氏　　　　名　　小　倉　　淳

学位（専攻分野）　　博士(理学)

学 位 記 番 号　　総研大甲第681号

学位授与の日付　　平成１５年３月２４日

学位授与の要件　　生命科学研究科　遺伝学専攻

　　　　　　　　　学位規則第４条第１項該当

学 位 論 文 題 目　　Comparative Analysis of Gene Expression in Camera

　　　　　　　　　Eye and its Implication to Genomic Diversification

　　　　　　　　　of Bilateria

論 文 審 査 委 員　　主　査　教授　　　　菅原　秀明

　　　　　　　　　　　　　　教授　　　　佐々木　裕之

　　　　　　　　　　　　　　教授　　　　城石　俊彦

　　　　　　　　　　　　　　助教授　　　藤澤　敏孝

　　　　　　　　　　　　　　助教授　　　山本　博章（東北大学）

It has been extremely difficult to study the molecular evolution of bilaterian animals that consist of Protostomes and Deuterostomes, because none of complete genome sequences were known for any bilaterian animals until a decade years ago. However, the advancement of the genome sequencing has changed this situation completely. Not only the genomic sequences but also the gene expression data have been obtained for various kinds of bilaterian organisms such as fly, nematode, ascidian, fish, rat, mouse and human. Thus, I conducted the present study for the purpose of studying the evolutionary process of bilaterian animals and the mechanisms of species diversity through the vast number of genomic data and gene expression data.

In chapter 1, I gave the overview of species diversity and evolution of bilaterian animals. In particular, I focused on the evolutionary diversification of organs such as eyes. In chapter 2, I then developed a novel method for estimating the ancestral gene set using all the available genome sequences and gene expression data.

In chapter 3, to understand the molecular mechanism of species divergence, I focused upon the camera eye structures that were known to be very similar because of "convergent evolution". In fact, because human and octopus belong to Deuterostomes and Protostomes, respectively, the camera eye, the one of the most elaborate eye, evolved the camera eye independently after their speciation, which is well known as a typical example of the convergent evolution in the level of morphology. Then, I conducted the comparative analysis of gene expression in the camera eyes between human and octopus. In practice, I collected the gene expression data of octopus eye using EST sequencing, and compared these EST sequences with the complete genome sequences of nematode, fly and human as well as the gene expression data of human eye. As a result, I found that 729 genes were expressed commonly between the camera eyes of human and octopus. Applying my method to the estimation of the ancestral gene set, I found that 933 genes had already existed at the common ancestor of Protostomes and Deuterostomes. I also found that 886 out of 933 genes were conserved between human and octopus although nematode and fly are closely related to octopus than to human. This suggests that the conservation of the ancestral gene set of camera eye is required for the evolution of the camera eye even after the speciation of human and octopus, implying that the evolution of the camera eyes of human and octopus is not subjected to the convergent evolution, but rather the divergent evolution derived

from the common ancestor.

In chapter 4, to understand the process of bilaterian evolution, I conducted the evolutionary analysis of bilaterian animals. The animals belonging to bilaterian phyla had started sudden species diversification in only 5 to 10 million years from 530 million years ago. This sudden diversification is inscribed in the fossil records such as Burgess Shale and called "Cambrian explosion". The molecular mechanism of this "Cambrian explosion" is studied by many researchers using commonly conserved genes such as 18S rRNA. However, taking into account that species evolved by the changing the usage or conservation of gene set as shown in chapter 3, it is reasonable that we study the molecular mechanism of this phenomena using all the available data including the genomic sequences. For the study of the evolution of the bilaterian animals, I estimated the ancestral gene set of common bilaterian ancestor to be 6,577 gene clusters. Then, I studied how this ancestral gene set had been maintained since the split of plant-animal-fungus at 1,070 million years ago, and also how it has changed to the extant animals through the Cambrian explosion. As a result, I found the number of gene clusters belonging to the ancestral gene set of bilateria had emerged from 2,469 gene clusters to 6,577 gene clusters. Comparing these gene clusters to the genome of fungi and plants, I found that fungi have had been conserved much more gene clusters in the extant fungi rather than plants have. This observation indicates that fungi are closely related to bilaterian animals than plants. Moreover, I found that the extant bilaterian animals might tend to maintain the ancestral gene set and differentiate their function instead of to emerge novel genes. I observed many events of gene loss in the genome of nematode, fly, mouse, human, suggesting that gene loss events may have an important role in the evolution since the pre-Cambrian period.

Finally, in chapter 5, I described the conclusion of the present study. Here, I conclude that the events of gene loss and the differentiation of usage of gene set may have an important role for the species diversity and organ diversity in the bilaterian evolution.

論文の審査結果の要旨

「カメラ眼における遺伝子発現の比較解析とその基礎となる左右相称動物のゲノム多様性の研究」と題した小倉淳君の博士論文は、爆発的に種が多様化し、現存する動物のほとんどの祖先が生まれたと考えられる先カンブリア時代におきた進化機構を遺伝子発現やゲノム上の遺伝子の構成の観点から理解しようとする研究の成果である。

論文は5章で構成されている。第1章では、序章として、ゲノムの進化の観点から器官レベルの多様性と種レベルの多様性を比較研究する展望をまとめた。具体的には、器官としてはカメラ眼に、種としては左右相称動物に注目した根拠と背景を述べている。第2章では、ゲノム配列データ、既知の遺伝子のデータおよび遺伝子発現データから祖先にあたる遺伝子セットを推定する手法を提案している。第3章は特定の器官としてのカメラ眼の進化について形態進化の観点からこれまで定説とされてきたタコとヒトにおける収斂進化を、タコの膨大な EST 配列を決定し、ヒト、線虫、ショウジョウバエのそれと比較することによって分子レベルで検証した。その結果、全体の共通祖先として推定した 933 個の遺伝子のうち 886 個の遺伝子がヒトとタコにともに保存されていることを示した。この遺伝子の数は、線虫とタコ、ショウジョウバエとタコがそれぞれ共有する遺伝子の数よりも多く、系統的に離れたヒトとタコのカメラ眼に発現している遺伝子群は、系統的に近い生物種相互の場合よりも、より多くの共通祖先遺伝子から由来したことが明らかになった。第4章では、左右相称動物を対象として種の多様化の機構を、冗長性を排除したタンパク質配列データベースから抽出した延べ 1,236,790 個の遺伝子のクラスタリングから検討し、カンブリア紀までにおよそ 6,000 種類という膨大な遺伝子の祖先クラスターが形成されていたことを示した。また、クラスター数が進化とともに減少していることから、遺伝子ネットワーク等が種の多様化の推進力であると結論付けている。第5章は、全体のまとめである。

このように小倉君の研究は、今後継続的に増加していくさまざまな生物種の遺伝子配列データ、ゲノム配列データならびに遺伝子発現データといった大量のデータを組み合わせて進化機構を解明する新しい手法を提示したものであることから、小倉淳君の論文は博士号授与の要件を満たすと審査員全員一致で判断した。