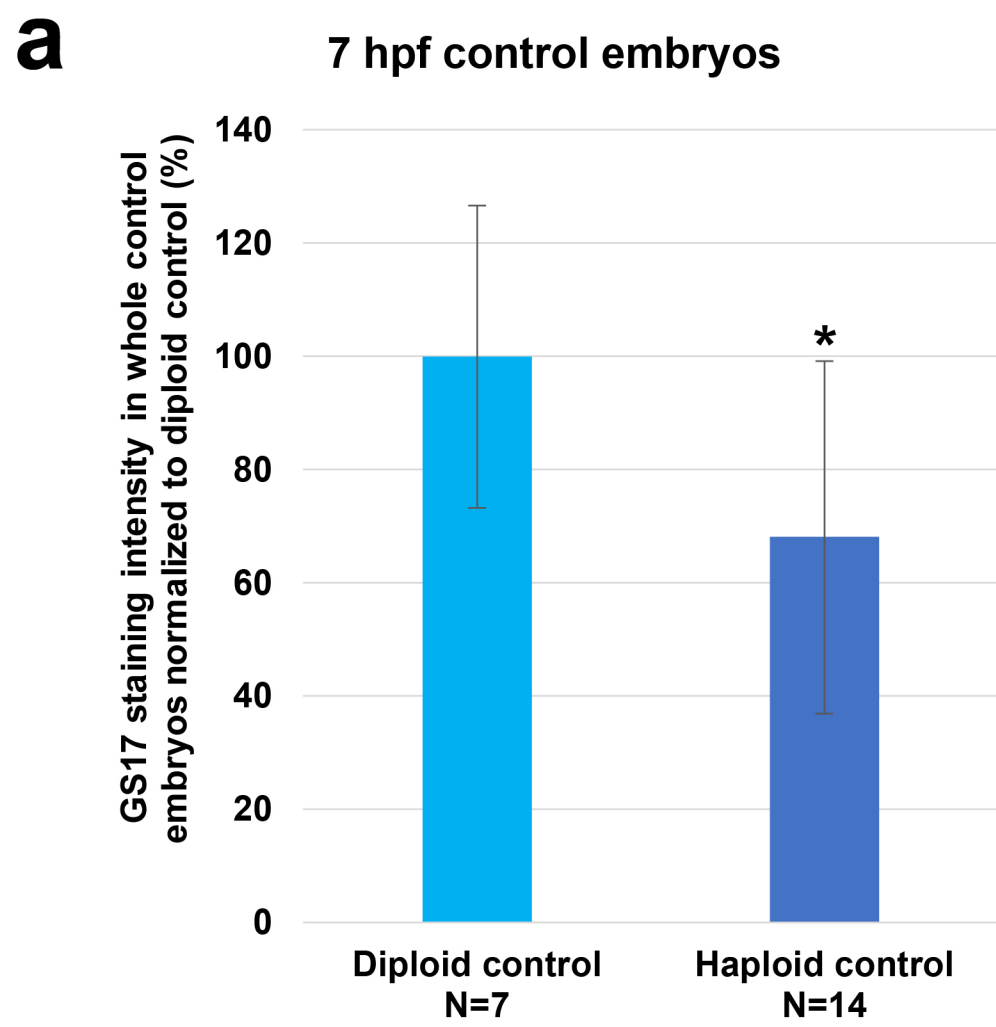
nuclear sizes were analyzed. The top graph shows average nuclear sizes, and the

bottom graph shows average GS17 staining intensity for those same cells, normalized

to the diploid. N = number of cells. Error bars represent SD. * $p < 0.05$; NS = not

significant.

(e) Comparisons were performed between diploid control embryos and haploid embryos

microinjected with mRNA to increase nuclear size. Cells in 7 hpf embryos that were

positive for GS17 staining and exhibited similar nuclear sizes were analyzed. The graph

on the left shows average nuclear sizes, and the graph on the right shows average

GS17 staining intensity for those same cells, normalized to the diploid control. N =

number of cells. Error bars represent SD. * $p < 0.05$; NS = not significant.

**Supplementary Figure 2. Cell cycle timing in embryos with altered nuclear size
and ploidy**

See Figure 4 legend for experimental approach. Raw data used to generate Figure 4B

are plotted as bar graphs with each bar representing the cell cycle length of an
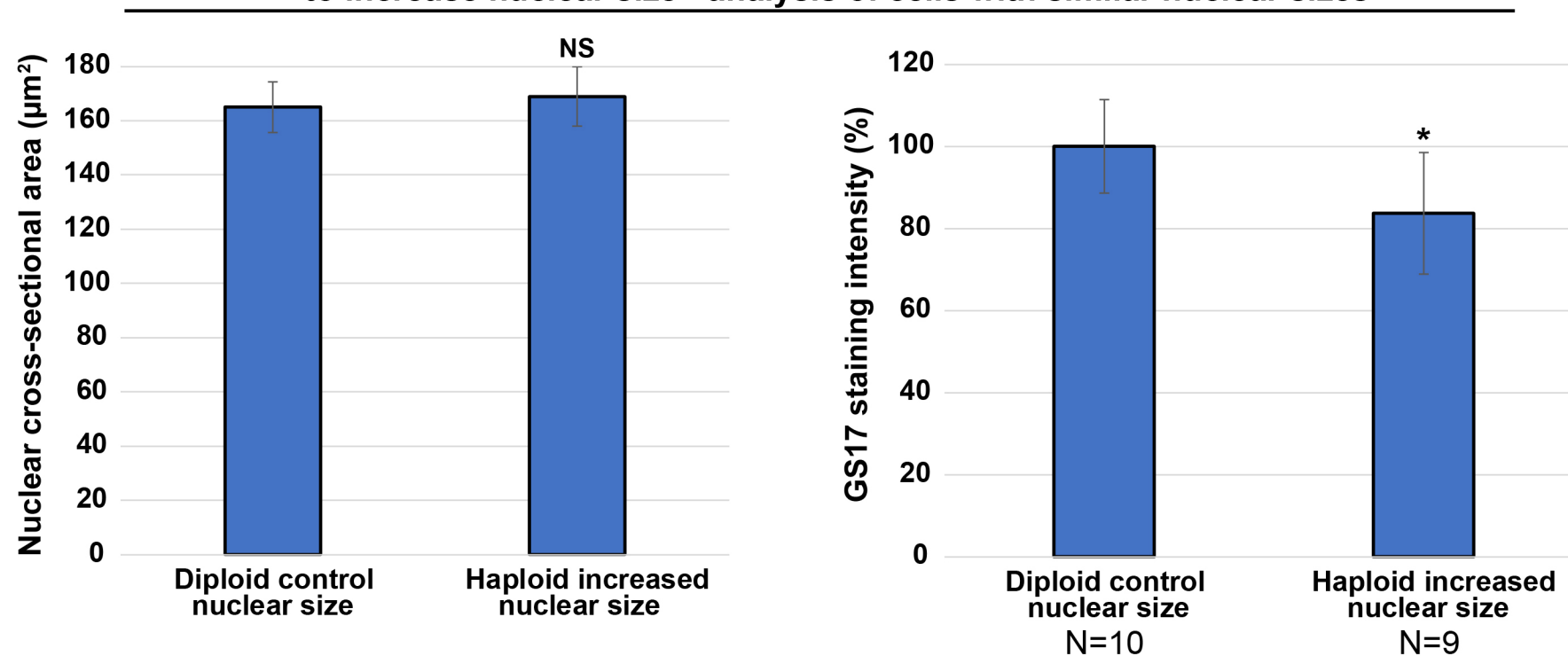
individual cell.

Supplemental Figure 1

a 7 hpf control embryos

b Pre-MBT embryos with increased nuclear size in injected half

c 4.5 hpf haploid embryos microinjected with mRNA to increase nuclear size

d 4.5 hpf diploid and haploid embryos microinjected with mRNA to increase nuclear size - analysis of cells with similar nuclear sizes
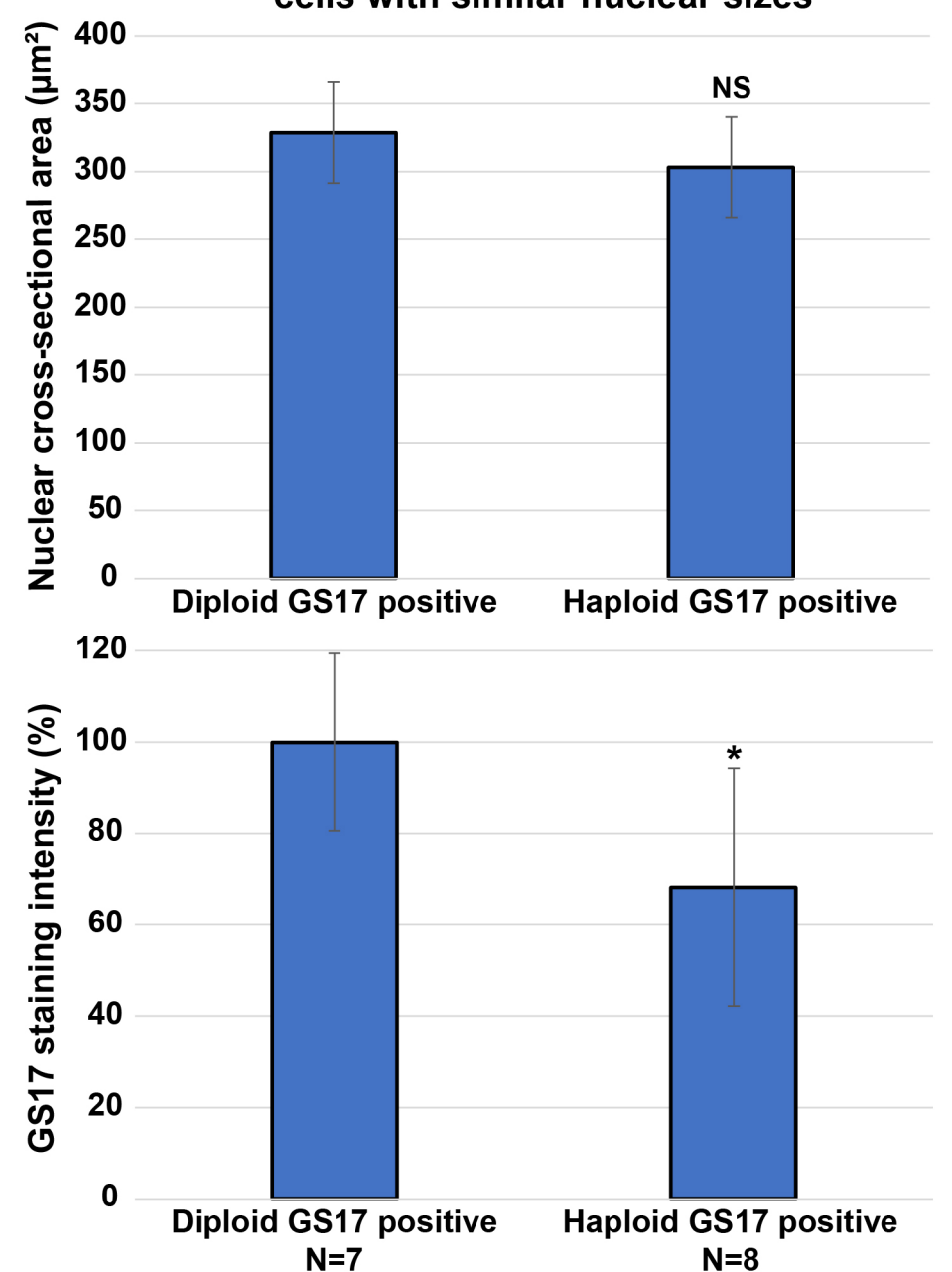
e 7 hpf diploid control embryos and haploid embryos microinjected with mRNA to increase nuclear size - analysis of cells with similar nuclear sizes

Supplemental Figure 2