Supplemental Figure 1



Supplemental Fig. S1. Expression of eGFP-H2B during pre-implantation development

Complementary RNA (cRNA) encoding eGFP-tagged H2B (eGFP-H2B) was injected into the cytoplasm of 1-cell stage embryos 2 h post-insemination (hpi). Embryos at the 1-, 2-, 4-, morula, and blastocyst stages were collected 13, 30, 48, 72, and 96 hpi, respectively. DNA was stained with 4', 6-diamino-phenylindole (DAPI). The area enclosed in the white rectangle is magnified and shown beneath the corresponding image. Euchromatin and heterochromatin regions are marked by white arrows and arrowheads, respectively. Asterisks indicate nucleolar precursor bodies. Bar = $10 \mu m$

Supplemental Figure. S2



Supplemental Fig. S2. Assessment of the unbound fraction of eGFP-H2B in 1- and 2-cell stage embryos.

cRNA encoding eGFP-H2B or eGFP was injected into the cytoplasm of embryos 2 h post-insemination (hpi). One-cell (**A**) and 2-cell (**B**) stage embryos were collected at 10 and 30 hpi, respectively. After the unbound fraction of eGFP or eGFP-H2B was washed away using 0.2% Triton X-100 (see Materials and Methods), the embryos were fixed and observed for fluorescence. A single experiment, in which at least 5 embryos were examined in each group, was conducted. Representative images are shown. Since there was no embryo in which male and female pronuclei were located at a single focal plane, two images were taken for each embryo at the plane at which the male or female pronuclei was clearly observed. Bar = 20 μ m.

Supplemental Figure. S3



Supplemental Fig. S3. Mobility of eGFP-H2B in the nucleolar precursor body (NPB)

cRNA encoding eGFP-H2B was injected into the cytoplasm of 1-cell stage embryos 2 h post-insemination (hpi). Mobility of eGFP-H2B at the NPB and euchromatin region in the nucleus of 1-cell stage embryos was examined using FRAP between 10-13 hpi. The recovery curve and mobile fraction are shown on the left and right, respectively. Three independent experiments were performed and the data were accumulated. In total, more than 17 pronuclei were examined. Bar = $10 \mu m$

Supplemental Figure. S4



Supplemental Fig. S4. Mobility of eGFP-H2B in euchromatin and heterochromatin regions.

cRNA encoding eGFP-H2B was injected into the cytoplasm of 1-cell stage embryos 2 h post insemination (hpi). Embryos at the 4-cell stage were collected 45 hpi. The mobility of eGFP-H2B in the regions of interest (ROIs; red quadrangle), which were randomly selected in chromatin (10.5 μ m²) (upper panel) and in euchromatin (middle panel) and heterochromatin (lower panel) (3.4 μ m² each) were analyzed using FRAP. The representative images taken before bleaching (left), soon after bleaching (middle), and after recovery (taken 150 s after bleaching; right) are shown. Bar = 10 μ m