electron transfer cofactor we performed extensive ultrafast optical pump-probe experiments on different preparations of RC complexes from *Chlorobium tepidum*, revealing energy/electron transfer rates between different groups of pigments. Surprisingly, we found that ~3 out of 4 Chl *a* pigments do not transfer excitation energy to the BChl *a* antenna or to P840, which indicates that these pigments must be >20Å away from any other BChl *a* pigment and thus argues against the suggested presence of 4 Chl *a* in the reaction center core complex.

2710-Pos Board B680

Biochemical and structural characterization of Photosystems from Galdieria sulphuraria

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Red alga (Rhodophyta) is one of the most ancient eukaryotic algae and its photosynthetic apparatus is in a transitional state between cyanobacteria and higher plants. Under rhodophyta, cyanidiales are group of asexual, unicellular red algae which thrive in acidic pH (0.5 - 3.0) and high temperature (50 to 55^0C). Cyanidiales are classified into three genera, Cyanidium, Cyaniodioschyzon and Galdieria. Within cyanidiales, Galdieria has been a considerable debate among researchers about its systematic position and it's an outlier in terms of habitat, reproduction and sequence similarity. There is also considerable difference in photosystems of cyanidiales. In case of photosystem I (PSI), cyanidium has a monomeric PSI with an intrinsic light harvesting complex attached to it. Also, in photosystem II (PSII), different cyanidiales have different lumenal PSII subunits: Cyanidium has PsbV not PsbP, whereas PsbV is replaced by PsbP in cyaniodioschyzon. But there is only minimal knowledge of PSI and PSII in Galdieria. In our study, we addressed these questions by use of high resolution mass spectrometry to identify the different subunits of PSI and PSII in *Galdieria sulphuraria*. For structural and functional aspects of both photosystems, we had studied the isolated complexes by electron microscopy and time resolved fluorescence spectroscopy. Initial results from these studies showed that PSI is a monomer and there are several pools of red-shifted chlorophyll with potentially complex kinetic relationships. Our work is supported from grants of National Science Foundation (MCB-0417142).

2711-Pos Board B681

Direct Photoelectrochemical Energy Transfer from Chlorosomes at Biohybrid Interfaces

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The electrogenic capacity of light antenna structures derived from *Chloroflexus aurantiacus* under light stimulation were explored in this study. Chlorosomes, which are unique light antenna structures composed of bacteriochlorophyll-c oligomers encased in a lipid monolayer, initiate the photoelectrochemical energy harvesting process in green photosynthetic bacteria at high quantum efficiencies (>92%). Previous work by this group suggest chlorosomes could be exploited for their fluorescence properties to enhance conventional silicon photovoltaics. Recent work suggests that chlorosomes can be functionally immobilized on conductive substrates. However, to date, chlorosomes have not been demonstrated to directly transduce light energy in an electrochemical system. In this study, chlorosomes are characterized in customized electrochemical cells using various electrochemical techniques, such as electrochemical impedance spectroscopy, chronoamperometry, cyclic voltammetry. The results obtained from chronoamperometric experimental studies demonstrate that isolated chlorosomes decoupled from their reaction centers are able to generate a measurable photocurrent when irradiated with light. In addition, the results indicate that only chlorosomes in proximity to the electrode participate in bioelectronic energy transfer. Electrochemical charge storage densities, also known charge injection capacities in neuroscience, show that when light stimulated, chlorosomes under a variety of conditions, i.e in bacterial fragments coupled to the photosynthetic apparatus, uncoupled colloidal solutions, and adsorbed systems, increase the charge stored near the electrode. The clear demonstration of the electrogenic capacity of chlorosomes at a heterogenous biohybrid interfaces may facilitate innovation in green technologies to novel biomedical therapeutics.

2712-Pos Board B682

Living Optical Elements in the Vertebrate Retina

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While cells are mostly transparent they are phase objects that differ in shape and refractive index. Any image that is projected through layers of cells will normally be distorted by refraction, reflection, and scattering. Strangely, the retina of the vertebrate eye is inverted with respect to its optical function and light must pass through several tissue layers before reaching the light-sensitive photoreceptor cells, with each photon having a chance of being scattered. Here we report how nature has optimized this apparently unfavourable situation. We investigated the optical properties of retinal tissue and individual Müller cells, which are radial glial cells spanning the entire thickness of the retina. Using confocal microscopy, quantitative refractometry, and a modified fiber-based dual-beam laser trap, we found that these cells act as optical fibers and guide light, which would otherwise be scattered, from the retinal surface to the photoreceptor cells. Their parallel arrangement in the retina is reminiscent of fiber-optic plates used for low-distortion image transfer. Behind the Müller cells, there seems to be a specific adaptation of the rod photoreceptor nuclei for improved light transmission through the outer nuclear layer of nocturnal animals. These nuclei have an inverted chromatin structure that turns them into microlenses channeling the light through the ONL. These findings ascribe a new function to glial cells, demonstrate the first nuclear adaptation for an optical function, and shed new light on the inverted retina as an optical system.

2713-Pos Board B683

Recording of Electrooculography in photo phobia patients

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Abstract

Photophobia is the condition which in accompanied by lack of color perception in human beings. Color perception is one of the characteristics of visual system in human beings. Retina in visual system is responsible for this characteristic. Electrooculogram(EOG) which is an electrophysiological technique has a contribution from cone cells in retina. Therefore EOG was examined in photophobia patients to search the possible disability of color perception. Fifty photophobia were selected & Electrooculography test was examined for all patients. Arden index (AI) was recorded in the population. SPSS a computerized program was used to analyze the data. The result of present study shows fall in Arden index. It is already reported that EOG has contribution from cone cells in addition to Retinal Pigment Epithelium (RPE) in Retina. Therefore the color vision is slightly distorted in patients suffering from photophobia.

KEYWORDS: Photophobia, Electrooculogram, Color Vision

2714-Pos Board B684

The Influence Of Rhodopsin Chromophore Binding On Protein Biosynthesis Examined In Vivo

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Over 100 mutations in the rhodopsin gene are associated with retinitis pigmentosa (RP) and other retinal disorders. A subset of mutations found in the N-terminus of rhodopsin cause sector RP in which the lower retina is preferentially affected, suggesting that in these cases retinal degeneration (RD) is influenced by light exposure. One such example, P23H, is the most prevalent RP-causing rhodopsin mutation in North America. Recently we have developed X. laevis (frog) models of RP based on human and bovine P23H rhodopsin which demonstrate light sensitivity. In these models, dark rearing either partially or completely rescues RD. We have shown that the rescuing effect of dark rearing is associated with chromophore binding, since blocking binding also prevents rescue. Light exposure is associated with decreased expression of P23H rhodopsin and decreased transport of P23H rhodopsin to the rod outer segment, suggesting a defect in export of the mutant protein from the ER. In order to define the role of chromophore binding in the rescue of P23H-induced RD, we have performed an extensive characterization of light sensitivity in these models. We raised transgenic F1 tadpoles in varying intensities, durations and wavelengths of light and determined the influence of these factors on RD. Our results suggest that the rescuing effects of dark rearing are not mediated by increased chromophore availablility, but rather by increased stability of rhodopsin in the secretory pathway. Our results have significant implications for the design of molecular chaperone therapies for RP.

2715-Pos Board B685

Photoreceptor ABC Transporter ABCA4: Its Role in the Visual Cycle and Retinal Degenerative Diseases

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ABCA4 is a member of the superfamily of ATP binding cassette (ABC) proteins that is localized in outer segment disc membranes of rod and cone photoreceptor cells. Mutations in the *ABCA4* gene are responsible for Stargardt macular dystrophy, cone-rod dystrophy and retinitis pigmentosa. Biochemical studies together with analysis of *abca4* knockout mice implicate ABCA4 in the transport of N-retinylidene-phosphatidylethanolamine across disk membranes. This transport process facilitates the complete removal of retinal derivatives from photoreceptors following the photobleaching of rhodopsin and cone opsin as part of the visual cycle. Loss in the activity of ABCA4 leads to the production of diretinal derivatives in disc membranes which accumulate in adjacent retinal pigment epithelial (RPE) cells as lipofuscin deposits following phagocytosis of outer segments. Progressive buildup of these toxic diretinal compounds causes the degeneration of RPE and photoreceptors and a loss in vision. Recently, we have investigated the effect of C-terminal deletion, including several disease related mutations, on the structural and functional properties of ABCA4. Our studies indicate that ABCA4 contains a conserved motif near the C-terminus that is crucial for proper protein folding and functional activity of ABCA4. Individuals missing this motif due to C-terminal truncation of ABCA4 exhibit a severe form of retinal degeneration known as cone-rod dystrophy.

2716-Pos Board B686

Formation Of All-*Trans* Retinol In Mouse Rod Photoreceptors

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Light detection destroys the visual pigment of vertebrate rod photoreceptors, rhodopsin, as its retinyl moiety is photoisomerized from 11-*cis* to all-*trans*. Rhodopsin is regenerated through a series of reactions that begin in the rod outer segment with the release of the all-*trans* retinal and its reduction to all-*trans* retinol. All-*trans* retinol is then transported to the neighboring retinal pigment epithelial cells where it is used to remake 11-*cis* retinal. The reduction of all-*trans* retinal to all-*trans* retinol is catalyzed by retinol dehydrogenase and requires metabolic input in the form of NADPH. We have used the fluorescence of all-*trans* retinol to monitor its concentration in isolated mouse rod photoreceptors. After the bleaching of rhodopsin, all-*trans* retinol formation proceeds with a rate of ~0.06 min^{-1}, which is faster than the rate of rhodopsin regeneration in whole animals; this would allow recycled chromophore to contribute to the 11-*cis* retinal used for regeneration. Inner segment metabolic pathways appear to make a significant contribution to the pool of NADPH needed for the reduction of all-*trans* retinal, as formation of all-*trans* retinol is suppressed in rod outer segments separated from the cell body. Finally, generation of all-*trans* retinol is suppressed in the absence of glucose, indicating a critical dependence of all-*trans* retinol formation on the level of metabolic activity.

2717-Pos Board B687

Control Of Sensitivity Following Pigment Bleaching By NADPH In Salamander Rods

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The recovery of sensitivity following photopigment bleaching requires the quenching of phototransduction, and the reduction of all-trans retinal is key. Retinol fluorescence increases after bleaching as a base to tip gradient in the rod outer segment and broadly matches the recovery of sensitivity. This gradient must result from a key component in retinal reduction, and we sought to determine how NADPH limits this process. Rod outer segment currents were recorded with suction electrodes, and responses were evoked by brief full-field flashes or by a narrow slit to stimulate selectively the base or tip of the outer segment. Simultaneous whole-cell recordings were made prior to bleaching to dialyze the cell with NADPH and track the recovery of sensitivity. After a 50% bleach rods remained in saturation for ~ 12 minutes. The base recovered sensitivity with tau ~ 160 s, but the tip recovered with tau ~ 450 s resulting in a tip-base tau ratio of ~ 3. Dialysis of 5 mM NADPH accelerated the recovery time by ~ 2 min, and eliminated the tip-base difference. Dialysis with 1.66 mM NADPH didn't influence recovery and failed to eliminate the tip-base difference. After a 90% bleach rods remained saturated for ~ 20 minutes with tip-base ratio ~ 10 (base tau ~ 250 s and tip tau ~ 2700 s). Thus 5 mM NADPH eliminates the gradient along the outer segment, while 1.66 mM fails to influence the recovery of sensitivity; suggesting intrinsic NADPH exceeds 1.66 mM. In addition, tau at the base following 50% or 90% bleach are remarkably equivalent, suggesting that NADPH availability is sufficient to reduce all-trans retinal. The slower tip tau following 90% bleach suggests NADPH originates predominantly near the base, which is adjacent to mitochondria.

2718-Pos Board B688

An Additional Retinoid Binding Site in Rhodopsin

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Recently we discovered that some dark-adapted salamander rods and cones generated an electrical response to the truncated retinal analogue, β-ionone. This finding was tempered by observations that exposure to β-ionone led to pigment bleaching, and that very high concentrations of β-ionone inhibited rod channels in patch experiments. Therefore we examined whether β-ionone could activate the visual pigments by attacking the chromophore-binding pocket or through an interaction at an alternate binding site. Microspectrophotometry showed an accumulation of β-ionone in green-sensitive rod outer segments that increased linearly with bath concentration indicating that β-ionone most likely partitioned into disk membranes. β-Ionone also went into blue-sensitive rod outer segments, however, uptake was higher than in green-sensitive rods at all bath concentrations tested, suggesting that β-ionone bound to at least one site on rhodopsin in addition to partitioning. X-ray diffraction of green-sensitive rod rhodopsin crystallized in the presence of millimolar concentrations of β-ionone revealed a binding site located near the extracellular/intradiskal side of rhodopsin. β-Ionone may have low efficacy and low binding affinity because rhodopsin with ligand bound retained the inactive state conformation. β-Ionone is not a native retinoid, so we also examined the effects of retinol. Dark-adapted green-sensitive rods exposed to retinol lost sensitivity to flashes due to direct rhodopsin activation. In addition, rods exhibited a relative increase in sensitivity to shorter wavelengths, consistent with the ability of retinol to act as an antenna chromophore. Similar effects were seen for a blue-sensitive rod. These results support the presence in opsins of a retinoid-binding site(s) in addition to the chromophore-binding pocket, and suggest that this alternate site(s) mediates a number of distinct ligand-specific effects.

2719-Pos Board B689

Normal Function of the Cone Visual System Requires the Interphotoreceptor Retinoid Binding Protein (IRBP)

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An adequate supply of 11-*cis* retinal is essential to the normal function and survival of photoreceptors. Rods and cones are activated when light isomerizes 11-*cis* retinal to all-*trans* retinal, and continuous function requires the recycling of all-*trans* photoproducts back into 11-*cis* retinal. Cones mediate color vision and are the daytime photoreceptors most important for human vision, and their ability to function in constant light may be linked to a novel cone-specific visual cycle. The interphotoreceptor retinoid-binding protein (IRBP) is a proposed retinoid transporter in the visual cycle, but retinoid metabolism in the rods of *Irbp*$^{-/-}$ mice is surprisingly normal. Our goal was to analyze the cone population in *Irbp*$^{-/-}$ mice and explore IRBP's contribution to normal cone function. *Irbp*$^{-/-}$ mice have cone densities equivalent to *C57Bl/6 (WT)* and express normal levels of cone opsins. However, cone function measured by electroretinogram (ERG) is reduced in *Irbp*$^{-/-}$ mice. Because a visual cycle disruptions could result in an 11-*cis* retinoid deficiency, cone ERGs were measured in *Irbp*$^{-/-}$ mice before and after injections of 9-cis retinal. Treatment with 9-*cis* retinal rescued the cone response in *Irbp*$^{-/-}$ mice, but had no effect on *Irbp*$^{-/-}$ rods or *WT* responses. These data show that the absence of IRBP results in an 11-*cis* retinal deficiency for cones but not rods and indicate that IRBP is essential to normal cone function.

Mitochondria in Cell Life & Death

2720-Pos Board B690

Apoptosis in FL5.12 cells is suppressed by inhibitors of the Mitochondrial Apoptosis-induced Channel MAC

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The Mitochondrial Apoptosis-induced Channel (MAC) forms early in apoptosis and orchestrates cell death by releasing cytochrome c from the