Sensory Receptors

3014-Pos Board B706

Mechanical Amplification by Non-Oscillating Saccular Hair Cell Bundles

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Hair cells from the inner ear are the mechano-electrical transducers for biological sound detection. Under *in vitro* conditions, hair bundles from bullfrog sacculus can exhibit spontaneous oscillations, which suggest the existence of an active process that might underlie its exquisite sensitivity to mechanical stimulations. However, it was shown that these bundles do not spontaneously oscillate under in *vivo* conditions: the overlying membrane applies not only elastic and mass loading, but also large mechanical offsets. While oscillating hair bundles were shown to amplify mechanical stimuli, it is still unknown whether or not a non-oscillating (quiescent) hair bundle benefits from its active process. Therefore, we focus on the response to mechanical sinusoidal stimulus of non-oscillating hair bundles under mechanical offsets. In this work, stimuli were applied to bundles with flexible glass fibers of stiffness 100-200 μN/m. Hair bundle motions were recorded with a high-speed CMOS camera at 500 frames per second. We found that, under large mechanical offsets, stochastic brief excursions (spikes) are observed in bundle motion, indicating the existence of an excitable regime. Application of a small sinusoidal stimulus entrains the occurrence of these mechanical spikes, leading to an amplified movement of the bundle with respect to the passive response.

3015-Pos Board B707

Barriers in the Brain: Morphology and Confinement as Barrier for Lateral Diffusion in Dendritic Spines

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Dendritic spines are the small-scale neuronal protrusions where the signal transmission between dendrites and axons is localized. The strength of such connections is regulated, among others, by a controlled concentration of AMPA receptors which are effectively confined to the spine's membrane.
The spine is able to retain these receptors in its functional domain for long times, but how does it do this? We show that the shape and curvature of the spine's membrane strongly influence the diffusive motion of receptor proteins on the spine's surface. These geometrical effects, together with crowding effects, hinder the lateral diffusion of particles on a membrane. We consider the lateral motion of receptors in the dendritic spine membrane, and find that geometrical confinement and crowding help sustain gradients in concentrations of receptors for very long times, in support of recent experiments. This suggests a deep relationship between shape and physiological function.
We present numerical and analytical results showing that the diffusion of receptors is increasingly hindered for decreasing neck sizes of the spine. Besides these geometrical effects, our simulations provide novel insights in crowding effects for interacting particles on curved surfaces. Both geometry and crowding dramatically increase the characteristic time scale for lateral diffusion, thus greatly suppressing the escape rate. This facilitates the confinement of receptors to their functional domain for very long times. These insights help to rationalize Fluorescence after Photobleaching (FRAP) and Single Particle Tracking (SPT) experiments - not only in dendritic spines, but also on bacteria, mitochondria, and other biological structures in which curved membranes feature.

3016-Pos Board B708

Charaterization of Calcium Current and Exocytosis in Zebrafish Lateral Line Hair Cells During Development

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Zebrafish offer a genetically manipulable system for studying hair cell signal transduction and exocytosis, but little is known about the physiological properties of hair cells in the developing zebrafish. To study these properties, we have developed an in vivo voltage clamp preparation that allows one to record from hair cells in the lateral line organs of living zebrafish. Here, we report the physiological properties of the hair cells of the zebrafish lateral line at 4 to 12 days post-fertilization (dpf). Hair cells were voltage clamped using a two-sine wave technique to characterize the calcium current and membrane capacitance change during exocytosis. In response to step depolarizations, hair cells exhibited maxima calcium current (8.6 +/-0.97pA) at −10mV beginning at 4-8 dpf, which were accompanied by near-linear increases in capacitance (117.4+/-32fF/s). At approximately 9-12 dpf, a shift in the I-V relationship of the calcium current occurs and hair cells begin to exhibit dramatically larger calcium current (18.3+/- 2.9 pA)) with peak current exhibited at 0-20 mV .The exocytosis (169+/-21 fF/s) also increased at −10mV. These results indicate a dramatic developmental switch in hair cell properties in the second week of life for zebrafish.

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Magnetic Field Effects on Geotactic Responses in Drosophila Melanogaster

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A wide range of organisms have the ability to sense Earth's magnetic field and use it as a method of orientation. The biophysical mechanism of this sixth sense is not well understood with two mechanisms currently discussed, one based on iron-oxide particles (magnetite mechanism) and, alternatively, a mechanism based on the sensitivity of electron spins (radical-pair mechanism). One of the challenges hindering progress towards conclusive identification of the magnetoreceptors is the lack of experimental assays with genetic models demonstrating sensitivity to the direction of earth-strength magnetic fields.
Here, we present results from two assays testing the effect of magnetic fields on geotactic responses of fruit flies, Drosophila Melanogaster. We are investigating Canton-S wild type fruit flies as well as two strains selected for positive ('Lo') and negative ('Hi') geotaxis. Experiments in a vertical nine-level geotaxis Hirsch maze assay show a significant effect of reversing the direction of the magnetic field on the responses of Canton-S strains. Under a double strength Earth field and a zero field, there was no significant effect. We also tested effects of a strong inhomogeneous field and one end of the maze. In a second assay, we began testing the Canton-S strain in a two-armed T-maze to allow for better comparison with previously published assays and we report preliminary results. Effects on geotaxis can potentially pave the way towards identifying a role of cryptochromes, a putative magnetoreceptor in the radical pair mechanism, because Hi and Lo strains have been shown to differ strongly in their cryptochrome expression.

3018-Pos Board B710

The Cloning and Expression of Whooping Crane Photopigments

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The whooping crane (*Grus Americana*) is an endangered species of bird native to North America and has yet to have its visual system characterized. Photopigments are a class of G-protein coupled receptors that respond to light. Each photopigment responds to a different wavelength of light. Our goal was to determine the sequences of the whooping crane's opsins and determine the spectral properties of the expressed visual pigments. With the rare gift of a whooping crane eye, we have cloned and sequenced all five opsin genes. We have then subcloned the genes into a mammalian expression vector and expressed them in a heterologous expression system. We then purified the reconstituted visual pigments and determined their spectral properties. The five visual pigments have the following absorbance maxima: SWS1, 404 nm; SWS2, 450 nm; RH, 500 nm; RH2 499 nm and LWS, 566 nm. The visual system of the whooping crane will be compared to that of other avian species.
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Electron Microscopy

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Quantitatively Imaging Stable Isotopes at Subcellular Level with Correlative Electron Microscopy and Nanosims Analysis

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Due to the complex of most biological problems, more and more correlative techniques have been developed to extract as much information as possible from the sample. In this presentation, we will report a reliable protocol and several applications of the correlative EM and NanoSIMS analysis on quantitatively imaging stable isotopes. The NanoSIMS is a secondary ion mass spectrometry instrument which can detect elemental and isotopic distributions with