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Supplementary Materials for

Ultrafast pulse shaping modulates perceived visual brightness in living animals

Geoffrey Gaulier, Quentin Dietschi, Swarnendu Bhattacharyya, Cédric Schmidt, Matteo Montagnese, Adrien Chauvet, Sylvain Hermelin, Florence Chiodini, Luigi Bonacina, Pedro L. Herrera, Ursula Rothlisberger, Ivan Rodriguez, Jean-Pierre Wolf*

*Corresponding author. Email: jean-pierre.wolf@unige.ch

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Figs. S1 to S4

Supplementary Materials

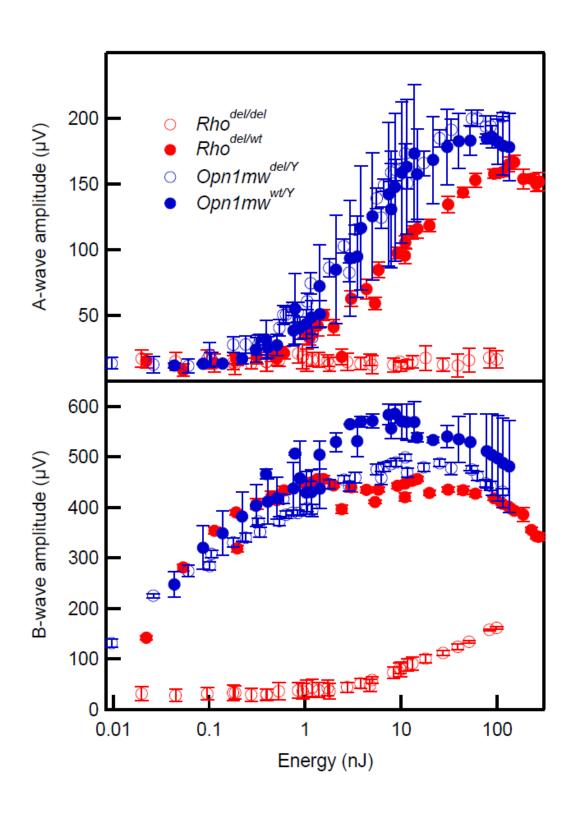
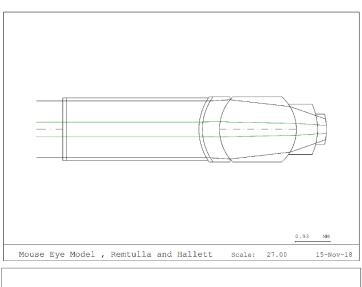


Figure S1: *ERG response of rods and cones at 535 nm.* The traces were obtained using a 5-pulse sequence plotted as a function of pulse energy. The Rho gene codes for the presence of rhodopsin. Rho^{del/del} mice lack both functional alleles of the rhodopsin gene, and therefore show no rhodopsin phenotype. The Rho^{del/wt} mice have one functional allele of the rhodopsin gene. Therefore, rhodopsin protein is present in their retina. The Opn1mw gene, coding for the protein of the M-opsin, is present only on the X- chromosome. Therefore, the Opn1mw^{del/Y} animals only translate functional rhodopsin and S-opsin proteins, while the Opn1mw^{wt/Y} animals possess the three functional receptor proteins. Mice lacking the rods (Rho^{del/del}) show no A-wave up to \sim 100 nJ per pulse, while the B-wave response sets in after 3 nJ. Other strains possessing both cones and rods show a similar response. The signal decrease at high energy levels ensues from the saturation of the number of excited rhodopsin molecules. Note that for these measurements contact lens electrodes were used instead of ring electrodes. The energy selected for the experiment (3nJ) was the highest experimentally determined such that the A-wave on the ERG signal was not saturated.



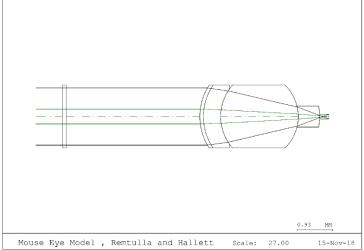


Figure S2: Ray tracing simulations. The calculations refer to a 450 μm diameter input beam propagating through the mouse eye in presence (top) or absence (bottom) of an index-matcher layer. The calculated spot size on the retina is 250 μm, and 5 μm, with and without index matcher, respectively. Values for the radii of curvature, indices of refraction, and thicknesses of the various eye components (cornea, crystalline lens and retina) are taken from Bawa, G. et al. (35). A tube filled with Methocel was used as an index matcher, in order to provide wider illumination of mouse retina. The arrangement was optimized using the software CODE V, as shown below. The spot size on the retina was verified experimentally ex-vivo, yielding an optimized spot diameter of 400 μm, with the tube and the Methocel index matching liquid.

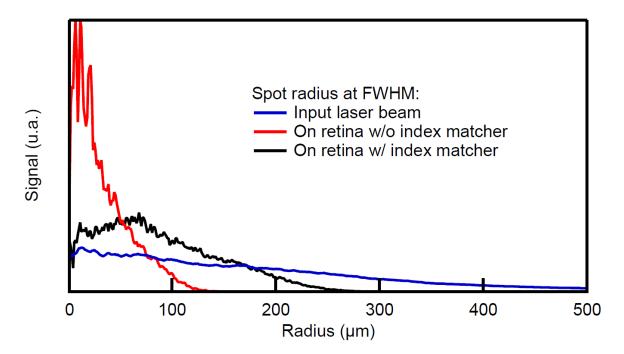


Figure S3: Measured beam size at the retina. Curves obtained with (black curve - 130 μ m radius) and without (red curve - 33 μ m radius) the index-matcher. The blue line corresponds to the beam profile before entering the eye (207 μ m radius). For these ex vivo measurements, the retina and the retinal pigment epithelium were delicately detached from the rest of the eye, leaving intact cornea, crystalline lens, and vitreous humour.

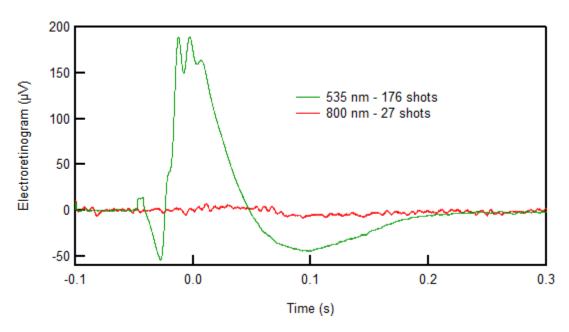


Figure S4: Effect on ERG of 800 nm excitation. Electroretinograms obtained upon 3 nJ excitation at 535 nm (green) and 1 μ J at 800 nm (red). The curves correspond to the average of 176 and 27 ERG traces, for 535 and 800 nm respectively.