

Implications of Dynamic Scaffolding for Signaling Efficiency in *Drosophila* Phototransduction

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***Drosophila* phototransduction converts information about light contrast into an electrical signal through G protein signaling. We recently identified that the visual scaffold InaD undergoes a light-driven conformational change which is predicted to affect binding of the activator molecule phospholipase C. This dynamic scaffolding event is predicted to affect the probability of generating a response to light using two different models.**

I. BACKGROUND

DROSOPHILA phototransduction converts light contrast information into an analog electrical signal. A single photon of light activates one rhodopsin receptor molecule, which then activates a few heterotrimeric G_Q proteins. These activated G proteins in turn activate a few phospholipase C (PLC) molecules, which break down phosphatidylinositol 4,5-bisphosphate (PIP₂) into diacylglycerol (DAG) and inositol triphosphate (IP₃). This reaction results in the opening of 15-25 cation channels of the transient receptor potential (TRP) family. Calcium then enters the cell, initially reinforcing the opening of channels but then subsequently inhibiting channel opening through multiple mechanisms, including phosphorylation of multiple targets by protein kinase C (PKC). Together these signaling reactions generate a stochastic, transient opening and closing of ion channels known as a “quantum bump” in response to a single photon of light.

Phototransduction takes place in a specialized light-sensing organelle of the photoreceptor cell known as the rhabdomere, which is composed of 30,000 microvilli. Each individual microvillus is thought to act as a single photon detector whose sensitivity and kinetics are coordinated primarily by the intracellular concentration of calcium.

Drosophila phototransduction is one of the fastest known signaling systems—the entire quantum bump is finished within 100 ms. A scaffolding protein, InaD, which binds to PLC, TRP, and PKC, among other signaling molecules, has been shown to be critical for ensuring fast, coordinated visual signaling. [1]

Recently we showed that InaD switches between two conformational states *in vivo* as a disulfide bond forms in

response to light in a PKC-dependent manner. The disulfide-bonded scaffold is predicted not to bind PLC. Mutant flies which are unable to form this disulfide bond lack a refractory period following quantum bump generation and display slow inactivation at higher light intensities. [2]

In order to understand what effects this possible dynamic binding of PLC by InaD may have on vision, we analyzed the behavior of a stochastic model of single quantum bumps and a simple model of continuous quantum bump generation under bright light conditions.

II. RESULTS AND CONCLUSIONS

Our stochastic model of the quantum bump revealed that the quantum bump consists of three phases: Initially, the presence of activated receptors is integrated by PLC as build up of an activator molecule. Upon the crossing of a dynamic threshold determined by the relative probabilities of activator molecule generation and channel opening, all the available channels open and close due to sequential positive and negative feedback. Additional oscillations are then suppressed by the build up of calcium-dependent inhibitory molecules. [3] Titration of PLC activity *in silico* predicts that dynamic scaffolding may affect the latency and probability of generating quantum bumps in response to light.

Quantum bump generation under bright light conditions was modeled as a Poisson process in order to compare the model with experimental data on the rate of quantum bump generation over a range of light conditions. This model showed that a fixed refractory period following a quantum bump was inconsistent with the experimental results, but a model in which the probability of generating quantum bumps in response to light varied with light intensity could explain the data.

REFERENCES

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