

Figure S1. Cells expressing low and moderate levels of APOL1 differentially localize G0 to LDs and risk variants G1 and G2 in a predominantly reticular pattern.

A) Micrographs of human primary podocytes transiently transfected with untagged APOL1 (G0 or G2 and labelled with BODIPY 493/503 and DAPI. Insets outlined in white borders. Scale bars 10 μ m. Arrows pointing to cells with highly overexpressed APOL1.

B) Human embryonic kidney cells transiently transfected with RFP-tagged APOL1 (G0 and G2) followed by labelling with BODIPY 493/503 and DAPI. Insets outlined in white borders. Scale bar 10 μ m. Arrow pointing to cells with highly overexpressed APOL1. Arrowheads pointing to lipid droplet.

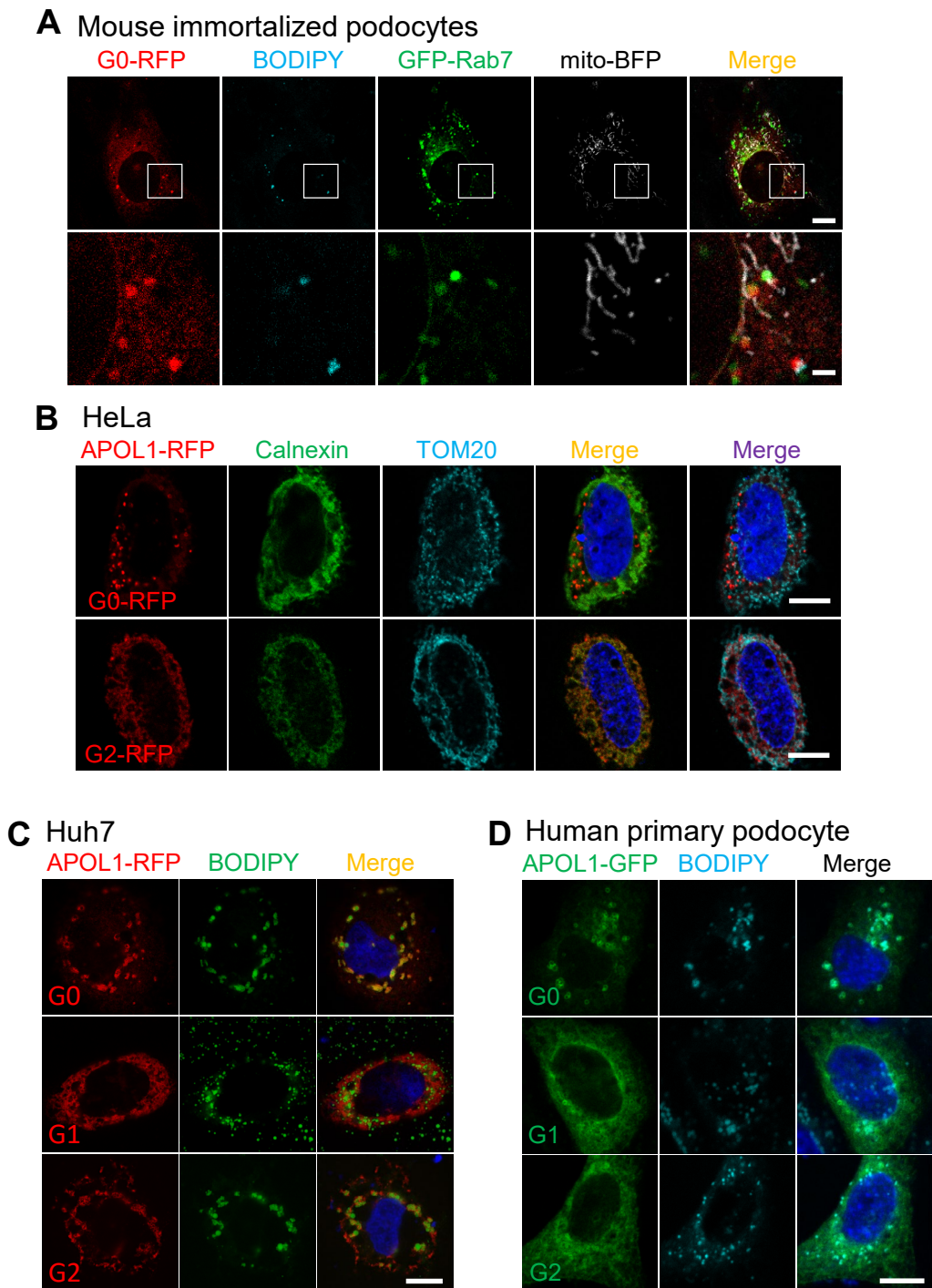


Figure S2. Fluorescent protein-tagged APOL1 localizes to lipid droplets in various cell types.

A) Confocal micrographs of representative mouse immortalized podocytes transiently co-transfected with APOL1 G0-RFP, GFP-Rab7 and mito-BFP, followed by labelling with BODIPY 493/503. Scale bar 10 μ m.

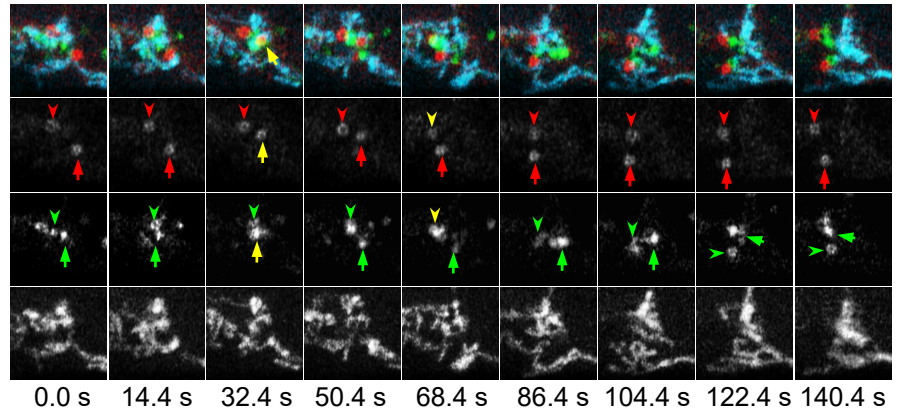
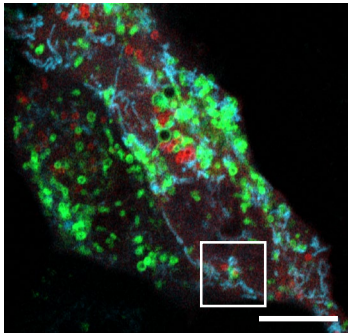
B) HeLa cells transiently transfected with RFP-tagged APOL1 (G0 or G2) and co-immunostained for ER marker calnexin, mitochondrial marker TOM20 and nuclear marker DAPI. Scale bar 10 μ m.

C) SR-SIM images of HuH7 cells transiently transfected with G0-RFP followed by labelling with BODIPY 493/503. Scale bar 10 μ m.

D) Confocal micrographs of representative human primary podocytes transiently co-transfected with APOL1 G0-GFP, G1-GFP, or G2-GFP followed by labelling with BODIPY 665/676. Scale bar 10 μ m.

A

APOL1-RFP GFP-Rab7 mito-BFP

**B**

APOL1-RFP GFP-PLIN2 BFP-KDEL

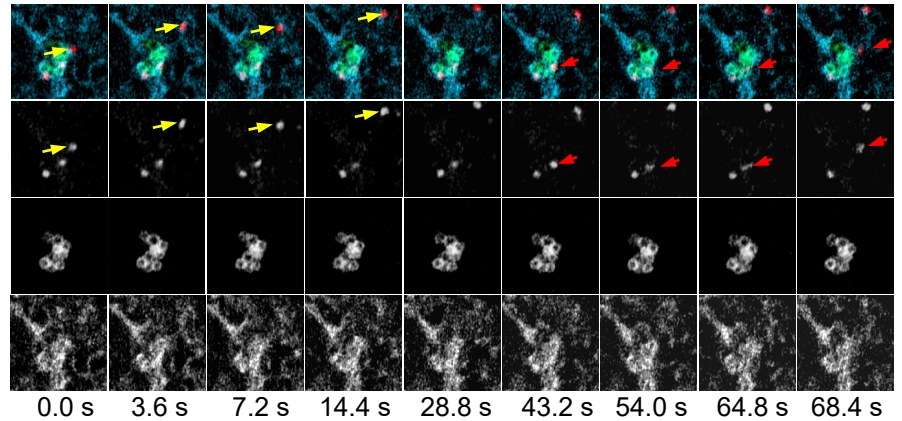
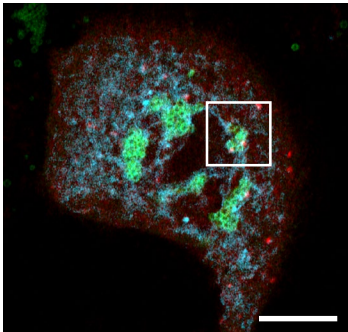


Figure S3. APOL1-positive lipid droplets dynamically interact with endosomes, mitochondria, and other organelles.

A) Time sequence stills of live primary human podocytes expressing fluorescent fusion proteins APOL1-RFP (G0) with GFP-Rab7 and mito-BFP. Airyscan images were acquired from the experiment of Fig. 2D. Scale bar 10 μ m. Inset 2 μ m.

B) Time sequence stills of live primary human podocytes expressing with GFP-PLIN2 and BFP-KDEL. Airyscan images were acquired from the experiment of Fig. 2E. Scale bar 10 μ m. Inset 2 μ m.

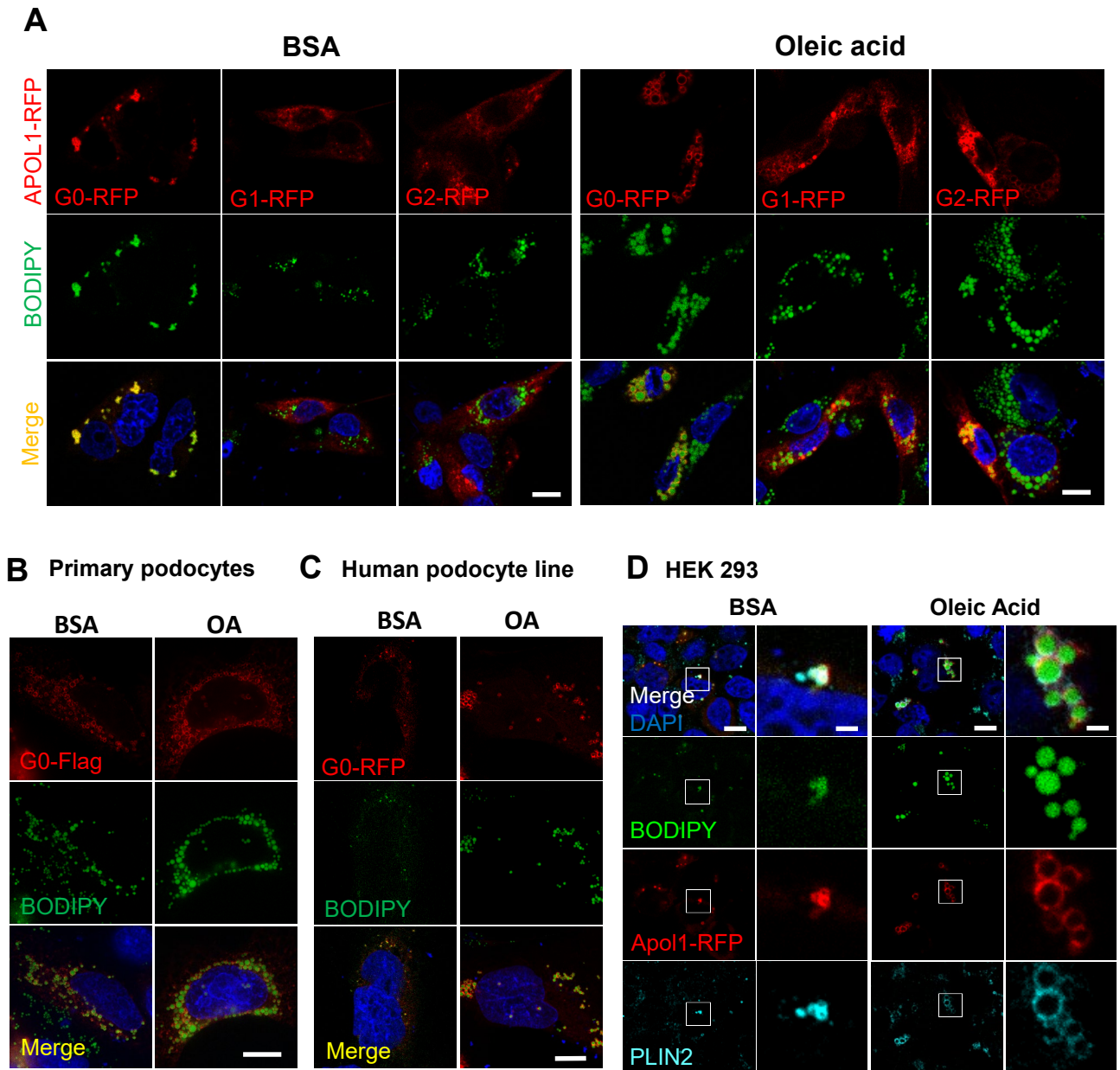


Figure S4. APOL1-RFP in T-Rex-293 cells and in human podocytes is readily detected on LDs after treatment with oleic acid.

A) Confocal micrographs of representative human primary podocytes transiently transfected with APOL1(G0)-RFP or B) APOL1(G0)-FLAG and treated with vehicle (fatty acid free BSA) or oleic acid (1 mM OA) and labelled with BODIPY 493/503 and DAPI. Scale bar 10 μ m.

C) Confocal micrographs of representative undifferentiated human immortalized podocytes transiently transfected with APOL1-RFP and treated 18.5 hrs with BSA or 0.5 mM OA; 0.5 mM and then labelled with BODIPY 493/503. Scale bar 10 μ m.

D) T-Rex-293 transiently transfected with APOL1(G0)-RFP and treated 18 hrs with vehicle or oleic acid (OA; 0.5 mM). Cell were labelled for BODIPY 493/503, perilipin 2 (PLIN2), and DAPI. Scale bars 10 μ m for full panel and 2 μ m for insets (white square outline).

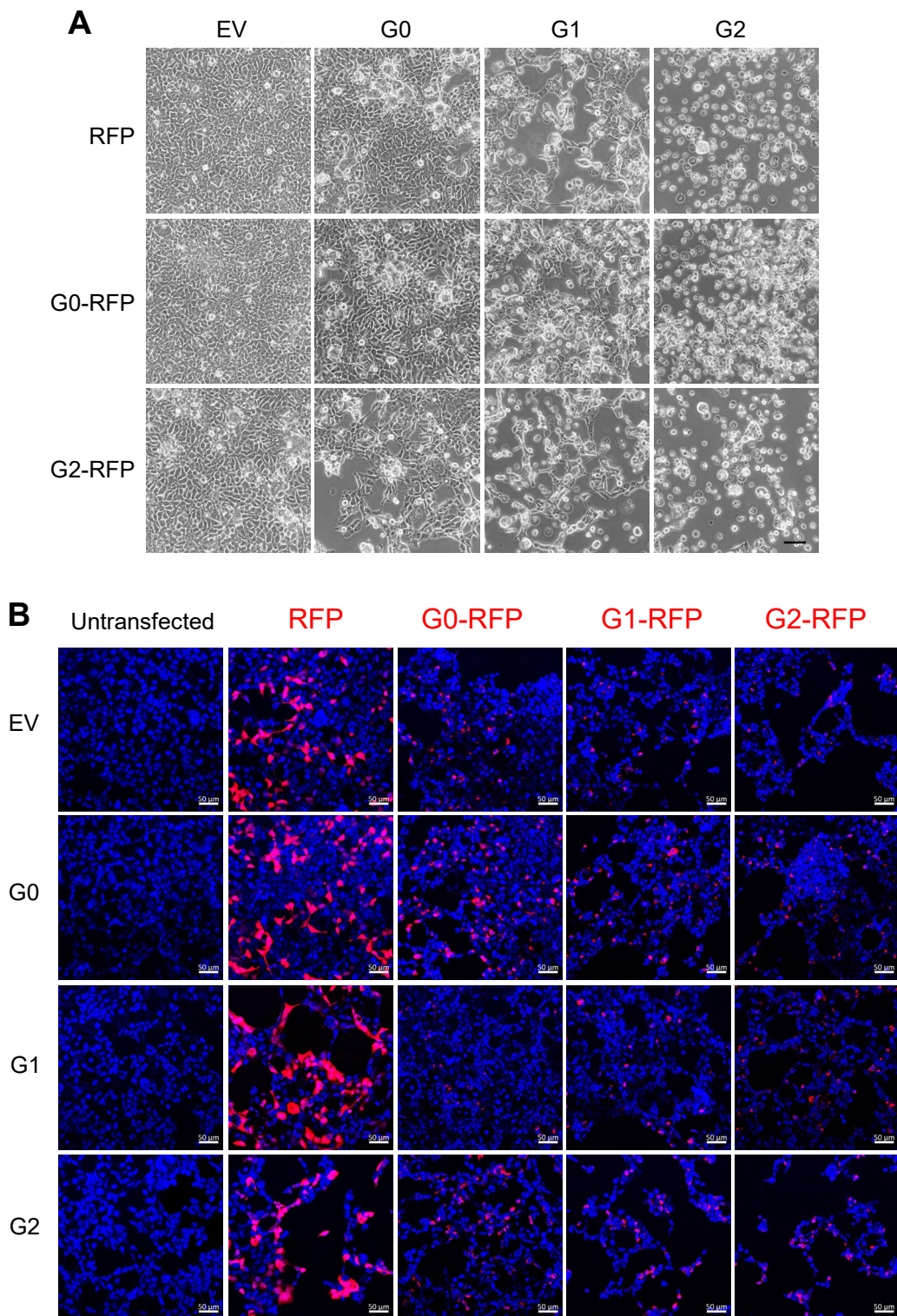


Figure S5. Transfection of wild-type APOL1 (G0) into APOL1 HEK 293 T-Rex stably expressing APOL1 risk variants (G1/G2) promotes cell survival.

A) Brightfield images of representative tetracycline-inducible transgenic APOL1 T-Rex-293 cells stably expressing empty vector, G0, G1, or G2 (C-terminal MYC and FLAG tags) transiently transfected with RFP, G0-RFP or G2-RFP four hours prior to tetracycline (10 ng/ml) induction of APOL1 for 16 hrs. Scale bar, 50 μ m.

B) APOL1-T-Rex stably expressing EV, G0, G1, or G2 that were untransfected or transiently transfected with RFP or RFP tagged APOL1 (G0, G1, or G2) and stained with DAPI. Scale bar 50 μ m.

Supplementary Movies

Supplementary Movie 1. Cropped inset of movie acquired for 140.4 sec from live primary human podocytes co-expressing APOL1-RFP (G0) with GFP-Rab7 and mito-BFP from Fig. 2D/ Fig. S3A using Airyscan.

Supplementary Movie 2. Cropped inset of movie acquired for 68.4 sec from live primary human podocytes co-expressing APOL1-RFP (G0) with GFP-PLIN2 and BFP-KDEL from Fig. 2E/ Fig. S3B using Airyscan.