P1.44

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PHYLOGEOGRAPHY OF *STERECHINUS NEUMAYERI* FROM SOUTH AMERICAN AND ANTARCTIC WATERS USING THE 16S MTDNA MARKER

Separation of South America and Antarctica, approximately 30 million years ago, led to the creation of the Drake Passage and the Antarctic Circumpolar Current, subjecting Antarctic benthic fauna to isolation for nearly 25 million years. Despite this isolation, some invertebrate taxa include more than 40% non-endemic species, suggesting present day gene flow across the Drake Passage maintains genetic continuity. Molecular phylogenetic work is currently being done using the 16S mitochondrial gene from *Sterechinus neumayeri*, the most abundant echinoid genus in Antarctica, and morphologically similar populations from South America. *S. neumayeri* is a model candidate for this study because it has a circumpolar distribution and planktotrophic larvae, which are abundant in the plankton during the short summer. Samples have been collected from South America, the Drake Passage, and Antarctica on a cruise during November-December 2004. With the mitochondrial data, we are assessing the level of genetic differentiation between the populations to identify genetic breaks, historical gene flow, and endemicity.

P1.136

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PHYLOGEOGRAPHY OF *HALOCARIDINA RUBRA*: DISCERNING RELATEDNESS BETWEEN POPULATIONS OF AN ENDEMIC HAWAIIAN SHRIMP

The Hawaiian Islands have the largest concentration of anchialine habitats in the world. These coastal ecosystems are typically small landlocked pools whose water level fluctuates with daily oceanic tidal oscillations. Water movement to and from such pools occurs beneath the surface via a hydrological vascular system. The numerically and geographically dominant organism of these environments is *Halocaridina rubra*, an endemic atyid shrimp. Although previous work has documented the life history of *H. rubra*, the genetic diversity, population structure and phylogeography of this shrimp has never been explored. Understanding the relationships between *H. rubra* populations is of importance in light of urbanization and exotic species introductions that are leading to the rapid destruction of anchialine habitats. We investigated the relationships between *H. rubra* populations of the Hawaiian archipelago using mitochondrial cytochrome oxidase I (COI) DNA sequences. A survey of 18 populations (422 individuals) spanning the islands of Hawaii, Maui and Oahu revealed 187 haplotypes. Significant genetic structure between populations suggests that little to no migration or gene flow is regularly occurring over large (>30 km) geographic distances. Seven genetically diverged (~3-10%) lineages were recovered from this sampling; although further work is required to define the taxonomic status of these lineages, these divergences suggest isolation over significant time-spans and the possibility of subsequent speciation. The intrinsic uniqueness of anchialine environments compounded by the genetic distinctiveness of most *H. rubra* populations has important implications for conservation of the species and their habitats.

P1.64

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IDENTIFYING THE SPECIFIC BASOLATERAL POTASSIUM CHANNEL THAT IS INVOLVED IN CL- SECRETION OF THE SPINY DOGFISH *(SQUALUS ACANTHIAS)*

The shark rectal gland is homologous to the thick ascending limb of the Loop of Henle in mammalian kidney. It serves as a model to study how chloride is secreted through Cystic Fibrosis Transmembrane Conductance Regulator channels. The dogfish shark has been in existence for over four hundred million years. The shark relies on a specialized organ, the shark rectal gland (SRG), to assist the kidney in maintaining salt balance. The SRG is made up of tubular epithelial cells. Therefore, when analyzing the effects of various compounds, the responses observed are those of the epithelial cells in the rectal gland. We sought to determine the identity of the specific basolateral potassium channel involved in Cl- secretion. Previously our laboratory showed that bupivacaine and quinine, which are specific inhibitors of 2P, 4TM K+ channels, inhibited chloride secretion in the *in vitro* perfused rectal gland of the *Squalus acanthias*. $BaCl_2$ (5 mM) completely blocked Cl- secretion in perfused glands. Rectal glands from male and female *S. acanthias* were cannulated and perfused. Volume of duct solution secreted, as well as the chloride concentration was measured. We examined the effects of Phentolamine (200 µ;M) and Charybdotoxin (50 nM), which are both inhibitors of the Ca+ sensitive Cl- channels of the 4TM, 2P K+ channel. We observed that they have no Ba+ affect to inhibit secretion. In further studies our lab has cloned and expressed a TASK-1 channel from the SRG. This channel is undergoing further characterization using electrophysiological methods.

P3.46

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MAPK IN THE D QUADRANT ORGANIZER OF THE SQUID EMBRYO *LOLIGO PEALEI*

Classic ablation and isolation experiments performed on gastropod embryos demonstrate that the D macromere plays a critical role in axial patterning as the founder cell of the dorsal lineage. More recently, MAP kinase (MAPK) signaling cascades in the gastropod *Ilyanassa obsolete* have been linked to cell fate specification within the D macromere lineage and micromeres of the A, B, and C quadrants (Lambert and Nagy, 2001). To determine whether a similar signaling mechanism is present in squid, early cleavage and blastoderm stage embryos were studied by immunocytochemistry. First cleavage bisects the squid embryo along its mid-line, while second cleavage separates the anterior and posterior regions. One and two cell stage embryos did not exhibit MAPK expression. Following second cleavage, MAPK was observed in the two posterior blastomeres. With third cleavage, MAPK was found exclusively within the two posterior mid-line macromeres and persists in these cells and their descendents until just before 6th cleavage when MAPK expression is restricted to the 6 central micromeres destined to form the shell gland primordia at the dorsal-most tip of the embryo. Blocking early MAPK expression with the inhibitor U0126 results in embryos deficient in many of the tissues normally associated with the D lineage including: shell gland, heart, gills and mantle. These results provide the first molecular support for the classic notion that squid possess mirror image A, B, C, and D quadrants organized along their first cleavage furrow as described in the *Cephalopoda* by Adolf Naef (1928) and link the fundamental developmental mechanisms of the celphalopods to the molluscs. Supported by an ROA to KC from the NSF.