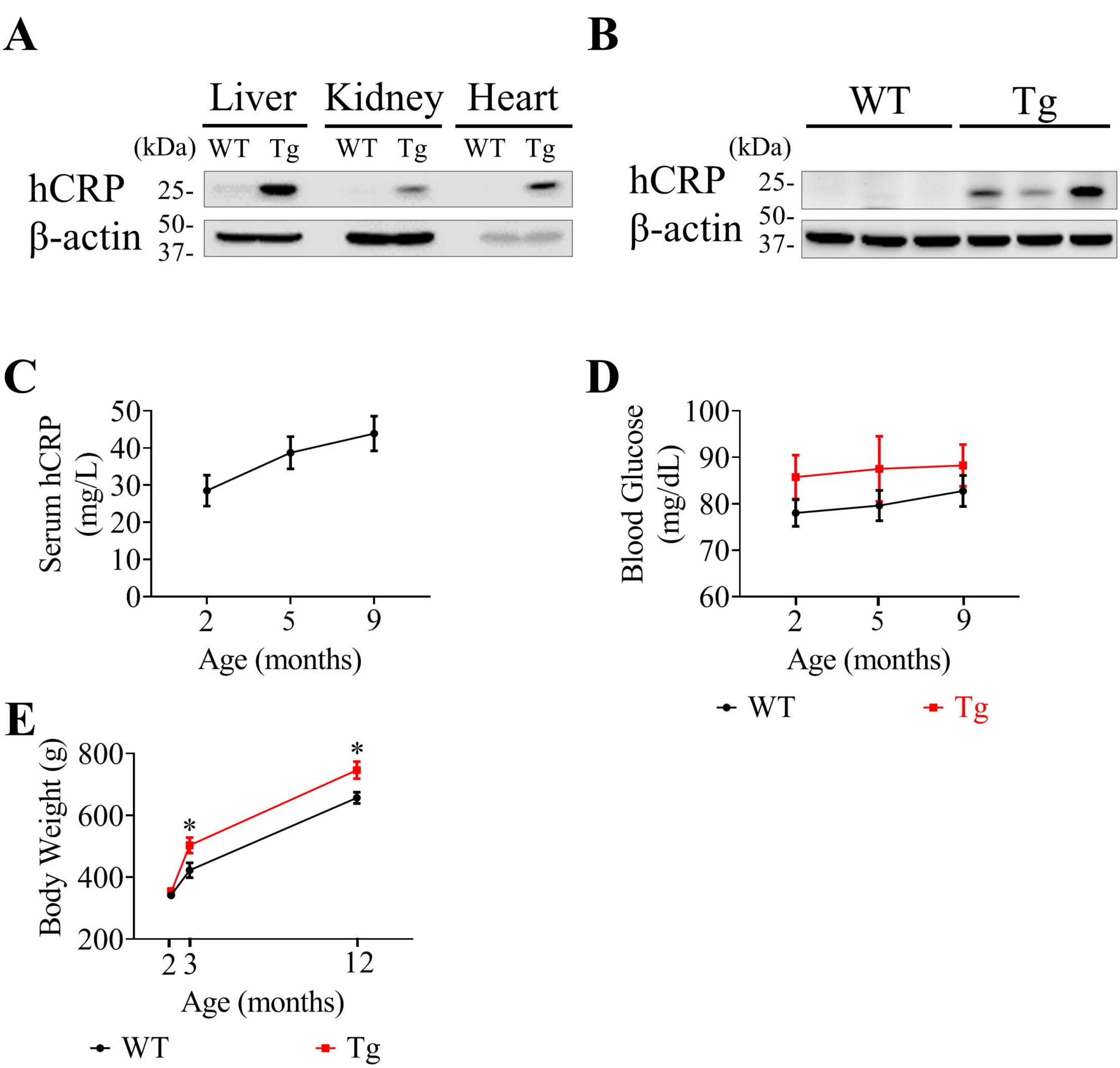
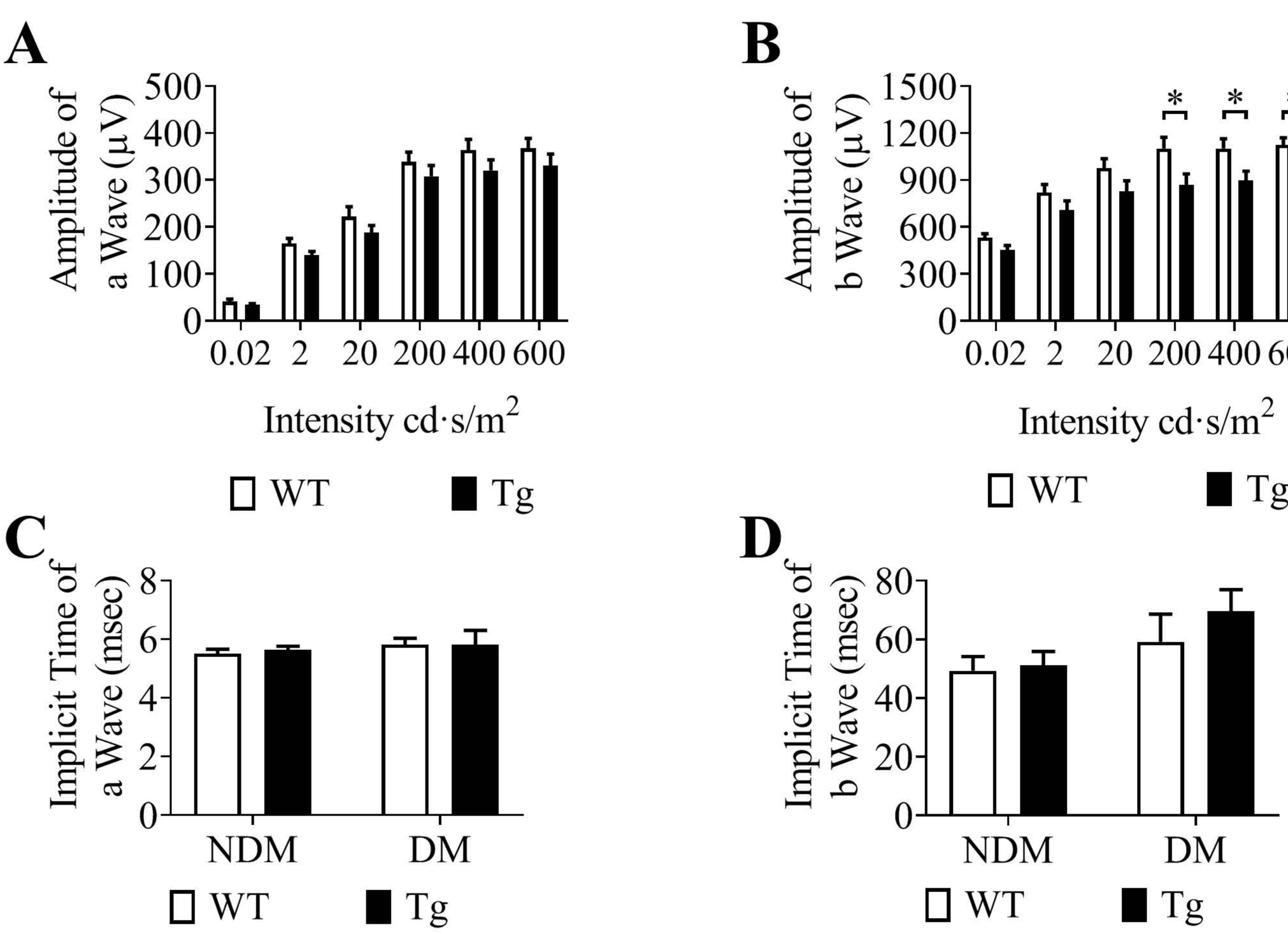


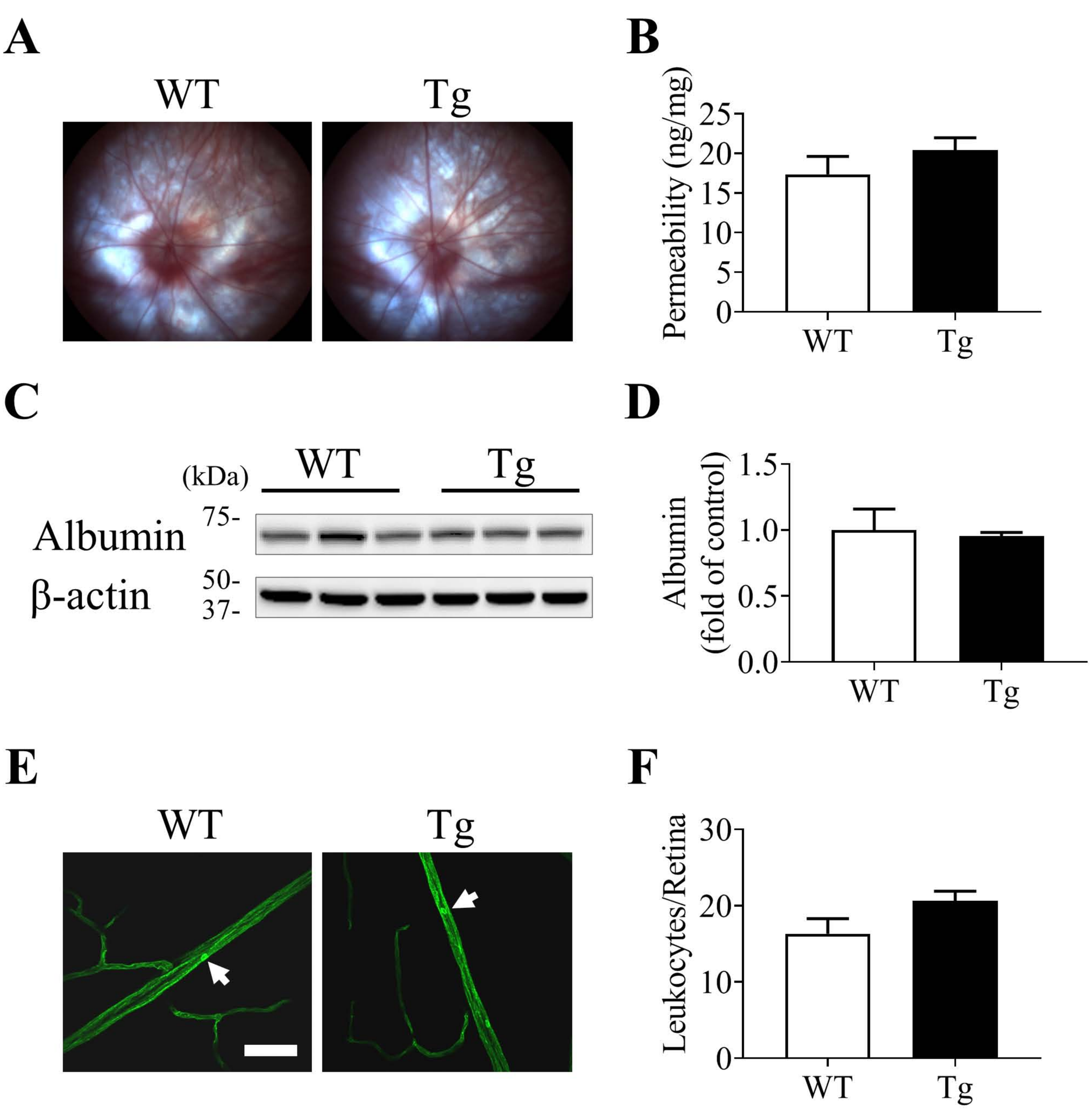
Supplementary Figure 1

Supplementary Figure 1. Levels of hCRP in the serum and tissues, body weight and blood glucose in age-matched WT rats and hCRP-Tg rats. *A*: Representative images of Western blotting for hCRP in the liver, kidney, and heart of 6-month-old WT rats and hCRP-Tg rats. *B*: Representative images of Western blotting for hCRP in the retina of 6-month-old WT rats and hCRP-Tg rats. *C*:hCRP levels in the serum of hCRP-Tg rats at the indicated ages. *D*: Blood glucose levels of age-matched WT rats and hCRP-Tg rats at indicated ages. *E*: Body weights of age-matched WT rats and hCRP-Tg rats at indicated ages. Data were presented as mean ± SEM (n = 10-12). * $P < 0.05$.



Supplementary Figure 2

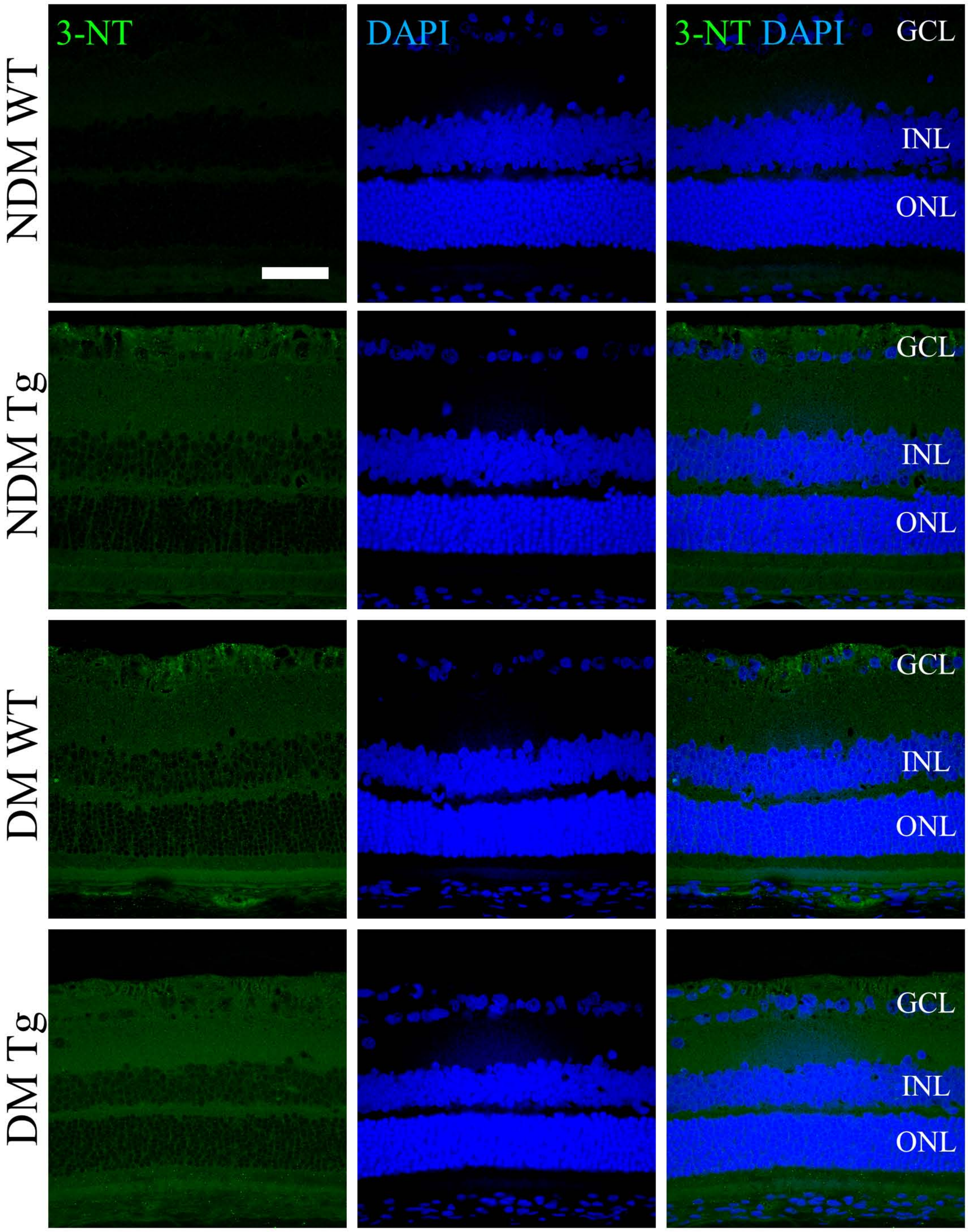
Supplementary Figure 2. ERG amplitudesand implicit time of scotopic a-wave and b-wave. *A* and *B*: Scotopic ERG a-wave (*A*) and b-wave (*B*) of 6-month-old WT and hCRP-Tg rats were measured in response to different intensities of light stimuli. *C*: The implicit time of scotopic a-wave of 6-month-old WT and hCRP-Tg rats (Tg) in the presence of diabetes (DM) or in the absence of diabetes (NDM). *D*: The implicit time of scotopic b-wave of 6-month-old WT and hCRP-Tg rats in the presence of diabetes or in the absence of diabetes. Data were presented as mean ± SEM (n=6). * $P < 0.05$.



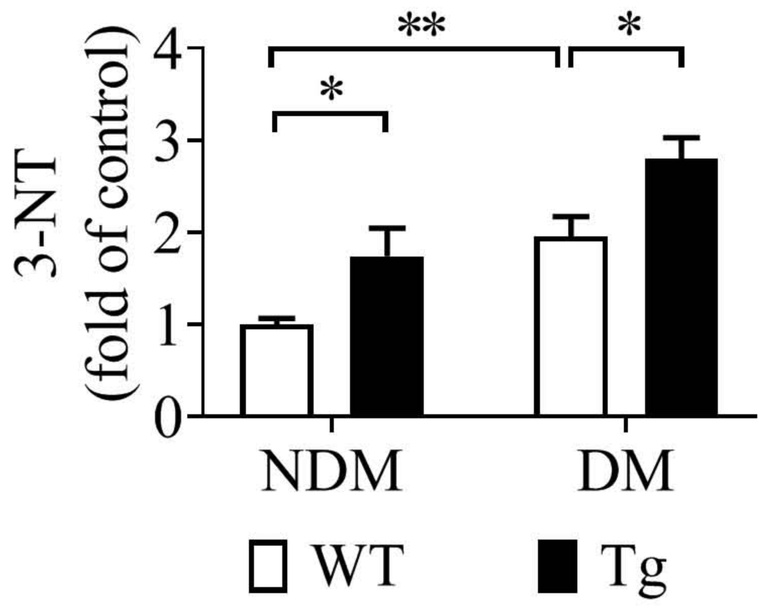
Supplementary Figure 3

Supplementary Figure 3. Elevated hCRP levels alone had no effect on retinal vasculature. *A*: Bright field fundus imaging of 6-month-old WT rats and hCRP-Tg rats. *B*: Retinal vascular permeability assay in 6-month-old WT rats and hCRP-Tg rats. *C*: Representative images of Western blotting for albumin in the perfused retinas of 6-month-old WT rats and hCRP-Tg rats. *D*: Levels of albumin in the retina were quantified by densitometry. *E*: Representative images of leukostasis assay in 6-month-old WT rats and hCRP-Tg rats. White arrows indicated adherent leukocytes. Scale bar: 10 μm. *F*: Adherent leukocytes were quantified. Data were presented as mean ± SEM (n = 6).

A

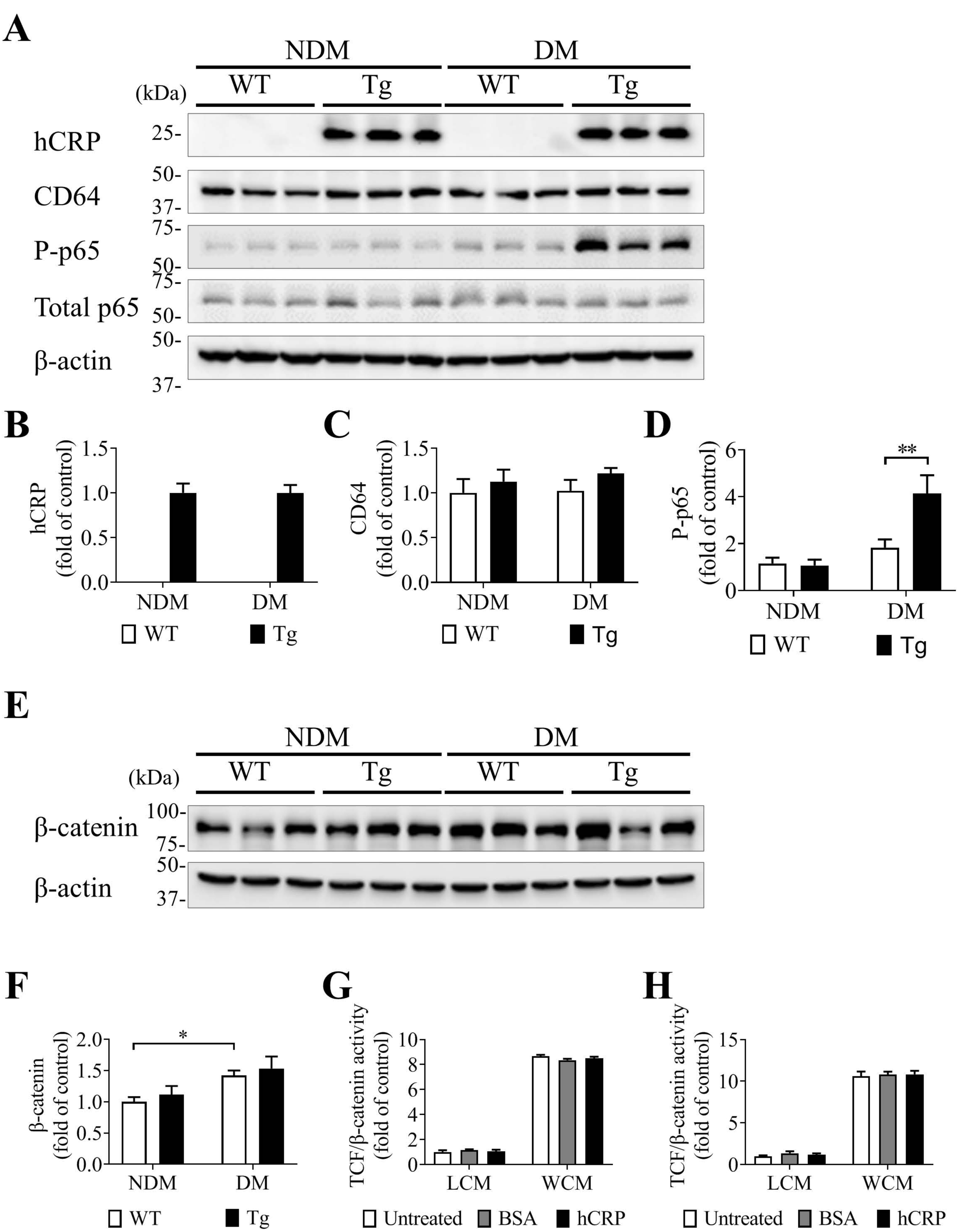


B



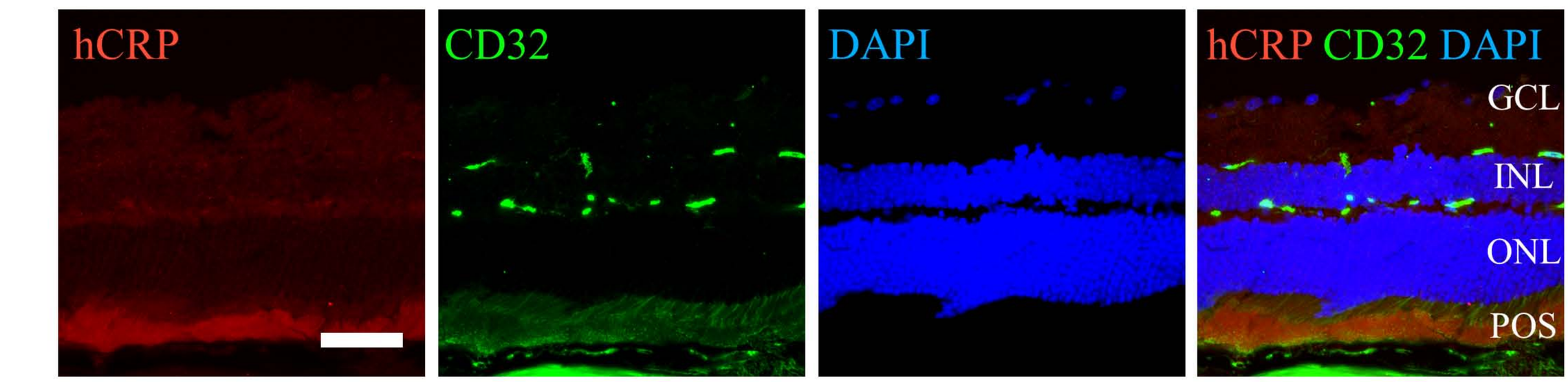
Supplementary Figure 4

Supplementary Figure 4. hCRP over-expression promoted diabetes-induced tyrosine nitration in retinal sections. *A*: Representative images of 3-nitrotyrosine (3-NT) immunostaining in retinal sections of 6-month-old non-diabetic WT rats (NDM WT), non-diabetic hCRP-Tg rats (NDM Tg), diabetic WT rats (DM WT), and diabetic hCRP-Tg rats (DM Tg). *B*: Quantification of 3-NT intensity.Scale bar = 25 μm. GCL: ganglion cell layer, INL: inner nuclear layer, ONL: outer nuclear layer. Data were presented as mean ± SEM(n = 5-6). * $P < 0.05$, ** $P < 0.01$.



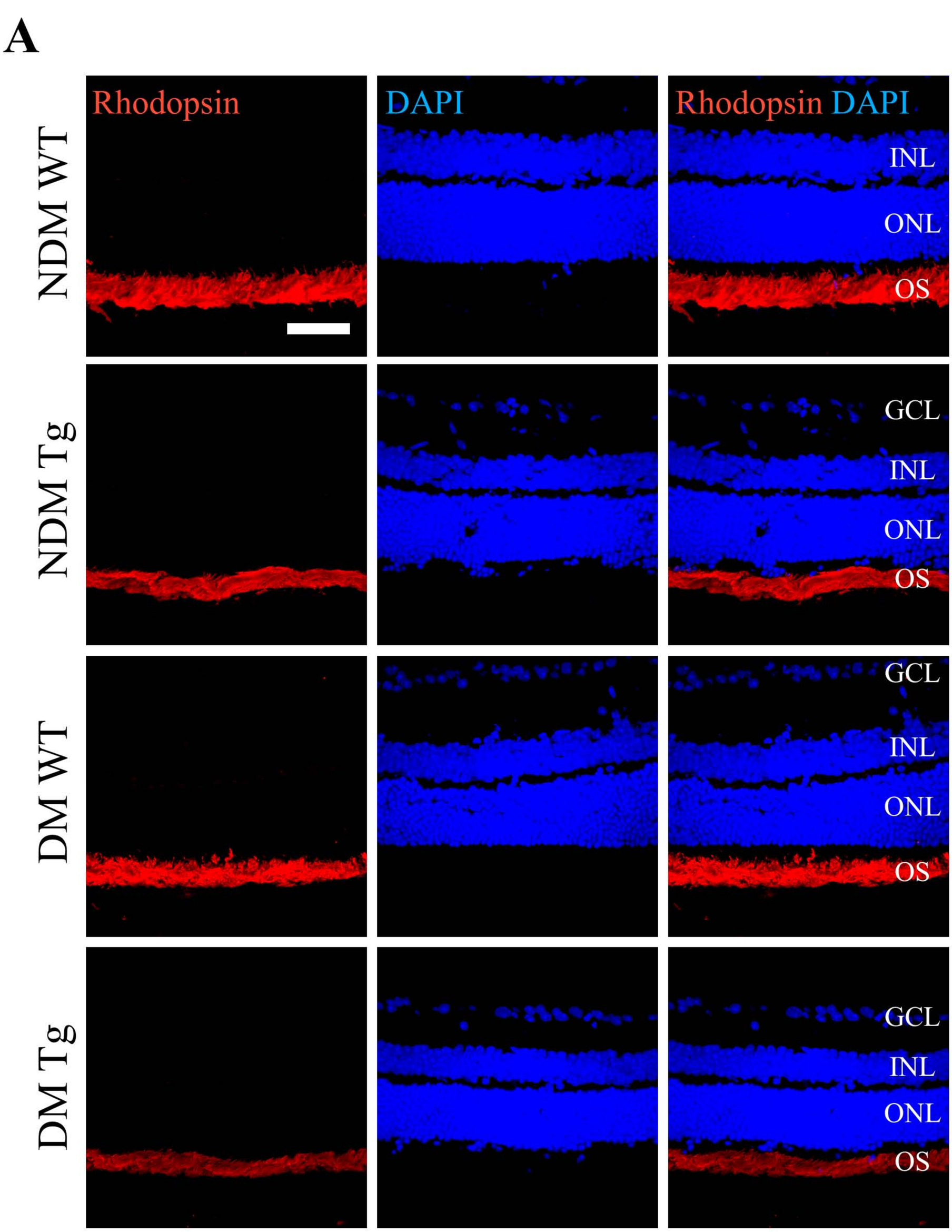
Supplementary Figure 5

Supplementary Figure 5. The effect of hCRP on NF-κBsignaling and Wnt pathway. *A*: Representative images of Western blotting for hCRP, CD64, phosphorylated p65 (P-p65) and total p65 in the retinas of 6-month-old diabetic WT rats and hCRP-Tg rats. *B*-*D*: Levels of hCRP (*B*), CD64 (*C*), and P-p65 (*D*) in *A* were quantified by densitometry. P-p65 were quantified and normalized to total p65. *E*: Representative images of Western blotting for β-catenin in the retinas of 6-month-old WT and hCRP-Tg rats in the presence or absence of STZ-induced diabetes. *F*:Levels of β-catenin in *A*were quantified by densitometry. *G*and *H*: The effect of hCRP on Wnt signaling was investigated in rMC-1 cells (*G*) and ARPE-19 cells (*H*). The cells were transiently transfected with TOPFLASH and pRL-TK plasmids. Then, the cells were treated with hCRP (10 μg/ml) were cultured for 16 hr in the presence of 20% L conditioned medium or Wnt3A conditioned medium. The untreated cells and BSA-treated cells were used as controls. Then, the cells were harvested for luciferase assays. Data were presented as mean ± SEM (n = 6). ** $P < 0.01$.



Supplementary Figure 6

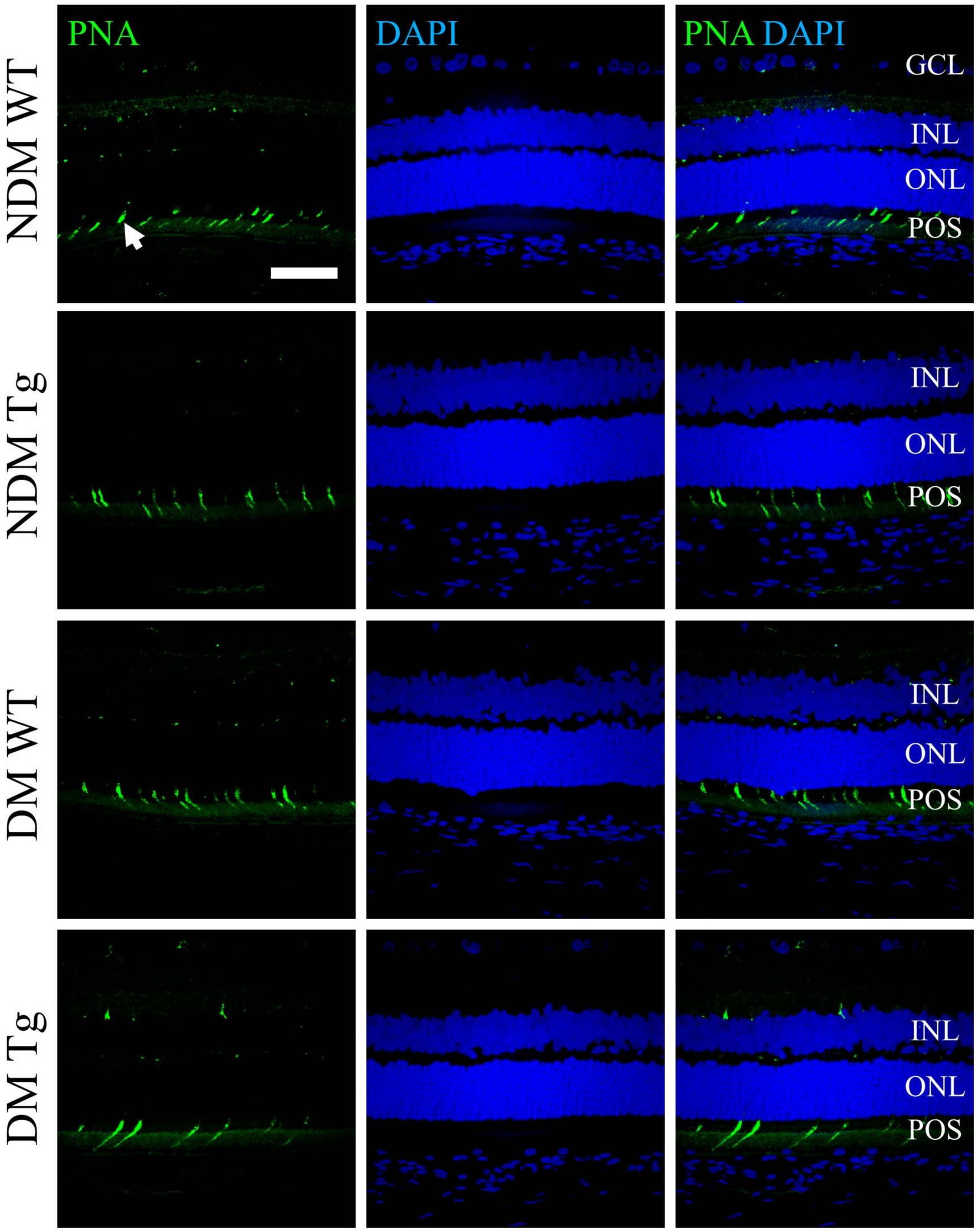
Supplementary Figure 6. Localization of hCRP and CD32a in the retinal sections.*A*: Representative images of hCRP(red) and CD32 (green) immunostaining in retinal sections of 6-month-old diabetic hCRP-Tg rats. The nuclei were counterstained with DAPI (blue).GCL: ganglion cell layer, INL: inner nuclear layer, ONL: outer nuclear layer, POS: photoreceptor outer segments. Scale bar = 25 μm.



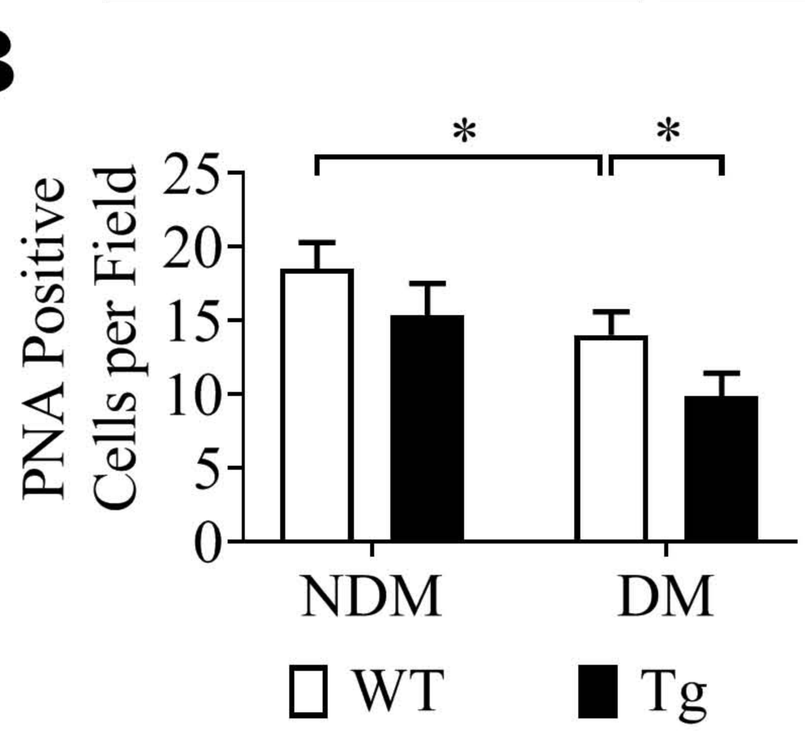
Supplementary Figure 7

Supplementary Figure 7. hCRP over-expression promoted diabetes-induced degeneration of rod photoreceptors. *A*: Representative images of rhodopsin (1D4) immunostaining in the retinal sections of 6-month-old non-diabetic WT rats (NDM WT), non-diabetic hCRP-Tg rats (NDM Tg), diabetic WT rats (DM WT), and diabetic hCRP-Tg rats (DM Tg). *B*: Quantification of rhodopsin signal intensity. GCL: ganglion cell layer, INL: inner nuclear layer, ONL: outer nuclear layer, OS: outer segment. Scale bar = 25 μm. Data were presented as mean ± SEM (n=6). * $P < 0.05$.

A



B



Supplementary Figure 8

Supplementary Figure 8. hCRP overexpression promoted diabetes-induced degeneration of cone photoreceptors. *A*: Representative images of peanut agglutinin (PNA) staining in the retinal sections of 6-month-old non-diabetic WT rats (NDM WT), non-diabetic hCRP-Tg rats (NDM Tg), diabetic WT rats (DM WT), and diabetic hCRP-Tg rats (DM Tg). *B*: Quantification of PNA-labeled cone cells. GCL: ganglion cell layer, INL: inner nuclear layer, ONL: outer nuclear layer, POS: photoreceptor outer segments. White arrowhead indicate the PNA-positive cell. Scale bar = 25 μm. Data were presented as mean ± SEM (n=6).

Supplemental Table 1: List of antibodies used in this study

Antibodies	Host & Type	Dilution	Company	Catalogue No.
Albumin	Goat	1:1000	Bethyl Laboratories	A90-134A
Brn-3a	Mouse	1:100	Santa Cruz	sc-8429
CD32	Goat	1:1000	R&D	AF1875
CD64	Rabbit	1:1000	Abcam	ab203349
Human CRP	Rabbit	1:1000	Abcam	ab32142
ICAM-1	Goat	1:200	Santa Cruz	sc-1511
Peanut Agglutinin	-	1:500	Vector Laboratories	FL-1071
Phospho-Ikkα/β	Rabbit	1:500	Cell Signaling	2697
Phospho-NF-κB p65	Mouse	1:500	Cell Signaling	3036
Prox-1	Rabbit	1:200	BioLegend	925201
Rhodopsin (1D4)	Mouse	1:1000	Abcam	ab5417
TNF-α	Rabbit	1:1000	Abcam	ab6671
Total β-catenin	Mouse	1:1000	Santa Cruz	sc-393501
Total Ikkα	Rabbit	1:1000	Cell Signaling	2682
Total Ikkβ	Rabbit	1:1000	Cell Signaling	2678
Total NF-κB p65	Rabbit	1:1000	Abcam	ab16502
VEGF	Rabbit	1:1000	Abcam	ab46154
β-actin	Goat	1:2000	Santa Cruz	sc-1616
3-nitrotyrosine	Mouse	1:100	Abcam	ab61392
Anti-goat IgG	Horse	1:3000	Vector Laboratories	PI-9500
Anti-mouse IgG	Horse	1:3000	Vector Laboratories	PI-2000
Anti-mouse IgG, Alexa Fluor 488	Donkey	1:250	ThermoFisher	R37114
Anti-rabbit IgG	Goat	1:3000	Vector Laboratories	PI-1000
Anti-rabbit IgG, Alexa Fluor 488	Donkey	1:250	ThermoFisher	R37118