Poster

Mechanoregulatory feedback loops in tissue morphogenesis: Functional analysis of genes and cis-regulator elements modulated by mechanical stress

del Pueyo Canelada, Ángela

Centro Andaluz de Biología del Desarrollo (CABD). Universidad Pablo de Olavide. Crta. De Utrera, km 1. 41013 Sevilla.

Tutor académico: Juan Ramón Martínez Morales. Jorge Corbacho Sánchez.

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ABSTRACT

Gastrulation is a key process in embryonic development, necessary for the formation of the trilaminar embryonic disc. This is the differentiation and redistribution of blastula cells to form the three embryonic layers that will give rise to different functional tissues (ectoderm, mesoderm and endoderm). This reorganization occurs through highly coordinated movements of specific groups of cells in which the entire embryo is involved.

The teleost medaka (Oryzias latipes) has been chosen as experimental animal model. In this species the gastrulation occurs simultaneously with the epibolia process. During this, the cells migrate from the animal pole to the vegetal pole leading to the formation of the embryonic axis, fundamental for the establishment of the vertebrate body plan. Little is known about the morphogenetic processes that occur during epiboly. However, the importance of some elements has been described for this process, as is the case of the members of the Yap family. These proteins are transcriptional regulators that receive signals and mechanical stimuli from the medium and integrate them together with genetic signals. This is necessary for cells to migrate correctly towards the midline of the embryo. If these signals are deregulated, gastrulation may not be properly developed which could even have lethal effects.

To learn more about the role of Yap in gastrulation, we will study the involvement of Yap downstream genes (Afap12, Akap12b, Efs, Glis2b, MarcksL1A/B, Rock2b, Synaptopodin and Ved) in the process of cytoskeletal reorganization, cell adhesion and interaction with the extracellular matrix. For this, the CRISPR-Cas9 system is used to generate knockout mutants of each gene. This genome editing mechanism is a tool adapted from a natural adaptive immune defense system of bacteria and archaea. This tool consists of two components: the sgRNAs, short fragments that match the target sequences of the genome, and the Cas9 endonuclease which cause a double-stranded DNA break in the same place. Afterwards, the cell repairs the affected area of the DNA, causing permanent modifications in the genome.

To perform data analysis we use the STATA statistical software. The preliminary data show special results in the study of Afap12, MarcksL1, Ved and Rock2b. In these cases, there seems to be a difference in the epiboly progress between controls and knockouts.

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