

Fig. S1. Loss of *nanog* induced apoptosis in oocyte and early embryo. (A) TUNEL staining of ovary sections of WT and *nanog*<sup>-/-</sup> adult fish. Obvious apoptotic signals are observed in Balbiani bodies and mitochondria in *nanog*<sup>-/-</sup> (c,d), but not WT (a,b). a' and b', enlarged regions of a and b; c' and d', enlarged regions of c and d. Scale bar, 100μm. (B) Immunostaining of active-Caspase3 in WT and M*nanog* embryos at 75% epiboly. Robust active-caspase3 expression was detected in M*nanog* embryo. Scale bar, 100 μm. (C) Detection of Nanog expression in ovary of WT, *Tg*(CMV:*nanog-myc*) with WT background, and *Tg*(CMV:*nanog-myc*) with *nanog*<sup>-/-</sup> background using anti-Myc antibody. DAPI was co-stained for DNA. Scale bar, 50μm. (D) Phenotype comparison of WT, WT, *Tg*(CMV:*nanog-myc*), M*nanog* and M*nanog*, *Tg*(CMV:*nanog-myc*) embryos at 8 and 24 hpf. Scale bar, 100 μm.

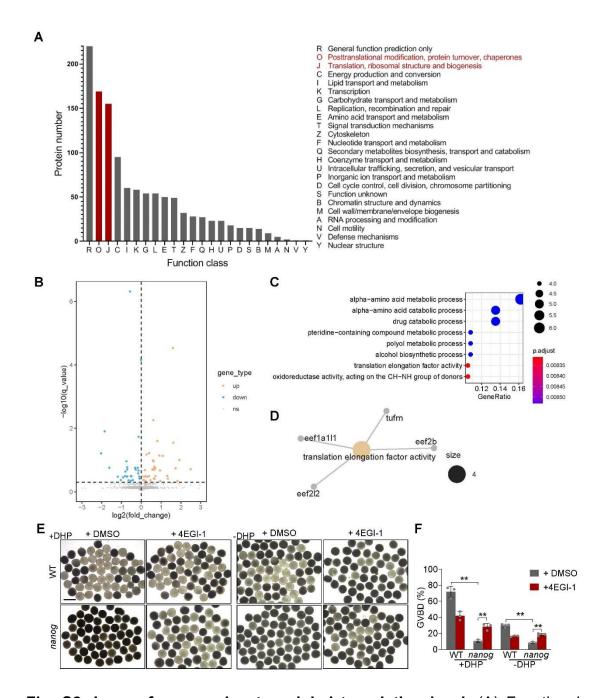
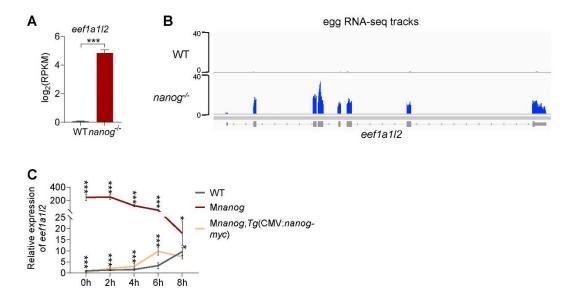
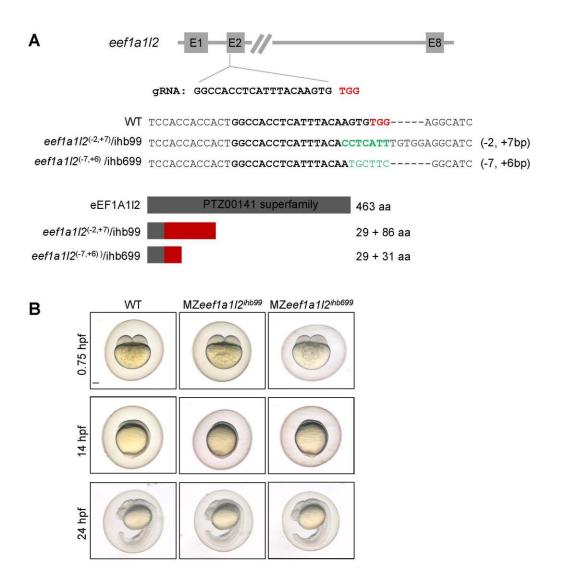


Fig. S2. Loss of *nanog* elevates global translation level. (A) Functional classification of the proteins identified in WT and *nanog* mutant eggs. The two most significant enriched categories which are related with translation were highlighted in red. (B) Volcano plot of upregulated and down-regulated proteins in *nanog* mutant egg. (C) GO analysis of upregulated proteins of *nanog* mutant. (D) Genes of translation elongation factors were enriched by Gene-Concept

Network analysis. (E) Morphology of stage IV follicles dissected from WT and *nanog*<sup>-/-</sup> ovaries and treated with or without 4EGI-1 (25ng/μL) in the present or absent of DHP after 2h incubation. 1μg/mL DHP was added to promote oocyte maturation. Scale bar, 1mm. (F) %GVBD comparison in WT and *nanog* mutant follicles treated with 4EGI-1. \*\**P*<0.01.



**Fig. S3.** eef1a1l2 transcription level is significantly upregulated in *nanog* mutant. (A) FPKM values of *eef1a1l2* expression in WT and *nanog* mutant eggs. \*\*\*P<0.001. (B) RNA-Seq reads mapped to *eef1a1l2* gene. Coverage tracks are displayed for WT and *nanog* mutant egg. (C) RT-qPCR analysis of *eef1a1l2* expression in WT and *Mnanog* embryos at 0, 2, 4, 6, 8 hpf. \*P<0.05, \*\*\*P<0.001.



**Fig. S4.** Generation of the *eef1a1l2* mutant allele using CRISPR/Cas9. (A) Top: the gRNA target site within the exon 2. Grey boxes and connecting lines represent the exons and introns, respectively. Middle: sequence of WT and *eef1a1l2* mutant alleles near the gRNA target site (bold). PAM sequence is in red, and the insert sequence in mutant alleles is in green. Two types of mutants, (-7, +6)/ihb99 and (-2, +7)/ihb699 were screened. Bottom: domain of eEF1A1l2 protein and predicted mutant protein. Grey box indicates truncated WT eEF1A1l2 protein, and red box indicate mutant protein by frameshift. (B) Phenotype of WT, MZ*eef1a1l2* ihb99, and MZ*eef1a1l2* ihb699 embryos at 0.75, 14, and 24 hours post- fertilization (hpf). Scale bar, 100 μm.

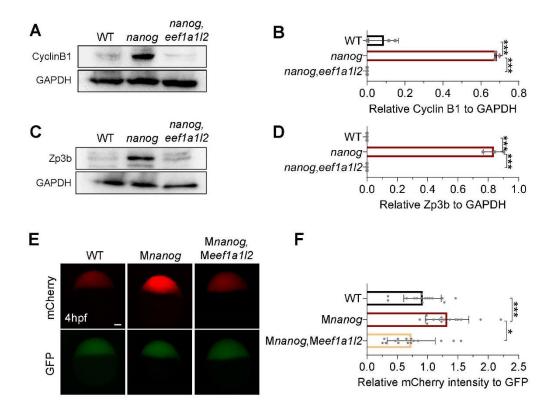


Fig. S5. Translation level evaluation of nanog and eef1a1/2 double mutant.

(A) Western blot analysis of Cyclin B1 in WT, *nanog* mutant, and *nanog*,*eef1a1l2* double mutant follicles at stage I/II. (B) Comparison of Cyclin B1 intensity in panel A. \*\*\**P*<0.001. (C) Western blot analysis of Zp3b in WT, *nanog* mutant, and *nanog*,*eef1a1l2* double mutant follicles at stage I/II. (D) Comparison of Zp3b intensity in panel B. \*\*\**P*<0.001. (E) Fluorescent images showing mCherry reporter levels with GFP protein control levels in WT, *Mnanog* and *Mnanog*,*Meef1a1l2* embryos at 4hpf. Scale bar, 100μm. (F) Measurement of mCherry reporter intensities relative to GFP in panel A. \*\**P*<0.01. n=15.

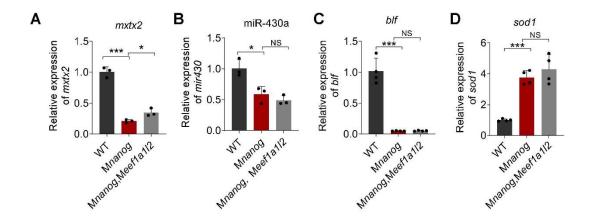


Fig. S6. Phenotype analysis of *nanog* and *eef1a1l2* double mutant. (A-D)

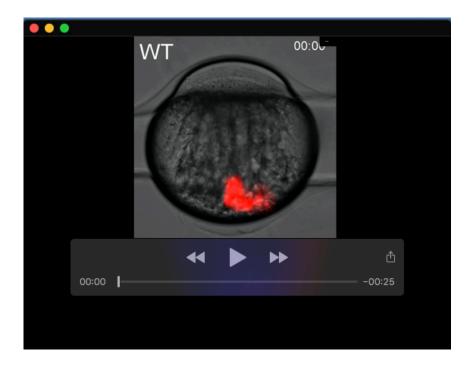
RT-PCR analysis of *mxtx2*, miR-430, *blf* and *sod1* in WT, Mnanog and Mnanog,Meef1a1l2 embryos. *mxtx2* and miR-430a were analyzed at 4 hpf, *blf* and *sod1* were analyzed at 6 hpf. \**P*<0.05, \*\*\**P*<0.001. NS means no significant difference.

Table S1. Differential proteins between nanog mutant and WT eggs.

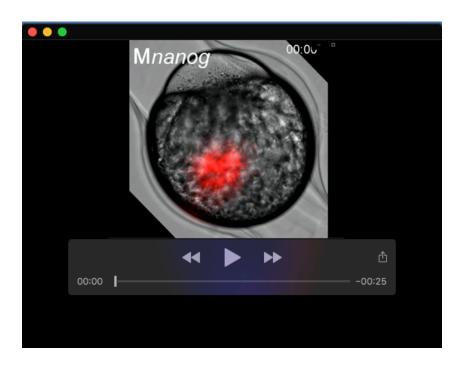
Click here to download Table S1

Table S2. Primers used in qRT-PCR and mutant screening.

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**Movie 1. Cytoplasmic movement in a WT embryo.** CM-Dil dye was injected at 20 mpf and embryos were imaged from 35 mpf to 55 mpf. The cytoplasmic streaming that transports CM-Dil dye to the animal pole can be visualized. 10 embryos were observed and this movie shows the representative result.



**Movie 2. Cytoplasmic movement in an M***nanog* **embryo.** CM-Dil dye was injected at 20 mpf and embryos were imaged from 40 mpf to 60 mpf. The CM-Dil dye remains stagnant in the yolk until 60 mpf, indicating cytoplasmic movement defect in M*nanog*. 10 embryos were observed and this movie shows the representative result.



**Movie 3. Cytoplasmic movement in an M***nanog*,**Meef1a1l2 embryo**. CM-Dil dye was injected at 20 mpf and embryos were imaged from 40 mpf to 60 mpf. The CM-Dil dye was continuously transported to the animal pole. 15 embryos were observed and this movie shows the representative result.