

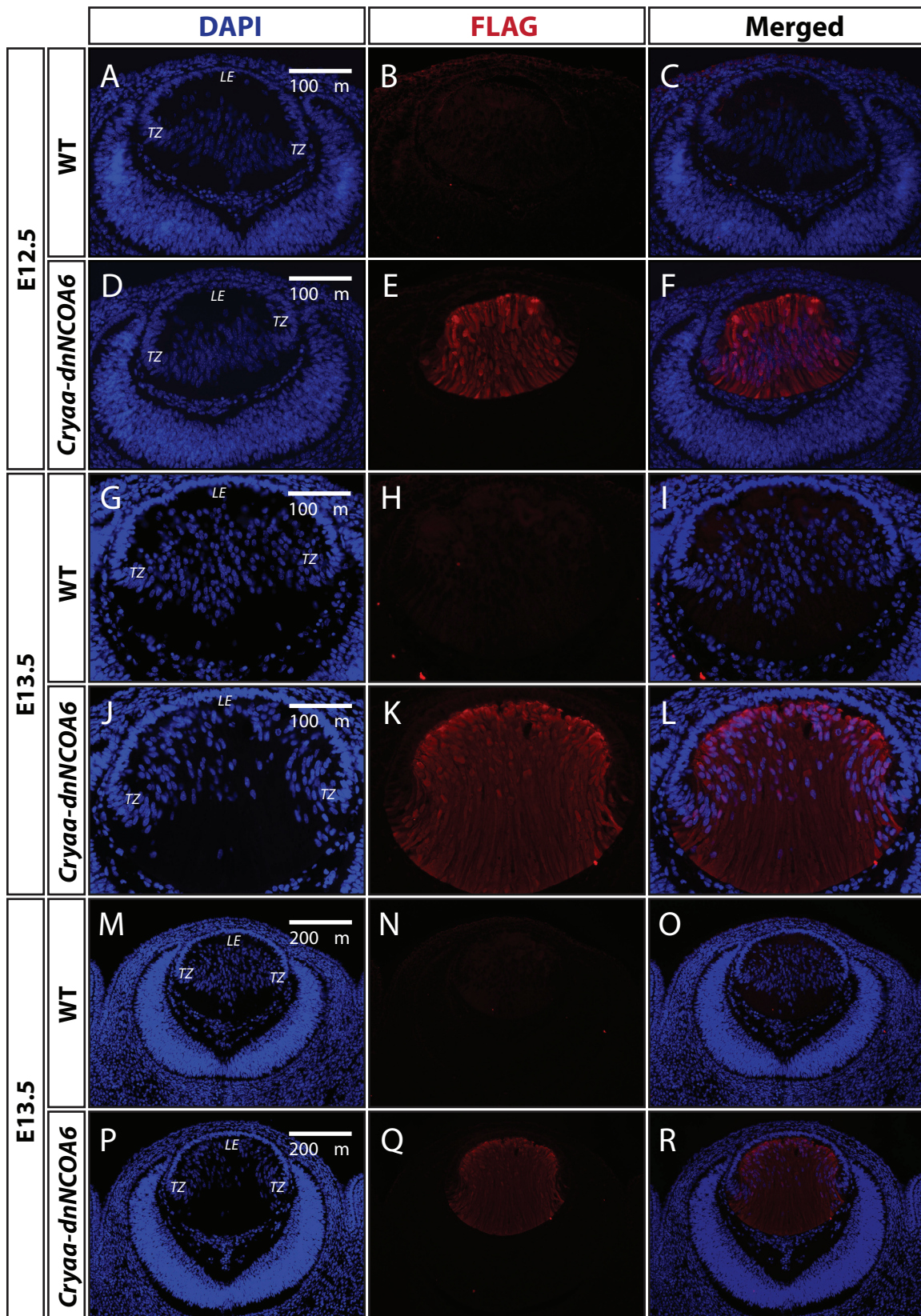
## SUPPLEMENTAL FIGURE LEGENDS

**Supplemental Figure S1. The onset and localization of the lens-specific *Cryaa-dnNCOA6* transgene.** (A-F) Expression of FLAG-dnNCOA6 (red signal) in E12.5 transgenic lens. (G-L) Expression of FLAG-dnNCOA6 in E13.5 transgenic lens. (M-R) The lens-specific expression of *dnNCOA6* transgene in E13.5 lenses. Nuclei (blue signal) were stained with DAPI. Lens epithelium, LE; transitional zone, TZ. Scale bar is shown in panels A, D, G, J and M.

**Supplemental Figure S2. Expression of NCOA6 proteins in WT, *Cryaa-dnNCOA6* transgenic mice and *Ncoa6* null embryos.** (A) Western blot analysis of WT and transgenic (AD5 and AD9) lens extracts from neonatal lenses to detect endogenous NCOA6 protein expression. (B) Western blot analysis of NCOA6 in extracts prepared from E11.5 embryos (heterozygous, +/-; null, -/-; WT, +/+) to demonstrate specificity of the NCOA6 antibody.

**Supplemental Figure S3. DSBs and DNA-PKcs activation in *Cryaa-dnNCOA6* lenses.**  $\gamma$ -H2AX antibody was used to label DSBs and DNA-PKcs T2609 antibody was used to label activated DNA-PKcs generated through DNA damage or normal denucleation process. DSBs and activated DNA-PKcs were detected as early as E13.5 only in the transgenic lens fiber cell compartment (E-H), whereas the WT lens was negative (A-D). Usually activated DNA-PKcs and DSBs signals overlap and the number of DSBs positive nuclei is higher. In the neonatal WT lens, fibers at the margin of the OFZ show  $\gamma$ -H2AX staining and some nuclei are also positive for DNA-PKcs T2609 (I-L and I'-L'). In contrast, fiber cells of neonatal transgenic animals show a dispersed pattern of DSBs and activated DNA-PKcs signals in the lens fiber cells (M-P and M'-P'). The OFZ is indicated by dashed circle. Nuclei (blue signal) were stained with DAPI. Non-specific signal outside of the lens is explained in the figure legend of Figure 6. Lens epithelium, LE; transitional zone, TZ. Scale bar is indicated for each panel.

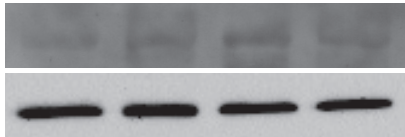
**Supplemental Figure S4. Identification of proliferating cells in WT and transgenic lenses.** (A) Immunofluorescence analysis of G2/M cells using phospho-histone H3 (red signal) antibody. Nuclei (blue signal) were stained with DAPI. (B) Immunohistochemical analysis of cells in S phase using Ki-67 antibody (brown). Nuclei (blue) were counterstained with hematoxylin. Lens epithelium, LE; transitional zone, TZ.



Supplymental Figure S1.

**A****Neonate Lens****WT AD5 WT AD9**

NCOA6

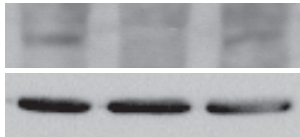
 $\beta$ -actin**B****E11.5 Embryo***Ncoa6*

+/-

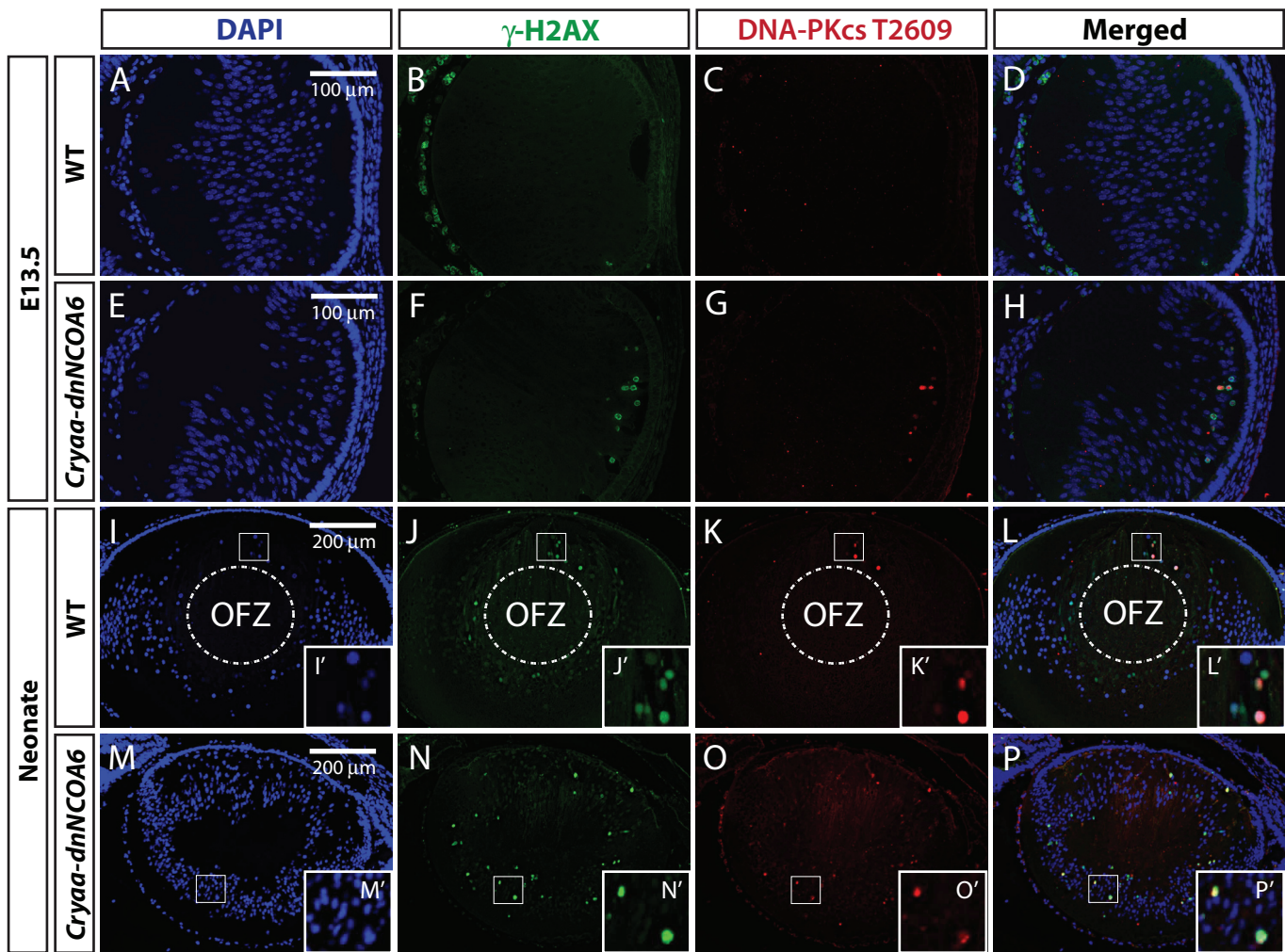
-/-

+/+

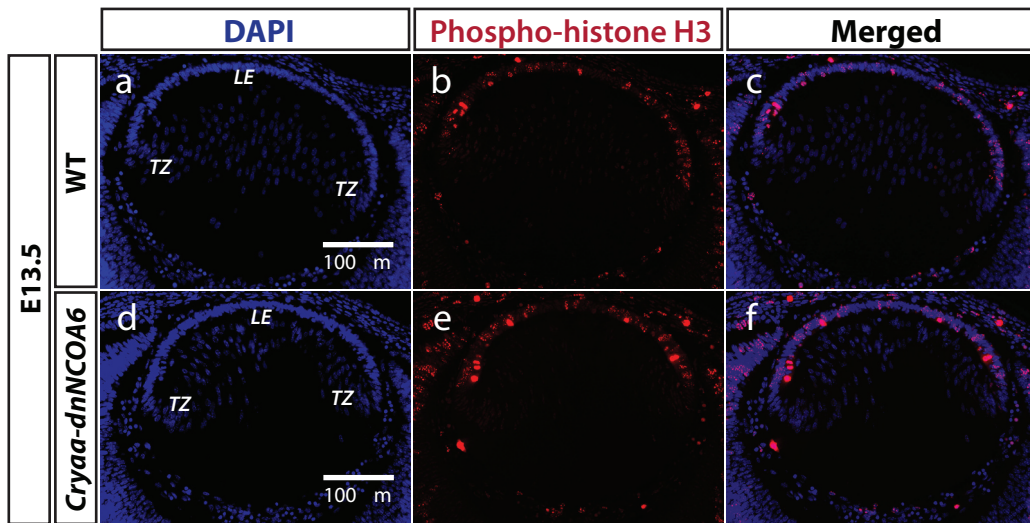
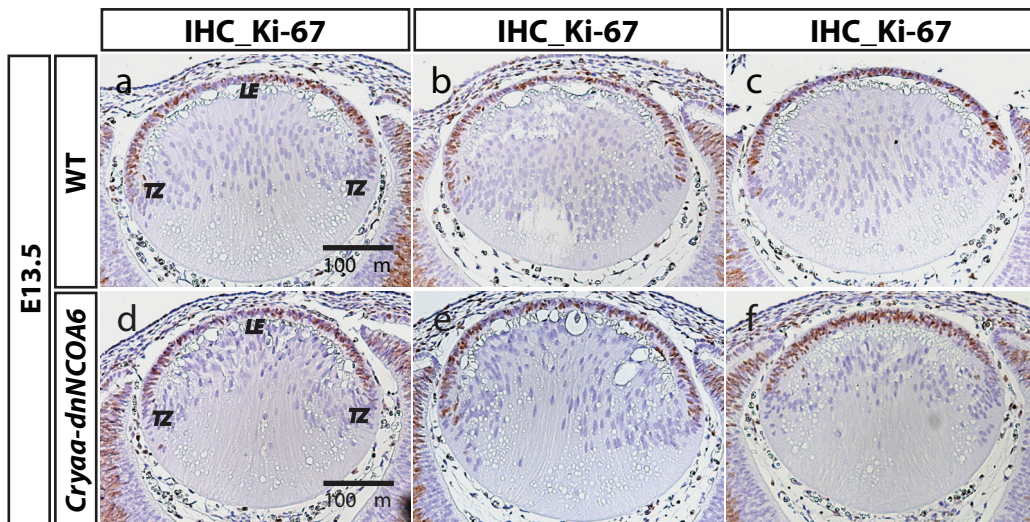
← ~230 kDa →



Supplemental Figure S2.



Supplemental Figure S3.

**A****B**

Supplemental Figure S4.