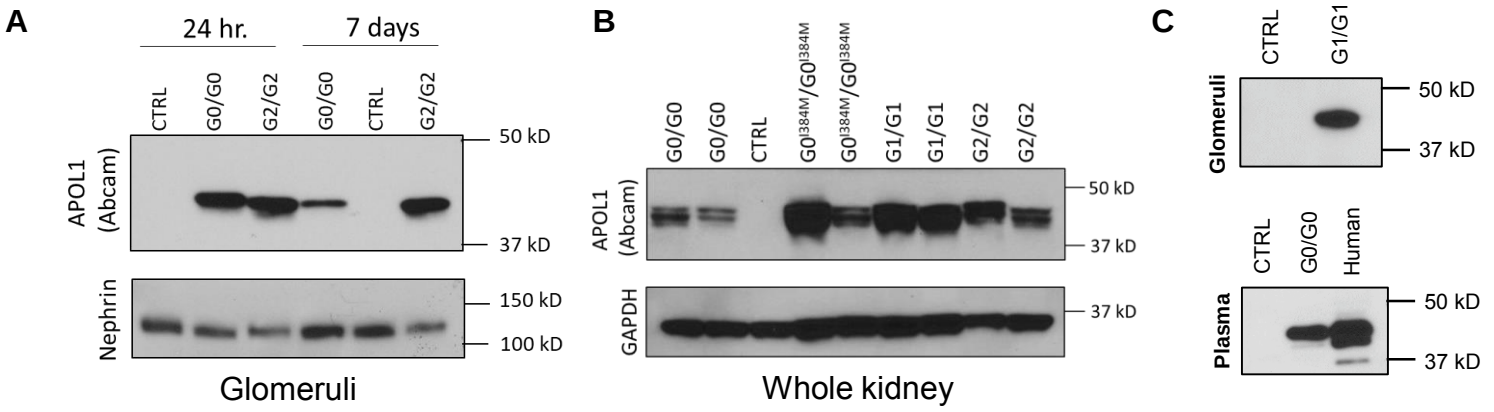


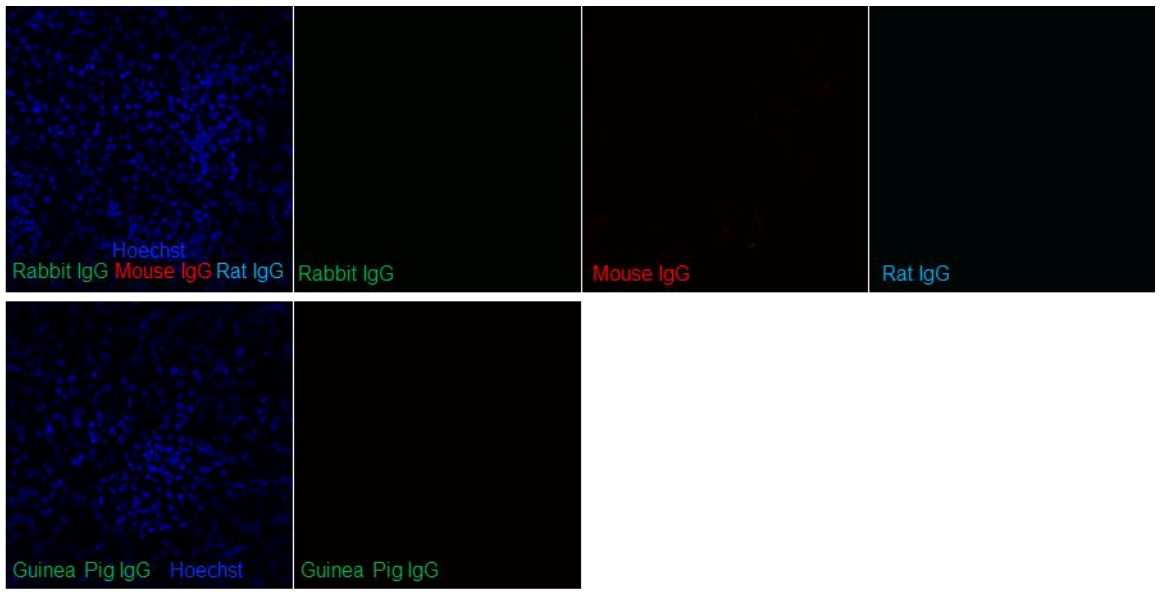


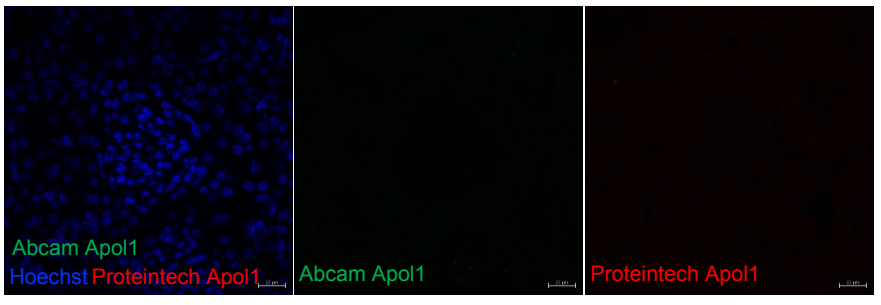


**Fig. S1. Antibody specificity**

The specificity of antibodies for western blot was verified using control FVB mice that do not contain the *APOL1* BAC. (A) Blot showing the Abcam APOL1 antibody in glomeruli from mice 24 hours or 1 week after pCpG-Muγ injection, Nephrin is shown as a loading control. (B) Blot showing the Abcam APOL1 antibody in whole kidney from untreated mice, GAPDH is shown as a loading control. (C) Blot showing the Sigma APOL1 antibody in mouse glomeruli 2-days post-injection and in untreated mouse and human plasma. The specificity of antibodies for immunofluorescence staining was verified using both IgG controls and by using control FVB mice. (D) Immunofluorescence antibody controls using rabbit, mouse, rat, and guinea pig IgG with the corresponding secondary antibodies. (E) Immunofluorescence staining of APOL1 (Abcam and Proteintech antibodies) in a control FVB mouse, 1 day after pCpG-Muγ injection.

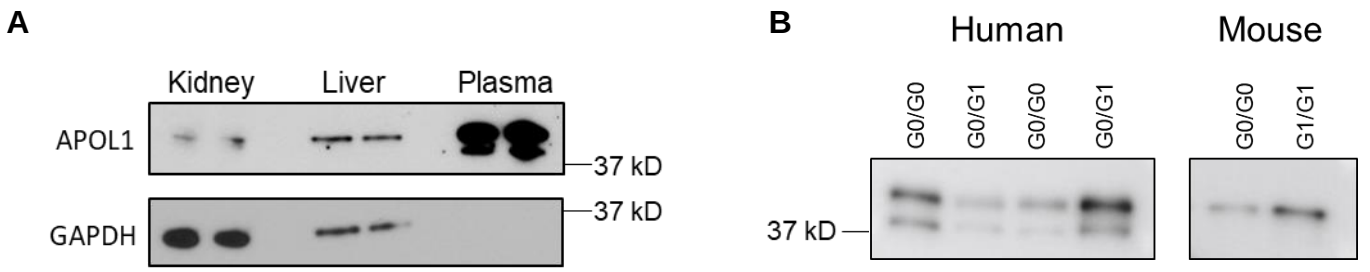


**Fig. S2. APOL1 expression in uninduced transgenic mouse lines**

APOL1 expression in the untreated transgenic mouse lines was measured by western blot. (A) APOL1 (Abcam) expression in the kidney (50 µg lysate), liver (20 µg lysate), and plasma (5 µg lysate) isolated from the same $G0^{I384M}$ mice 1 day post-injection; n =2. GAPDH is shown as a loading control for kidney and liver lysates. (B) APOL1 (Sigma) expression in 20 ug of plasma from 4 different humans and 2 different mice (untreated), genotypes indicated above the sample.

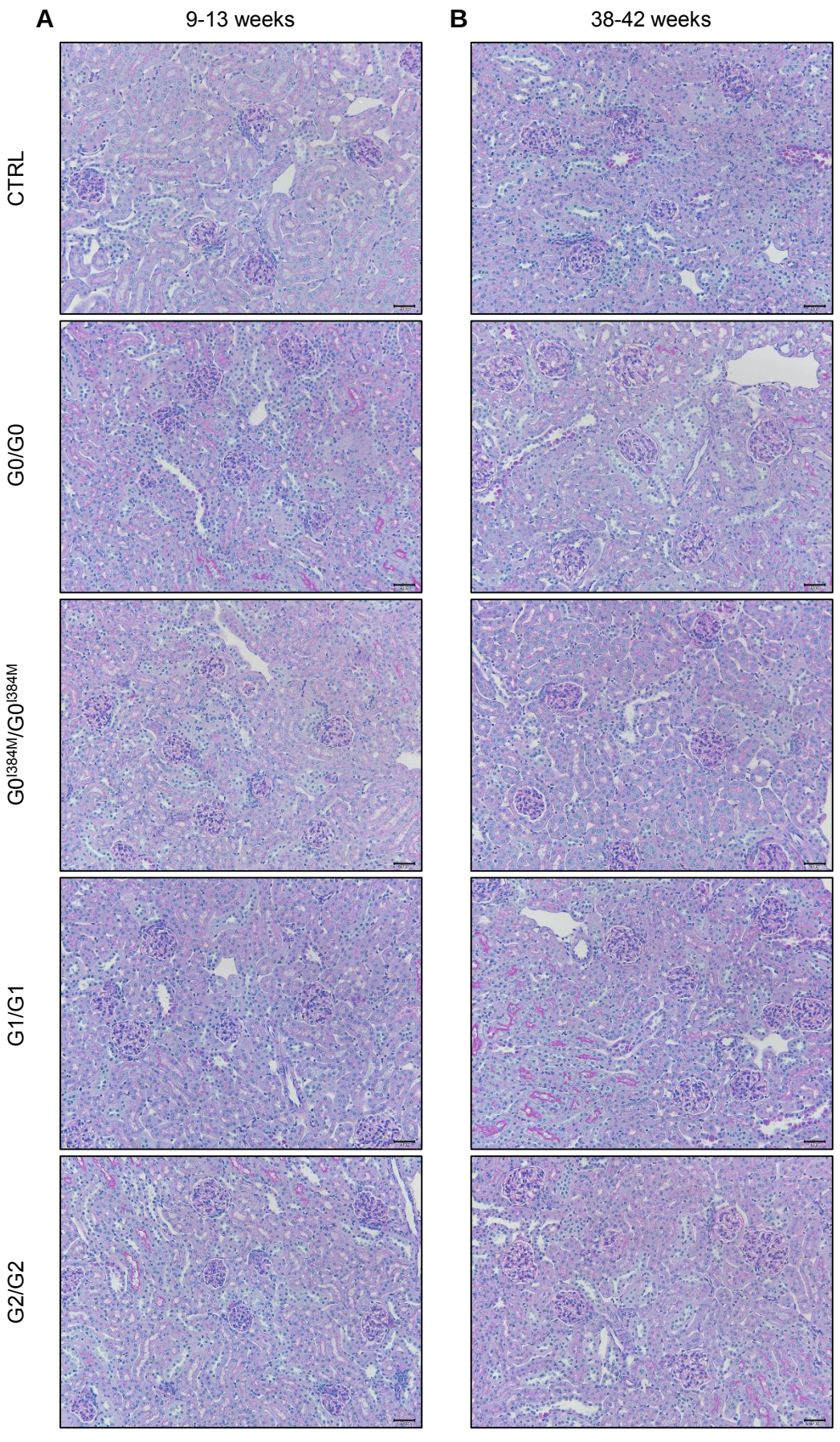


**Fig. S3. Untreated APOL1- transgenic mice show no histological kidney damage**
Representative PAS images from the kidney cortex of control and APOL1-transgenic mice at 9-13 (A) and 38-42 (B) weeks old. Scale bar is 20 microns. No morphological differences were observed

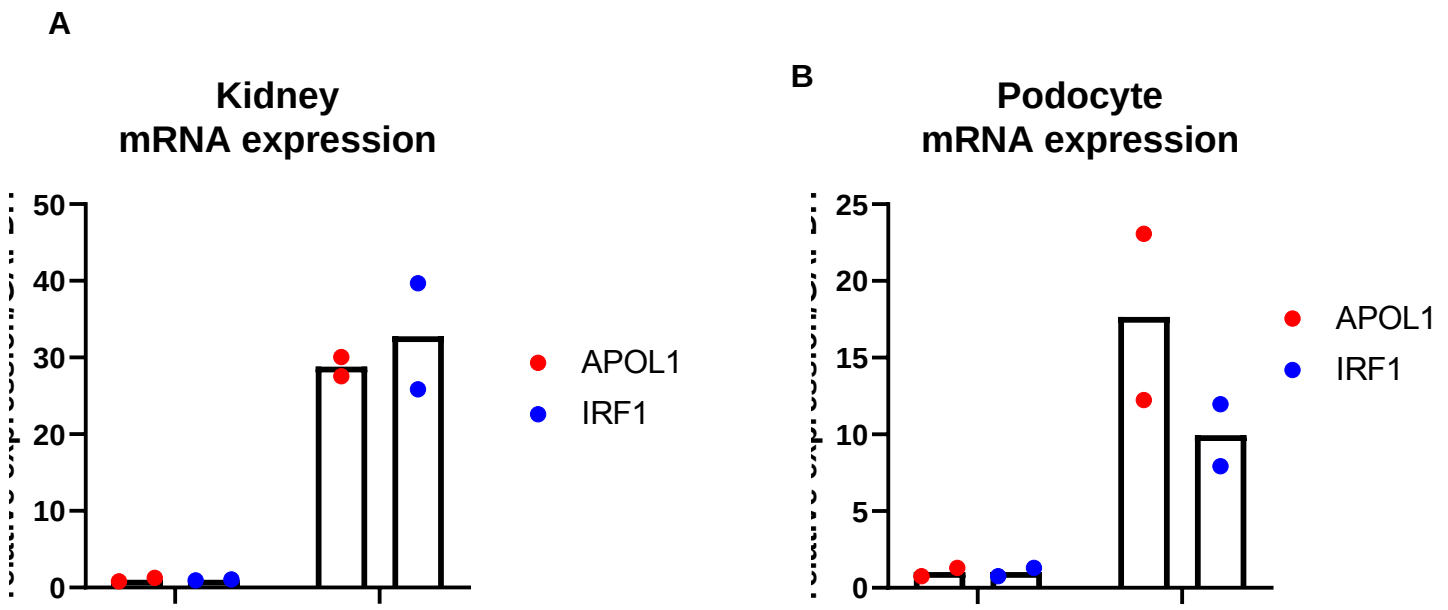


**Fig. S4. IFN-γ effect on mRNA levels**

mRNA expression of APOL1 and IRF1 in (A) G2/G2 mouse kidney at baseline or 24-hours after pCpG-Muγ injection and (B) G0/G0 mouse podocytes 6-hours after IFN treatment. Expression is normalized to Gapdh, n= 2 per time point.

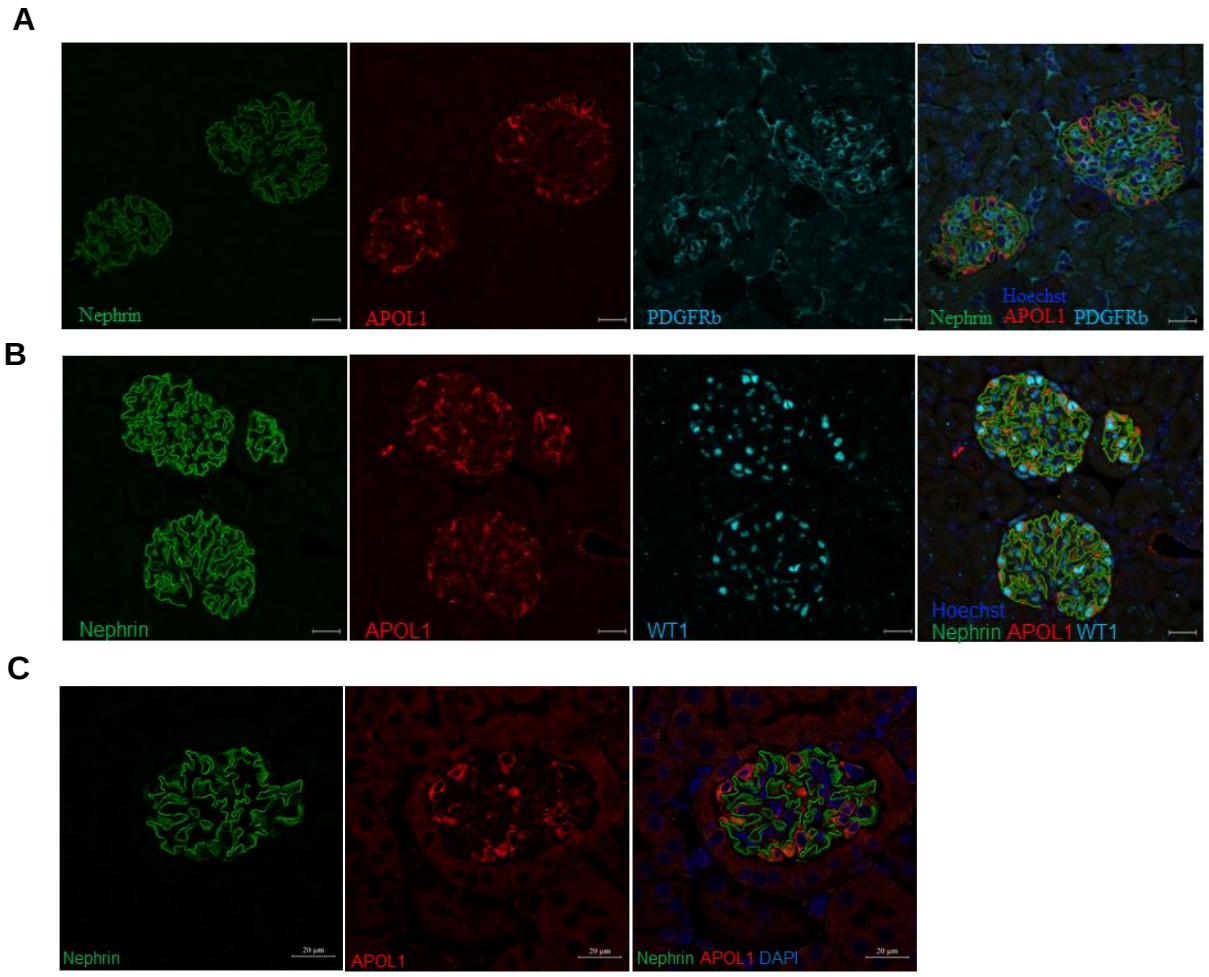


**Fig. S5. APOL1 is induced in podocytes following pCpG-Muγ injection**

Immunohistochemistry in kidney from G1 mice 24 hours after plasmid injection. Co-staining with Nephrin, APOL1 (Proteintech), and PDGFRb (A) or WT1 (B). Scale bar = 20 μM. (C) Co-staining with Nephrin and APOL1 (Sigma). APOL1 cell-type localization did not differ by APOL1 genotype.

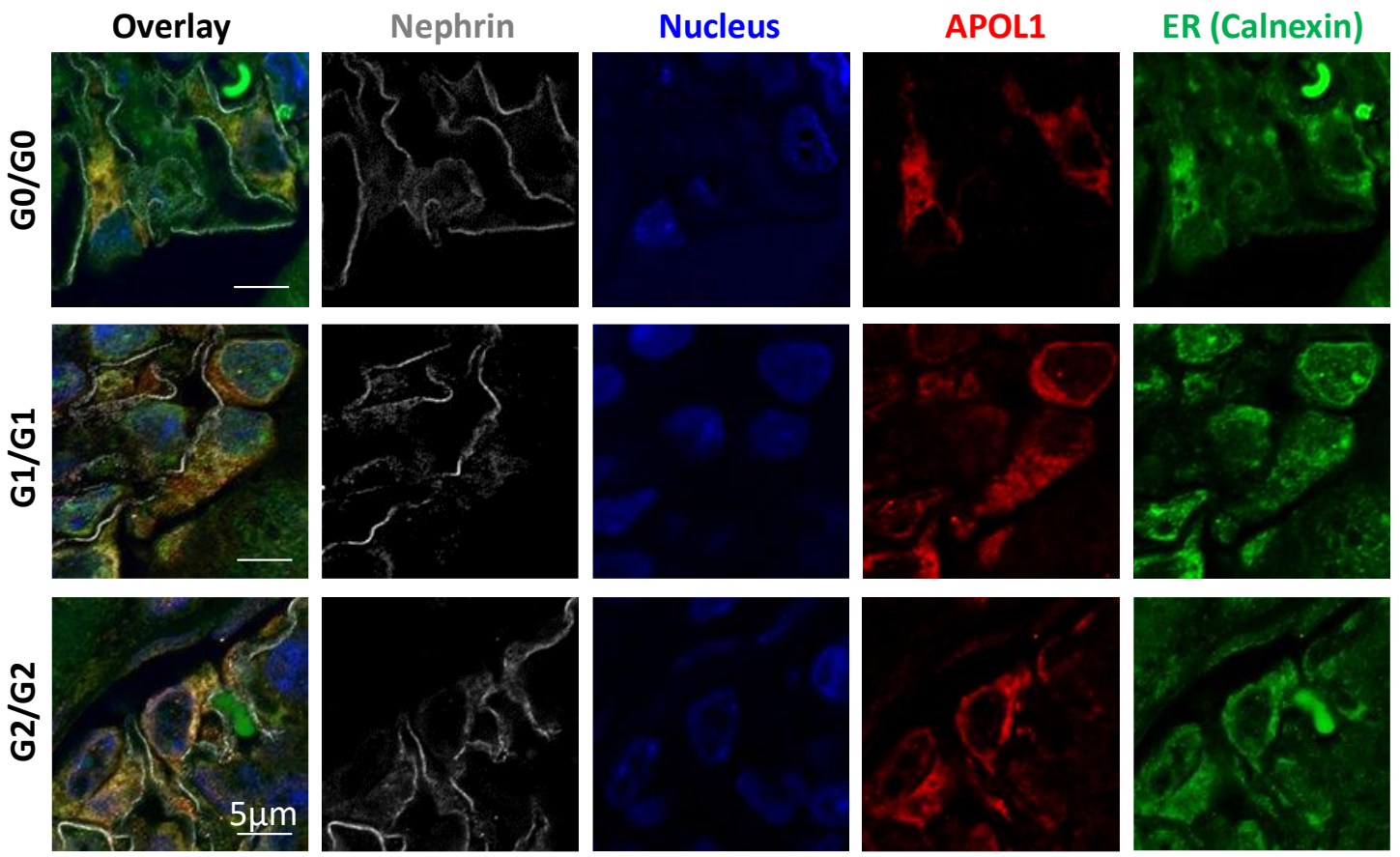


**Fig. S6. APOL1 co-localization with the Endoplasmic Reticulum (ER)**

Kidney sections from G0/G0, G1/G1 and G2/G2 mice 2 days post-injection co-stained with APOL1 (Abcam 1:50), the ER marker Calnexin (1:50) and Nephrin (1:200). The overlay shows some co-localization between APOL1 and the ER. Nephrin is shown to outline the podocytes. Image acquisitions were performed using a Zeiss Confocal system with airy scan processing as described previously (Chun et al., PNAS 2019). Airy scan processing was performed using ZEN Black (Zeiss) software with default 2D airy scan processing parameter settings.

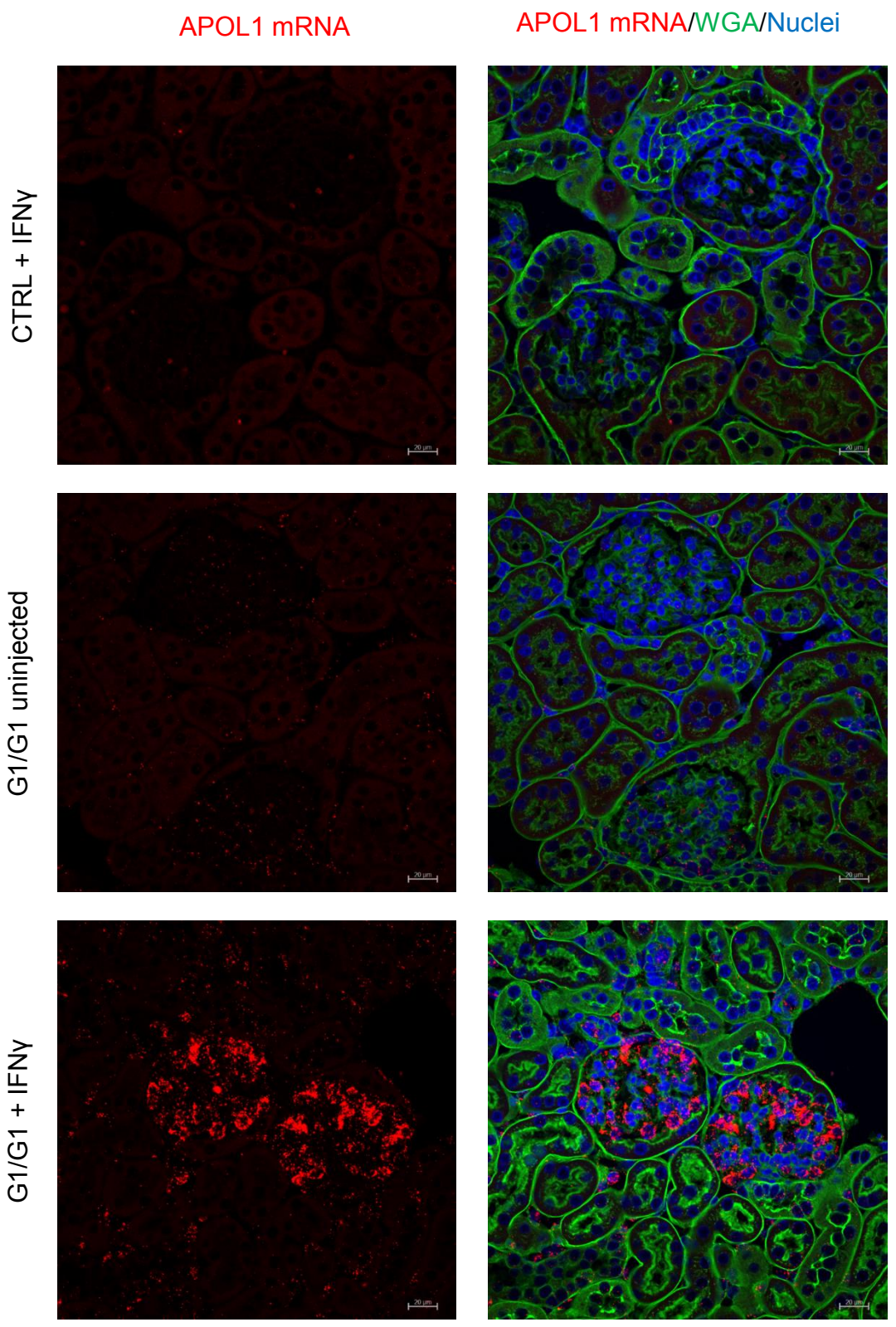


**Fig. S7. Apol1 mRNA is localized within the glomeruli**

Apol1 *in situ* hybridization on kidney from Control and G1/G1 mice either uninjected or injected with pCpG-Muγ (24 hours). Tissue was co-stained with the lectin WGA (green) and Hoescht (blue). RNA is primarily localized within the glomeruli of injected G1/G1 mice, but there is some weaker staining in the tubules.

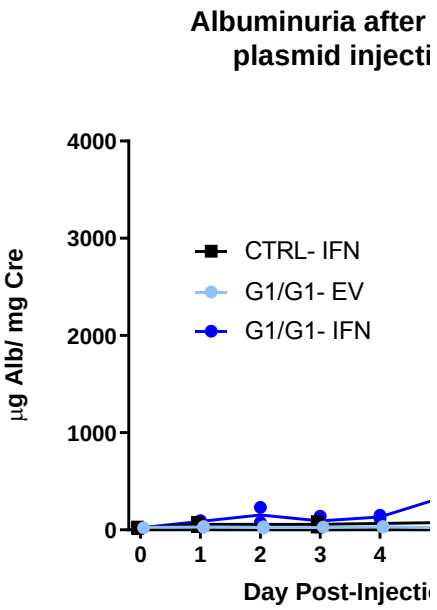


**Fig. S8. Albuminuria after pCpG plasmid injection**

Albumin to creatinine ratios following pCpG-Muγ (IFN) or pCpG-EV (EV) injection in control or G1/G1 mice. Day 0 shows baseline levels pre-injection. N = 2 per group, individual data points shown with line connecting the means.

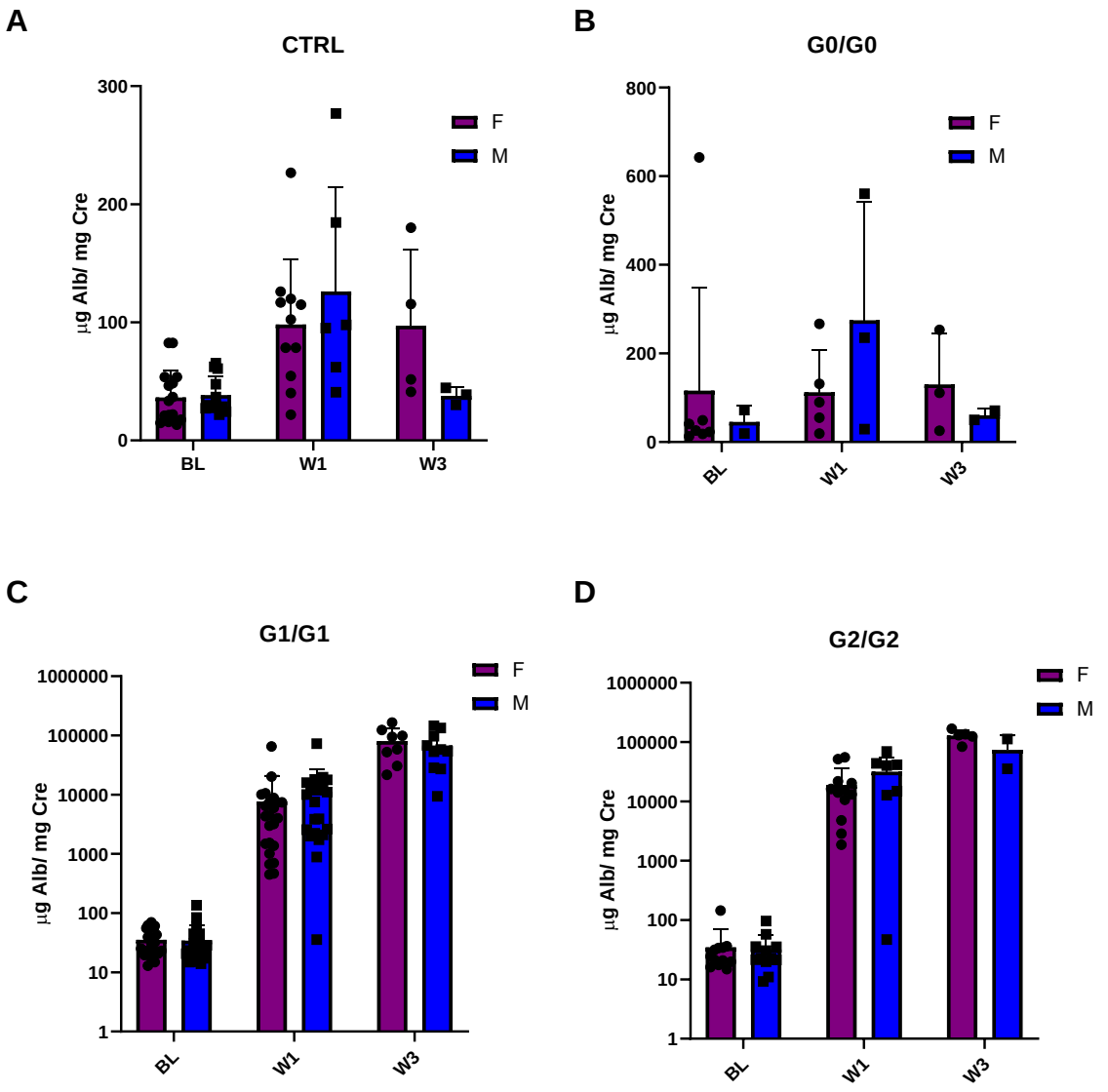


**Fig. S9. Albuminuria in APOL1 transgenic mice is not sex dependent**

Albumin to creatinine ratio for control (A), G0/G0 (B), G1/G1 (C), and G2/G2 (D) mice following injection with pCpG-Muγ plasmid. Individual data points from several experiments are shown at baseline (BL), week 1 (W1) and week 3 (W3). No significant differences were found using a one-way ANOVA with Sidak's multiple correction's test.

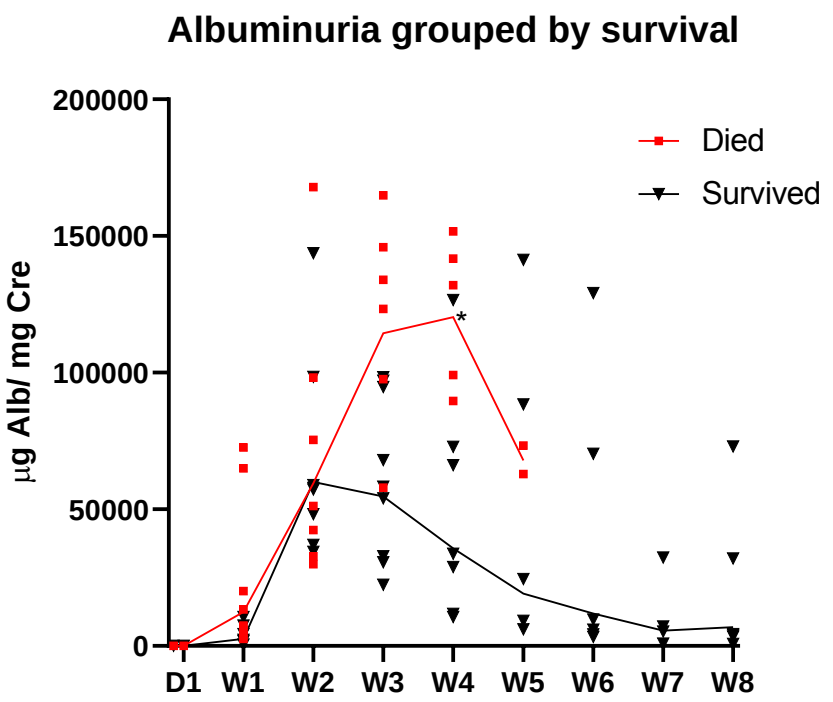


**Fig. S10. Difference in proteinuria between G1 mice that survived and those that died.**

Albuminuria over time for G1 mice that either died before of survived until 8-weeks post-injection. Individual data points are shown with a line connecting the geometric means. * p value < 0.05, mixed-effects analysis with Sidak's multiple comparisons test.

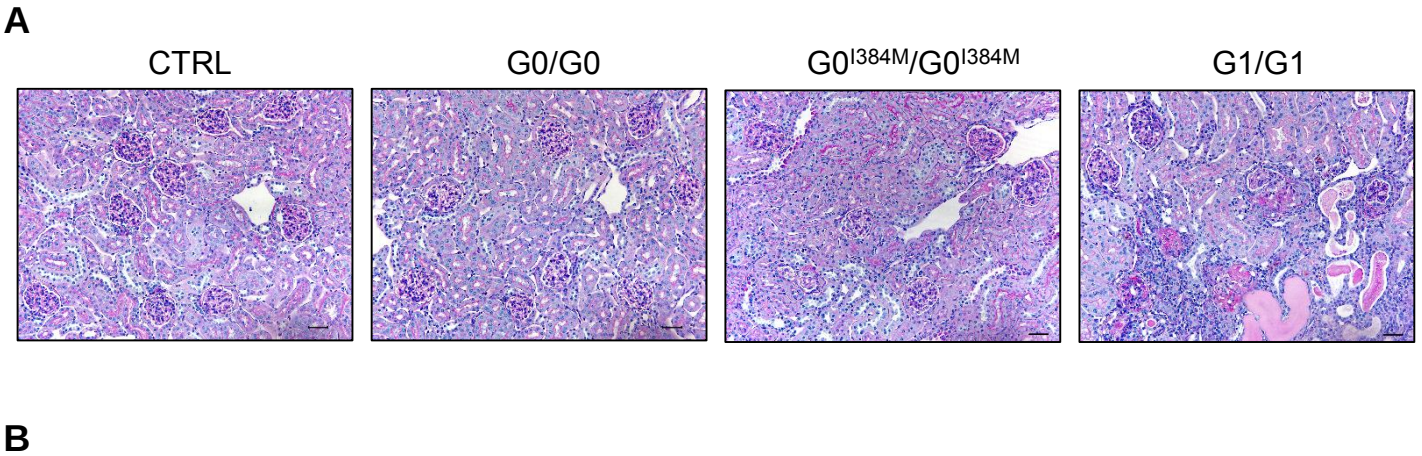


**Fig. S11. PAS images 8-weeks post-injection**

Representative PAS kidney images 8-weeks post-injection from (A) CTRL, G0/G0, $G0^{I384M}/G0^{I384M}$, and G1/G1 mice. (B) G1 and (C) G2 hemizygous and heterozygous mice. (D) $G0^{alt}$, G1/G1, and G1/G1/ $G0^{alt}$ mice. Scale bar = 20 µM

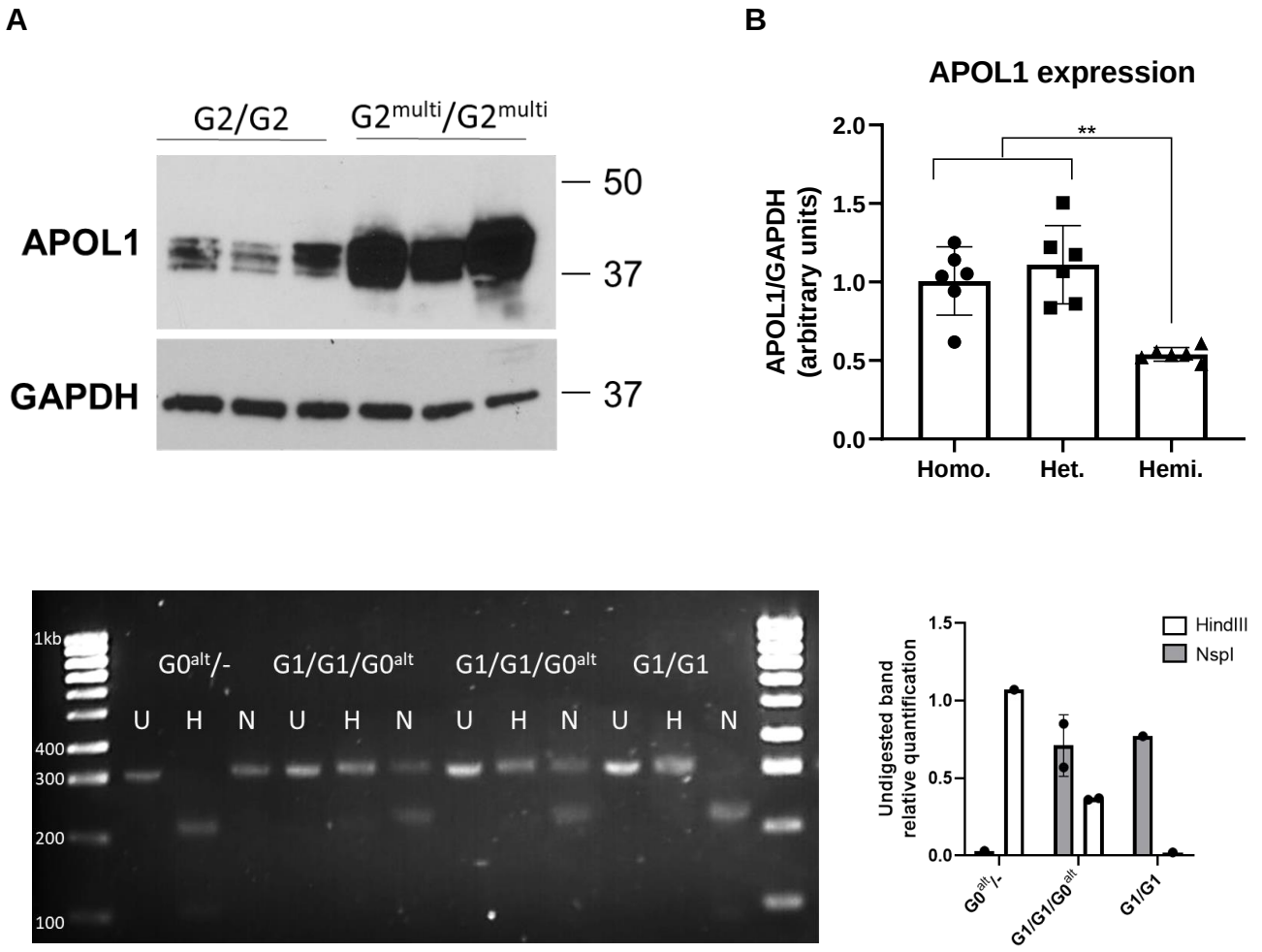


**Fig. S12. APOL1 protein expression**

(A) Representative western blot showing APOL1 expression in kidney from G2/G2 and $G2^{multi}/G2^{multi}$ mice 24 hours after injection with pCpG-Muγ, n=3 per group. GAPDH is shown as a loading control. (B) Relative glomerular APOL1 expression between homozygous, heterozygous and hemizygous mice 24-hours after injection. ** $p < 0.01$, One-way ANOVA with Tukey's multiple comparisons test. (C) Gel from allelic discrimination assay showing undigested product (U), product digested with *HindIII* (H) which only cuts G0, and product digest with *NspI* (N) which only cuts G1. Quantification for the undigested product with each enzyme relative to undigested sample is shown to the right. Glomerular cDNA samples (1-day post-injection) are from two $G1/G1/G0^{alt}/-$ mice, with $G0^{alt}/-$ and G1/G1 mice shown as controls. $G1/G1/G0^{alt}$ mice express ~2X more G1 RNA (undigested with *HindIII*) than G0 RNA (undigested with *NspI*).

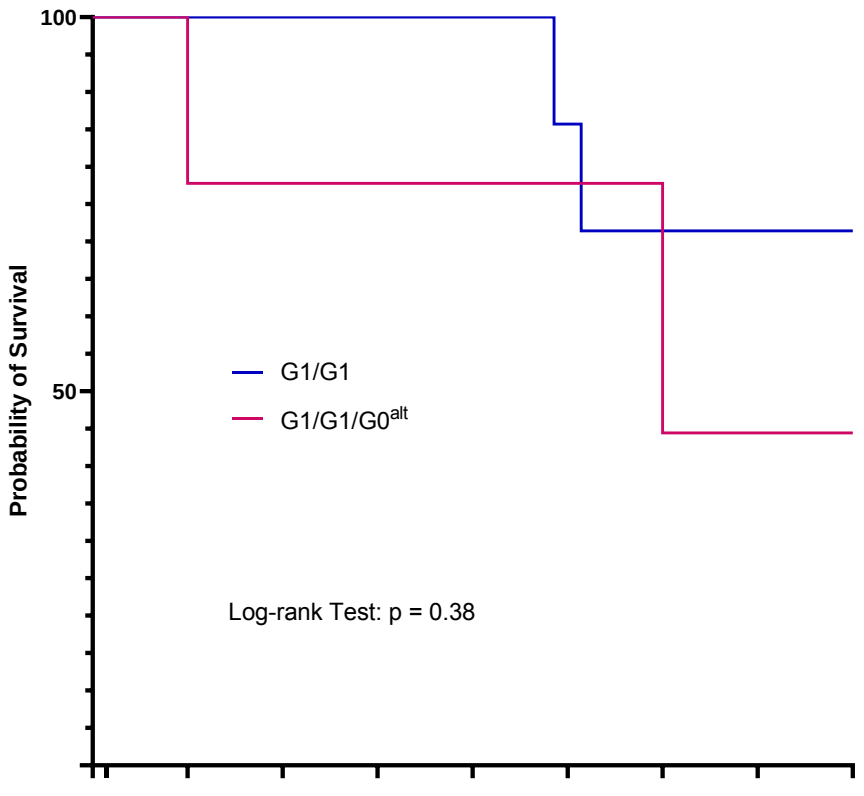


**Fig. S13. Survival of G1/G1 and G1/G1/G0alt mice**

Survival curves for G1/G1 and G1/G1/G0$^{alt}$ mice over the course of an 8 week experiment. No significant difference in survival was seen (Log-rank test). N= 9 for G1/G1/G0$^{alt}$; n=7 for G1/G1 . D indicates day, W indicates week post-injection.

**Table S1. Guide RNAs used for CRISPR mutagenesis of transgenic mouse lines and potential off-target cut sites**

| | | | | off-target sites with fewer than 3 mismatches | | | | | | primers used for PCR/sequencing | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| From | To | Mutation site(s) changed | gRNA(s) | Sequence (mismatch in lower case, PAM sequence underlined) | Chrom. | location | strand | # of mismatches | annotated gene | Forward primer | Reverse primer |
| G1 | G2$^{S342G}$ | 384/388 | TAATTATAAGATTCTGCAGG | TAATTATAtGATgCTGCAGGAGG | chr3 | 126,000,486 | + | 2 | none | 5'- CAGAGAAGCCAACAGTCATACA- 3' | 5'- CATCCCAGAGATATCTGAGCATAAG- 3' |
| G2$^{S342G}$ | G2 | 342 | AGGAGTCAAGCTCACGGATG | | | | | | | | |
| G1 | G0$^{I384M}$ | 342 | TGTAAGCTTCTTTCTTGTGC | TGTAAGCTTCTagCTTGTGCAGG | chr4 | 131,530,700 | - | 2 | none | 5'- GACCCATGCATGACAGACCT- 3' | 5'- GTTTGAGCGGGTTTTGTGGG- 3' |
| | | | CTGCTCATTTCCAGGATGCT | CaGCTCcTTTCCAGGATGCTGGG | chr6 | 94,403,336 | - | 2 | none | 5'- GGAAGAGGAAGAGGAAGAAGAAG- 3' | 5'- GTGGGAAGTGGTACTTGTAGTC- 3' |
| G0$^{I384M}$ | G0 | 384/388 | TAATTATAAGATTCTGCAGG | TAATTATAtGATgCTGCAGGAGG | chr3 | 126,000,486 | + | 2 | none | 5'- CAGAGAAGCCAACAGTCATACA- 3' | 5'- CATCCCAGAGATATCTGAGCATAAG- 3' |

## Table S2. Transgenic mouse nomenclature used

| Symbol | Description |
|---|---|
| CTRL | FVB mouse with no transgene |
| G0/G0 | Homogzyous BAC transgenic mouse with G0 APOL1 integrated into chromosome 4 |
| $G0^{I384M}/G0^{I384M}$ | Homozygous BAC transgenic mouse with G0I384M APOL1 integrated into chromosome 4 |
| G1/G1 | Homozygous BAC transgenic mouse with G1 APOL1 integrated into chromosome 4 |
| G2/G2 | Homozygous BAC transgenic mouse with G2 APOL1 integrated into chromosome 4 |
| G1/- | Hemizygous BAC transgenic mouse with one copy of G1 APOL1 |
| G0/G1 | Heterozgyous BAC transgenic mouse with one copy of G0 and one copy of G1 |
| $G0^{I384M}/G1$ | Heterozgyous BAC transgenic mouse with one copy of $G0^{I384M}$ and one copy of G1 |
| G2/- | Hemizygous BAC transgenic mouse with one copy of G2 APOL1 |
| G0/G2 | Heterozgyous BAC transgenic mouse with one copy of G0 and one copy of G2 |
| $G0^{I384M}/G2$ | Heterozgyous BAC transgenic mouse with one copy of $G0^{I384M}$ and one copy of G2 |
| $G2^{multi}/G2^{multi}$ | Homozygous BAC transgenic mouse with multiple copies of G2, integration sites unknown |
| $G0^{alt}$/- | Hemizygous BAC transgenic mouse with one copy of G0, integration site unknown |
| $G1/G1/G0^{alt}$ | BAC transgenic mouse homozygous for G1 (chromosome 4) with an additional single copy of $G0^{alt}$ (unknown integration site) |

## Table S3. Antibodies used for western blot

| **Primary antibodies** | | | | | |
|---|---|---|---|---|---|
| | | | | | |
| **Antibody target** | **Species** | **Manufacturer** | **Catalog number** | **Dilution used** | **diluent** |
| APOL1 | Rabbit | Abcam | ab252218 | 1:1000 | 5% NFDM |
| APOL1 | Rabbit | Sigma | HPA018885 | 1:1000 | 5% NFDM |
| PhosphoSTAT1 (Y701) | Rabbit | Cell Signaling | 58D6 | 1:500 | 5% BSA |
| Transferrin | Sheep | abcam | ab9033 | 1:5000 | 5% NFDM |
| Nephrin | Goat | R&D | AF3159 | 1:500 | 5% NFDM |
| Vinculin | mouse | sigma | V9131 | 1:5000 | 5% NFDM |
| GAPDH-HRP | | Genetex | GT239 | 1:5000 | 5% NFDM |
| | | | | | |
| **Secondary antibodies** | | | | | |
| | | | | | |
| **Antibody target** | | **Manufacturer** | **Catalog number** | **Dilution used** | **diluent** |
| Rabbit anti-Goat IgG, HRP | | Thermo Scientific | 31402 | 1:5000 | 5% NFDM |
| Donkey anti-sheep IgG, HRP | | Thermo Scientific | A16041 | 1:5000 | 5% NFDM |
| Anti-Rabbit IgG, HRP | | Cell Signaling | 7074 | 1:5000 | 5% NFDM |
| Anti-mouse IgG, HRP | | Cell Signaling | 7076 | 1:5000 | 5% NFDM |
| | | | | | |
| NFDM- Non-fat dry milk | | | | | |

## Table S4. Antibodies used for immunofluorescence

| Antibody target | Manufacturer | Catalog number | Dilution used | Secondary Antibody | Secondary Manufacturer | Secondary Catalog number | Secondary Dilution |
|---|---|---|---|---|---|---|---|
| Wilms Tumor (WT1) | Abcam | ab89901 | 1:300 | Goat anti-rabbit IgG, Alexa Fluor 647 | Thermo Fisher | A-21244 | 1:250 |
| APOL1 | Abcam | ab252218 | 1:500 | Goat anti-rabbit IgG, Alexa Fluor 546 | Thermo Fisher | A-11035 | 1:250 |
| APOL1 | Proteintech | 66124-1-Ig | 1:500 | Goat anti-mouse IgG, Alexa Fluor 546 | Thermo Fisher | A-11030 | 1:250 |
| APOL1 | Sigma | HPA018885-100UL | 1:500 | Goat anti-rabbit IgG, Alexa Fluor 546 | Thermo Fisher | A-11035 | 1:250 |
| Nephrin | Progen | GP-N2 | 1:200 | Goat anti-Guinea Pig IgG, Alexa Fluor 488 | Thermo Fisher | A-11073 | 1:250 |
| Endomucin | Life Technologies | 14-5851-81 | 1:200 | Goat anti-Rat IgG, Alexa Fluor 647 | Thermo Fisher | A-21247 | 1:250 |
| PDGRFb | Cell Signaling | 3169 | 1:200 | Goat anti-Rabbit IgG, Alexa Fluor 647 | Life Technologies | A-21244 | 1:250 |
| Calnexin | Cell Signaling | C5C9 | 1:50 | Goat anti-Rabbit IgG, Alexa Fluor 488 | Life Technologies | A-11008 | 1:250 |
| FITC conjugated Lectin from wheat (WGA) | Sigma | L4895 | 1:100 | N/A | | | |
| Rabbit IgG | Santa Cruz | sc-2027 | 1:250 | Goat anti-Rabbit IgG, Alexa Fluor 488 | Invitrogen | A-11008 | 1:250 |
| Guinea Pig IgG | Sino Biological | CR4 | 1:400 | Goat anti-Guinea Pig IgG, Alexa Fluor 488 | Invitrogen | A-11073 | 1:250 |
| Rat IgG | Sigma Aldrich | I4131-10MG | 1:400 | Goat anti-Rat IgG, Alexa Fluor 647 | Invitrogen | A-21247 | 1:250 |
| Mouse IgG | Santa Cruz | sc-2025 | 1:150 | Goat anti-rabbit IgG, Alexa Fluor 546 | Invitrogen | A-11035 | 1:250 |