

CROWN REGENERATION FROM RADIAL PLATES OF A STALKED CRINOID, *METACRINUS ROTUNDUS*

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Crown regeneration of a stalked crinoid, *Metacrinus rotundus*, was examined. The crown of each specimen was dissected at a muscular articulation between radial plates and first arm plates. The remaining part comprised the radial plates, basal plates, infrabasal plates and the entire stalk. The specimens that had lost their crowns regenerated them on the remaining stalks. Tiny buds of five arms appeared on the top of the radial plates within 2 months after dissection of the crown. Each arm was formed on each radial plate, in contrast to crown regeneration from the basal plates in that a bundle of five arms were formed in the central area at the top of the basal plates. The growth rate of the arms from the radial plates was slower than that from the basal plates. The results showed that the radial plates had the potential to regenerate a crown when the upper plates were removed by dissection at a muscular articulation.

NEURAL AND HORMONAL EFFECTS ON SETTLEMENT AND METAMORPHOSIS OF LARVAE OF THE SEA URCHIN *ANTHOCIDARIS CRASSISPINA*Y. Ishizuka^{1,2}, T. Tanaka², K. Yamasu² and T. Suyemitsu²¹Dept. of Biol. Sci., Grad. Sch. of Sci., Univ. of Tokyo, Tokyo.²Dept. of Reg. Biol., Fac. of Sci., Saitama Univ., Urawa.

L-β-3,4-dihydroxyphenylalanine (L-DOPA) and its derivatives, that is, dopamine, norepinephrine and epinephrine, which are known as neurotransmitters, induced settlement and metamorphosis of larvae of the sea urchin *Anthocardaris crassispina*, while serotonin, acetylcholine and γ-aminobutyric acid (GABA) had no effect. Furthermore, thyroid hormones, which induce metamorphosis in several animals, also induced the settlement and metamorphosis. Actinomycin D inhibited the effect of thyroid hormones but not those of neurotransmitters.

These results suggest that dopaminergic and adrenergic neurons are active during larval metamorphosis of *Anthocardaris crassispina* and that thyroid hormones mediate the transcription of some genes which influence the settlement and metamorphosis of the sea urchin larvae.

CROWN REGENERATION FROM BASAL PLATES OF A STALKED CRINOID, *METACRINUS ROTUNDUS*.S. Amemiya¹ and T. Oji² ¹Dept. of Biol. Sci. and ²Geol. Inst., Grad. Sch. of Sci., Univ. of Tokyo, Hongo, Tokyo 113

Regenerating potential of a stalked crinoid (sea lily), *Metacrinus rotundus*, was examined. Some specimens autotomized their crown, and the crowns of others were removed by cutting at the junction of the radial and basal plates. The remaining part of the specimens comprised the basal plates, infrabasal plates and the entire stalk. The specimens which lost their crowns, regenerated them on the remaining stalks. In the central area at the top of the basal plates, tiny buds of five arms with several arm plates were distinguishable within 3-7 months after loss of the crown. The oral disk including mouth, intestine, anus and chambered organ were completed by this stage. The growth rate of the longest arm in the regenerating crown was almost linear from the appearance of the arm buds to a length of 1.5 cm, then declined and the arm reached 2.1 cm at 385 days post-autotomy. The specimen completely reautotomized the regenerated crown at 2 years after the first autotomy, and regenerated it again.

REGULATIVE POTENTIAL IN DEVELOPMENT OF A DIRECT DEVELOPING ECHINOID.

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The effects of LiCl have been investigated on many animals. LiCl had important effects on specific phases of development including differentiation of germ layers and determination of body axis. In the present study, the regulating potentials of a direct developing echinoid, *Peronella japonica*, were investigated using LiCl as a tool for analysis of the mechanisms of differentiation in development.

Intact embryos treated with LiCl were exogastrulated and evaginated the amniotic cavity depending on the concentration of LiCl. The stages sensitive to LiCl for evagination of the amniotic cavity were earlier than those for exogastrulation. Some of the larvae with an evaginated amniotic cavity and/or evaginated archenteron had the potential to metamorphose into juveniles. It has been known for the typical sea urchins that animal caps (presumptive ectoderm) isolated from 16-cell stage embryos treated with LiCl have the potential to develop into pluteus-like larvae. The animal caps of *Peronella* embryos treated with LiCl developed into pluteus-like larvae. Moreover, the larvae were shown to have the potential to metamorphose into juveniles with five-fold radial symmetry.

The expression patterns of the oral and aboral ectoderm-specific molecules in the larvae were examined using an oral or an aboral ectoderm-specific antibody. The results suggested that the amniotic cavity in the *Peronella* larvae corresponded to the stomodaeum in the typical sea urchins. Some modifications were found between the larvae of *P. japonica* and those of typical sea urchins in the expression patterns of the molecules specific to the oral and aboral ectoderm.

The regulative potential of the sea urchin embryos was shown in the present study to be much larger than the potential shown previously.

DISTRIBUTION OF SMALL MICROMERE DERIVATIVES BETWEEN LEFT-RIGHT COELOMIC POUCHES IN SEA URCHIN EMBRYOS TREATED WITH ACTIVIN.

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It has been reported that activin treatment caused an increase in oral-ectoderm area in sea urchin embryos. Increase in the oral-ectoderm area in the embryos depended on activin concentration. The embryos treated with activin at a concentration of 15 ng/ml sea water developed into radially symmetrical bell-shaped larvae without establishing the oral-aboral axis. In the present study, distribution of small micromere derivatives between left-right coelomic pouches in activin treated embryos was examined. The distribution pattern of the small micromere derivatives in the embryos treated with lower concentration of activin was almost the same as that in the normal embryos. The more oral-ectoderm area was increased, the more distribution pattern of the micromere derivatives was randomized. The small micromere derivatives in the bell-shaped larvae distributed radially. These results indicate that activin influences on left-right polarity as well as increase in the oral-ectoderm territory of the larvae.

INDUCTIVE AND DIFFERENTIATIVE POTENTIAL OF *veg2* TIER IN SEA URCHIN EMBRYOS

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Macromeres in 16-cell stage embryos of sea urchin divide horizontally at the 6th cleavage to produce *veg1* tier locating just beneath the equator and *veg2* tier adjacent to the micromeres locating in the vegetal pole.

The developmental fate of blastomeres in the *veg2* tier in normal embryos is secondary mesenchyme and endoderm. Hörstadius (1973) reported that the partial embryos derived from the *veg2* tier isolated from the 64-cell stage embryos failed to develop into pluteus, and formed embryoids composed of three germ layers of ecto-, meso- and endoderm. It was also reported that a chimeric embryo derived from a *veg2* tier recombined with an animal half (presumptive ectoderm) isolated from a 64-cell stage embryo developed into an apparently normal pluteus.

In the present study, a recombinant embryo consisting of a fluorescence-labeled *veg2* tier and an animal half was produced in order to examine the inductive and differentiative potential of the *veg2* tier.

The results showed that the descendants of the *veg2* tier differentiated into mesenchyme cells and that a fraction of the animal half cells differentiated into endoderm, indicating that the *veg2* tier had the potential to induce endoderm differentiation of the presumptive ectoderm.