

# Genome Sequence of *Corynebacterium glutamicum* ATCC 14067, Which Provides Insight into Amino Acid Biosynthesis in Coryneform Bacteria

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**We report the genome sequence of *Corynebacterium glutamicum* ATCC 14067 (once named *Brevibacterium flavum*), which is useful for taxonomy research and further molecular breeding in amino acid production. Preliminary comparison with those of the reported coryneform strains revealed some notable differences that might be related to the difficulties in molecular manipulation.**

Several *Brevibacterium* species, such as *Brevibacterium lactofermentum* and *Brevibacterium flavum* (isolated as an L-glutamate and an L-lysine overproducing bacterium, respectively), being also non-spore-forming, Gram-positive bacteria, had been considered to be related to *Corynebacterium glutamicum* (10). Although the complete genome sequences of two variants of *C. glutamicum* ATCC 13032 have been published (5, 6), some molecular manipulation of *Brevibacterium flavum*, especially gene knockout strategy, still has been reported to be relatively difficult (14); this might be related to specific or unclear genetic information about *C. glutamicum*, such as its gene-restricted modification mechanism (3, 13). Here we report the genome sequence of the above-mentioned coryneform bacteria ATCC 14067 (once named *Brevibacterium flavum*) for deep insight into the genetic background of *Brevibacterium flavum*, which might be useful for taxonomy research and further molecular breeding in amino acid production.

The genome was sequenced using the Illumina Solexa GA IIX instrument at Beijing Genomics Institute (BGI) (Shenzhen, China). A library containing 500-bp inserts was constructed. Sequencing was performed with the paired-end strategy of 90-bp reads to produce 400 Mb of filtered sequences, representing a 121.95-fold coverage of the genome. The sequences were assembled into 104 contigs and 60 scaffolds using the SOAPdenovo package (9). Some remaining gaps in scaffolds were closed by Sanger sequencing of PCR products. The chromosome of *Brevibacterium flavum* ATCC 14067 is 3,273,044 bp in length, with an average G+C content of 54.13%.

Genome annotation was performed at the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP), in which 3,143 open reading frames (ORFs), 64 tRNA genes, and 7 rRNA genes were identified by Glimmer 3.02 (4), Genemark (2), tRNAscan-SE (11), and RNAmmer (8). The GenBank NR database and the KEGG (7) and COG (12) databases were employed for BLASTP identification (1). A total of 24 pseudogenes were also detected.

A brief comparative analysis of some genes related to the glutamate, lysine, and arginine synthesis pathway and the gene-restricted modification system was conducted between ATCC 14067, ATCC 13032, and *Corynebacterium glutamicum* R. The preliminary results showed that some notable sequence differences and specific arrangements might lead to difficulties in amino acid pathway reconstruction. The abundant gene-restricted modification system being annotated might be the obstacle in gene knockout manipulation. The gap in the draft of ATCC 14067 is now being closed by a new 454 sequence

method using a 5K library. A complete genome sequence will be included in a future publication, which will also include the organization of some typical amino acid biosynthesis pathways and functional validation of some gene restriction mechanisms.

**Nucleotide sequence accession numbers.** This Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number [AGQQ00000000](https://www.ncbi.nlm.nih.gov/nuccore/AGQQ00000000). The version described in this paper is the first version, AGQQ01000000.

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