

氏 名	OLA ALESSA
授与した学位	博 士
専攻分野の名称	農 学
学位授与番号	博甲第 6 6 5 4 号
学位授与の日付	2 0 2 2 年 3 月 2 5 日
学位授与の要件	環境生命科学研究科 農生命科学専攻 (学位規則第 4 条第 1 項該当)
学位論文の題目	The taxonomic and phenotypic diversity of <i>Methylobacterium</i> species (<i>Methylobacterium</i> 属細菌の分類及び表現型の多様性)
論文審査委員	教授 佐藤和広 教授 鈴木信弘 教授 ガリス イバン 准教授 谷明生
学位論文内容の要旨	
<p>The pink pigmented facultative methylotrophs (PPFMs) are a major bacterial group found in the plant phyllosphere. They represent around 10-20% of the bacterial communities and mostly encountered as an endophyte, directly carried to new plants through the seeds. <i>Methylobacterium</i> species can enhance the plant growth and increase its tolerance to multiple stresses. The number of validated species have been increasing rapidly and reached 63 species recently. These species were separated based on multi-locus sequence analysis (MLSA) into two genera <i>Methylorubrum</i> and <i>Methylobacterium</i>. Within these genera, however, some species lack either pigmentation or methylotrophy, which raises the question of what actually defines the PPFMs. In the present study, a comprehensive comparative genomics approach was employed to explore the phylogenetic relationship and to explain their genotypic differences that confer this group phenotypes. We newly sequenced the genomes of 29 relevant strains to complete a dataset for almost all PPFMs type strains, 33 type strains assemblies were downloaded from NCBI and JGI data bases. To clarify unique genes found only in <i>Methylobacterium</i> species, 16 species from the family <i>Methylobacteriaceae</i> were added. Totally a set of 78 species were analyzed. The phylogeny of this bacterial group was constructed with three approaches, genome blast-based phylogeny, the alignment of 711 core genes, and presence and absence matrix of all genes in the pan genome. Using these approaches, I was able to identify that some species were heterotypic synonyms and divided the PPFMs into eight homogenous sub-clades. Most members in the same sub-clade had a close phenotyping characteristic, these phenotypes included ability to utilize C1 compounds, complex sugars, and mediums.</p> <p>Pan genome analysis have shown that at least one methanol dehydrogenase cluster was conserved in the core genome of this bacterial group, and other important genes related to C1 utilization were conserved. These genes sets were found to be one of the unique features of the PPFM species compared to <i>Microvirga</i> sp. This highlights the importance of methanol as a source of energy and carbon for this bacterial group.</p> <p>Number of secondary metabolites cluster were found in their genomes, and these gene clusters might be important for the PPFMs activity as a biocontrol agent. All PPFM strains were able to produce indole acetic acid, they also did not induce any immune response in rice cells, and some were able to inhibit the growth of pathogenic fungi. This is the first research that approaches all the PPFM type strains and explains in detail their unique features which are related to C1 metabolism and their symbiotic interaction with the plant.</p>	

論文審査結果の要旨

植物が放出するメタノールを利用して生育出来る*Methylobacterium*属細菌は、植物葉上の微生物群集の中で優占化し、多くはピンク色の色素を持っているため、Pink-Pigmented Facultative Methylophs (PPFM) と呼ばれる。本属細菌は様々な環境から分離され約60種が知られているが、メタノールの資化性を示さないものや色素を持たないものも存在する。またメチルアミン資化性、メタノールの代謝経路などにも多様性が見られている。しかし、本属細菌のゲノム情報は60種のうち半分程度の種しか存在せず、どのような遺伝的性質が本属細菌を定義づけるのかは不明であった。

本研究では*Methylobacterium*属細菌の基準株全てのゲノム情報を完全に整備し、配列情報から導き出される他の属の細菌との分類学的な違い、及び属内での種の違い、遺伝子・機能モジュールの構成の違い、メタノール代謝酵素遺伝子のレパートリーの違いを情報科学的に明らかにした。さらに実際の菌株を使って可能な限りのフェノタイプの解析を行い、ジェノタイプで定義されるフェノタイプとの対応を実験的に明らかにした。

本研究の成果はFrontiers in Microbiology誌に掲載された。シーケンシングからアセンブリ、アノテーションまでを60株もの細菌ゲノムについて詳細な解析を行い、ドライ解析だけでなくフェノタイプとの対応まで明らかにしていることが学術的に高く評価できる。国内・国際学会で研究成果を発表しており、演習科目での他の研究分野の発表にも積極的に質問議論し、学位発表も複雑なデータを簡潔にまとめて発表できた。

学位論文の内容は論文提出者が論文発表会において口頭発表し、質問およびコメントに対して的確に回答しており、学位審査委員会において博士の学位を授与するに値すると判断された。