

**COMPARATIVE HISTOLOGICAL STUDY OF THE HEPATIC ARCHITECTURE IN AMPHIBIAN LIVERS IN RELATION TO PHYLOGENY**

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The 46 amphibian livers showed variety of histological images, but anurans were the same image, as in mammalian livers. The hepatocyte sinusoidal structures of the amphibian livers were classified into three different types: cord like form, tubular form, and solid form. The hematopoietic cells were observed in the connective tissue regions of the perihepatic subcapsular and Glisson's sheath, but not anurans except *Bombina* and *Xenopus*. As phylogenetic advancement is graded from low to high, the parenchymal arrangement progressed from solid or tubular form to cord like form, but the myeloid structures were not involved anurans. We demonstrate that this study is the first to investigate amphibian livers phylogenically, and their architectural differences are shown in the route of hepatic ontogenesis. In hepatic ontogenesis, the formation of the parenchymal arrangement is acquired phylogenically, but the occurrence of the myeloid cells may be related with the development of the immune system in spleen and bone marrow. This study presents a detailed description of hepatic architecture in 46 amphibian livers by light microscopy and extensively discusses the phylogenetic viewpoint.

**COMPARATIVE HISTOLOGICAL STUDY IN THE CONNECTIVE TISSUE OF TELEOST LIVERS IN RELATION TO PHYLOGENY**

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This study presents a detailed description of hepatic architecture, the connective tissue: a) collagen fibers and b) reticular fibers, in 60 teleost livers by light microscopy and extensively discusses the phylogenetic viewpoint. The livers were perfusion-fixed via the heart or the portal vein with 4% paraformaldehyde. The specimens were embedded in paraffin. Serial 4 μm sections were obtained, and some of these were stained with hematoxylin-eosin. And these specimens were also sectioned into 8 μm slices, and stained with Silver to detect the collagen fibers and the reticular fibers. The abundant of the collagen fibers of the Grison sheath was usually deficient compared with the vertebrate including amphibians, but as phylogenetic advancement is graded from low to high, the abundant of the collagen fibers of the Grison sheath has increased. The abundant of the reticular fibers in the hepatic lobule was well developed except of Cypriniformes and Gobioidae.

**PHYLOGENIC STUDY ON STORAGE OF NEUTRAL LIPID TO 86 TELEOST LIVERS**

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Using 86 Teleost livers, we examined the correlation between storage of neutral lipid to fish livers and phylogenetic advancement of fishes. Moreover, it made comparative study of the correlation of the sinusoidal structures and the biliary system which are hepatic architecture. In hepatocyte, the quantity to be stored of neutral lipid was almost changeless by seasonal change, and as phylogenetic advancement is graded from low to high, tended to increase. The sinusoidal structures of Perciformes which is a high rank phylogenically and Tetraodontiformes is not developed, there were many quantities to be stored of neutral lipid, and since the biliary system was developed, having characteristic metabolism about storage of neutral lipid to hepatocyte was suggested.

**ULTRASTRUCTURE OF AMPHIOXUS COLLECTED OFF CAPE NOMAMISAKI**○Sachiko Morisawa<sup>1</sup>, Kaoru Kubokawa<sup>2</sup><sup>1</sup>St. Marianna University, School of Medicine, Kawasaki, Kanagawa, 216-8511, Japan, <sup>2</sup>Ocean Research Institute, The University of Tokyo, Nakano, Tokyo 164-8639, Japan

Lancelets collected off cape Nomamisaki in Kagoshima prefecture under a project of JAMSTEC in 2003 were identified as a new species and named *Asymmetron inferum* by Nishikawa in 2004. Here, surface of the body and fine structure of the spermatozoa of the new species were studied by scanning electron microscopy and transmission electron microscopy, respectively. One of the types of cilia on the body surface was different from but structure of the spermatozoa was similar to those known in the genus *Branchiostoma*.

**MOLECULAR PHYLOGENY OF CYCLOSTOMES AND ITS EVOLUTIONARY TIMESCALE**

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Cyclostomata (extant agnathans) consists of hagfishes and lampreys. Since the cyclostomes diverged from the lineage leading to gnathostomes at the earliest phase of vertebrate evolution, various kinds of biological studies have been conducted at the molecular level. In this study, we performed a series of molecular evolutionary analyses using the nucleotide sequences already reported in cyclostomes. We first review the phylogenetic studies regarding the evolutionary position of cyclostomes, and report the results of our own analyses focusing on GC-content of cyclostome genomes and evolutionary distances among cyclostomes. In addition, we provide perspectives towards the cyclostome genomics in relation to 2R (two-round) genome duplication hypothesis that is assumed in the earliest part of the vertebrate evolution.

**HH SIGNALING IN BIVALVE MOLLUSCAN DEVELOPMENT AND THE ORIGINE OF BIVALVE MOLLUSCAN HINGE**○Shota Kakoi<sup>1</sup>, Hiroshi Wada<sup>2</sup><sup>1</sup>Graduate School of Science, Kyoto University, Sakyo-ku, Kyoto 606-8503, Japan, <sup>2</sup>Graduate School of Environmental and Life Science, Tsukuba University, Tsukuba, Ibaraki 305-8572, Japan

The bivalve mollusca is originated from monoplacophoran-like single shell plate ancestor by dividing its shell plate at dorsal midline, hinge. Our research aims to reveal the origin of bivalvian hinge formation mechanism. We hypothesized that bivalvia hinge was originated from gastropod trochophora larvae ventral midline. *hh* expression was detected at gastropod trochophora larvae ventral midline cells so we obtained bivalvia *hh* expression pattern data to reveal whether hinge formation mechanisms was originated from ventral midline or not. *hh* expression was detected at the posterior tip of the hinge just before hinge appearance and anterior and posterior tip of the hinge with appearance of hinge. These expression patterns suggest that HH signaling play roles of hinge formation. So we promote functional analysis of HH signaling at bivalvia hinge formation by *hh* signaling inhibition experiment. The origine of bivalvia hinge from gastropod ventral midline was unclear because ventral midline like expression was not shown at bivalvia. Whether bivalvia hinge was originated from ventral midline will be solved by another ventral midline expressing gene, *brachyury*, expression analysis.

**THE EXPRESSION OF *Ets* AND *Alx* AND THE EVOLUTION OF SPICULE IN ECHINODERM**○Mioko Matsubara<sup>1</sup>, Koji Akasaka<sup>2</sup>, Miéko Komatsu<sup>3</sup>, Hiroshi Wada<sup>4</sup><sup>1</sup>SMBL, Nishimuro-gun, Wakayama 649-2211, Japan, <sup>2</sup>MMBS, Graduate School of Science, University of Tokyo, Kanagawa 238-0225, Japan, <sup>3</sup>Department of Biology, Faculty of Science, Toyama University, Toyama 930-8555, Japan, <sup>4</sup>Institute of Biological Sciences, University of Tsukuba, Tsukuba, Ibaraki 305-8572, Japan

In Echinoderm, the larvae show morphological variation. Echinoid, ophiuroid, crinoid and holothurian have spicules, while starfish larvae do not form any spicule. In this study, we tried to understand molecular evolutionary background for these variations in larval spicule. *Ets* and *Alx*, transcriptional factor, are known to be involved in skeletogenesis in sea urchin embryo. These expression is restricted to the skeletogenic primary mesenchyme, and these genes activate expression of spicule matrix proteins. We investigated expression of the *Ets* and *Alx* homologue of starfish, *Asterina pectinifera*, and brittle star, *Amphipholis kochii*, and compared these expression with sea urchin embryo. In starfish and brittle star embryo, *Ets* transcripts were restricted in mesenchyme cell and archenterons to be invaginated. Brittle star *Alx* is expressed in only mesenchyme cell, while its expression is detected in cells of the tip of archenteron and the left coelomic vesicle in starfish embryo. The expression of *Ets* and *Alx* was restricted in endoderm cell in brittle star, starfish and sea urchin. We will report evolutionary history of larval spicule in echinoderm larvae.

**TERRESTRIAL ISOPODS ON HACHIJU-JIMA AND MIKURA-JIMA ISLANDS**

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The senior author investigated diversity and distribution of terrestrial isopods in Hachijo-jima and Mikura-jima islands in Tokyo. The five specific terrestrial isopods are known from Izu islands up to now. However, both soil and seaside species occur for which there are few previous investigations of species diversity. Therefore, the diversity of terrestrial isopods in Izu islands has not been accurately clarified. Especially, terrestrial isopods have not been investigated previously on Mikura-jima island. To existing survey of Izu islands and Ogasawara islands is added the result of this investigation, and we report the diversity of terrestrial isopods in these islands.

**EVOLUTION OF HATCHING ENZYME GENES IN TELEOST**○Mari Kawaguchi<sup>1</sup>, Shigeki Yasumasu<sup>1</sup>, Junya Hiroi<sup>2</sup>, Masaki Miya<sup>3</sup>, Mutsumi Nishida<sup>4</sup>, Ichiro Iuchi<sup>1</sup><sup>1</sup>Life Science Institute, Sophia University, Tokyo 102-8554, Japan, <sup>2</sup>St. Marianna University School of Medicine, Kanagawa 216-8511, Japan, <sup>3</sup>Department of Zoology, Natural History Museum and Institute, Chiba, Chiba 260-8682, Japan, <sup>4</sup>Ocean Research Institute, University of Tokyo, Tokyo 164-8682, Japan

Full length cDNAs for hatching enzymes were amplified from RNA of Ayu (*Plecoglossus altivelis altivelis*) and Japanese anchovy (*Engraulis japonicus*) embryos by RACE-PCR. Whole mount *in situ* hybridization analysis revealed that those genes were specifically expressed in the hatching gland cells. The genomic clones of hatching enzyme genes (HCE) of Ayu (AyHCE), Japanese anchovy (AcHCE), milkfish (*Chanos chanos*) (MfHCE), neon tetra (*Parachanna innesi*) (NeHCE), electric eel (*Electrophorus electricus*)