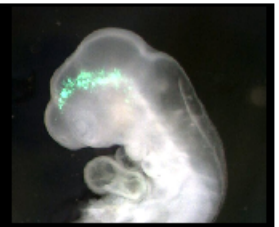
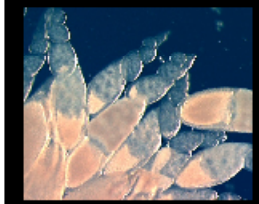


Aims/History

Course Schedule Course Program



Information

Frontiers in Developmental Biology: Concepts, Techniques and Model Organisms

Instituto Leloir

Buenos Aires, Argentina

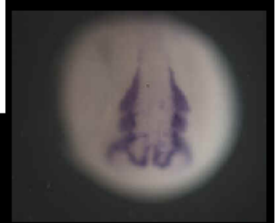
October 23-November 1, 2008

Participants

Photos - Coming Soon

Instruction Materials

Pan American Advanced Studies Institute 2008



Sponsors

The beginning




Work Hard !!!

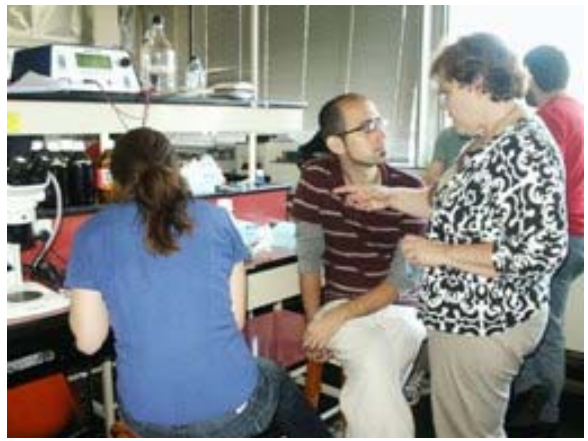


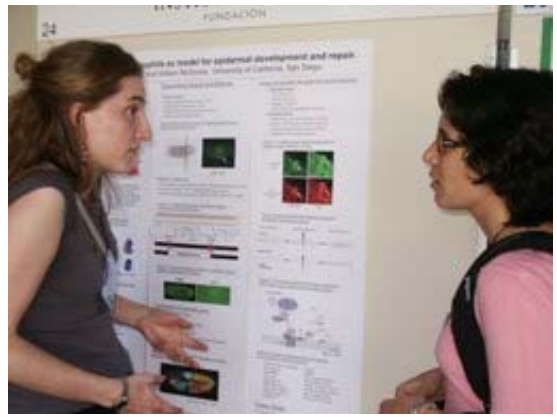













HAVANNA

STUDENT POSTER AWARDS

- **1st - Michelle T. Juárez:** ***Wound healing: Drosophila as model for epidermal development and repair***

- **2nd - Otto C. Guedelhoefer IV:** ***Stem cell migration in the planarian Schmidtea mediterranea***

- **3rd - Hozana A. Castillo:** ***Cis-regulation of RALDH2 (ALDH1A2): An evolutionary-conserved intronic enhancer for the developing dorsal spinal cord, epicardium, kidneys and gut***

INSTITUTO LELOIR
FUNDACIÓN

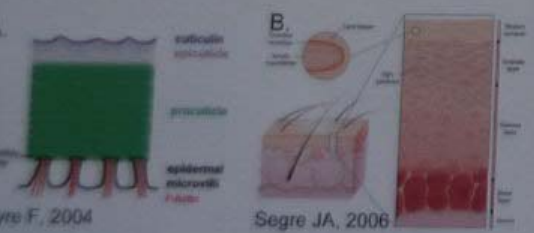
Wound Healing: Drosophila as model for epidermal development and repair.

Michelle T. Juárez and William McGinnis. University of California, San Diego.

ıtroduction

As some chordate and arthropod animals moved from aquatic to errestrial environments 400 million years ago, they required the development of an impermeable barrier to protect the organism nd maintain homeostasis of internal fluids. The contrasts between he monolayer of cuticle-secreting epidermis found in *Drosophila* nd the multilayer keratinized epidermal tissue found in mammals uggested independent evolution of the two protective barriers. owever, both mammal and arthropod epidermal barriers depend n the cross-linking of proteins and lipids to form an impermeable arrier, and recent evidence suggests that the development and pair of this protective barrier from epidermal cells evolved using a nilar genetic system in both animal systems.

e 1. Epidermal Barriers. (A) *Drosophila* (B) Mammalian.



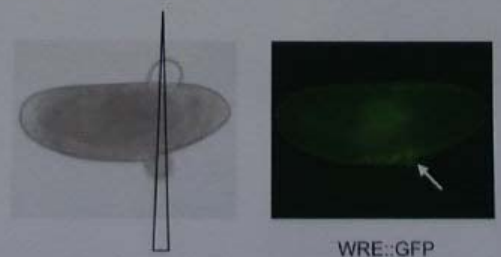
yre F, 2004 Segre JA, 2006

Experimental Design and Methods

Wound assay

- Collect and age embryos (stage 15-17)
- Microinjection with aseptic needle
- Detect wound response enhancer activity with fluorescent reporter gene

Figure 6. Micro-injection assay to detect epidermal wound response enhancer activity.



WRE::GFP

Deletion Collections

Genes that regulate the epidermal wound response:

- **Confirmed Genes**
  - Grh **Activator**
  - Fos-D **Activator**
  - Reggie1 **Inhibitor**
  - Mak3 (N-acetyl transferase) **Inhibitor**
- **Candidate Genes**
  - DNAJ-B15, heat shock protein **Inhibitor**
  - Sec24C, ER to golgi transport **Activator**
  - Sodium phosphate symporter **Inhibitor**
  - Ubiquitin protein ligase **Inhibitor**

Figure 11. Global wound response activation in reggie1 mutant embryos (*in situ*).

Stem Cell Migration in the Planaria *Schmidtea mediterranea*

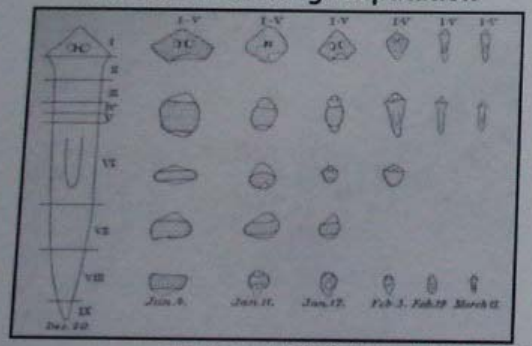
Otto C. Guedelhoefer IV[1] and Alejandro Sánchez Alvarado[1,2]

[1]Department of Neurobiology and Anatomy and [2]Howard Hughes Medical Institute, The University of Utah, Salt Lake City, Utah 84132

Abstract

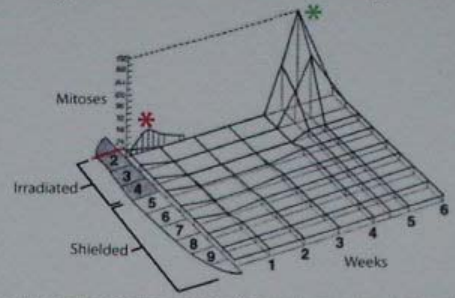
The free-living planarian *Schmidtea mediterranea* is uniquely suited to advance our understanding of key biological aspects of adult stem cells (ASCs). This organism possesses a high degree of developmental plasticity that is facilitated by a large and experimentally accessible pool of pluripotent ASCs. These stem cells (also known as neoblasts) proliferate in order to self-renew and produce division progeny that will go on to differentiate into all of the 30-40 morphologically different cell types found in planarians, including the germ line. Classical experiments suggest that planarian ASCs may have the ability to migrate long distances to reach a wound site, and that wounding may induce a signal that modulates ASC migration. However, basic questions including the active or passive nature of ASC migration have not been thoroughly investigated on the cellular or molecular level. Because they are undergoing cell division, planarian stem cells can be readily eliminated using ionizing radiation. Irradiated planaria display an inability to maintain tissue homeostasis, a loss of regenerative capacity, and eventual death. Indeed, transplantation of an ASC-containing non-irradiated graft into a lethally irradiated animal rescues homeostasis and regeneration in the host. This rescue of the host implies that stem cells and/or their division progeny may be migrating into the irradiated tissue. Because little is known about the migratory properties of stem cells in adult organisms, this transplantation technique coupled with ASC and mitotic cell markers provides an experimental paradigm to define and dissect stem cell migration after their reintroduction into an irradiated host. Following transplantation, we observed a rapid repopulation of irradiated host tissue with mitotically active ASCs that appears to be influenced by host wounding. Cell tracking and established ASC progeny markers indicate that these proliferating cells are also activating specific differentiation programs as they repopulate irradiated tissues. Presently, we are interrogating the migratory properties of planarian stem cells via RNAi screening of both candidate genes and cDNA libraries. By carefully describing and functionally testing ASC migration in vivo, we hope to better understand how stem cells are recruited out of the niche and move to damaged tissues to restore their functions, thus further elucidating the mechanisms underlying how stem cells drive regeneration in this and other multicellular organisms.

Planaria Rapidly Regenerate Their Complex Structure Following Amputation

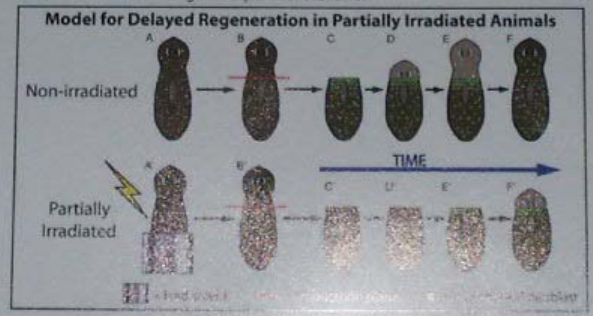


T. H. Morgan (1898) *Arch. Entw. Mech. Org.* **7** 364-97

Early Evidence for Neoblast Migration



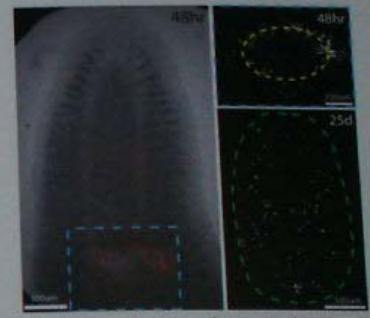
Anterior irradiation and cephalic amputation followed by scoring mitotic indices in worm sections (2-9) over six weeks, suggests that neoblasts migrate from the shielded tissue, through the irradiated tissue, and to the amputation plane. The burst of proliferation at the amputation plane in the irradiated case (green asterisk) is delayed and increased as compared to the burst seen at the amputation plane of a non-irradiated worm (red asterisk). A model describing this experiment follows:



Transplantation Rescues Lethally Irradiated Hosts

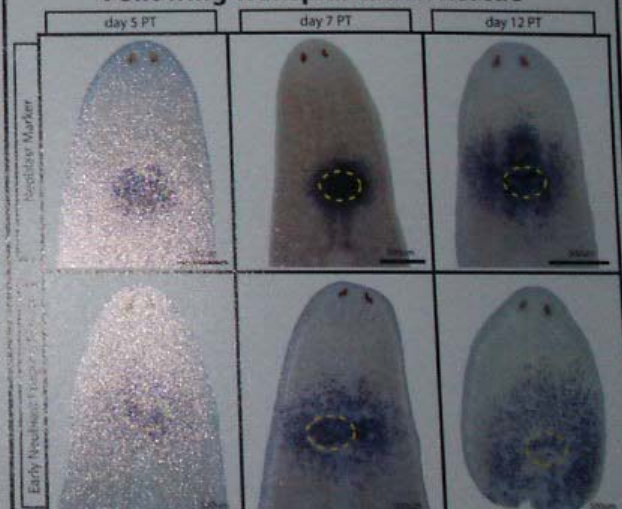
Using [illegible] this experiment [illegible] transplantation of neoblasts into a neoblast devoid host.

Transplantation Rescues Mitotic Activity in Irradiated Host Tissue



α-H3P staining following transplantation of a non-irradiated graft into an irradiated host reveals mitotic cells (overlaid on the brightfield image, left panel). Magnification of the graft area (blue box, top right panel) shows mitotic cells are present outside the graft boundary (yellow dotted line) by 48 hours post-transplant. By 25 days post transplant (bottom right panel) mitotic activity is restored throughout the animal (green dotted outline).

Repopulation Dynamics of Irradiated Host Tissue Following Transplantation Rescue



CIS-REGULATION OF RALDH2 (ALDH1A2): AN EVOLUTIONARY-CONSERVED INTRONIC ENHANCER FOR THE DEVELOPING DORSAL SPINAL CORD, EPICARDIUM, KIDNEYS AND GUT

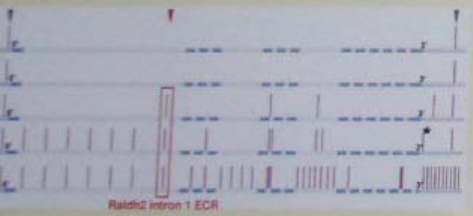
#66

Hozana A. Castillo[1]; Roberta Mascioli Cravo[1]; Ana Paula Azambuja[1], Marcos Sawada Simões-Costa[1] and José Xavier-Neto[1].
[1]Laboratório de Genética e Cardiologia Molecular, Instituto do Coração- InCor - HC. FMUSP, 05403-000;
hozanaac@yahoo.com.br

Background

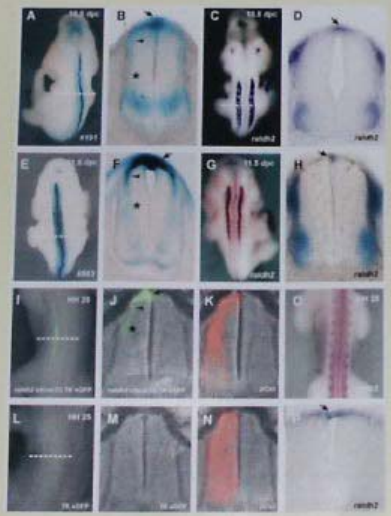
Retinoic acid signaling is crucial for correct ryonic development. Raldh2/Aldh1A2, an hyde dehydrogenase that converts aldehyde into RA , is the major enzyme lved in retinoic acid synthesis during early lopment. Its expression pattern is very mic, suggesting that *raldh2* might be ated in a highly modular fashion. To rstand how *raldh2* is regulated we screened gene for evolutionary-conserved regions s). Here we describe a 840 bp *raldh2* ECR in 1 that is conserved in tetrapods such as ibians, avians and humans, is partially rved in the agnathan *Petromyzon marinus*, absent from teleost genomes.

utionary conservation of non-ng sequences in *raldh2* gene



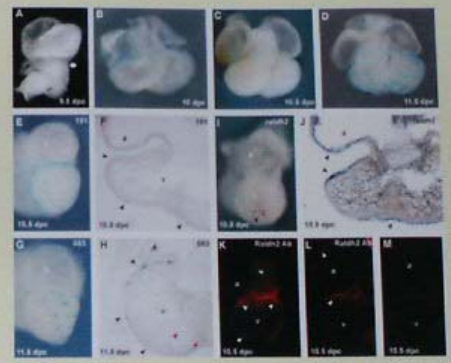
dh2 intron 1 ECR activates cZ expression in transient transgenic mice

Raldh2 intron 1 ECR is a dorsal neural tube enhancer in mice and chicken

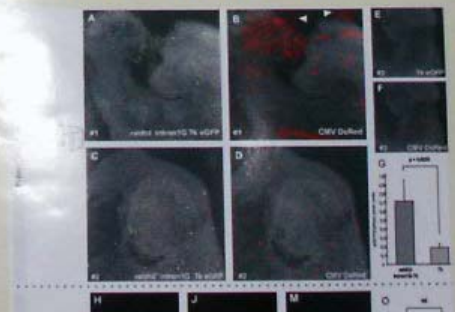


The Raldh2 intron 1 ECR is a conserved roof plate enhancer in tetrapods

Raldh2 intron 1 drives expression in derivatives of the proepicardium in mice



The epicardial function of the Raldh2 intron 1 enhancer is conserved in chicken



Looking for *cis* regulatory elements in the roof plate enhancer

Honorable Mentions

- **Andrés Romero Carvajal**: ***Early development in foam-nesting frogs genus Engystomops***
- **Ahmet Zehir**: ***Different members of BMP signaling pathway have different roles in sympathetic nervous system development***
- **Hana Paula Masuda**: ***ABAP1 is a novel pre-replication complex binding protein with a role in plant developmental control***
- **Ashley Seifert**: ***Sonic Hedgehog regulates cell cycle kinetics during morphogenesis***

Pablo and all his fantastic crew - GRACIAS !!!