



# Complete genome sequence of *Variovorax* sp. PMC12, a plant growth-promoting bacterium conferring multiple stress resistance in plants

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## 다양한 스트레스에 대한 식물의 내성을 유도하는 식물생육촉진 세균 *Variovorax* sp. PMC12 균주의 유전체 염기서열

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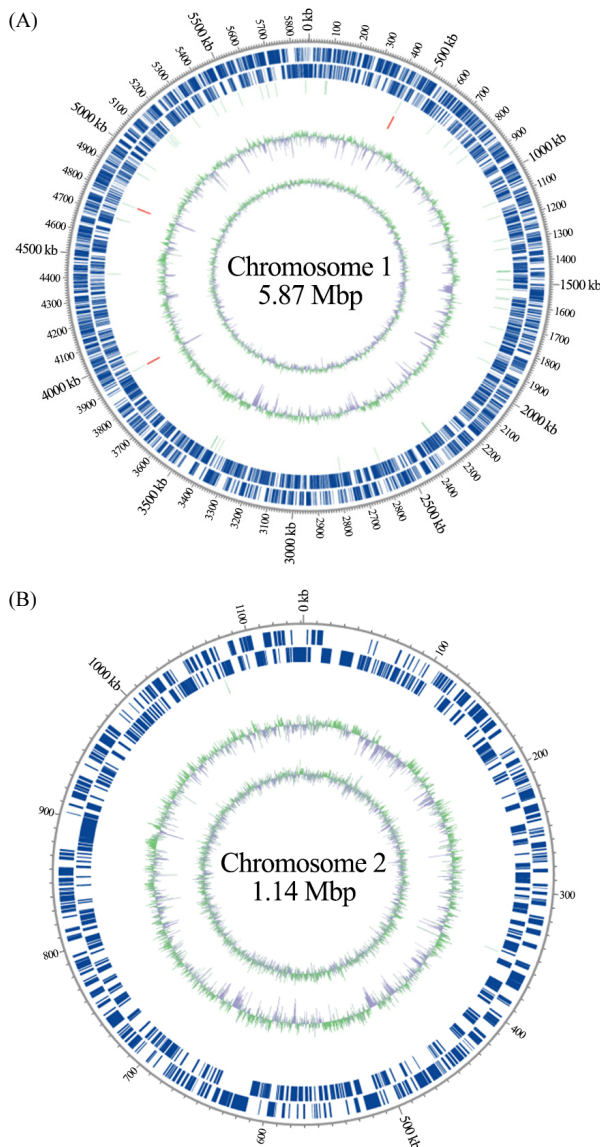
*Variovorax* sp. PMC12 is a rhizobacterium isolated from tomato rhizosphere and enhanced the plant resistance to abiotic and biotic stresses. Here we present the complete genome sequence of strain PMC12. The genome is comprised of two circular chromosomes harboring 5,873,297 bp and 1,141,940 bp, respectively. A total of 6,436 protein-coding genes, 9 rRNAs, 64 tRNAs, 3 ncRNAs, and 80 pseudogenes were identified. We found genes involved in 1-aminocyclopropane-1-carboxylate (ACC) deaminase, antioxidant activity, phosphate solubilization, and biosynthesis of proline and siderophore. Those genes may be related to capability of improving plant resistance to various stresses including salinity, cold temperature, and phytopathogen.

**Keywords:** *Variovorax*, plant growth-promoting rhizobacteria, plant stress

Members of *Variovorax* are a Gram-negative bacteria belonging to the family *Comamonadaceae*, the class *Betaproteobacteria* (Willems *et al.*, 1991). They were initially isolated

from contaminated soil and tolerate to various metals including Cd, Co, Cu, Ni, and Zn and capable of degrading a wide range of substrates (Willems *et al.*, 1991; Belimov *et al.*, 2005; Satola *et al.*, 2013). In addition, some strains of *Variovorax* inhabit interior and exterior of plant roots and confer beneficial effect on plant growth (Han *et al.*, 2011). It has been reported that *V. paradoxus* 5C-2 with 1-aminocyclopropane-1-carboxylate (ACC) deaminase activity enhanced plant growth in arid soil (Belimov *et al.*, 2009) and *V. paradoxus* S110 isolated from potato plant has been well studied its metabolic diversity and various properties associated with host plants (Han *et al.*, 2011). In the previous study, strain PMC12 was isolated from tomato rhizosphere and exhibited potential for application as biostimulant and biocontrol agent; promoting tomato (*Solanum lycopersicum* L.) plant growth in abiotic stress conditions such as salinity and low temperature (10°C) and enhancing the resistance to bacterial wilt disease caused by *Ralstonia solanacearum* (Kim *et al.*, 2018). To gain better knowledge about molecular mechanisms of *Variovorax*, we carried out complete genome sequencing of strain PMC12 and found out genes that

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**Fig. 1. Circular maps of chromosomes 1 (A) and 2 (B) of *Variovorax* sp. PMC12.** Each circle from outside to the center represents the GC skew, the black CDS on forward strand, CDS on reverse strand, tRNA, rRNA, GC content, and GC skew.

are predicted to be involved in plant growth promotion under abiotic and biotic stress conditions.

The genome of strain PMC12 was sequenced using the PacBio RSII and Illumina HiSeq platforms at Microgen Inc. The sequences generated by PacBio RSII were assembled *de novo* using RSHGAP assembly version 3.0 (Chin *et al.*, 2013) and HiSeq reads were subsequently used for error correction of the draft genome assemblies by using Pilon version 1.21 (Walker *et al.*, 2014). Gene prediction and functional annotation

**Table 1. Genome features of *Variovorax* sp. PMC12**

Genome feature	Chromosome 1	Chromosome 2	Total
Accession number	CP027773	CP027774	
Genome size (bp)	5,873,297	1,141,940	7,015,237
G + C content (%)	67.6	67.7	67.61
Protein-coding genes	5,438	998	6,436
tRNAs	62	2	64
rRNAs (5S, 16S, 23S)	9 (3, 3, 3)	-	9 (3, 3, 3)
Pseudogenes	56	24	80

were carried out using the NCBI prokaryotic genomes annotation pipeline (Tatusova *et al.*, 2016) and the RAST server (Aziz *et al.*, 2008). The genes involved in the secondary metabolite production were analyzed using antiSMASH version 4.0.0 (Blin *et al.*, 2017).

The complete genome of strain PMC12 is composed of two circular chromosomes; one is a 5,873,297 bp chromosome with 67.6% G + C content and the other is an 1,141,940 bp chromosome with 67.7% G + C content (Fig. 1). The genome contains 6,436 protein-coding genes, 9 rRNAs, 64 tRNAs, 3 ncRNAs, and 80 pseudogenes (Table 1). Functional annotation reveals that the genome of strain PMC12 possesses a numbers of genes associated with plant growth promotion and stress tolerance. ACC deaminase gene (*acdS*) is well-known to promote plant growth by reducing phytohormone ethylene. The genes involved in proline biosynthesis (*proA*, *proB*, and *proC*) are related to produce organic osmolytes increasing plant stress tolerance. There are 7 genes encoding the antioxidant enzymes, superoxide dismutase (SOD), catalase (CAT), and peroxidase (PRX), that scavenge reactive oxygen species. Strain PMC12 possesses *phnA* and *phnX* genes involved in hydrolysis of phosphonoacetate and phosphonoacetaldehyde, respectively that could increase biologically available phosphate for plants. The homologous gene cluster for biosynthesis of variochelin, a lipopeptide siderophore (Kurth *et al.*, 2016), were found in the chromosome 2 of strain PMC12 genome. This study will provide basic information for understanding of molecular mechanisms where rhizobacteria enhances plant growth under environmental stresses and resistance to bacterial wilt disease.

#### Nucleotide sequence accession numbers

The genome sequences of two chromosomes described in

this study were available under the accession numbers CP027773 and CP027774 in GenBank database.

## 적 요

본 연구에서 생물 및 비생물학적 스트레스에 내성을 유도하는 식물 생육 촉진 세균인 *Variovorax* sp. PMC12 균주의 유전체 염기서열을 분석하였다. PMC12 균주의 유전체는 5,873,297 bp와 1,141,940 bp 크기의 원형 염색체 2개로 구성되었다. 총 6,436개 단백질 유전자, rRNA 9개, tRNA 64개, ncRNA 3개와 유사유전자 80개가 확인되었다. 유전체상에서 발견된 1-aminocyclopropane-1-carboxylate (ACC) deaminase, 항산화 활성, 인산 가용화, 프롤린 생합성, 시드로포어 생합성과 관련된 유전자들은 PMC12 균주가 염, 온도, 병원균에 대한 스트레스에 대한 식물의 내성 유도와 관련되어 있을 것으로 판단된다.

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## References

- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formisano K, Gerdes S, Glass EM, Kubal M, *et al.* 2008. The RAST server: rapid annotations using subsystems technology. *BMC Genomics* **9**, 75.
- Belimov AA, Dodd IC, Hontzeas N, Theobald JC, Safronova VI, and Davies WJ. 2009. Rhizosphere bacteria containing 1-aminocyclopropane-1-carboxylate deaminase increase yield of plants grown in drying soil via both local and systemic hormone signalling. *New Phytol.* **181**, 413–423.
- Belimov AA, Hontzeas N, Safronova VI, Demchinskaya SV, Piluzza G, Bullitta S, and Glick BR. 2005. Cadmium-tolerant plant growth-promoting bacteria associated with the roots of indian mustard (*Brassica juncea* L. Czern.). *Soil Biol. Biochem.* **37**, 241–250.
- Blin K, Wolf T, Chevrette MG, Lu X, Schwalen CJ, Kautsar SA, Suarez Duran HG, de Los Santos ELC, Kim HU, Nave M, *et al.* 2017. antiSMASH 4.0-improvements in chemistry prediction and gene cluster boundary identification. *Nucleic Acids Res.* **45**, W36–W41.
- Chin CS, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, *et al.* 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. *Nat. Methods* **10**, 563–569.
- Han JI, Choi HK, Lee SW, Orwin PM, Kim J, Laroe SL, Kim TG, O’Neil J, Leadbetter JR, Lee SY, *et al.* 2011. Complete genome sequence of the metabolically versatile plant growth-promoting endophyte *Variovorax paradoxus* S110. *J. Bacteriol.* **193**, 1183–1190.
- Kim HS, Lee SA, Kim Y, Sang MK, Song J, Chae JC, and Weon HY. 2018. Enhancement of tomato tolerance to biotic and abiotic stresses by *Variovorax* sp. PMC12. *Res. Plant Dis.* **24**, 221–232.
- Kurth C, Schieferdecker S, Athanasopoulou K, Seccareccia I, and Nett M. 2016. Variochelins, lipopeptide siderophores from *Variovorax boronicumulans* discovered by genome mining. *J. Nat. Prod.* **79**, 865–872.
- Satola B, Wubbeler JH, and Steinbuchel A. 2013. Metabolic characteristics of the species *Variovorax paradoxus*. *Appl. Microbiol. Biotechnol.* **97**, 541–560.
- Tatusova T, DiCuccio M, Badretdin A, Chetvermin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, and Ostell J. 2016. NCBI prokaryotic genome annotation pipeline. *Nucleic Acids Res.* **44**, 6614–6624.
- Walker BJ, Abeel T, Shea T, Priest M, Abouelliel A, Sakthikumar S, Cuomo CA, Zeng Q, Wortman J, Young SK, *et al.* 2014. Pilon: An integrated tool for comprehensive microbial variant detection and genome assembly improvement. *PLoS One* **9**, e112963.
- Willems A, Deley J, Gillis M, and Kersters K. 1991. *Comamonadaceae*, a new family encompassing the acidovorans rRNA complex, including *Variovorax-paradoxus* gen. nov., comb. nov., for *Alcaligenes-paradoxus* (Davis 1969). *Int. J. Syst. Bacteriol.* **41**, 445–450.