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研究課題名（和文）Shedding light on genome evolution of a 220-million-year old's cockroach friend: Blattabacterium cuenoti

研究課題名（英文）Shedding light on genome evolution of a 220-million-year old's cockroach friend: Blattabacterium cuenoti

研究代表者
ブギーニョン　トマ（Bourguignon, Thomas）

沖縄科学技術大学院大学・進化ゲノミクスユニット・准教授

研究者番号：４０８１７５５８

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研究成果の概要（和文）：私は内部共生細菌におけるゲノム縮退の起因となる要因を識別するために５０以上のブラタバクテリウムゲノムについて配列決定ならびにアノテーションを行った。ブラタバクテリウムにおけるゲノム縮小の主要因が突然変異率（の増加）であったことを見出した。遺伝子欠損を駆動するその他の要因としては、選択圧の緩和および代謝機能的に関連した遺伝子群が欠損するドミノ効果が含まれる。集団ボトルネックによる遺伝的浮動については遺伝子欠損と相関がなく、ブラタバクテリウムのゲノム縮小におけるマラーのラチェット役割は有意ではないことが示唆された。ゆえに本研究は昆虫内部共生細菌の縮小ゲノム進化に対する我々の理解を深めるものである。

研究成果の学術的意義や社会的意義
We already published one paper in Genome Biology and Evolution (IF: 3.73). For another paper, we resubmitted a revised version that addressed the comments of the reviewers and is now under reevaluation for publication in Current Biology (IF: 9.19). I expect one last paper to be submitted this year.

研究成果の概要（英文）：I sequenced, assembled and annotated >50 Blattabacterium genomes that I used to identify the factors at the origin of genome erosion in endosymbionts. I found that the main factor linked to genome reduction in Blattabacterium was mutation rate. Other factors driving gene loss include relaxed selection, and the domino effect caused by the loss of metabolically related genes. However, genetic drift resulting from population bottlenecks showed no correlation with gene loss, suggesting that the Muller’s Ratchet plays no significant role in Blattabacterium genome erosion. This project therefore improved our understanding of the evolution of small genomes in insect endosymbionts.

研究分野：Evolutionary biology

キーワード：cockroach　Blattabacterium　symbiosis　endosymbiont

様　式　Ｃ－１９、Ｆ－１９－１、Ｚ－１９（共通）

１．研究開始当初の背景

Blattabacterium are cockroach endosymbionts that assist their host with their nitrogen metabolism. All cockroaches, with the exception of non-Mastotermitidae termites and *Nocticola*, host *Blattabacterium* in their fat body cells. Cockroaches acquired *Blattabacterium* >200 million years ago and the bacterium has been transmitted across generations of their cockroach hosts since then. Like it is generally the case for intracellular endosymbionts, *Blattabacterium* have small genomes, typically <650kb. Previous studies have shown that genome size is not entirely stable and continues to shrink as genes are lost by *Blattabacterium* over millions of years of evolution. In consequence of their small genome size, *Blattabacterium* largely rely on their cockroach hosts for their own metabolism, as they lost a great many genes required for a free-living life style. A few genomes of *Blattabacterium* have been sequenced in the course of the last decade, but they are not representative of the diversity of cockroaches, and genomes are missing for rare cockroach families. No *Blattabacterium* genome is available for most lineages of cockroaches. In consequence, it is still unclear how these genomes have evolved during the last 200 million years. More genomes are required to determine how *Blattabacterium* have evolved, and what is their faith. Additionally, a robust phylogeny of *Blattabacterium* can be used as a proxy for the termite phylogeny, as cockroaches and *Blattabacterium* strictly coevolve.

２．研究の目的

The main goal of this project was to sequence new *Blattabacterium* genomes in order to study how they evolve. With a high number of *Blattabacterium* genomes, we were planning to determine the cause of genome reduction in these endosymbionts. The classical approach to study endosymbiont genome evolution is to compare endosymbiont genomes to the genomes of their free-living relatives. While this approach is very relevant, it only allows to determine how genomes evolve during the initial stage of endosymbiogenesis. However, genomes of endosymbionts continue to evolve following this initial stage, and continue to erode, albeit at an extremely slow pace that give a false impression of stability. A large number of endosymbiont genome sequences are needed to study this second phase of genome evolution and determine how genes are lost in endosymbionts. Our main goal was therefore to test how the genomes of strict endosymbionts evolve. More precisely, we tested the effect of the Muller's Ratchet, high mutation rate, relaxed selection, and domino effect on gene loss. This allowed us to determine the proximal cause of endosymbiont genome reduction for the first time.

３．研究の方法

We obtained a total of 67 *Blattabacterium* genomes, which is unprecedented for any endosymbiont lineage. We found that about 350 genes have been preserved across all these strains and used these orthologous genes to reconstruct a phylogenetic tree of Blattabacterium, and therefore a phylogenic tree of their cockroach host. This phylogenetic tree allowed us to estimate a series of parameters that are relevant to genome reduction. We estimated across the tree: dN values, dS values, dN/dS values, mutation rates and rates of gene loss. We then modelled these rates on the tree in order to determine the cause of gene loss. We repeated the analyses using various estimations of dN values, dS values, dN/dS values and mutation rates, with the aim of controlling for any bias inherent to the analyses. All analyses largely concur and showed that our results were particularly robust.

４．研究成果

We found that the Muller's Ratchet does not explain gene loss in *Blattabacterium*. This was largely unexpected, as the Muller's Ratchet hypothesis has been the dominant hypothesis for the last 25 years. Instead, we found that gene loss in endosymbionts is primarily caused by mutation rate. This suggested two possibilities: either (1) *Blattabacterium* is a special case that does not follow the same trend as other endosymbiotic bacteria, or (2) the general approach that has been used to test the effect of the Muller's Ratchet on other endosymbionts wasn't completely appropriate. To determine which scenario is correct, we carried the same analyses on another eight lineages of bacteria. We included other lineages of endosymbionts and free-living

bacteria as a comparison. We did not sequence the genomes of these bacteria, but instead opted to work on genomes available from online depositories. We tested whether mutation rate also affect gene loss in other bacterial lineages and found that it does in seven out of nine lineages of bacteria. This included several lineages of free-living bacteria, and therefore showed that the mechanism of genome erosion we found for *Blattabacterium* is valid for all prokaryotes. In other words, we have demonstrated that mutation rate is one of the primary cause of genome reduction in bacteria. Our results are important for the understanding of genome size evolution.

５．主な発表論文等

〔雑誌論文〕　計2件（うち査読付論文　0件／うち国際共著　0件／うちオープンアクセス　0件）

１．著者名 Kinjo Yukihiro、Bourguignon Thomas、Tong Kwei Jun、Kuwahara Hirokazu、Lim Sang Jin、Yoon Kwang Bae、Shigenobu Shuji、Park Yung Chul、Nalepa Christine A、Hongoh Yuichi、Ohkuma Moriya、Lo Nathan、Tokuda Gaku	４．巻 10
２．論文標題 Parallel and Gradual Genome Erosion in the Blattabacterium Endosymbionts of Mastotermes darwiniensis and Cryptocercus Wood Roaches	５．発行年 2018年
３．雑誌名 Genome Biology and Evolution	６．最初と最後の頁 1622～1630
掲載論文のDOI（デジタルオブジェクト識別子） https://doi.org/10.1093/gbe/evy110	査読の有無 無
オープンアクセス オープンアクセスではない、又はオープンアクセスが困難	国際共著 －

１．著者名 Arab Daej A.、Bourguignon Thomas、Wang Zongqing、Ho Simon Y. W.、Lo Nathan	４．巻 16
２．論文標題 Evolutionary rates are correlated between cockroach symbionts and mitochondrial genomes	５．発行年 2020年
３．雑誌名 Biology Letters	６．最初と最後の頁 20190702
掲載論文のDOI（デジタルオブジェクト識別子） https://doi.org/10.1098/rsbl.2019.0702	査読の有無 無
オープンアクセス オープンアクセスではない、又はオープンアクセスが困難	国際共著 －

〔学会発表〕　計1件（うち招待講演　0件／うち国際学会　1件）

１．発表者名 Thomas Bourguignon
２．発表標題 過去２億３千万年のゴキブリ－ブラタバクテリウムのゲノム浸食
３．学会等名 Entomological Society of Japan（国際学会）
４．発表年 2018年

〔図書〕　計0件

〔産業財産権〕

〔その他〕

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６．研究組織

	氏名 （ローマ字氏名） （研究者番号）	所属研究機関・部局・職 （機関番号）	備考