

アミノアシル tRNA 合成酵素と翻訳伸長因子の分子系統解析：古細菌と真正細菌の系統学的位置の再検討

Molecular phylogenetic analyses of aminoacyl tRNA synthetases and translational elongation factors

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Woese ら (1990, PNAS 87: 4576) は細菌、古細菌、真核生物のそれぞれが単系統群であるとする 3 ドメイン説を発表した。しかし、系統解析の方法や、解析する遺伝子の種類によって、異なる結果も数多く得られている。例えば、Rivera と Lake (1992, Science 257: 74) は翻訳伸長因子 EF-Tu/1 と EF-G/2 の配列の欠失/挿入の比較から、古細菌の一門である Crenarchaeota (Lake らは Eocyte と呼称) が真核生物に近縁であり、真核生物は古細菌の内部系統になることを示唆した。

近年のゲノム配列情報の爆発的な増加により、ゲノム情報に基づく分子系統解析が進められている。例えば、Ciccarelli ら (2006, Science 311: 1283) は、31 の蛋白質遺伝子に基づく無根系統樹を作製し、古細菌は真核生物とは独立したグループであると示唆した。また、古細菌に関しても多くのゲノム配列情報が得られるようになってきた。それに加え、Woese らの提案による Crenarchaeota と Euryarchaeota に加え、Nanoarchaeota、Thaumarchaeota、Korarchaeota、Aigarchaeota の門が提案されてきた。よって、改めて真正細菌、古細菌、真核生物の系統関係を検討することは、重要である。

しかし、ゲノムレベルでの分子系統解析であっても、全生物をカバーできる遺伝子の数は限られ、解析法によって結果も異なる。我々は、全生物の初期の進化を理解するためには、個々の遺伝子の進化を、細胞活動の中での位置づけを考慮しつつ理解し、積み上げていくこと重要であると考えている。

以上を踏まえ、我々は、現在翻訳に関わる蛋白質遺伝子の系統解析を積み重ねて、全生物の初期の進化経路を理解しようとして研究を進めている。我々の進めているアミノアシル tRNA 合成酵素並びに翻訳伸長因子の分子系統解析の進捗状況を紹介し、特に古細菌と真核生物の関係について議論する。

キーワード: 古細菌, 真正細菌, 真核生物, 全生物の最後の共通祖先, アミノアシル tRNA 合成酵素, 翻訳伸長因子

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

シアノバクテリアの進化とクロロフィルの多様化 Diversification of antenna chlorophylls in the Cyanobacteria

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シアノバクテリアは、酸素発生型の光合成を行う原核生物である。歴史的には、その生理学的特徴から藻類（藍色植物門）と位置づけられ藍藻（blue-green-algae, ラン藻）と呼ばれてきた。しかし、現在では真正細菌（バクテリア）上界に帰属するという系統学的知見により、主にシアノバクテリアと呼ばれている。細胞体制には、単細胞、単細胞群体（平板状、サルシノイド状、パルメラ状）、糸状体（分岐なし、擬分岐、棍棒状、針状、群体など）がある。また、糸状体を形成するものなかには、ヘテロシスト、アキネートなど特異に分化した細胞を形成するものがある。分裂様式には、二等分裂に加え、外生孢子形成（バディングを含む）、孢子形成、ホルゴニア形成などが知られている。このうち、ヘテロシスト形成や1000個近くの娘細胞をつくることもある内生孢子形成は、シアノバクテリアに特有にみられるものである。バクテリアとしてほぼ唯一、20億年以前の化石記録として認知できる生物であり、20億年前までには既に多様な形態に分化していたことが明らかにされている。植物分類学のうえでは2000種以上が報告されており、ひととき多様な生物群である。

シアノバクテリアは、長い間、光合成色素としてChl *a*とフィコビルン色素をもつと特徴付けられてきた。しかし、1975年にChl *b* (1)、1988年にDVChl *a* (2)、1994年にMgDVP (3)、1996年にChl *d* (4)、2010年にChl *f* (5)を光合成のアンテナとして含むものが見出された。シアノバクテリアにはこれまでにアンテナとして機能するクロロフィルとして、Chl *a*、DVChl *a*、Chl *b*、DVChl *b*、Chl *d*、Chl *f*、MgDVPの7つが知られており、陸上植物と他の藻類に見られるクロロフィルを合計しても3種（Chl *a*、Chl *b*、Chl *c*）であることと比較すると、シアノバクテリアの進化過程において激しいクロロフィルの多様化（アンテナの多様化）が起こっていることがわかってきた。

シアノバクテリアは、環境適応範囲の広いジェネラリストであり、一般の水圏・土壌環境のほか、貧栄養水圏、砂漠・極域・高地などの過酷な光・温度・水分環境においても生育し、一次生産に一定の役割を果たしている。また、共生体として宿主に栄養源を供給しているものもある。効率的な無機炭素輸送、優れた光質適応、高い温度耐性、高い凍結・融解耐性などの環境適応能力は、遺伝子の変異・獲得・欠失と自然選択によってもたらされ、多様な環境への適応の結果として、ジェネラリストとして、あるいは特殊な耐性や代謝機能を獲得したスペシャリストとしての能力を獲得したものと考えられている。クロロフィルの多様化も、多様な環境に適応する変異の選択によって形成されてきたものであると考えられる。

本発表では、シアノバクテリアにおけるクロロフィルの多様化をシアノバクテリアの分子系統関係、クロロフィルの吸収特性、シアノバクテリアの生育環境に基づき、環境適応によるニッチ獲得の視点で考察する。

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キーワード: シアノバクテリア, クロロフィル, 進化, 多様化

Keywords: Cyanobacteria, chlorophyll, evolution, diversification

複数遺伝子を用いた浮遊性有孔虫遺伝子型の分岐年代推定 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 *Neoglobobadrina 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 *Neoglobobadrina 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.

キーワード: 浮遊性有孔虫, 分岐年代推定, LSU, SSU

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

軟体動物比較ゲノム学 たかがゲノム、されどゲノム Comparative 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 evidence 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.

キーワード: 軟体動物, ホメオボックス

Keywords: Molluscs, Homeobox

海洋環境適応と鯨類の嗅覚の進化

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.

キーワード: ヒゲクジラ, ハクジラ, 始新世鯨類, 嗅覚受容体遺伝子

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

ソフトアブレーション-LA-ICPMS法を用いた生体試料中の微量金属元素のイメージング分析 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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