



Figure S3

**Figure S3. CTSD KO ARPE-19 cell line validation and AFG formation.** (A) Schematic representation of the CTSD<sup>-/-</sup> CRISPR/Cas9 strategy targeting at CTSD exon 4. The scheme represents the wild-type endogenous human chromosome sequence. The sgRNA target sequence is highlighted in light grey and the PAM sequence is in red, on the fourth exon. (B) CTSD protein levels in ARPE-19 wild-type (WT) and mutant 19-CTSD-KO (CTSD<sup>-/-</sup>) cells assessed by Western blot. (C) Representative confocal immunofluorescence microscopy images of AFG (green) in ARPE-19 wild type and 19-CTSD-KO cells pulsed with POS and chased for different time periods. Staining with an anti-ZO-1 antibody was used to monitor cell limits and a Z-projection is shown. Images are representative of three independent experiments, where at least 10 random fields were analyzed per assay. Scale bar: 20 μm.