

the nucleus pulposus may shed light on disk formation, maintenance, and degeneration.

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#### Program/Abstract # 502

##### Control of scapular development by *Emx2* and *Pbx* transcription factors

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Development of the pectoral girdle is a poorly understood process. To date, it is known that for the scapula, its blade derives from mesenchyme of dermomyotomal origin and expresses genes such as *Alx1*, *Emx2*, and *Tbx15*. In contrast, its neck and head derive from the somatopleure and express genes such as *Pbx1* and *Hoxc6*. Among studies on the molecular basis of scapular development, the phenotypes of *Pbx* and *Emx2* mutant mice have been described; *Pbx1* and compound *Pbx1/Pbx2* mutants have severe alterations in scapular blade, neck, and head structures, while *Emx2*<sup>-/-</sup> mutants lack the blade entirely. We explore the genetic and biochemical interactions of *Pbx* and *Emx2* in scapular development. We first establish that *Pbx* and *Emx2* are expressed in embryonic blade progenitors in the body wall mesenchyme and proximal limb. We next reveal their genetic interaction: *Pbx1/Pbx2*, *Pbx1/Pbx3* and *Pbx/Emx2* mutants display novel and more severe scapula blade and neck/head phenotypes than single mutants. We demonstrate that in these mutants, the expression of two regulators of posterior blade formation, *Tbx15* and *Gli3*, is relatively unperturbed, while regulators of anterior and overall blade condensation formation, *Alx1* and *Sox9*, respectively, are severely reduced. In this context, we show that a *Pbx/Emx* heterodimeric complex binds to a potential *Alx1* regulatory element *in vivo* and *in vitro*, and thus may be capable of regulating *Alx1* expression during blade formation.

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#### Program/Abstract # 503

##### Sonic hedgehog signaling in the apical ectodermal ridge is essential for proper patterning of the vertebrate limb

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#### Program/Abstract # 504

##### A reevaluation of X-irradiation induced phocomelia and proximodistal limb patterning

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#### Program/Abstract # 505

##### Flotillin2 controls the spread of epidermal wound response in *Drosophila*

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*Drosophila* wound healing is a localized process of regeneration around a wound site. Sensing a wound and limiting the initial response, only to the surrounding epidermal cells, provide a challenge to the organism. The epidermis is the largest organ of the body for most animals, and the first line of defense against invading pathogens. A breach in the epidermal cell layer triggers a rapid but poorly understood response that results in the repair of the wound. In *Drosophila*, this process includes transcriptional activation of genes involved in cross-linking epidermal cuticle, e.g. the enzyme Dopa-decarboxylase (*Ddc*). We performed a genetic screen to identify genes that regulate the activation of the wound response and have uncovered several candidates that function to inhibit the spread of *Ddc* gene expression around wound sites. One such gene, *Flotillin2*, encodes for a membrane bound protein that has been shown to be localized in lipid raft signaling centers. *flo2* mutant embryos are viable and survive after epidermal wounding. We are currently testing alternative assays to determine the role of *Flo2* in wound regeneration. One such assay is based on imaginal disc regeneration. During this process, activation of cell division and outgrowth of the fragmented wing disc are limited to the blastema site. We aim to determine if *Flo2* is required to limit the wound response and outgrowth during imaginal disc regeneration. Understanding the role of *Flo2* during cellular processes in *Drosophila* may provide further insight into the mechanisms controlling the localization of the epidermal wound responses in a wide variety of animals including humans.

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#### Program/Abstract # 506

##### Fluorescence activated cell sorting and transgenic reporters for measuring gene expression profiles in embryonic epidermis of the ascidian *Ciona intestinalis*

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Transgenic GFP reporter constructs can be transformed into large numbers of *Ciona intestinalis* single-cell embryos by electroporation. Using regulatory fragments upstream of the *Dll-B* (*Dlx*) and *FoxA-a* transcription factor genes driving GFP reporter constructs, we have labeled ectodermal and non-ectodermal cell populations. Cells of these labeled embryos are then dissociated and sorted using fluorescence activated cell sorting (FACS). Quantitative PCR was then applied to measuring gene expression levels in cDNA pools from the sorted cells. This method will be used to compare gene expression levels of epidermis-specific genes between wild-type embryos and embryos co-transformed with transgenic knock-down constructs targeting the putative epidermal developmental regulator *Dll-B*. The technique may also be generally applicable to other developmental gene profiling problems.

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