

# Notice of the Final Oral Examination for the Degree of Master of Science

of

## THOMAS IWANICKI

BSc (University of Victoria, 2013)

"The Visual Opsins of the Starry Flounder (*Platichthys stellatus*), a New Model for Studying the Physiological and Molecular Basis of Fish Vision and Light Sensitivity"

Department of Biology

Thursday, July 28, 2016 12:30 P.M. Hickman Building Room 120

### **Supervisory Committee:**

Dr. John Taylor, Department of Biology, University of Victoria (Supervisor)
Dr. Juergen Ehlting, Department of Biology, UVic (Member)
Dr. Bob Chow, Department of Biology, UVic (Member)
Dr. John Dower, Department of Biology, UVic (Additional Member)

#### **External Examiner:**

Dr. Robert Burke, Department of Biochemistry and Microbiology, UVic

Chair of Oral Examination:

Dr. Frank van Veggel, Department of Chemistry, UVic

Dr. David Capson, Dean, Faculty of Graduate Studies

#### **Abstract**

Ray-finned fish from a diversity of distantly related lineages have remarkably large repertoires. Starry flounder (Platichthys stellatus) development, morphology, life history, and behavior make this species especially suitable for experiments designed to determine why fish have so many opsins. Human and bird colour vision is mediated using three and five opsins, respectively. Fish often have many more opsins. We sequenced an eye transcriptome to characterize the starry flounder opsin repertoire, and used High Performance Liquid Chromatography to determine the chromophore content of the retina. We found eight visual opsins and that those opsins utilize only 11-cis-retinal (vitamin A1). This species' entire visual opsin toolkit appears to be functional. The number of distinct cone and rod cell absorbance profiles determined using microspectrophotomery corroborated the number of visual opsins in the transcriptome. The density of cones, cone type, and outer segment morphology varied across the retina. RH2 transcripts were expressed more and SWS1 and SWS2 transcripts were expressed less in the dorsal retina, where cone density was highest, outer segments the longest, and where we observed unequal M527/M545 and M527/L557 double cones. Regions of fish retinas appear to be specialized and we predict that this fine-tuning is enhanced by photoreceptor plasticity and opsin gene duplication and divergence.

Studies that compare opsin expression patterns among individuals, populations, or species typically assume that the differences observed influence vision. Direct connections between opsin expression and quantitative behaviours are rare. This thesis aimed to test this assumption by modifying opsin expression and characterizing vision in starry flounder. We held starry flounder in aquaria exposed to either broad spectrum sunlight or green-filtered light. We tested vision by quantifying the visuallymediated camouflage response and we measured opsin expression using digital-PCR. Granularity analysis of photographs of the camouflage response revealed higher overall pattern energy at each of the seven spatial frequency bands in fish exposed to broad spectrum sunlight compared to the green-filtered fish. However, no statistical difference in typical measurements of pattern or contrast (e.g., maximum filter size, the standard deviation of pattern energy, and the proportional power) was observed between the two groups. Opsin expression was different between fish held in the green light environment compared to those exposed to broad spectrum light. SWS1 (UV sensitive) and SWS2B (blue sensitive) were significantly down regulated in response to the green light environment. Surprisingly, this difference was lost after only three hours under a white LED light (the duration of the behavioural assay), suggesting rapid plasticity of opsin expression in response to the light environment. We found tantalizing, albeit not statistically significant evidence that fish with higher UV- and blue-wavelength sensitive opsins expressed could see more colour contrast on bluegreen checkerboards.