Supplementary Materials

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Supplementary Figure Legends

FIGURE S1. Carnosic acid **(**CA)-induced upregulation of anti-oxidant and ARE-related genes by microarray analysis. ARPE-19 cell lines were treated with CA or vehicle, and total RNA was extracted and used for expression analysis of ARE-related genes. Fold-change after CA treatment compared to vehicle is plotted.

FIGURE S2. Effect of CA on Nrf2 translocation into the nucleus of ARPE-19 cells. Retinal cells were treated with vehicle (**A**) or CA (**B**) for 24 hours. Cells were then fixed, processed for immunostaining, and Nrf2 protein visualized by FITC-conjugated secondary antibody. DNA was counterstained with Hoechst.

FIGURE S3. Concentration of CA in plasma and retina after intraperitoneal (i.p.) injection in rats. Liquid chromatography/mass spectrometry (LC/MS/MS) analysis of carnosic acid in plasma (**A**) and neural retina (**B**). Rats received one or two doses of CA at a concentration of 25 mg/kg/d by i.p. injection. Then, 24 hours after the last injection, plasma and neural retina were collected and analyzed for CA (n = 3 for each group).

FIGURE S4. Retinal morphology in control animals treated with CA but not exposed to light. (**A**) Retinal ONL thickness in rats not exposed to light damage (LD) or drug. Values are mean + SEM. (**B**) ONL thickness for 3 groups of control rats: no LD-no treatment (n = 4), no LD-Vehicle treatment (n = 4), and no LD-CA treatment (n = 4). Values represent mean ± SEM. Groups were compared by two-way ANOVA and found not to be significantly different ($P > 0.05$).

FIGURE S5. Cell death detected by TUNEL analysis in the ONL of rat retinas. (**A** and **B**) Photomicrographs of retinal sections from rats exposed to damaging light and treated with vehicle (**A**) or CA (**B**). Apoptotic TUNEL-positive cells (red), DNA stained with Hoechst (blue). (**C**) Bar graph of Tunel-positive cells in the retinal ONL. Rats were divided into the following groups: (i) not exposed to light damage (LD) and non-treated, (ii) not exposed to LD and treated with CA, (iii) exposed to LD and treated with CA, and (iv) exposed to LD and treated with vehicle (n = 8 total). TUNEL-positive cells were counted in 14 targeted fields within each retina chosen to cover the area most susceptible to light damage. Values represent mean ± SEM (****$P < 0.0001$ by one-way ANOVA). GraphPad Prism 5 software was used for data analysis and graphical representation.

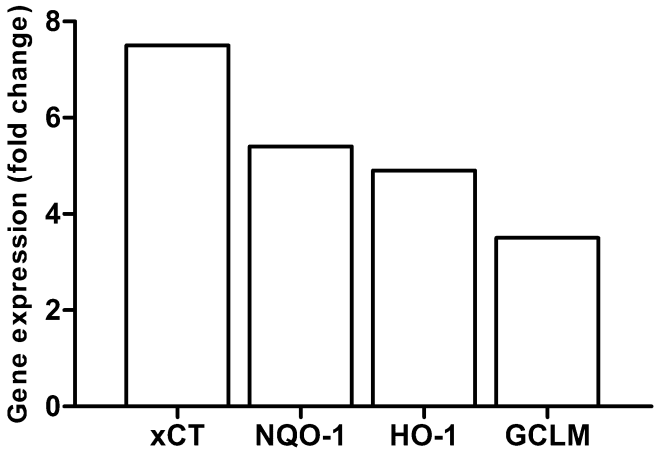

Figure S1

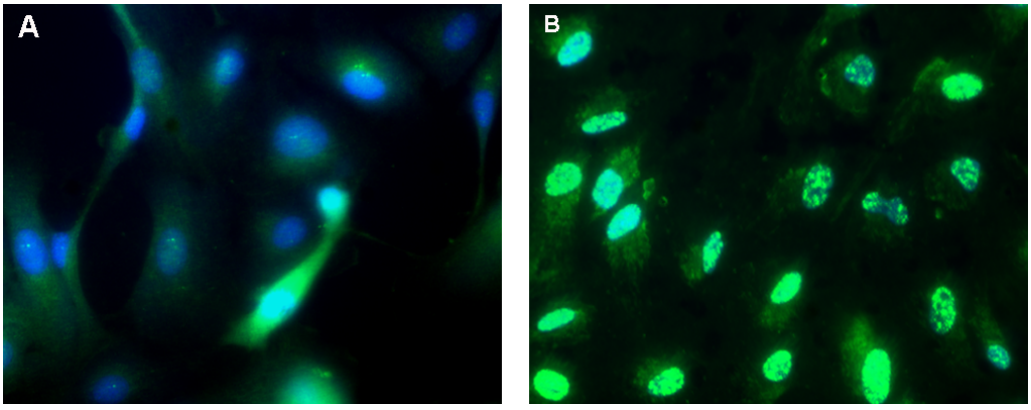

Figure S2

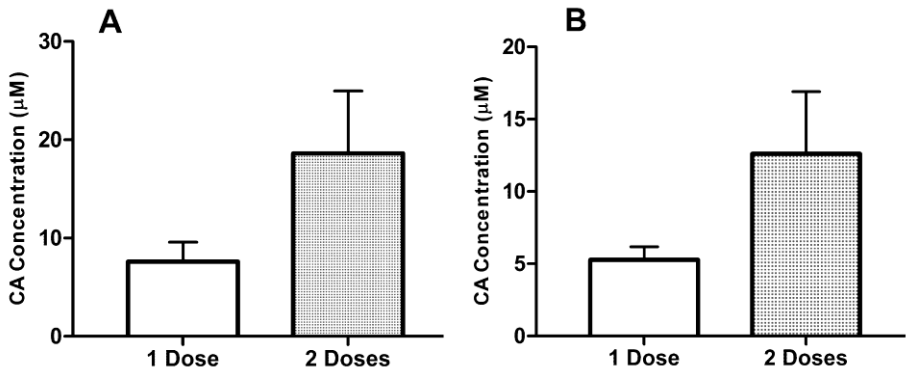

Figure S3

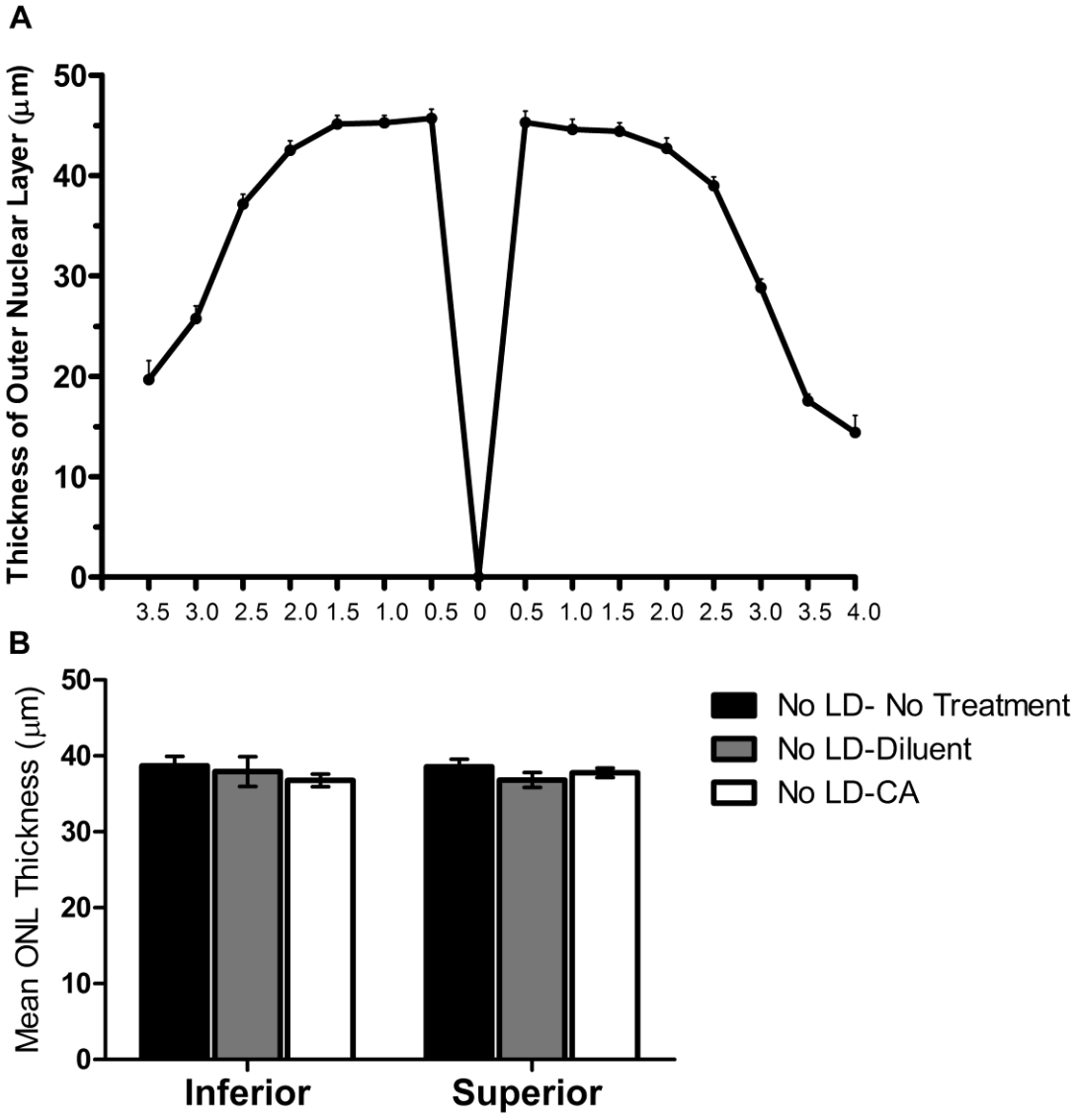

Figure S4

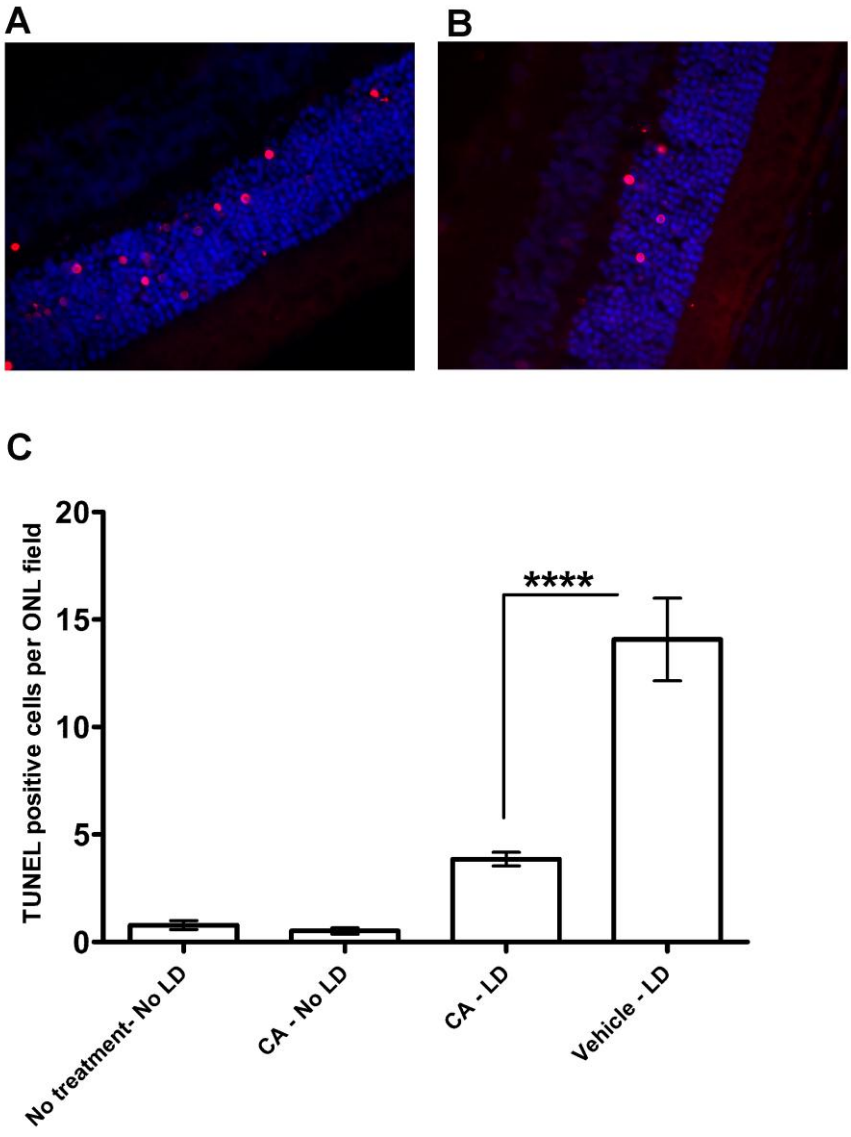
A
B
C
TUNEL positive cells per ONL field
20
15
10
5
0
****
No treatment- No LD
CA - No LD
CA - LD
Vehicle - LD

Figure S5