Japan Geoscience Union Meeting 2013
(May 19-24 2013 at Makuhari, Chiba, Japan)

BPT26-01 Room:102A Time:May 19 16:30-16:45

Molecular phylogenetic analyses of aminoacyl tRNA synthetases and translational elongation factors

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Woese et al. (1990, PNAS 87: 4576) divided all extant life into three groups (Domains), Bacteria, Archaea, and Eukarya. However, Rivera & Lake (1992, Science 257: 74) suggested that Eukarya is a sub-group of Archaea as the sister group of Eocyte (Crenarchaeota) from the comparative analysis of indels in the alignment of translational elongation factors, EF-Tu/1a and EF-G/2. Thus, the results of phylogenetic analyses are different depending on the tree reconstruction methods and genes used for analysis.

Recent accelerated increase of genome information has promoted genome-wide phylogenetic analyses. The accelerated increase of archaeal genome information is also apparent in recent years. When Woese et al. (1990) proposed 3 domain hypothesis, only two archaeal phyla, Cerenarcheaota and Euryarchaeota, were known. However, Nanoarcheaota, Thaumarchaeota, Korarchaeota, and Aigarchaeota have recently been proposed as novel archaeal phyla. Therefore, re-analysis of relationship among Bacteria, Archaea, and Eukarya by using current data is important.

The number of genes used in the genome-wide phylogenetic analysis is, however, limited. In addition, different methods for inferring phylogenetic trees often give different results. We think that careful curation and careful phylogenetic analysis of each gene is important prior to the multi-gene based phylogenetic analyses.

We are performing molecular phylogenetic analyses of various protein genes related to translation, to understand early evolution of life. In this paper, we will report the progress of our phylogenetic analyses on some aminoacyl tRNA synthetases (ARS) and translational elongation factors, and discuss relationship between Archaea and Eukarya.

Keywords: Archaea, Bacteria, Eukarya (Eucarya), Last Universal Common Ancestor, Aminoacyl tRNA synthetase, Translational Elongation Factor

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Diversification of antenna chlorophylls in the Cyanobacteria

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Cyanobacteria are oxygenic photosynthetic prokaryotes. While they had been called blue-green algae based on their physiological characteristics, in recent decades, they are called cyanobacteria based on the knowledge that they are accommodated in the domain Bacteria. Cyanobacteria have extensive morphological diversity in their cell organization, ranging from single-celled to differentiated multicellular or filamentous forms with or without branching patterns. Part of filamentous cyanobacteria develops unique differentiated cells called heterocyst which carries out atmospheric nitrogen fixation and akinate which is resting-state cells. They also have very diverse cell division patters including binary fission (including budding), multiple fission forming baeocytes and hormogonia formation. Heterocyst formation and the baeocyte formation with more than thousand of baeocytes are unique in part of cyanobacteria. Cyanobacteria are rare bacteria whose ancestors can be observed in the fossil records which are estimated more than 2.0 billion years old. Diverse morphology was observed even in those records. More than 2,000 species has been described under Botanical Code in the Cyanobacteria.

Cyanobacteria had long been characterized to contain Chl *a* and phycobiliprotein until the mid of nineteen-seventies. However, chlorophylls, such as Chl *b* (1), DVChl *a* (2), MgDVP (3), Chl *d* (4) and Chl *f* (5) were found to act as antenna in certain cyanobacteria at 1975, 1988, 1994, 1996 and 2010, respectively. A total seven chlorophylls, Chl *a*, DVChl *a*, Chl *b*, DVChl *b*, Chl *d*, Chl *f* and MgDVP, are used as antenna chlorophyll in cyanobacteria. Comparing those diversities with those in eukaryotic photosynthetic organisms which has only three types of chlorophylls, Chl *a*, Chl *b* and Chl *c*, diversification of antenna chlorophyll has been occurred more frequently in the cyanobacterial lineage.

Cyanobacteria are generalists that have huge range of ecological habitat not only in aquatic environments from marine to freshwater but also in terrestrial environments, ranging from polar to tropical zone. They also found in hot or cold, highly eutrophic or oligotrophic, acidic and alkaline and symbiotic environments. They seem to be able to grow in almost all environments where liquid water and sunlight are available. This broad range habitat is due to the acquisition of the effective inorganic carbon transport system, the adaptation mechanism to light quality, the tolerance mechanism to high/low temperatures and so on though the accumulation of mutation on their genome followed by natural selection. The chlorophyll diversification is also seemed to be the result of natural selection to survive in various niche with various light quality.

In this presentation, I would like to discuss the chlorophyll diversification in cyanobacteria on the viewpoint of niche adaptation based on the current reports on the molecular phylogeny of cyanobacteria, the properties of antenna chlorophylls and niche of cyanobacteria which have unusual antenna chlorophylls.

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Keywords: Cyanobacteria, chlorophyll, evolution, diversification

Japan Geoscience Union Meeting 2013
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BPT26-03 Room:102A Time:May 19 17:00-17:15

Divergence dates for planktic foraminiferal cryptic species estimated from multi-genes

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Accurate estimation of divergence dates advances understanding of organismal evolution and assesses the effects of climatic and geological conditions on patterns of speciation and geographic distribution among organisms. Planktic foraminifera is one of the model organisms for divergence time estimation due to good fossil records, which are applicable to give time constraints during the dating analysis. In recent molecular phylogenetic studies, multiple genetic types have been found in a single morphospecies. In present study, we focused on a species *Pulleniatina obliquiloculata*, which mainly distribute in the subtropical-tropical water of the Indo-Pacific Oceans, having three genetic types (types I, IIa, and IIb). Intriguingly, these three types show longitudinal clines in frequencies within a narrow latitudinal range in the Indo-Pacific Warm Pool (IPWP) area. We estimate divergence time of these three genetic types corresponding with geological events that generated the oceanic circulation system in the IPWP area.

Firstly, we increased sequence data, which are complete small and large subunit ribosomal DNAs (SSU, LSU), of three genetic types of *P. obliquiloculata* and two outgroup species *Neogloboquadrina dutertrei* and *Globorotalia inflata*. Each single gene data-set was applied to maximum likelihood estimation through the program multidivtime (Bayesian molecular dating using PAML). Three patterns with single-gene data-sets (SSU and LSU) and multi-gene data set (SSU + LSU) were conducted for MCMC analyses using one maximum constrain (the first appearance date of *Neogloboquadrina acostaenesis* as the common ancestral lineage for both *P. obliquiloculata* and *N. dutertrei*). Divergence ages based on multi-gene analysis were estimated with more narrow credibility intervals (CI) than single-gene analyses. Estimated ages were similar among these data-sets: divergence of the lineages *P. obliquiloculata* and *N. dutertrei*, between 10.5 and 10.1 Ma, first divergence of genetic types (I and II) between 4.0 and 3.7 Ma, and latest divergence of genetic types (IIa and IIb) between 2.0 and 1.1 Ma. Divergence time of three genetic types suggests that their longitudinal clines have been established according to development of the IPWP system: distinct water masses were formed between the Pacific and Indian-sides and subtropical gyre system was emphasized in this area. The present results indicate that changes of the oceanic circulation system impact on the geographic patterns of migration and divergence in pelagic organisms.

Keywords: planktic foraminifera, divergence time estimation, LSU, SSU

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BPT26-04 Room:102A Time:May 19 17:15-17:30

Comaparative genomics of molluscs

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Homeobox genes are involved in various aspects of the development of multicellular animals, including anterior-posterior patterning of the body plan. We performed a genomic survey of homeobox genes in the Japanese pearl oyster, Pinctada fucata. We annotated 92 homeobox-containing genes and 5 homeobox-less Pax genes. This species possesses ten or eleven Hox genes. However, most of them are encoded in different scaffolds, and thus we did not obtain evindence for clustering of these genes. We annotated another homeobox genes that cover 77 out of the 111 gene families identified in the amphioxus genome. Investigation of these repertoires of homeobox genes will shed new light on the relatively less well known lophotrochozoan development.

Keywords: Molluscs, Homeobox

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BPT26-05 Room:102A Time:May 19 17:30-17:45

Aquatic adaptation and the evolution of olfaction in cetaceans

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Transition between the sea and land is one of the most striking types of evolutionary event in the history of life. Vertebrates originated in the sea, and a group of vertebrates became terrestrial during the Devonian period. This transition is well documented in the fossil record, and the land vertebrates that newly emerged are called tetrapods. Tetrapods include the modern amphibians, reptiles, birds and mammals. Amphibians still need a humid environment, while amniotes (reptiles, birds and mammals) have acquired keratin-covered waterproof skin and dehydration-protected embryos, allowing them to be independent of aquatic habitats.

Full terrestrial adaptation caused profound changes in the olfactory sensory modality in amniotes. Marine fish smells water-soluble molecules, whereas terrestrial amniotes need not smell underwater, but instead need to smell volatilized odorants in the air. The repertoires of the olfactory receptors (ORs) in amniotes differ greatly from those in marine fish. The ORs are encoded by intronless OR genes which constitute one of the largest multigene families in vertebrate genomes. It has been reported that the OR gene family in fish is much more diverse than that in amniotes, but two OR gene subfamilies have expanded explosively in the amniote genomes. These two OR subfamilies are called class I and class II, respectively. Interestingly, the OR gene repertoire in amphibians is as diverse as that in fish, but the class II OR gene subfamily expansion has also been confirmed in amphibian genomes. These findings suggest that the amniote class I and II OR subfamilies have been expanded to detect airborne molecules, and that large-scale degeneration of ORs that detect underwater odorants had occurred by the time when the transition to land was complete.

Amniotes have returned to the sea many times in their evolution, and a number of modern amniotes are living in the marine environment. Especially among such amniotes, cetaceans (whales, dolphins and porpoises) are one of the most perfectly adapted modern aquatic groups. Cetacea is an order of mammals that originated in the early Eocene epoch and that was derived from artiodactyls. Extant cetaceans are classified into two suborders -Mysticeti (baleen whales) and Odontoceti (toothed whales)- and both of them are fully aquatic. How did cetaceans re-adapt their olfactory systems to their underwater lives? Anatomical evidences strongly suggest that toothed whales have no sense of olfaction, whereas baleen whales have it but they can smell in air, not underwater. In my talk, I will provide my recent studies about the evolution of whale olfaction from the anatomical and genomic points of view.

Keywords: baleen whale, toothed whale, Eocene whale, olfactory receptor gene

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BPT26-06 Room:102A Time:May 19 17:45-18:00

Quantitative Imaging for Trace-elements in Biochemical Samples using Laser Ablation-ICPMS coupled with Soft Ablation Tec

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In biological systems, many trace-elements play important roles to control numerous biochemical reactions. With the improvement of the analytical technique, nutritional status, distribution, metabolism and function of the trace-elements have been well investigated. To obtain further detailed information for elemental metabolism or function of the trace-elements, changes in concentration and distribution of the trace-elements at microscopic or histologic scales, such as tissue or cell, are highly desired. The combination of laser ablation sample introduction technique and ICP-mass spectrometry (LA-ICPMS) has now become a fast, accurate, versatile and user-friendly analytical tool for elemental and isotopic analysis of solid geochemical and biochemical samples [1]. One of the great advantage to use the LA-ICPMS technique is that sample is analyzed under the atmospheric pressure, and neither coating with conductive materials nor time-consuming evacuation procedures is required, and therefore the LA-ICPMS technique has a capability to accept the most biochemical samples including wet tissue or cell samples without any complicated sample preparation procedures.

For the conventional LA-ICPMS technique, abundance values for the trace-elements have been calibrated by means of comparison in the signal intensity data for analytes between the sample and standard. However, for the biochemical samples, availability of the homogeneous and well-calibrated matrix matched standard was very limited. Moreover, because of the heterogeneity in hardness or color within the sample piece, the amount of sample ablated can vary significantly even at the laser sampling under the identical ablation pit sizes and fluence, and the changes in ablation volume (weight) can become a major source of analytical error. In this study, we have developed a new quantification technique for the LA-ICPMS analysis. It is widely recognized that laser ablation can be achieved when the energy fluence exceeds the critical value (energy threshold). The ranges of energy fluence required to ablate the organic components is generally lower than those required for most glass, crystal or metallic samples, and therefore, only the organic components can be ablated when the fluence was carefully controlled (soft ablation [2]). With the soft ablation technique, the sliced sample (1um thickness) can be totally ablated or evaporated through the laser ablation without any damage or ablation of substrate (slide glass). This suggests that the resulting sampling depth (i.e., volume) for the samples can be kept constant despite the local heterogeneity in hardness or color of the samples, and therefore, reliable quantitative elemental analysis or mapping can be made. In this study, we will discuss the unique feature and the versatility of the present calibration protocol for the elemental determination using the LA-ICPMS technique based on imaging of Cu and Zn in the cross section of blood vein and also on the time-changes in element distribution of Ce and Eu in mices alveolus. With the high-sensitivity LA-ICP-MS technique with newly developed calibration technique, the LA-ICPMS technique has immediate potential as a reconnaissance method for reliable technique for quantitative imaging for trace-elements in biochemical samples.

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Keywords: Elemental Imaging, Essential Trace-elements, Tissue Samples, Alveolus Samples, Laser Ablation-ICPMS, Soft Ablation