**Table S1: Backgrounds and references for MEF cells used in this study.** The MEF genotypes, backgrounds of mice used to generate each MEF line, and references reporting these reagents are shown.

| Genotype | Reference | Mouse Strain |
|---|---|---|
| eIF2α mutants (SS and AA) | Scheuner et al. 2001 | 129/Sv |
| *PERK*$^{+/+}$ and *Perk*$^{-/-}$ | Zhang et al. 2002 | 129/Sv |
| *PKR*$^{+/+}$ and *Pkr*$^{-/-}$ | Abraham et al. 1999 | 129/Ter |
| *HRI*$^{+/+}$ and *Hri*$^{-/-}$ | Han et al. 2001 | 129/Bl6 mixed |
| *GCN2*$^{+/+}$ and *Gcn2*$^{-/-}$ | Zhang et al. 2002 | 129/Sv |

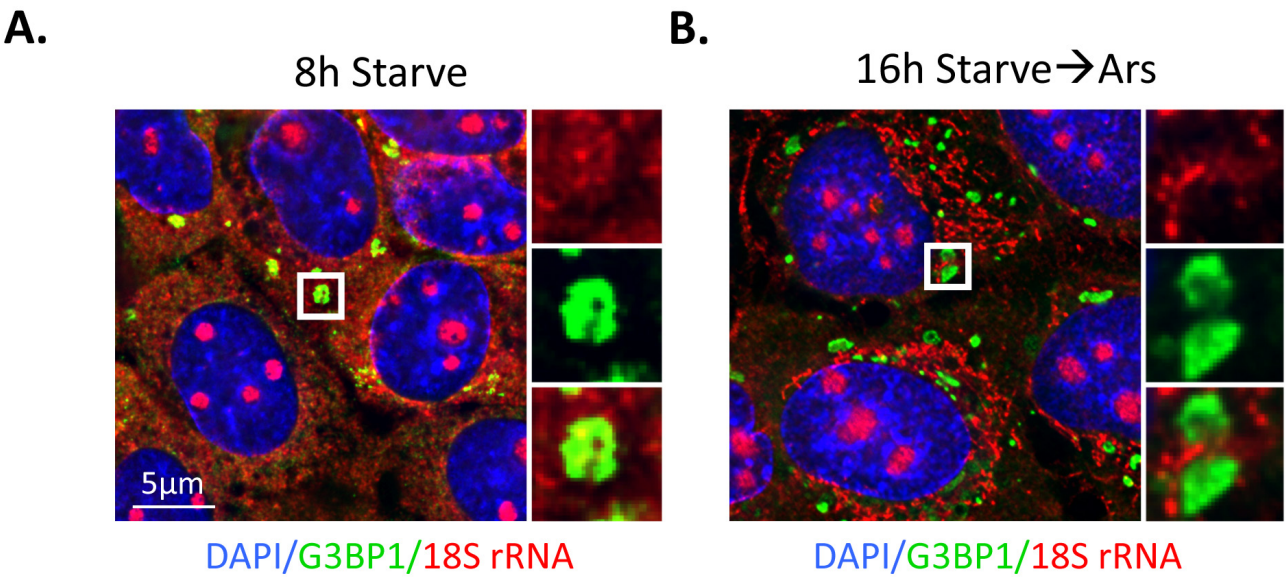


**Figure S1: 18S rRNA does not concentrate in stSG.** *A,* U2OS cells were starved for 8 h prior to fixation and staining against G3BP1 (green) and 18Ss rRNA (red). *B,* U2OS cells were starved for 16 h and arsenite was added at 250 µM for the last hour. Cells were stained as in A.

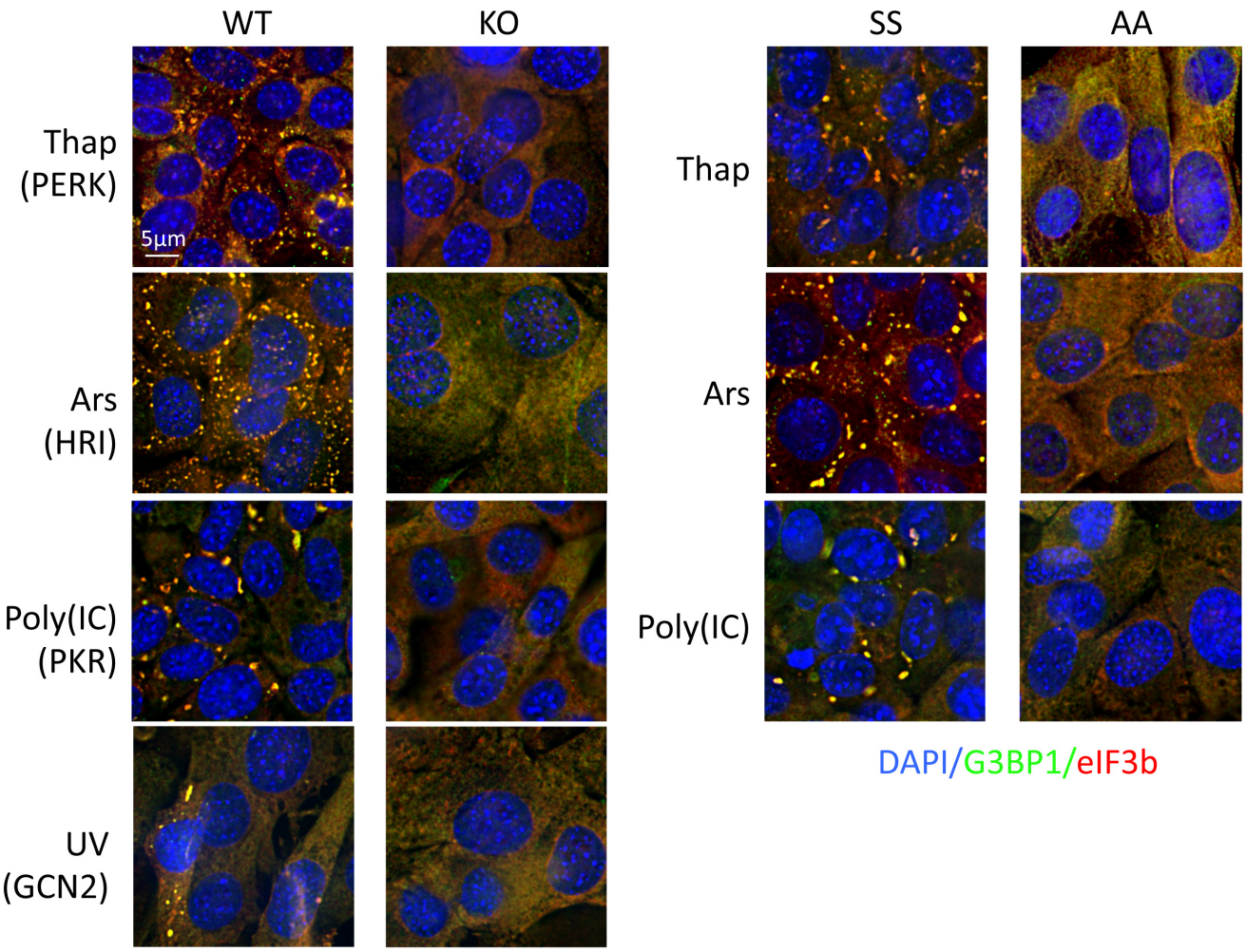


**Figure S2: EIF2AK wild type and knockout MEFs were characterized to confirm they respond in accordance with known properties reflecting genotype.** Each EIF2AK wild type and KO MEF pair was exposed to the indicated stress (2µM thapsigargin for PERK and S51A MEFs, 2h; 250 µM arsenite, 1h for HRI and S51A MEFs; transfection of 100ng poly(IC), 6h for PKR and S51A MEFs; or 200J/m2 UV, 2 h recovery for GCN2 MEFs) followed by staining for G3BP1 (green) and eIF3b (red) to mark SG. Several fields of cells per condition were acquired, and representative images are shown.

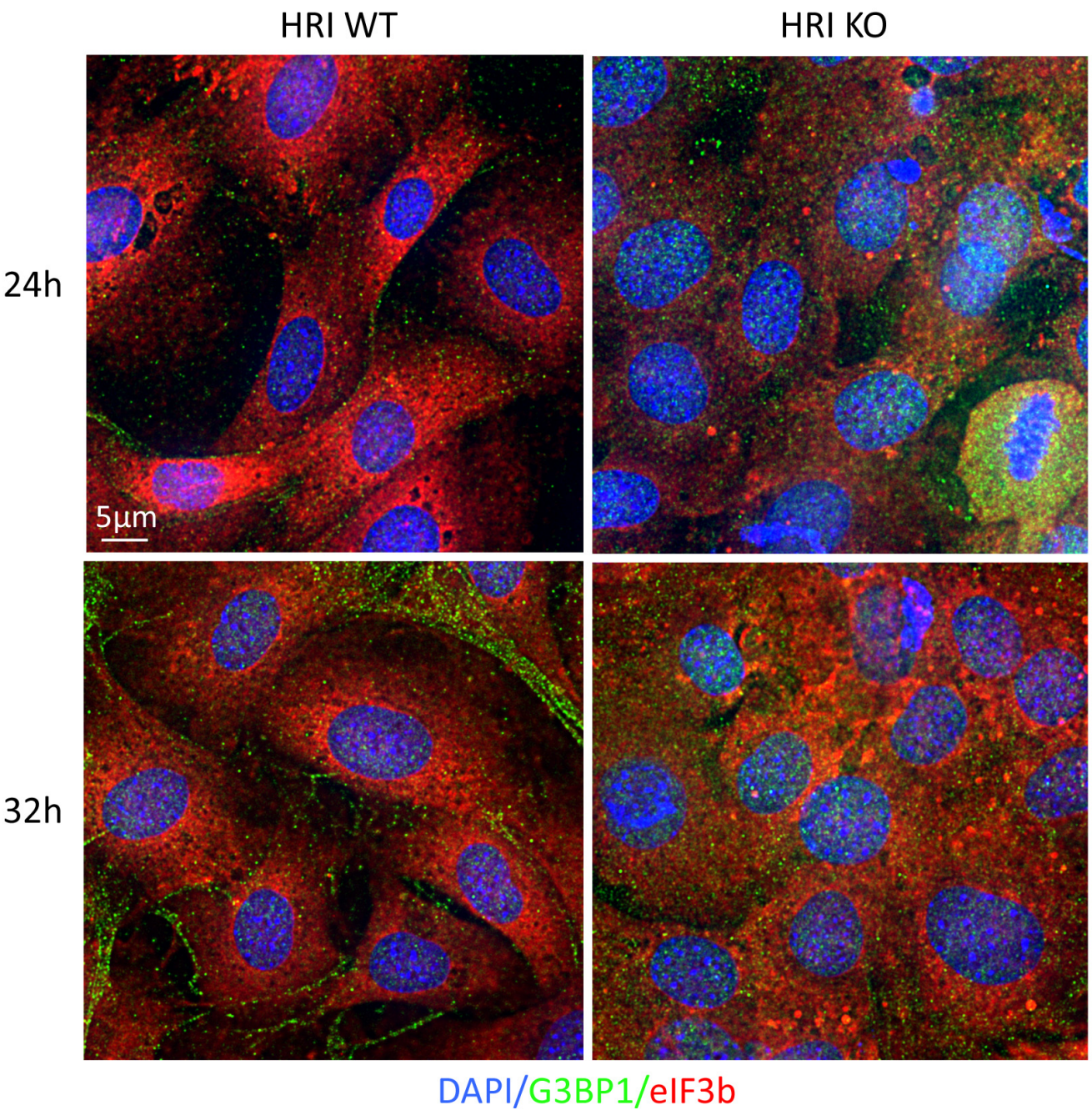


**Figure S3: HRI wild type and knockout MEFs are intrinsically resistant to SG induction.** HRI wt and KO MEFs were stained for the stSG markers G3BP1 (green) and eIF3b (red) at 24 and 32h of starvation. No stSG are visible. Red puncta represent some type of puncta that are extracellular as indicated by staining outside of the cell area across multiple fields.

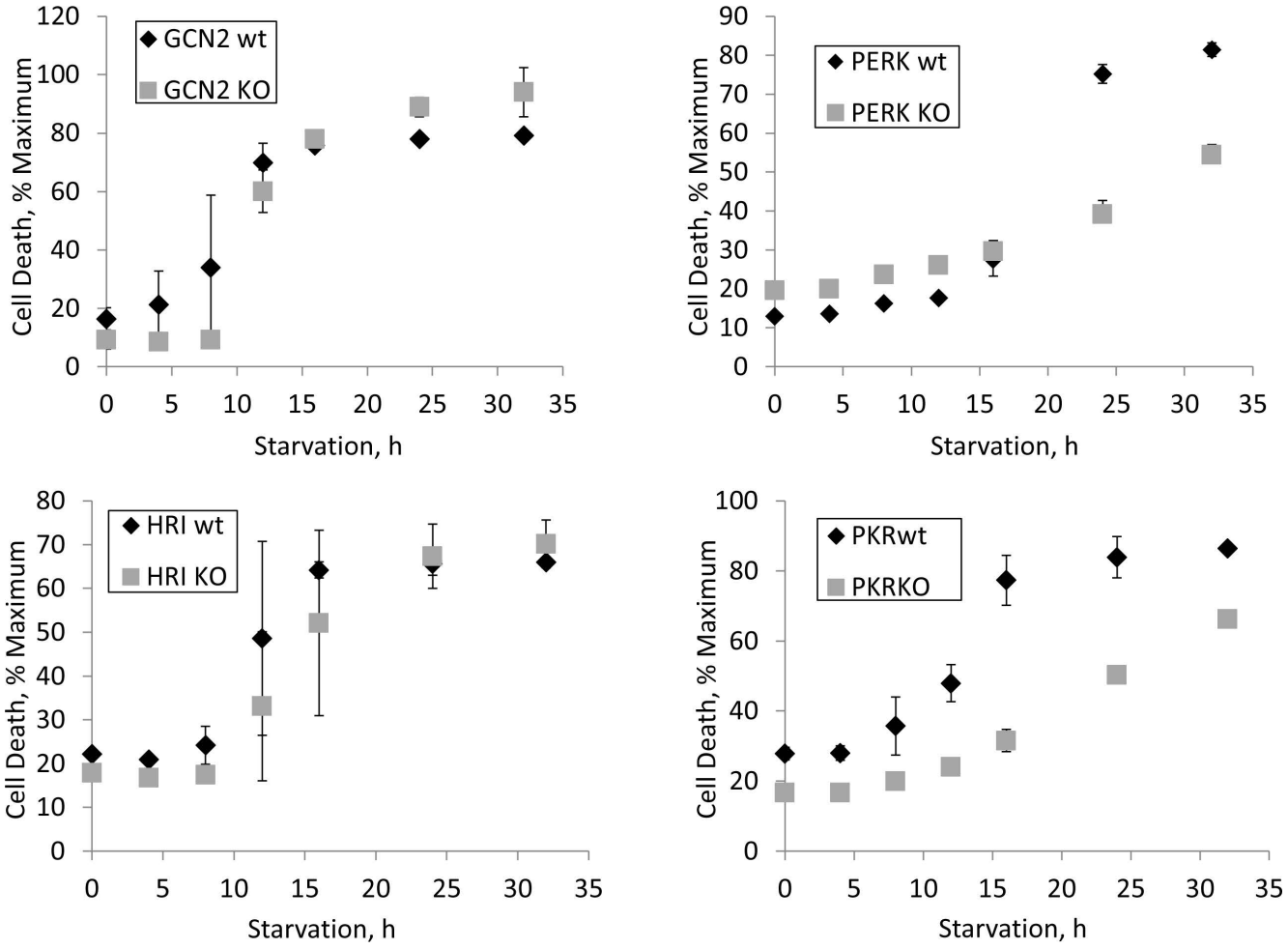


**Figure S4: Survival Curves of EIF2AK wild type and knockout MEFs.** Cells were starved for the indicated time points in the presence of ethidium homodimer-1. Time points were measured with a fluorescent plate reader as described in the Methods. Black diamonds represent wild type and gray squares represent paired KO cell lines. Bar graphs are presented as mean±s.d..

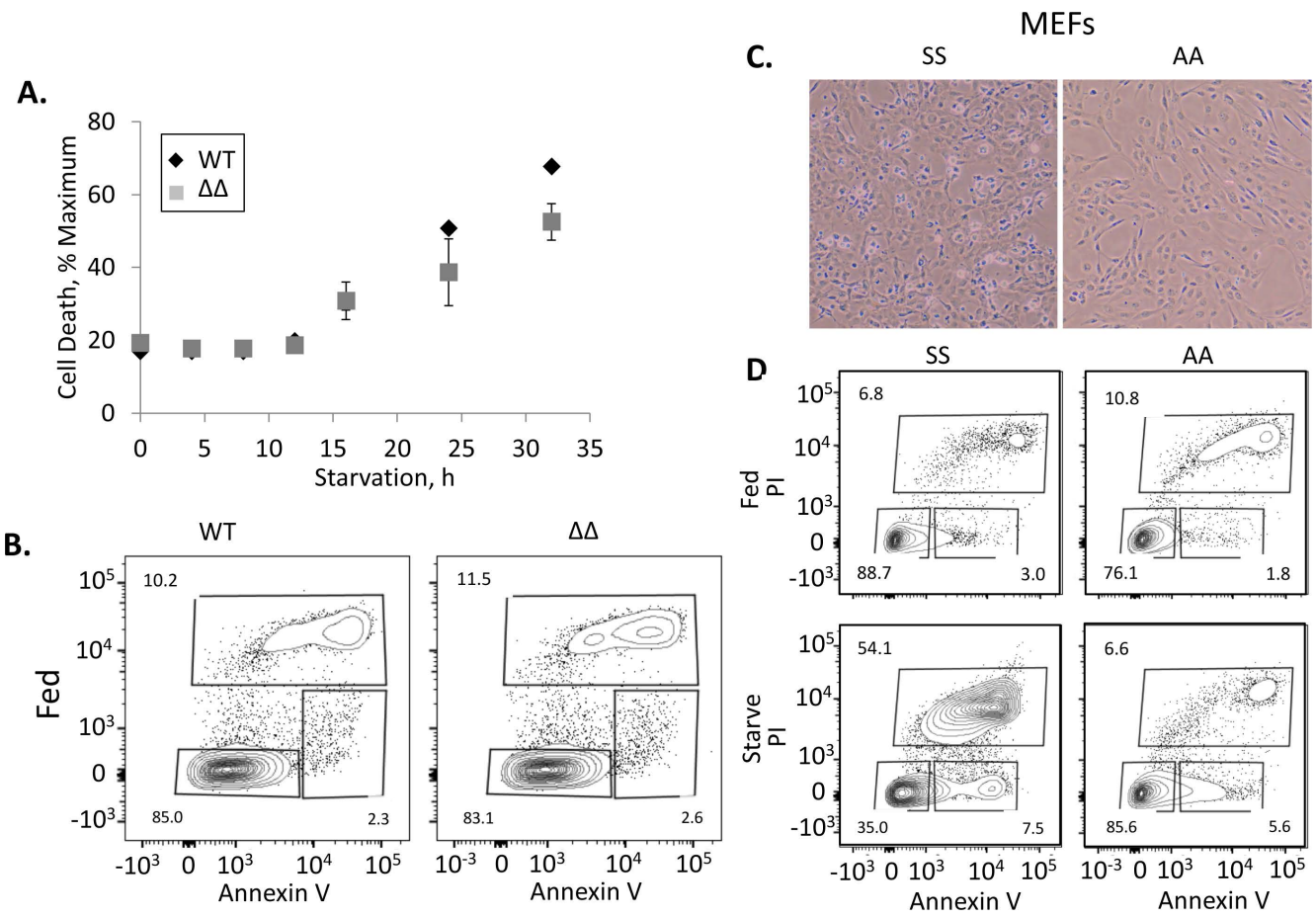


**Figure S5: Cell death analysis to complement Figure 5.** *A,* Ethidium homodimer-1 staining of wt and ΔΔ U2OS cells as described in Figure S2 to show similar results as those shown by cell monolayers and flow cytometry. *B,* Flow cytometry with AnnexinV and propidium iodide staining of fed wt and ΔΔ U2OS showing that cells are healthy. *C,* Cell monolayer for SS and S51A MEFs showing that more dead/dying cells are present in SS MEFs as compared to the S51A mutant consistent with ethidium homodimer-1 staining and flow cytometry. *D,* Flow cytometry with AnnexinV and propidium iodide staining of wild type (SS) and S51A (AA) MEFs. This data was repeated thrice and is summarized in Figure 5f. Bar graphs are presented as mean±s.d..