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Neuropsin, TRPV4 and intracellular calcium mediate intrinsic photosensitivity in corneal epithelial cells

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ABSTRACT

Purpose: To investigate intrinsic phototransduction in the corneal epithelium and its role in intracellular and inflammatory signaling.
Methods: Optical imaging in isolated corneal epithelial cells (CECs) and debrided epithelia was combined with molecular, biochemical, pharmacological assays and gene deletion studies to track UVB-induced calcium signaling and release of cytokines, chemokines and matrix remodeling enzymes. Results from wild type mouse CECs were compared to data obtained from *Opn5*$^{-/-}$ and *Trpv4*$^{-/-}$ cells.
Results: UVB stimuli and TRPV4 activity induced epithelial release of IL-1β, IL-17, matrix metalloproteinases MMP-3/MMP-9, and thymic stromal lymphopoietin (TSLP). UVB stimuli evoked $[Ca^{2+}]_i$ elevations in dissociated mouse CECs that were partially reduced by inhibition of TRPV4 channels, *Trpv4* knockdown and replacement of control saline with Ca^{2+}-free saline. UVB-induced Ca^{2+} responses were significantly suppressed by OPN5 deletion and by inhibition of phospholipase C signaling, and responses were abrogated in cells with depleted intracellular Ca^{2+} stores.
Conclusions: Mammalian CECs are intrinsically and constitutively photosensitive. UVB photons are transduced by neuropsin, phospholipase C and CICR signaling, with mouse but not human CE transduction exhibiting a UVB-sensitive TRPV4 component. TRPV4 activity and UVB transduction are linked to cell-autonomous release of proinflammatory, matrix remodeling and nociceptive interleukins and MMPS. TRPV4-induced cytokine release may contribute to the pain induced by mechanical injury of the cornea and CEC photosensing may alert and protect the visual system from ultraviolet B (UVB) radiation -induced snow blindness, injury, vision loss and cancer.

1. Introduction

Our environment is permeated by ultraviolet (UV) photons that cover ~2 % of the solar spectrum, are invisible to us yet profoundly affect our health [1–3]. Biological effects of UV radiation are predominantly mediated by the UVB band covering the 280–315 nm range. Overexposure may induce skin and eye injuries such as sunburn, snow blindness, *xeroderma pigmentosum*, pain, vision loss, cancers as well as accelerate aging [4–7]. Due to larger pupils and more transparent ocular media, children are especially vulnerable to UVR with up to 80 % of a person's lifetime exposure to UV light reached before the age of 18 [8]. Pigmentation (tanning) and sunscreen formulations help prevent UV-induced aging and carcinogenesis [9–12] but cannot prevent injuries of the eye.

The visual system is protected from UV radiation by the cornea and the conjunctiva. ~2 % of UVB photons reach the lens and ~1 % the retina, with ~95 % of absorption taking place within the stratified nonkeratinizing corneal epithelium (CE) and anterior stroma [5,13,14]. The CE is particularly vulnerable to UVB injury [15], with a single UVB dose sufficient to induce genetic and cellular remodeling [16–18] whereas chronic stimulation forces senescence of the limbal epithelium,

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Abbreviations

CE	corneal epithelium
CICR	Ca^{2+}-induced Ca^{2+} release
CPA	cyclopiazonic acid
ECM	extracellular matrix
GSK101	GSK1016790A (TRPV4 agonist)
HC-06	HC067047 (TRPV4 antagonist)
IP_3R	inositol triphosphate receptor
MMP	matrix metalloproteinase
TRPV4	transient receptor potential vanilloid isoform 4
UVB	ultraviolet light B spectrum

increases the risk for *pterygium* and *pingueculae* (conjunctival overgrowth) and climatic droplet keratopathy (corneal opacity) [19,20], and may result in partial or total blindness due to photokeratitis (snow blindness), cataract formation, corneal edema, loss of angiogenic privilege, and ocular melanomas [4,20–22]. Photokeratitis, the corneal analog of sunburn, is associated with edema and opacification from damaged CE, stroma and endothelium [23]. CECs respond to UVB radiation with nuclear translocation of NF-kB [24], release of matrix metalloproteinases (MMPs), production of reactive oxygen species, inflammasome activation, mitochondrial dysregulation, CEC sloughing, extracellular matrix (ECM) degradation, barrier disruption, edema formation and afferent overexcitation [25–31]. UVB-induced CE injury is experienced as cytokine-induced pain mediated by subepithelial afferents from the ophthalmic branch of the trigeminal nerve and may be associated with temporary vision loss [32,33] yet despite the pervasiveness and clinical burden associated with UVB -induced pathologies, the identity of UVB sensors within the CE, their downstream effectors and relationship to inflammatory and nociceptive pathways remain poorly understood [20].

The past two decades have linked vertebrate light sensing to an astounding complexity of transduction mechanisms in which canonical phototransduction, mediated by OPN1-SW cone opsins and OPN2 (rhodopsin) occurs alongside nonvisual transduction mediated through TRP channels [34], cryptochromes [35], OPN2 [36], OPN3 (panopsin) [37,38], OPN4 (melanopsin) [34,39,40], and OPN5 (neuropsin) [41–45] molecules. TRPV1, TRPA1 and/or TRPV4 channels have been implicated in keratinocyte, melanocyte, fibroblast and skin afferent UV sensing and inflammation [46–51] and opsins mediate photosensitivity in keratinocytes, lymphocytes, Jurkat cells, fibroblasts, melanocytes, adipocytes and subsets of retinal, spinal cord and hypothalamic neurons [37,41,52,53]. Examples of opsin- TRP collaboration include blue-light-induced melanogenesis mediated through OPN3 - TRPV1 interactions [38] and OPN4 - TRPC6/7 coupling that subserves ipRGC phototransduction [34,54]. OPN5 contributes to retinal and corneal circadian rhythmicity, wound healing and vascular development [43,44,55] but it is not known whether OPN5-based signaling coexists with TRP-based photosensing.

Inspired by the report that activation of the polymodal Ca^{2+}-permeable channel TRPV4 (Transient Receptor Potential Vanilloid isoform 4) is obligatory for UVB transduction in skin keratinocytes [47] and taking into account strong TRPV4 expression in mammalian CEs [56–58] and the similarity between molecular and functional properties of keratinocytes and CECs [59], we investigated the TRPV4-dependence of UVB sensing in corneal epithelia. We found that CECs are intrinsically photosensitive but mainly rely on rhabdomeric-like signaling to mediate the effects of clinically relevant UVB dosing.

2. Methods

2.1. Ethical approval and animals

Animal handling and experiments followed institutional guidelines set by University of Utah IACUC (Protocol #22–02005) and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals, and the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research. The experiments involving human tissue followed The Declaration of Helsinki and The Utah Lions Eye Banks protocols. C57BL/6, *Trpv4*$^{-/-}$ mice (obtained from Dr. Wolfgang Liedtke, Duke University) [60,61] and *Opn5*$^{-/-}$ mice [55] were maintained in a pathogen-free facility with a 12-h light/dark cycle and unrestrained access to food and water. KO animals were phenotyped as described previously. Data were gathered from 1- to 3-month-old male and female animals with no noted gender differences.

2.2. Reagents

The TRPV4 agonist GSK1016790A (GSK101), antagonist HC-067047 (HC-06) and U-7322 were purchased from Sigma (St. Louis, MO, USA) or Cayman Chemical (Ann Arbor, MI, USA). Cyclopiazonic acid (CPA) was obtained from Tocris (Bristol, UK), and other salts and reagents were obtained from Sigma, VWR (West Chester, PA, USA), Acros Organics (Pittsburgh, PA, USA), or ThermoFisher (Waltham, MA, USA).

2.3. Tissue preparation

Corneas were dissected from enucleated eyeballs and placed in Dulbecco's Modification of Eagle's Medium (DMEM)/F12 (1:1 mixture, GIBCO (Grand Island, NY, USA)/Thermo Fisher) containing Dispase II (15 mg/mL, Sigma) and 1 % penicillin/streptomycin for 1 h at 4 °C and an additional hour at room temperature [56]. Epithelial sheets were peeled off and used *in situ*. Dissociated mouse CECs (mCECs) were incubated in DMEM (GIBCO/Thermo Fisher) containing papain (15 U/mL, Worthington (Columbus, OH, USA)) for 30 min, rinsed with D-PBS containing 0.5 % bovine serum albumin (BSA, Genesee Scientific) and triturated. Human corneas were handled similarly, with Dispase II incubation prolonged to overnight at 4 °C.

2.4. Real-time PCR

Semiquantitative RT-PCR followed protocols described in Refs. [56,62,63]. Total RNA was isolated using the Arcturus PicoPure RNA isolation kit (Thermofisher Scientific) and cDNA generated using qScript XLT cDNA Supermix (Quanta Biosciences). Real-time PCR was conducted with 2X GREEN Master Mix (Apex Bioresearch Products). *Gapdh* was used as an endogenous control to normalize fluorescence signals. Gene expression relative to *Gapdh* was measured using the comparative CT method ($2^{-[\Delta CT(gene)- \Delta CT(GAPDH)]}$). Genes were assessed from 6 individual CEs isolated from WT mouse eyes, with data normalized relative to OPN1SW expression. Primer sequences, expected product length, and gene accession # are provided in STable 2.

2.5. Cytokine detection

Cytokine release from mouse CE was tracked with the C-Series Mouse Cytokine Antibody Array C1000 (RayBiotech, Peachtree Corners, GA, USA), with the membranes, antibody cocktails, buffers and streptavidin used per manufacturer's instructions. Following dissociation, cells were evenly distributed among vials and DMEM/F12 was added to each vial together with the NPTDase inhibitor ARL 67156 (100 μM) and protease inhibitor cocktail (15 μL/mL). The cells' exposure to pharmacological agents was 30 min and UVB irradiation 3 min. The membranes were visualized by photographic film and the intensity was quantified using ImageJ software (NIH, Bethesda, MD, USA). The samples were

evaluated as duplicates in two independent experiments.

2.6. UVB irradiation and optical imaging

Detached corneal epithelial sheets and dissociated cells were placed in the recording chamber (Warner Instruments (Hamden, CT, USA)) and loaded with Fura-2 AM (5–10 μM, Life Technologies) for 30–45 min. The chamber was placed under an upright Nikon (Tokyo, Japan) microscope with 40x (0.80 NA water) objective. The coverslip at the bottom of the recording chamber was fused quartz as per GE 124 standard (VWR, West Chester, PA, USA) with high 260–300 nm transmittance. The UVB light source (UVTOP290H LED, Roithner Lasertechnik, Vienna, Austria) was placed underneath the recording chamber, with irradiation density at the corneal epithelial sheet ~150 μW/mm^2. CE sheets were superfused with extracellular saline solution containing (in mM): 133 NaCl, 10 HEPES hemisodium salt, 10 glucose, 2.5 KCl, 2 CaCl2, 1.5 MgCl2, 1.25 NaH2PO4, 1 pyruvic acid, 1 lactic acid, and 0.5 glutathione at pH 7.4 with solution exchanges performed via an electronically controlled multibarrel inlet port (Warner Instruments, Hamden, CT). CE cells were irradiated by 295 nm light for 3 min. For calcium imaging, 340 and 380 nm excitation was delivered via a Xe lamp (Lambda DG-4; Sutter Instruments, Novato, CA, USA) and the emission collected at 510 nm with 14-bit CoolSNAPHQ2 camera (Photometrics, Tucson, AZ, USA) [56,64,65] with image acquisition paused for the duration of UVB irradiation. Backgrounds were subtracted and F340/F380 ratios computed with NIS-Elements software (Nikon, Lockbourne, OH); ΔR/R (peak F340/F380 – baseline/baseline) was used to quantify the amplitude of Ca^{2+} signals, in which R is the ratio of emission intensity at 510 nm evoked by 340 nm excitation versus emission intensity at 510 nm evoked by 380 nm excitation.

2.7. Statistical analysis

Data analysis and statistical tests were performed using Origin Pro 8.5 (Northampton, MA, USA). Unless otherwise stated, data were acquired from at least three independent experiments. Results are given as mean ± SEM. Unpaired sample *t*-test was used to compare two means, and 1- or 2-way ANOVA with Tukey's test to analyze three or more means. $P > 0.05$ = nonsignificant (N.S.), $P \leq 0.05$ = *, $P \leq 0.01$ = **, $P \leq 0.001$ = ***, $P \leq 0.0001$ = ****.

3. Results

3.1. UVB drives CEC release of proinflammatory molecules

UVB -induced inflammation may compromise the integrity of the CE barrier via fibrosis and photokeratitis and contribute to corneal pain [18,30,57,66]. To circumvent keratocyte and afferent involvement, we experimentally debrided the CE layer and tracked UVB -dependent cytokine/chemokine release with a chemiluminescence assay. UVB evoked >50 % increase in release for 30/96 tested proteins that included interleukins IL-1β ($P < 0.001$), IL-2β ($P < 0.05$), IL-17 ($P < 0.01$), metalloproteinases MMP-3 ($P < 0.05$), proMMP-9 ($P < 0.005$), and thymic stromal lymphopoietin (TSLP) ($P < 0.001$) (Fig. 1A and STable I). In addition, we observed upregulation of the TNF receptor superfamily member 8 (CD30/TNFRS8), Cluster of Differentiation 40 (CD40), Insulin-like Growth Factor-binding Protein 3 (IGF-BP3), leptin, Macrophage Inflammatory Protein (MIP), Monokine Induced by Gamma/chemokine Ligand 9 (MIG/CXCL9), Chemokine (C-C motif) Ligand 5 (CCL5/RANTES), E-selectin, osteopontin, Sonic hedgehog (Shh-N), and vascular endothelial growth factor receptors 1 and 3 (VEGFR1, VEGFR3) proteins (STable I). UVB stimuli thus drive CE-autonomous release of multiple proinflammatory cytokines, chemokines, proangiogenic and ECM remodeling enzymes.

3.2. TRPV4 activation induces corneal epithelial cytokine release

Secretion of neuroactive factors from epithelia generally reflects changes in the intracellular concentration of calcium, a 2nd messenger that regulates signaling, proliferation and differentiation, with TRP channels constituting an important venue for Ca^{2+} influx [67–70]. We previously reported that exposing mouse CECs to the selective TRPV4 agonist GSK1016790A (GSK101) induces $[Ca^{2+}]_i$ elevations that reach the peak within ~3 min and are followed by a gradual decrease to a stable plateau [56]. To test the role of TRPV4 in cytokine release [47], we stimulated debrided mouse CE sheets with 3 min exposure to GSK101 (25 nM). The agonist facilitated >50 % release of 24/96 tested proteins (STable 1), with 12 proteins, including IL-1β ($P < 0.05$), IL-2 ($P < 0.0001$), IL-3 ($P < 0.05$) and TSLP ($P < 0.05$) showing augmented release in response to both GSK101 and UVB stimuli (Fig. 1B) (orange fields; STable 1). The specificity of TRPV4 activation was validated by blocking GSK101-evoked release with the selective antagonist HC067047 (HC-06; 1 μM) (Fig. 1B).

3.3. Mouse and human corneal epithelia are intrinsically photosensitive

To explore the UVB-dependence of CEC Ca^{2+} homeostasis under conditions that minimize contributions from purinergic signaling, afferent, and cell-cell interactions, we dissected CEs from mouse and human corneas, plated dissociated cells onto UV-permeant quartz glass (*inset* Fig. 2B) and loaded them with the ratiometric Ca^{2+} indicator Fura-2-AM (5–10 μM). 3 min exposure to 295 nm light consistently elevated $[Ca^{2+}]_i$ (Fig. 2Ai-iii), with average peak fluorescence increasing ~4-fold (from 0.06 ± 0.00 to 0.21 ± 0.01; n = 68 cells, N = 4) ($P < 0.0001$) (Fig. 2). We conducted a proof-of-principle experiment in cells isolated from 3 human donors. Supplemental Fig. 1 shows that human

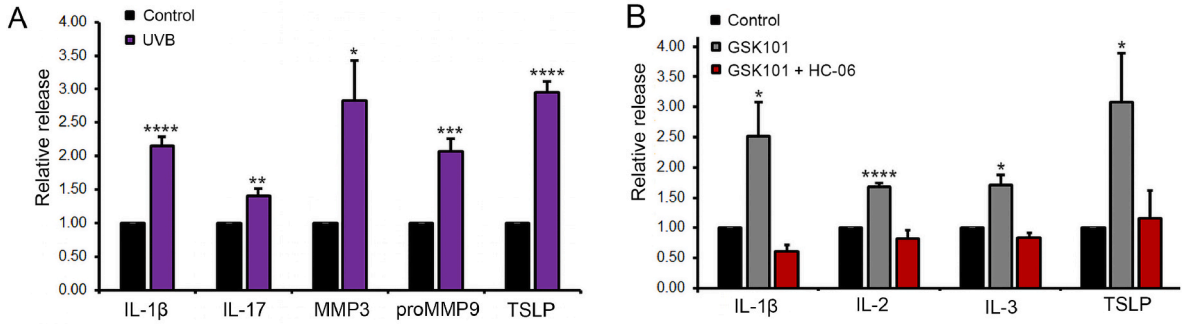

Fig. 1. Debrided mouse CE. (A) UVB stimulation promotes release of inflammatory mediators and matrix remodeling enzymes. Debrided mouse CE, chemiluminescent dot assay. Averaged data from 2 independent experiments conducted in duplicate, each sample contained 6 isolated CE. UVB stimulates release of interleukin isoforms, MMPs and TSLP. (B) The TRPV4 agonist GSK101 evokes release of inflammatory mediators and matrix remodeling enzymes. The antagonist HC-06 was co-applied in parallel experiments to validate the specificity of TRPV4 activation. ± SEM, ****$P < 0.001$, ***$P < 0.005$, **$P < 0.01$, *$P < 0.05$.

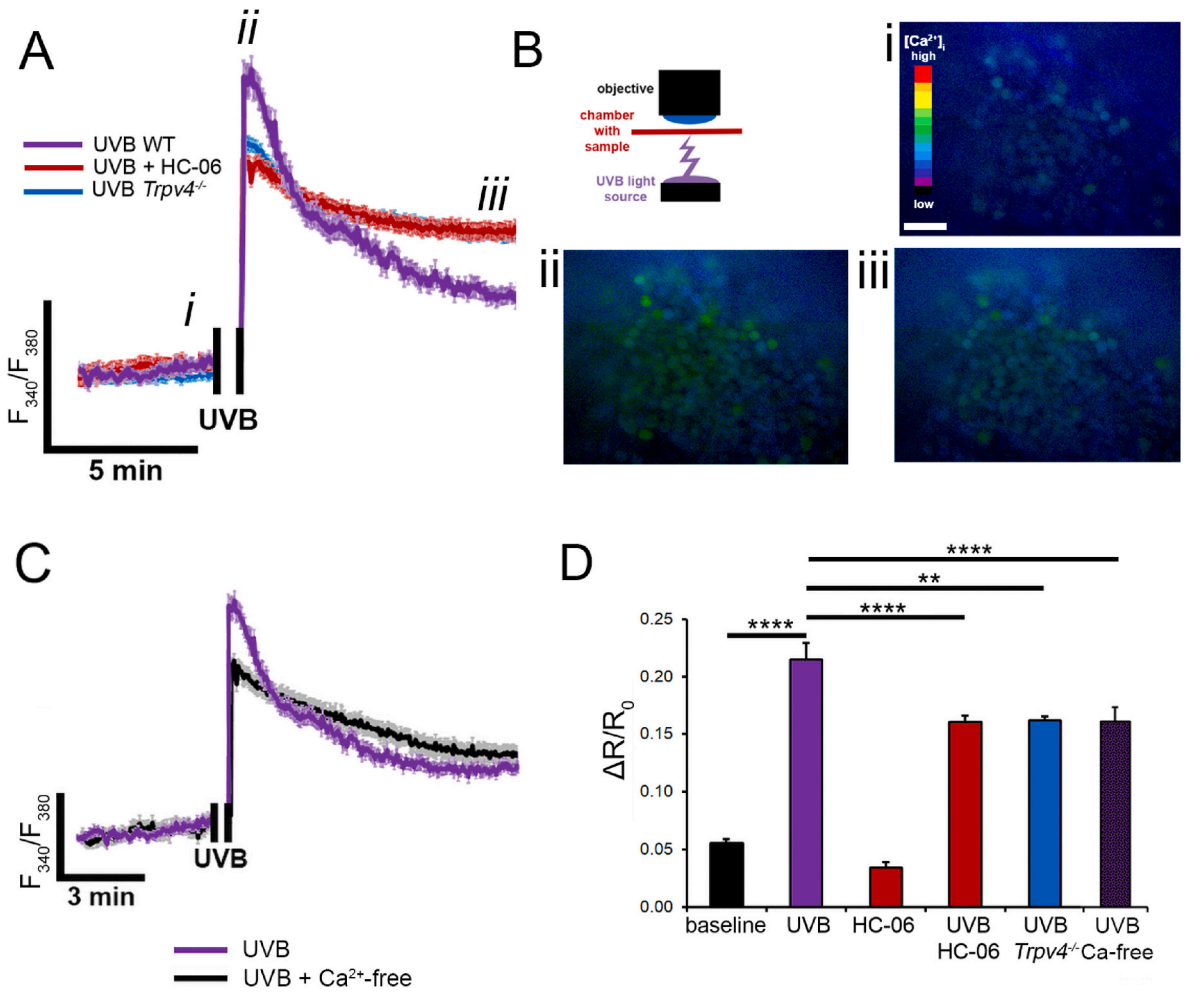

Fig. 2. Mouse CE loaded with Fura-2 AM. UVB stimulation induces $[Ca^{2+}]_i$ increases consisting of peak response that relaxes into a plateau. (A) Representative Ca^{2+} trace from ROIs placed on somata before (i), immediately after (ii) and 10 min after UVB stimulus, with superposed traces from control, HC-06-treated and *Trpv4*$^{-/-}$ CE. Fluorescence acquisition was paused during UVB stimulation. (B) Raw images depicting the spatiotemporal changes in the fluorescence ratio at i-iii time-points. Inset, Schematic representation of the experimental configuration. (C) Averaged data show ~4-fold $[Ca^{2+}]_i$ increase in UVB-stimulated eyes ($P < 0.001$), with TRPV4 inhibition (HC-06), deletion (*Trpv4*$^{-/-}$) and Ca^{2+}-free saline inducing comparable reductions in $[Ca^{2+}]_{CE}$. ****$P < 0.001$, ***$P < 0.005$, **$P < 0.01$, *$P < 0.05$.

preparations respond to UVB with an increase in Fura-2 fluorescence (from 0.05 ± 0.01 to 0.19 ± 0.02; n = 46; $P < 0.0001$). Hence, mammalian corneal epithelia are intrinsically sensitive to UVB light.

We tested whether the UVB-dependence of $[Ca^{2+}]_i$ signals requires TRPV4 involvement using the selective antagonist HC067047 (HC-06; 1 μM), by testing signals in *Trpv4*$^{-/-}$ cells and by replacing the control saline with Ca^{2+}-free saline. HC-06 reduced the amplitude of the UVB-evoked $[Ca^{2+}]_i$ response by ~25 % (to 0.16 ± 0.01; N = 4; n = 76; P

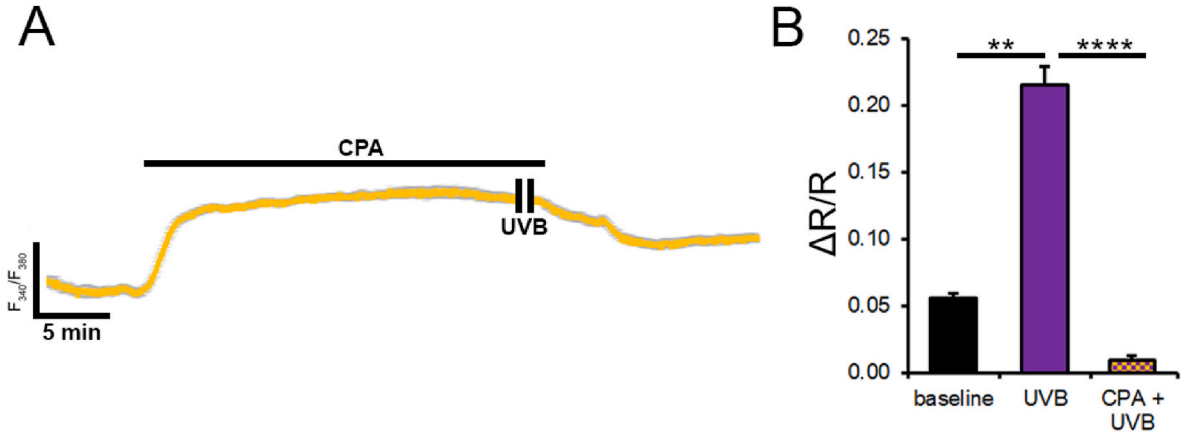

Fig. 3. CICR is obligatory for UVB-evoked Ca^{2+} signaling. Dissociated cells. (A) Representative trace of the UVB-evoked Ca^{2+} response in CPA-treated cells. Store depletion is associated with abolition of UVB-evoked. (B) Averaged UVB-evoked response amplitude under control conditions and in the presence of CPA. **$P < 0.01$.

< 0.0001), with CECs isolated from *Trpv4*$^{-/-}$ corneas showing comparable reductions in the response amplitude ($\Delta R/R = 0.16 \pm 0.01$; $N = 2$; $n = 45$; $P < 0.01$) (Fig. 3). The average response in Ca^{2+}-free saline was comparable to signals in HC-06-treated and *Trpv4*$^{-/-}$ CECs ($\Delta R/R = 0.16 \pm 0.01$) ($N = 2$; $n = 45$, $P < 0.01$) (Fig. 2A and B), indicating that (i) TRPV4 constitutes the principal component of UVB-induced transmembrane Ca^{2+} influx but (ii) mediates a fraction of the overall calcium response. In contrast to the mouse preparation, pilot experiments in human CECs suggest that UVB -induced Ca^{2+} elevations does not involve TRPV4 activation ($P = 0.079$) (SFig. 1).

3.4. UVB -induced [Ca^{2+}]i elevations require release from intracellular stores

The induction of UVB-evoked Ca^{2+} signals in the absence of extracellular Ca^{2+}suggests that phototransduction may be upstream from intracellular Ca^{2+} release. We tested the involvement of the ER Ca^{2+} pool using the SERCA transporter blocker cyclopiazonic acid (CPA, 10 μM). As expected [71,72], SERCA inhibition increased cytosolic [Ca^{2+}] (to 0.22 ± 0.01; $n = 64$, $P < 0.0001$) (Fig. 3, yellow bar). Depletion of ER stores also abrogated the UVB-evoked response ($\Delta R/R = 0.01 \pm 0.01$; $n = 64$, $N = 2$; $P > 0.05$), indicating an obligatory role for Ca^{2+}-Induced Ca^{2+} Release (CICR) (Fig. 3).

3.5. The CE-intrinsic photoresponse is mediated by neuropsin and phospholipase C signaling

Results depicted in Figs. 1–3 show that UVB signaling in the mouse CE requires CICR, with TRPV4 channels mediating a residual fraction of the overall evoked Ca^{2+} signal. We considered involvement of neuropsin, a photopigment encoded by the OPN5 gene that has been implicated in corneal circadian photoentrainment and wound healing [43,44]. Comparison of UVB-evoked signals in CECs isolated from wild type and *Opn5*$^{-/-}$ animals revealed a significant ($P < 0.005$) reduction of UVB response amplitude in KO cells (Fig. 4; SFig. 2). Phospholipase C (PLC) inhibition participates in phototransduction in neurons, melanocytes and keratinocytes [34,41,47,73]. Consistent with the presence of rhabdomeric signaling, we found that inhibition of phosphatidylinositol-specific G_q-coupled phospholipases with U-73122 (1 μM) reduces the amplitude of UVB-evoked by ~85 %, an extent roughly comparable to the effect of OPN5 knockdown (Fig. 4; SFig. 2). OPN5 knockdown and PLC inhibition did not affect resting $[Ca^{2+}]_i$ ($P < 0.001$) (SFig. 2), indicating that OPN5-PLC signaling quiescent in the absence of appropriate photic stimuli.

Mouse cornea was suggested to express OPN3 and OPN5 genes [74], with additional OPN4 expression observed in trigeminal neurons [40]. We analyzed expression of genes encoding photosensitive opsins in the mouse epithelium to gain an impression of the overall landscape of CE photosensing. The overall expression pattern was dominated by rhodopsin (OPN2) and melanopsin (OPN4) transcripts, with OPN5 levels comparable to OPN3 (panopsin) (Fig. 5). While TRPV4 expression was

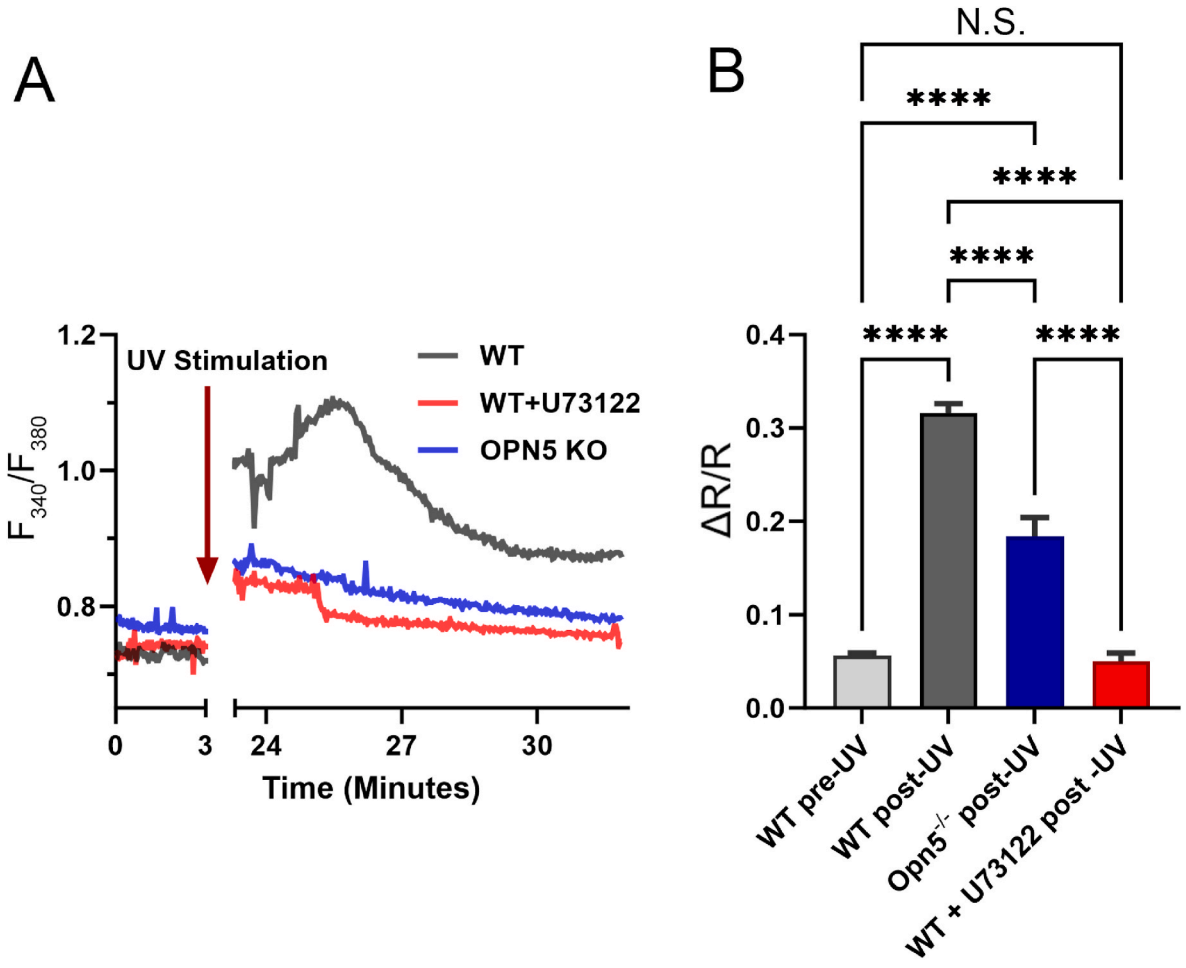

Fig. 4. UVB-evoked Ca^{2+} signals require neuropsin and phospholipase C pathways. Dissociated cells. (A) Representative traces for UVB-evoked Ca^{2+} response in control wild type, *Opn5*$^{-/-}$ CECs and cells treated with the PLC antagonist U-73122 (1 mM). (B) Averaged data for experiments shown in A. ****$P < 0.001$.

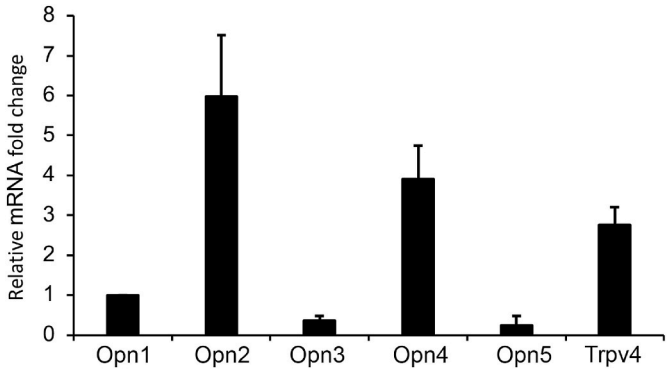

Fig. 5. Relative photo-opsin and *Trpv4* gene expression in the mouse corneal epithelium. Semi-quantitative RT-PCR normalized to Opn1SW (blue cone opsin) mRNA expression levels = 1. (n = 6 epithelia, N = 3 mice).

markedly higher compared to OPN5, these data suggest that light sensing in the CE might be complex, multifactorial and context dependent.

4. Discussion

Photokeratitis induced by UVB radiation is characterized by corneal pain and photophobia that protect the eye from photodamage while constraining behavior through temporary vision impairment or loss. The present study identifies epithelial neuropsin and TRPV4 channels as transducers that couple UVB exposure to release of proinflammatory modulators known to mediate aversive behavior. We show that the photoresponse is intrinsic to the epithelium and requires OPN5 signaling that acts in parallel to TRPV4 activation to promote Ca^{2+} signaling and release of proinflammatory cytokines, chemokines and matrix remodeling enzymes (Fig. 5). We propose that the OPN5/TRPV4-PLC-CICR-cytokine axis functions as a rapid alert system that protects vision from high energy photons. Acute overactivation of this mechanism may lead to snow blindness that protects the eye from photoinjury, while its dysregulation or chronic stimulation could contribute to climatic droplet keratopathy, endothelial dysfunction and angiogenesis. This pathway could be targeted acutely to mitigate the debilitating effects of UVB-induced inflammation and pain.

The UVB stimulus utilized in our experiments (150 μW/mm^2; 3 min) corresponds to flux density of ~27 mJ/mm, approximating ~20 h of outdoor exposure at UV index 10 without sunlight reflection and matching doses used in animal studies [1,75–77] and clinical studies of non-occupational photokeratitis [29,78]. Our observation that a single UVB exposure suffices to evoke [Ca^{2+}] elevations in debrided CEs and/or dissociated CECs (i.e., cells separated from stroma and trigeminal feedback) indicates thatphotosensitivity is an intrinsic property of the corneal epithelium that is tied to intracellular 2nd messenger signaling. Taking into account the TRPV4-dependence of UVB-evoked calcium signals in keratinocytes [47], our initial goal was to assess the involvement of TRPV4 channels in CE photosensing. We found the amplitude of UVB-evoked Ca^{2+} responses in mouse preparations to be partially reduced by inhibition of TRPV4 channels and deletion of the *Trpv4* gene. Because Ca^{2+} removal from extracellular saline reduced mCEC responses to an extent comparable to TRPV4 inhibition/knockdown, TRPV4 likely constitutes the principal effector of UVB-sensitive transmembrane Ca^{2+} influx, with the predominant fraction (~80 %) of the UVB-evoked response independent of TRPV4 activation. While human CECs express TRPV4 [58] and respond to UVB stimuli with calcium elevations (SFig. 1), their insensitivity to HC-06 argues against TRPV4 involvement in UVB transduction. Light sensitivity of the fly TRP channel requires association with rhodopsin (OPN2) and PLC-dependent formation of inositol-1,4,5-trisphosphate (IP_3) and diacylglycerol (DAG) [79,80] whereas studies of vertebrate TRPV1 and TRPA1 isoforms suggest the possibility that the channels might be directly responsive to UV stimuli [48,51,81,82] through mechanisms that are poorly understood but could involve generation of reactive oxygen species, lipid oxydation, lipid peroxidation and covalent modification of cysteine residues [46,47,50,83].

The absorption spectrum of 11-cis-retinal-bound OPN5, a bistable, highly conserved noncanonical opsin (Upton et al., 2022), resembles rod and SWS1/UV (OPN1/2) spectra, with a peak in blue-violet UVA (380 nm λ_{max}) that extends into the UVB (λ_{max} 297 nm) range [41,42]. We've previously observed that *Opn5*$^{-/-}$ mice exhibit normal optokinetic responsiveness [43] and SCN-dependent circadian corneal photoentrainment [44] whereas debriding induces a CE-intrinsic circadian clock ~ 3–4 days after corneal wounding [44]. In contrast to the injury-dependence of CE photoentrainment, our imaging and biochemical experiments suggest that isolated CECs and intact epithelia may be constitutively sensitive to violet light through the OPN5-PLC-CICR axis and auxiliary TRPV4 signaling. Historically considered within the context of bidirectional photoconversion, G_i activation and cAMP lowering [45,84], OPN5 photosensing has been recently associated with the G_q -PLC - IP_3 receptor pathway [85] that resembles rhabdomeric photosignaling in fly photoreceptors, M1-M7 ipRGCs, melanocytes, choroidal endothelial cells and fibroblasts [34,54,86,87]. PLC activation cleaves phosphatidylinositol 4,5-bisphosphate (PIP_2) into IP_3 and diacylglycerol (DAG) to stimulate calcium release from IP_3R-sensitive ER compartments. Consistent with CICR/IP_3R involvement, we found that UVB-evoked [Ca^{2+}]$_i$ signaling largely resists removal of extracellular Ca^{2+} but is abrogated by SERCA and PLC inhibition. The preservation of the photoresponse under Ca^{2+}-free conditions excludes major involvement of Orai/TRPC channels, DAG-sensitive TRPC3/6/7 channels, and $G\beta\gamma_q$-adenylate cyclase signaling. Unexpectedly, CE showed strong expression of rhodopsin and melanopsin genes, and panopsin mRNA levels that were comparable neuropsin expression. Given that rhodopsin was almost certainly desensitized under our experimental conditions, the difference between the suppression of UVB-evoked calcium signals by thePLC inhibitor and OPN5 knockdown (Fig. 4) may reflect auxiliary contributions from OPN1/3/4 mechanisms that remain to be investigated in future work. In any case, our data suggest that UVB-evoked calcium signaling in the corneal epithelium requires an intermediary G_q-PLC step that is downstream from opsin transduction and reminiscent of signaling steps documented for the rhabdomeric pathway (Fig. 6).

The biological function of CE non-image forming opsins might be to align tissue responsiveness to daily intensity variation of UV radiation by photoentraining peripheral circadian rhythmicity in the absence of SCN inputs [41] to protect the organism from DNA/protein photodamage and cancer [88]. Our findings additionally bring insight into cellular mechanisms that contribute to corneal pathology induced by overexposure to UVB radiation. High energy photons impact the cornea through direct nucleotide damage, by compromising intarcellular signaling that regulates CE function and survival, and through downstream effects on keratocytes and afferents that promote recruitment of immune cells, nociception and photophobia [16–33]. Specifically, disrupted CE Ca^{2+} homeostasis may compromise NF-κB signaling and inflammasome activity, cell proliferation/apoptosis and oncogenesis [24], impair epithelial desquamation and corneal repair [89], compromise the barrier [90], promote edema [91], drive excitation of trigeminal afferents and regulate viability of stromal keratocytes [92] via increased secretion of morphogens and inflammatory mediators. TRPV4-induced purinergic signaling [56] may contribute to cytotoxicity [93] while OPN expression itself could influence the risk for oncogenic induction and progression [94]. The mass release of proinflammatory cytokines and ECM enzymes (e.g., interleukins, MMPs and TSLP) downstream from opsin and TRPV4-evoked increases [Ca^{2+}]$_i$ are likely to subserve both induction and maintenance of corneal pain and light avodiance. As one of the most densely innervated avascular tissues in mammals, with ~2500 nerve endings per mm^2 including nociceptive (C) and mechanoreceptive (Aδ) afferents, the cornea is exquisitely sensitive

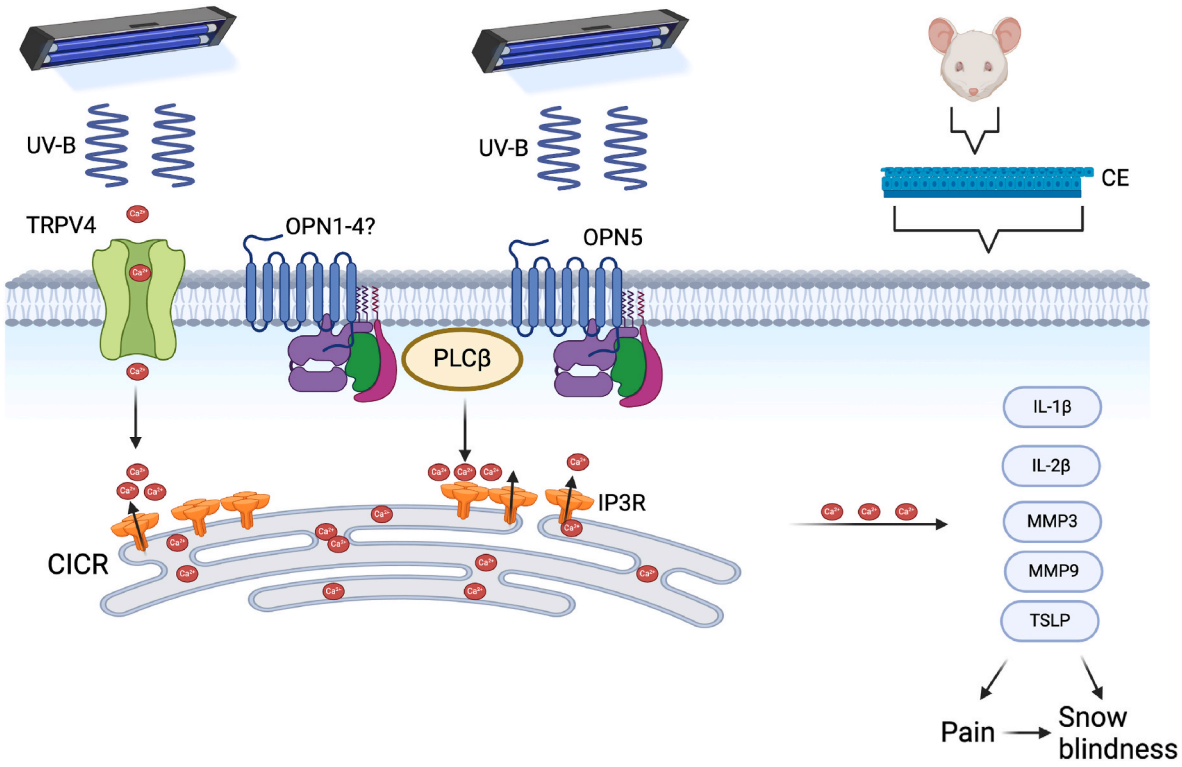

Fig. 6. Schema of UVB-induced signaling in the mouse corneal epithelium. Ultraviolet photons stimulate a rhabdomeric-like pathway composed of OPN5, phospholipase -C - G_q signaling and attendant CICR mediated largely by endoplasmic reticulum IP_3 release. OPN1, 3 and 4 may contribute residual components to UVB-induced calcium signals while OPN2 is likely to be largely desensitized during daylight but may contribute to signaling under mesopic conditions. $[Ca^{2+}]_i$ changes downstream from opsin activation promote gene expression, intracellular signaling and release of inflammatory factors that drive corneal pain and light aversion.

to mechanical, osmotic, inflammatory and photic stimuli [57,95,96]. UVB and GSK101 induced >2-fold increase in release of IL-1β (Fig. 1), implicating this master regulator of corneal injury in UV-induced inflammasome activation and apoptosis as well as macrophage infiltration, epithelial barrier dysfunction, nociception, angiogenesis, mechanical hyperalgesia and stimulation of downstream MMP, TNF-α, MCP-1 and collagenase pathways [30,31,51,97]. IL-2, IL-17, MMP-3/9 and TSLP (Fig. 1, STable I) are likely to promote excitation of afferent fibers via monocyte recruitment and ECM degradation [98]. MMP3/9 stromelysins [18,97,99,100] drive stromal thinning, pterygium and corneal neovascularization in addition to feedback damage of the CE while TSLP, an IL-17-like alarmin molecule produced in keratinocytes and epithelia, promotes immune cell infiltration, innate/adaptive immunity and ocular surface inflammation [51,98,101]. Given that pharmacological blockade of TRPV4 channels mitigates edema, inflammation and fibrosis across the body [102] and that TRPV4 blockers alleviate pathological conditions associated with mechanical/osmotic injury of the cornea, it is possible that UVB-induced corneal edema include a TRPV4 component. Moreover, the release of VEGF molecules downstream from UVB and TRPV4 activation suggests a novel mechanistic link between TRPV4 and OPN overactivation and angiogenesis. Keratinocytes similarly respond to UVB light with increased secretion of cytokines and matrix enzymes (IL-1, IL-6, IL-8, TNFα, TSLP, and MMPs) [51,100,103] and it is not inconceivable that noncanonical opsin pathways contribute to snow blindness in addition to sunburn. When these pathways malfunction or are eliminated (as in OPN4/5 KO mice), tissues are at risk for apoptosis, ROS formation, lipid oxydation, inactivated DNA repair, H3K79 methylation and suppression of protective signaling [73].

In conclusion, our findings place non-canonical phototransduction in mammalian corneas within the ever expanding range of extraretinal cells and tissues - cranial nerves, heart, skin and GI tract, RGCs, preoptic area of the hypothalamus (OPN4), pineal gland, skin and testis [36–38,41,42,54,70,104,105] that utilize photoreactive opsins to inform and modulate cellular and organismal homeostasis outside of image-forming vision. Constituting a fraction of the overall UV spectrum, UVB radiation is phototoxic/genotoxic and a main cause of sunburn, snow blindness and most skin and eye cancers [3,8,106,107]. The order-of-magnitude amplification of input signals mediated by opsin signaling [107] equips cells and organisms with warning systems that alert us about the need to avoid injury from high-energy photons, mechanical, chemical and thermal stimuli. We propose that corneal epithelial opsin pathways work with afferent OPN4 transduction [40,95] to drive nociceptive and inflammatory calcium, protein and lipid signals [102] that underlie allodynia, nociception, photophobic behavior and SCN-independent photoentrainment in the presence of UVB overexposure. Our findings open many questions regarding corneal biology, such as the significance of epithelial coexpression of distinct photo-opsins, the control of RGR (retinal G-protein-coupled receptor) signaling together with the stability, and photoconversion of chromophores (i.e., 11-cis vs. all-trans retinals) and the role of $G_o/G_i/G_q$ cGMP vs. PLC-dependent calcium signaling under homeostatic and pathological conditions. While the function of neuropsins is poorly understood, it may be worthwile to investigate the role of OPN4/5-mediated violet light-sensing in myopia prevention [108] and to delve deeper into the role of both TRPV4 [109] and ciliary body opsins [74] in circadian intraocular pressure rhythmicity. Patients with open angle glaucoma tend to be, for example, particularly susceptible to UVA/B damage [110]. Given that cornea, ciliary body, ciliary muscle, trabecular meshwork, Schlemm's canal and the retina express pressure-sensing TRPV4 channels [56,64,65,90,111–113] it may also be of interest to investigate ocular photosensing and mechanotransduction within the context of chronic ocular hypertension.

CRediT authorship contribution statement

Luka Lapajne: Resources, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Monika Lakk:** Methodology, Investigation, Formal analysis, Data curation. **Christopher N. Rudzitis:** Validation, Investigation, Data curation. **Shruti Vemaraju:** Resources. **Richard A. Lang:** Resources. **Marko Hawlina:** Supervision, Resources. **David Križaj:** Writing – review & editing, Writing – original draft, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization.

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Appendix A. Supplementary data

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References

[1] Madronich S. Analytic formula for the clear-sky UV index. Photochem Photobiol 2007;83:1537–8.
[2] Bais AF, et al. Environmental effects of ozone depletion, UV radiation and interactions with climate change: UNEP Environmental Effects Assessment Panel, update 2017. Photochem Photobiol Sci 2018;17:127–79.
[3] Bernard JJ, Gallo RL, Krutmann J. Photoimmunology: how ultraviolet radiation affects the immune system. Nat Rev Immunol 2019;19:688–701.
[4] Bergmanson JP, Soderberg PG. The significance of ultraviolet radiation for eye diseases. A review with comments on the efficacy of UV-blocking contact lenses. Ophthalmic Physiol Opt 1995;15:83–91.
[5] de Gruijl FR, et al. Health effects from stratospheric ozone depletion and interactions with climate change. Photochem Photobiol Sci 2003;2:16–28.
[6] Nishigori C. Cellular aspects of photocarcinogenesis. Photochem Photobiol Sci 2006;5:208–14.
[7] Wang F, et al. Risk of eye damage from the wavelength-dependent biologically effective UVB spectrum irradiances. PLoS One 2012;7:e52259.
[8] Ivanov IV, Mappes T, Schaupp P, Lappe C, Wahl S. Ultraviolet radiation oxidative stress affects eye health. J Biophot 2018;11:e201700377.
[9] Halliday GM. Inflammation, gene mutation and photoimmunosuppression in response to UVR-induced oxidative damage contributes to photocarcinogenesis. Mutat Res 2005;571:107–20.
[10] Green AC, Wallingford SC, McBride P. Childhood exposure to ultraviolet radiation and harmful skin effects: epidemiological evidence. Prog Biophys Mol Biol 2011;107:349–55.
[11] Green AC, Williams GM, Logan V, Strutton GM. Reduced melanoma after regular sunscreen use: randomized trial follow-up. J Clin Oncol 2011;29:257–63.
[12] Hughes MC, Williams GM, Baker P, Green AC. Sunscreen and prevention of skin aging: a randomized trial. Ann Intern Med 2013;158:781–90.
[13] Kolozsvari L, Nogradi A, Hopp B, Bor Z. UV absorbance of the human cornea in the 240- to 400-nm range. Invest Ophthalmol Vis Sci 2002;43:2165–8.
[14] Yam JC, Kwok AK. Ultraviolet light and ocular diseases. Int Ophthalmol 2014;34:383–400.
[15] Ringvold A. Corneal epithelium and UV-protection of the eye. Acta Ophthalmol Scand 1998;76:149–53.
[16] Fris M, Cejkova J, Midelfart A. Changes in aqueous humour following single or repeated UVB irradiation of rabbit cornea. Graefes Arch Clin Exp Ophthalmol 2007;245:1705–11.
[17] Delic NC, Lyons JG, Di Girolamo N, Halliday GM. Damaging effects of ultraviolet radiation on the cornea. Photochem Photobiol 2017;93:920–9.
[18] Ardan T, Cejkova J. Immunohistochemical expression of matrix metalloproteinases in the rabbit corneal epithelium upon UVA and UVB irradiation. Acta Histochem 2012;114:540–6.
[19] Fletcher AE, et al. Sunlight exposure, antioxidants, and age-related macular degeneration. Arch Ophthalmol 2008;126:1396–403.
[20] Volatier T, et al. Short-term UVB irradiation leads to persistent DNA damage in limbal epithelial stem cells, partially reversed by DNA repairing enzymes. Biology 2023;12.
[21] Meyer LM, et al. Ultrastructure of UVR-B-induced cataract and repair visualized with electron microscopy. Acta Ophthalmol 2014;92:635–43.
[22] Clahsen T, et al. The novel role of lymphatic vessels in the pathogenesis of ocular diseases. Prog Retin Eye Res 2023;96:101157.
[23] Bashir H, Seykora JT, Lee V. Invisible shield: review of the corneal epithelium as a barrier to UV radiation, pathogens, and other environmental stimuli. J Ophthalmic Vis Res 2017;12:305–11.
[24] Lee DH, Kim JK, Joo CK. Translocation of nuclear factor-kappaB on corneal epithelial cells induced by ultraviolet B irradiation. Ophthalmic Res 2005;37:83–8.
[25] Taylor HR, et al. Corneal changes associated with chronic UV irradiation. Arch Ophthalmol 1989;107:1481–4.
[26] Cejkova J, et al. UV Rays, the prooxidant/antioxidant imbalance in the cornea and oxidative eye damage. Physiol Res 2004;53:1–10.
[27] Juge R, et al. Quantification and characterization of UVB-induced mitochondrial fragmentation in normal primary human keratinocytes. Sci Rep 2016;6:35065.
[28] Boersma PM, Haarsma LD, Schotanus MP, Ubels JL. TNF-R1 and FADD mediate UVB-Induced activation of K(+) channels in corneal epithelial cells. Exp Eye Res 2017;154:1–9.
[29] Singleton KR, et al. Elevated extracellular K+ inhibits apoptosis of corneal epithelial cells exposed to UV-B radiation. Exp Eye Res 2009;89:140–51.
[30] Korhonen E, et al. UV-B-Induced inflammasome activation can Be prevented by cis-urocanic acid in human corneal epithelial cells. Invest Ophthalmol Vis Sci 2020;61:7.
[31] Maugeri G, et al. Regulation of UV-B-induced inflammatory mediators by activity-dependent neuroprotective protein (ADNP)-Derived peptide (NAP) in corneal epithelium. Int J Mol Sci 2023;24.
[32] McKay TB, et al. Corneal pain and experimental model development. Prog Retin Eye Res 2019;71:88–113.
[33] Lasagni Vitar RM, Bonelli F, Rama P, Ferrari G. Immunity and pain in the eye: focus on the ocular surface. Clin Exp Immunol 2022;207:149–63.
[34] Xue T, et al. Melanopsin signalling in mammalian iris and retina. Nature 2011;479:67–73.
[35] Hoang N, Bouly JP, Ahmad M. Evidence of a light-sensing role for folate in Arabidopsis cryptochrome blue-light receptors. Mol Plant 2008;1:68–74.
[36] Kim HJ, et al. Violet light down-regulates the expression of specific differentiation markers through Rhodopsin in normal human epidermal keratinocytes. PLoS One 2013;8:e73678.
[37] Nayak G, et al. Adaptive thermogenesis in mice is enhanced by opsin 3-dependent adipocyte light sensing. Cell Rep 2020;30:672–86. e678.
[38] Yu E, et al. The pigmentation of blue light is mediated by both melanogenesis activation and autophagy inhibition through OPN3-TRPV1. J Invest Dermatol 2024. in the press.
[39] Provencio I, Jiang G, De Grip WJ, Hayes WP, Rollag MD. Melanopsin: an opsin in melanophores, brain, and eye. Proc Natl Acad Sci U S A 1998;95:340–5.
[40] Matynia A, et al. Light aversion and corneal mechanical sensitivity are altered by intrinscally photosensitive retinal ganglion cells in a mouse model of corneal surface damage. Exp Eye Res 2015;137:57–62.
[41] Andrabi M, Upton BA, Lang RA, Vemaraju S. An expanding role for nonvisual opsins in extraocular light sensing physiology. Annu Rev Vis Sci 2023;9:245–67.
[42] Guido ME, et al. Non-visual opsins and novel photo-detectors in the vertebrate inner retina mediate light responses within the blue spectrum region. Cell Mol Neurobiol 2022;42:59–83.
[43] Buhr ED, et al. Neuropsin (OPN5)-mediated photoentrainment of local circadian oscillators in mammalian retina and cornea. Proc Natl Acad Sci U S A 2015;112:13093–8.
[44] Diaz NM, Lang RA, Van Gelder RN, Buhr ED. Wounding induces facultative opn5-dependent circadian photoreception in the murine cornea. Invest Ophthalmol Vis Sci 2020;61:37.
[45] Yamashita T. Unexpected molecular diversity of vertebrate nonvisual opsin Opn5. Biophys Rev 2020;12:333–8.
[46] Mendez F, Penner R. Near-visible ultraviolet light induces a novel ubiquitous calcium-permeable cation current in mammalian cell lines. J Physiol 1998;507(Pt 2):365–77.
[47] Moore C, et al. UVB radiation generates sunburn pain and affects skin by activating epidermal TRPV4 ion channels and triggering endothelin-1 signaling. Proc Natl Acad Sci U S A 2013;110:E3225–34.
[48] Bellono NW, Kammel LG, Zimmerman AL, Oancea E. UV light phototransduction activates transient receptor potential A1 ion channels in human melanocytes. Proc Natl Acad Sci U S A 2013;110:2383–8.
[49] Fialho MFP, et al. Topical transient receptor potential ankyrin 1 antagonist treatment attenuates nociception and inflammation in an ultraviolet B radiation-induced burn model in mice. J Dermatol Sci 2020;97:135–42.
[50] Camponogara C, et al. Neuronal and non-neuronal transient receptor potential ankyrin 1 mediates UVB radiation-induced skin inflammation in mice. Life Sci 2020;262:118557.

[51] Camponogara C, Oliveira SM. Are TRPA1 and TRPV1 channel-mediated signalling cascades involved in UVB radiation-induced sunburn? Environ Toxicol Pharmacol 2022;92:103836.
[52] Garcia-Fernandez JM, et al. The hypothalamic photoreceptors regulating seasonal reproduction in birds: a prime role for VA opsin. Front Neuroendocrinol 2015;37:13–28.
[53] Feuda R, Menon AK, Gopfert MC. Rethinking opsins. Mol Biol Evol 2022;39.
[54] Li G, Chen L, Jiang Z, Yau KW. Coexistence within one cell of microvillous and ciliary phototransductions across M1- through M6-IpRGCs. Proc Natl Acad Sci U S A 2023;120:e2315282120.
[55] Nguyen MT, et al. An opsin 5-dopamine pathway mediates light-dependent vascular development in the eye. Nat Cell Biol 2019;21:420–9.
[56] Lapajne L, et al. Polymodal sensory transduction in mouse corneal epithelial cells. Invest Ophthalmol Vis Sci 2020;61:2.
[57] Okada Y, Sumioka T, Reinach PS, Miyajima M, Saika S. Roles of epithelial and mesenchymal TRP channels in mediating inflammatory fibrosis. Front Immunol 2021;12:731674.
[58] Pan Z, et al. Dependence of regulatory volume decrease on transient receptor potential vanilloid 4 (TRPV4) expression in human corneal epithelial cells. Cell Calcium 2008;44:374–85.
[59] Bukowiecki A, Hos D, Cursiefen C, Eming SA. Wound-healing studies in cornea and skin: parallels, differences and opportunities. Int J Mol Sci 2017;18.
[60] Liedtke W, Friedman JM. Abnormal osmotic regulation in trpv4-/- mice. Proc Natl Acad Sci U S A 2003;100:13698–703.
[61] Ryskamp DA, et al. The polymodal ion channel transient receptor potential vanilloid 4 modulates calcium flux, spiking rate, and apoptosis of mouse retinal ganglion cells. J Neurosci 2011;31:7089–101.
[62] Lakk M, et al. Membrane cholesterol regulates TRPV4 function, cytoskeletal expression, and the cellular response to tension. J Lipid Res 2021;62:100145.
[63] Yarishkin O, et al. Emergent temporal signaling in human trabecular meshwork cells: role of TRPV4-TRPM4 interactions. Front Immunol 2022;13:805076.
[64] Ryskamp DA, et al. TRPV4 regulates calcium homeostasis, cytoskeletal remodeling, conventional outflow and intraocular pressure in the mammalian eye. Sci Rep 2016;6:30583.
[65] Lakk M, Krizaj D. TRPV4-Rho signaling drives cytoskeletal and focal adhesion remodeling in trabecular meshwork cells. Am J Physiol Cell Physiol 2021;320:C1013–30.
[66] Yamanaka O, Liu CY, Kao WW. Fibrosis in the anterior segments of the eye. Endocr, Metab Immune Disord: Drug Targets 2010;10:331–5.
[67] Weber EW, Muller WA. Roles of transient receptor potential channels in regulation of vascular and epithelial barriers. Tissue Barriers 2017;5:e1331722.
[68] Brodskiy PA, Zartman JJ. Calcium as a signal integrator in developing epithelial tissues. Phys Biol 2018;15:051001.
[69] Wiernas TK, Davis TL, Griffin BW, Sharif NA. Effects of bradykinin on signal transduction, cell proliferation, and cytokine, prostaglandin E2 and collagenase-1 release from human corneal epithelial cells. Br J Pharmacol 1998;123:1127–37.
[70] Zhang F, et al. Transient receptor potential vanilloid 1 activation induces inflammatory cytokine release in corneal epithelium through MAPK signaling. J Cell Physiol 2007;213:730–9.
[71] Krizaj D, Lai FA, Copenhagen DR. Ryanodine stores and calcium regulation in the inner segments of salamander rods and cones. J Physiol 2003;547:761–74.
[72] Szikra T, Krizaj D. Intracellular organelles and calcium homeostasis in rods and cones. Vis Neurosci 2007;24:733–43.
[73] Sua-Cespedes C, et al. Melanopsin (OPN4) is a novel player in skin homeostasis and attenuates UVA-induced effects. J Photochem Photobiol, B 2023;242:112702.
[74] Tsuchiya S, Buhr ED, Higashide T, Sugiyama K, Van Gelder RN. Light entrainment of the murine intraocular pressure circadian rhythm utilizes non-local mechanisms. PLoS One 2017;12:e0184790.
[75] Chandler HL, Reuter KS, Sinnott LT, Nichols JJ. Prevention of UV-induced damage to the anterior segment using class I UV-absorbing hydrogel contact lenses. Invest Ophthalmol Vis Sci 2010;51:172–8.
[76] Cejka C, et al. Hydration and transparency of the rabbit cornea irradiated with UVB-doses of 0.25 J/cm(2) and 0.5 J/cm(2) compared with equivalent UVB radiation exposure reaching the human cornea from sunlight. Curr Eye Res 2011;36:607–13.
[77] Ren H, Wilson G. The effect of ultraviolet-B irradiation on the cell shedding rate of the corneal epithelium. Acta Ophthalmol 1994;72:447–52.
[78] Sliney DH. Geometrical assessment of ocular exposure to environmental UV radiation–implications for ophthalmic epidemiology. J Epidemiol 1999;9:S22–32.
[79] Krizaj D, Cordeiro S, Strauss O. Retinal TRP channels: cell-type-specific regulators of retinal homeostasis and multimodal integration. Prog Retin Eye Res 2023;92:101114.
[80] Katz B, Minke B. The Drosophila light-activated TRP and TRPL channels - targets of the phosphoinositide signaling cascade. Prog Retin Eye Res 2018;66:200–19.
[81] Cao L, et al. Mechanisms of broad-band UVB irradiation−induced itch in mice. J Invest Dermatol 2021;141:2499–508. e2493.
[82] Pantke S, Fricke TC, Eberhardt MJ, Herzog C, Leffler A. Gating of the capsaicin receptor TRPV1 by UVA-light and oxidants are mediated by distinct mechanisms. Cell Calcium 2021;96:102391.
[83] Babes A, et al. Photosensitization in porphyrias and photodynamic therapy involves TRPA1 and TRPV1. J Neurosci 2016;36:5264–78.
[84] Yamashita T, et al. Opn5 is a UV-sensitive bistable pigment that couples with Gi subtype of G protein. Proc Natl Acad Sci U S A 2010;107:22084–9.
[85] Wagdi A, et al. Selective optogenetic control of G(q) signaling using human Neuropsin. Nat Commun 2022;13:1765.
[86] Beiert T, Bruegmann T, Sasse P. Optogenetic activation of Gq signalling modulates pacemaker activity of cardiomyocytes. Cardiovasc Res 2014;102:507–16.
[87] Spoida K, et al. Melanopsin variants as intrinsic optogenetic on and off switches for transient versus sustained activation of G protein pathways. Curr Biol 2016;26:1206–12.
[88] Upton BA, et al. Evolutionary constraint on visual and nonvisual mammalian opsins. J Biol Rhythm 2021;36:109–26.
[89] Sun CC, et al. Targeting Ca(2+)-dependent pathways to promote corneal epithelial wound healing induced by CISD2 deficiency. Cell Signal 2023;109:110755.
[90] Phuong TTT, et al. Calcium influx through TRPV4 channels modulates the adherens contacts between retinal microvascular endothelial cells. J Physiol 2017;595:6869–85.
[91] Baumann JM, et al. TRPV4 and chloride channels mediate volume sensing in trabecular meshwork cells. Am J Physiol Cell Physiol 2024;327:C403–14.
[92] Podskochy A, Gan L, Fagerholm P. Apoptosis in UV-exposed rabbit corneas. Cornea 2000;19:99–103.
[93] Mankus C, Rich C, Minns M, Trinkaus-Randall V. Corneal epithelium expresses a variant of P2X(7) receptor in health and disease. PLoS One 2011;6:e28541.
[94] Zhang W, et al. Integrated analysis of the prognostic and oncogenic roles of OPN3 in human cancers. BMC Cancer 2022;22:187.
[95] Delwig A, et al. Melanopsin expression in the cornea. Vis Neurosci 2018;35:E004.
[96] Belmonte C. Pain, dryness, and itch sensations in eye surface disorders are defined by a balance between inflammation and sensory nerve injury. Cornea 2019;38(Suppl 1):S11–24.
[97] Chandler HL, Kusewitt DF, Colitz CM. Modulation of matrix metalloproteinases by ultraviolet radiation in the canine cornea. Vet Ophthalmol 2008;11:135–44.
[98] Smolinska S, Antolin-Amerigo D, Popescu FD, Jutel M. Thymic stromal lymphopoietin (TSLP), its isoforms and the interplay with the epithelium in allergy and asthma. Int J Mol Sci 2023;24.
[99] Kozak I, Klisenbauer D, Juhas T. UV-B induced production of MMP-2 and MMP-9 in human corneal cells. Physiol Res 2003;52:229–34.
[100] Tang SC, et al. Glycolic acid attenuates UVB-induced aquaporin-3, matrix metalloproteinase-9 expression, and collagen degradation in keratinocytes and mouse skin. Biochem J 2019;476:1387–400.
[101] Lin J, et al. Interleukin-32 induced thymic stromal lymphopoietin plays a critical role in the inflammatory response in human corneal epithelium. Cell Signal 2018;49:39–45.
[102] Peng S, Poole DP, Veldhuis NA. Mini-review: dissecting receptor-mediated stimulation of TRPV4 in nociceptive and inflammatory pathways. Neurosci Lett 2022;770:136377.
[103] Keshari S, et al. Butyric acid from probiotic Staphylococcus epidermidis in the skin microbiome down-regulates the ultraviolet-induced pro-inflammatory IL-6 cytokine via short-chain fatty acid receptor. Int J Mol Sci 2019;20.
[104] Toh PP, et al. Expression of peropsin in human skin is related to phototransduction of violet light in keratinocytes. Exp Dermatol 2016;25:1002–5.
[105] Wicks NL, Chan JW, Najera JA, Ciriello JM, Oancea E. UVA phototransduction drives early melanin synthesis in human melanocytes. Curr Biol 2011;21:1906–11.
[106] Yang Y, et al. UVB drives different stages of epigenome alterations during progression of skin cancer. Cancer Lett 2019;449:20–30.
[107] Stryer L. Cyclic GMP cascade of vision. Annu Rev Neurosci 1986;9:87–119.
[108] Jiang X, et al. Violet light suppresses lens-induced myopia via neuropsin (OPN5) in mice. Proc Natl Acad Sci U S A 2021;118.
[109] Rudzitis CN, et al. TRPV4 overactivation enhances cellular contractility and drives ocular hypertension in TGFbeta2 overexpressing eyes. bioRxiv 2024.11.05.622187.
[110] Sacca SC, Izzotti A. Oxidative stress and glaucoma: injury in the anterior segment of the eye. Prog Brain Res 2008;173:385–407.
[111] Lapajne L, et al. TRPV4: cell type-specific activation, regulation and function in the vertebrate eye. Curr Top Membr 2022;89:189–219.
[112] Yarishkin O, et al. Piezo1 channels mediate trabecular meshwork mechanotransduction and promote aqueous fluid outflow. J Physiol 2021;599:571–92.
[113] Redmon SN, et al. TRPV4 channels mediate the mechanoresponse in retinal microglia. Glia 2021;69:1563–82.