Program/Abstract # 368

Analysis of the mechanisms that determine tail regenerative ability in Xenopus laevis tadpoles

Yuko Naoraa, Kota Kanekoa, Yuko Hishidaa, Taro Fukazawab, Takekazu Kuniedaa, Takeo Kuboa
aDept. of Biol. Sci, Grad. Sch. of Sci., Univ. of Tokyo, Bunkyo-ku, Tokyo, Japan
bRIKEN BRC, Tsukuba, Ibaraki, Japan

Xenopus laevis tadpoles possess high regenerative ability of amputated tails, except during the ‘refractory period’ when regenerative ability is transiently lost. We recently reported that treatment with an immunosuppressant (FK506) restores the regenerative ability, suggesting that immune responses act to impair regenerative ability during the refractory period. An expression analysis of immune-related genes suggested that immature cytotoxic T cells infiltrate regenerating tail tissues during the refractory period, which eventually impairs the regenerative ability. To identify the molecular processes affected by the immune response, we screened for genes whose expression in the tail stumps was altered by FK506 treatment during the refractory period, and identified three candidate gene fragments. Expression of these three candidate genes was significantly increased after tail amputation during the refractory period. Expression of two of the genes was downregulated by FK506 treatment, whereas expression of the third gene was upregulated. These findings suggest that the two genes downregulated by FK506 treatment are involved in immune responses that impair regenerative ability during the refractory period. Alternatively, these genes might encode autoantigens that are expressed in regenerating tail tissues and targeted by the immune system. The candidate gene that was upregulated by FK506 treatment might be involved in the regenerative processes, which are restored by FK506 treatment.

doi:10.1016/j.ydbio.2010.05.375

Program/Abstract # 369

Na\textsubscript{v}-mediated sodium transport is required for vertebrate appendage regeneration

Kelly Ai-Sun Tsenga, Wendy S. Beanea, Joan M. Lemirea, Alessio Masiab, Michael Levinab
aCenter for Reg. and Dev. Biology, Tufts Univ., Medford, MA, USA
bForsyth Institute, Boston, MA, USA

Mammals have a limited ability to regenerate tissues. In contrast, amphibians such as frogs can restore lost developmental structures, including the lens and tail. Thus a detailed understanding of natural regeneration is important for developing repair therapies. Recently, ion transport has been implicated as a regulator of regeneration. While voltage-gated sodium channels play a well-known and important role in propagating action potentials in excitable cells, we identify a novel requirement for the ion transport function mediated by the voltage-gated sodium channel Na\textsubscript{v}1.2 (Na\textsubscript{v}) in initiating regeneration. Expressed in the Xenopus tail regeneration bud by 18 after amputation (hpa), Na\textsubscript{v} induces an increase in intracellular sodium. Its inhibition abolishes sodium influx, resulting in regenerative failure by greatly reducing the expression of downstream genes that drive regenerative outgrowth, leading to decreased proliferation and altered axonal patterning. Na\textsubscript{v} is absent under non-regenerative conditions but ectopic expression of human Na\textsubscript{v}1.5 can rescue regeneration during these states. Remarkably, pharmacological induction of a transient sodium current into an amputated tail, even after a non-regenerative wound epithelium has formed, is sufficient to restore full regeneration. Our data reveal a previously undetected competency window in which cells retain their intrinsic regenerative program, identify a novel endogenous role for Na\textsubscript{v} in regeneration, and show that modulation of sodium transport represents an exciting new approach to organ repair.

doi:10.1016/j.ydbio.2010.05.451

Program/Abstract # 370

Satellite cells originate from the lateral plate mesoderm in Xenopus laevis

Randy Daughters, Ying Chen, Jonathan M. Slack
Stem Cell Institute, Univ. of Minnesota, Minneapolis, MN, USA

Satellite cells are myogenic progenitors found in the basal lamina surrounding adult skeletal muscle fibers. Satellite cells are the source of new muscle during postnatal muscle growth and are responsible for generating new fibers following muscle damage. They can self-renew and generate differentiated progeny, so satisfy the normal definition of a tissue-restricted stem cell. We have previously shown that satellite cells form the new muscle in the regenerating tail of Xenopus tadpoles. In this study we examined the embryonic origin of satellite cells prior to somite segmentation in Xenopus laevis. Medial (paraxial) or lateral (plate) mesoderm was microdissected from labeled (nucGFP) donors and orthotopically grafted into non-labeled hosts. Analysis of the percentage of donor derived (nucGFP) satellite cells (Pax7 positive) in host tail muscle suggests that satellite cells arise from lateral plate mesoderm and not paraxial mesoderm as previously thought. Based on this data we took a candidate gene approach to identifying factors that regulate the formation of satellite cells. We identified three factors: Bmp4, Msi1 and Fgfb, that will all increase the number of satellite cells formed when overexpressed in lateral plate mesoderm, both in intact embryos and in appropriate tissue explants. Furthermore, induction of Noggin expression in hsp90-noggin transgenic tissue inhibits the formation of satellite cells that normally occurs in lateral plate explants. Taken together, these data suggest that satellite cells originate from lateral plate mesoderm in response to the ventral to dorsal gradient of BMP activity found in the early embryo.

doi:10.1016/j.ydbio.2010.05.376