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Draft genome sequence of *Fusobacterium polymorphum* KCOM 1001 isolated from a human subgingival dental plaque of gingivitis lesion

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사람 치은염 병소 치은연하치면 세균막에서 분리된 *Fusobacterium polymorphum* KCOM 1001의 유전체 염기서열 해독

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Recently, *Fusobacterium nucleatum* subsp. *polymorphum* was reclassified as *Fusobacterium polymorphum* based on the average nucleotide identity and genome-to-genome distance analyses. *F. polymorphum* is a Gram-negative, anaerobic, and filament-shaped bacterium. *F. polymorphum* is a part of normal flora of oral cavity and causative agent of periodontal diseases. *F. polymorphum* KCOM 1001 (= ChDC F119) was isolated from a human subgingival plaque of gingivitis lesion. Here, we present the complete genome sequence of *F. polymorphum* KCOM 1001.

Keywords: *Fusobacterium polymorphum*, gingivitis, human

Fusobacterium nucleatum subsp. *polymorphum* was classified as one of four subspecies of *Fusobacterium nucleatum* by DNA-DNA hybridization (DDH) and polyacrylamide gel electrophoresis (PAGE) protein pattern of whole-cell proteins or electrophoretic patterns of glutamate dehydrogenase and 2-oxoglutarate reductase (Dzink *et al*., 1990; Gharbia and Shah, 1989, 1990). Recently, *F. nucleatum* subsp. *polymorphum* was reclassified as *Fusobacterium polymorphum* based on the average nucleotide identity and genome-to-genome distance analyses which were introduced as a replacement for DDH to delineate bacterial species (Kook *et al*., 2017). *F. polymorphum* is a Gram-negative, anaerobic, and filament-shaped bacterium (Strauss *et al*., 2008). *F. polymorphum* is a part of normal flora of oral cavity and causative agent of periodontal diseases (Haffajee and Socransky, 1994; Han, 2015). *F. polymorphum* KCOM 1001 (= ChDC F119) was isolated from a human subgingival plaque of gingivitis lesion. In this report, we present the complete genome sequence of *F. polymorphum* KCOM 1001.

The *F. polymorphum* KCOM 1001 was grown in brain heart infusion (BHI, Difco Laboratories) medium supplemented with 0.5% yeast extract, 0.05% cysteine HCl-H_2O, 0.5 mg/ml of hemin, 2 μg/ml of vitamin K_1, and 5% sheep blood in an anaerobic chamber (Model Bactron I) was maintained using a gas mixture of 10% H_2, 5% CO_2, and 85% N_2 (Park *et al*., 2013). The bacterial genomic DNA was prepared as described previously and DNA concentration was determined by the Epoch™ Microplate Spectrophotometer (BioTek Instruments

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Inc.) at wavelengths of 260 and 280 nm (Cho *et al*., 2015).

The genomic DNA of *F. polymorphum* KCOM 1001 was sequenced using the Illumina Hiseq 2000 platform by Macrogen Inc. The library of 5 kb mate-pair was sequenced which reached coverage of 1,370 x. The de novo assembly was performed by SPAdes (version: 3.8.2) (Bankevich *et al*., 2012) and AlignGraph (Bao *et al*., 2014). All gaps among the scaffolds were filled by GapCloser (Luo *et al*., 2012; http://sourceforge.net/projects/soapdenovo2/files/GapCloser). And we confirmed the scaffolds were placed at gaps on the largest scaffold by dot plot analysis. Finally, the assembly was polished by iCORN2 (Otto *et al*., 2010). Genome annotation was conducted by the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (https://www.ncbi.nlm.nih.gov/genome/annotation_prok/).

The complete genome of *F. polymorphum* KCOM 1001 is 2,523,644 bp in length and has a G + C content of 26.7% (Table 1). A total of 2,296 protein-coding sequences (CDSs), 13 rRNAs including one partial 16S rRNA, and 45 tRNAs were annotated (Table 1). The genome sequence contained virulence factors such as hemolysin A, hemolysin transporter protein ShlB, metalloprotease LoiP/ YpwA, ATP-dependent zinc metalloprotease FtsH, protease HtpX, sialic acid TRAP transporter permease protein SiaT, outer membrane porin F, multidrug export ATP-binding/permease protein, multidrug export protein MepA, putative multidrug export ATP-binding/permease protein, multi-drug resistance protein MexA/MdtK/NorM, metallo-beta-lactamase superfamily protein, beta-lactamase CARB-6 precursor, macrolide export ATP-binding/permease protein MacB, antitoxin HicB/YwqK/YefM/RelB/DinJ, and transport protein TonB. The genome contained SPBc2 prophage-derived glycosyltransferase SunS and putative DNA-invertase from lambdoid prophage Rac. The complete genome encodes for involving the biofilm formation, autoinducer-2 (AI-2) modifying protein LsrG, toxin-antitoxin biofilm protein TabA, glycosyltransferase family 9 (heptosyl-transferase), and fibronectin-binding protein A N-terminus (FbpA). It also contained type II secretion system protein, type IV secretion system protein virB4/virB9/PtlH/PtlG, preprotein translocase subunit YajC, and protein translocase subunit SecA/SecD/SecE/SecF/SecY.

Table 1. Genome features of *Fusobacterium polymorphum* KCOM 1001

Attribute	Value
Genome size (bp)	2,523,644
GC content (%)	26.7
No. of contigs	3
Total genes	2,454
Protein-coding genes	2,296
tRNA	45
Complete rRNA (5S, 16S, 23S)	12 (4, 2, 4)
Partial rRNA (16S)	1
ncRNA	3
Pseudogene	97

The genome also contained the oxidative stress-response genes such as anaerobic nitric oxide reductase flavorubredoxin, glutaredoxin, thioredoxin reductase, nitroreductase A, peptide methionine sulfoxide reductase MsrA/MsrB, pyruvate-flavodoxin oxidoreductase, thiol-disulfide oxidoreductase ResA, and putative oxidoreductase. The genome contained the four two-component systems (YpdA/YpdB, PdtaS/PdtaR, putative sensor histidine kinase/PhoB, and YehU/putative response regulatory protein).

The *Fusobacterium polymorphum* KCOM 1001 strain was deposited in the Korean Collection for Oral Microbiology (Gwangju, Korea).

Nucleotide sequence accession number

This Whole Genome Shotgun project has been deposited at DDBJ/ENA/GenBank under the accession NHRT00000000. The version described in this paper is version NHRT01000000.

적 요

최근 *Fusobacterium nucleatum* subsp. *polymorphum*는 average nucleotide identity 및 genome-to-genome distance 분석법에 의해 *Fusobacterium polymorphum*로 재분류 되었다. *F. polymorphum* 그람 음성이면서, 혐기성 및 가는 섬유 모양의 세균이다. *F. polymorphum*은 사람의 구강 내 정상세균총의 하나이고, 치주질환의 원인 인자이다. *F. polymorphum* KCOM 1001 (= ChDC F119) 균주가 사람 치은염 병소의 치은연하치면세균막에서 분리되었다. *F. polymorphum* KCOM 1001 균주 유전체 염기서열을 해독하여 보고한다.

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