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An improved high-quality draft genome sequence of *Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup>

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**Abstract**

Despite their ubiquity and their involvement in food spoilage, the genus *Carnobacterium* remains rather sparsely characterized at the genome level. *Carnobacterium inhibens* K1<sup>T</sup> is a member of the *Carnobacteriaceae* family within the class *Bacilli*. This strain is a Gram-positive, rod-shaped bacterium isolated from the intestine of an Atlantic salmon. The present study determined the genome sequence and annotation of *Carnobacterium inhibens* K1<sup>T</sup>. The genome comprised 2,748,608 bp with a G + C content of 34.85 %, which included 2621 protein-coding genes and 116 RNA genes. The strain contained five contigs corresponding to presumptive plasmids of sizes: 19,036; 24,250; 26,581; 65,272; and 65,904 bp.

**Keywords:** *Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup>

**Introduction**

The genus *Carnobacterium* was proposed in 1987 to encompass a group of closely related bacteria originally classified as unusual species of *Lactobacillus* [1, 2]. The genus *Carnobacterium* includes heterofermentative, facultatively anaerobic, psychrotolerant, either motile or non-motile, Gram-positive rod-shaped lactic acid bacteria that produce mostly L-lactic acid by fermentation from glucose [3]. At present the genus contains 11 species with validly published names, which can be roughly divided into two groups. As the genus name implies, most *Carnobacterium* species (*Carnobacterium divergens, Carnobacterium gallinarum, Carnobacterium inhibens, Carnobacterium jeotgali, Carnobacterium maltaromaticum, Carnobacterium mobile, Carnobacterium viridans*) belong to a group that were originally isolated from biological sources such as living fish or foods derived from animal sources [4]. A second group of *Carnobacterium* spp. has been isolated from cold, low-nutrient environments such as Antarctic ice lakes (*C. funditum, C. alterfunditum, C. iners*) [5, 6] or Arctic permafrost (*C. pleistocentrum, C. inhibens* subsp. *gilichinskiyi*) [7, 8]. Owing to an upsurge in investigations involving *Carnobacterium* strains isolated from novel environments, at present genome sequences have been published for the following *Carnobacterium* environmental strains: *Carnobacterium* sp. 17–4 isolated from permanently cold sea water [9]; *C. maltaromaticum* strain ATCC 35586 isolated from a diseased salmon [10]; *C. maltaromaticum* strain LMA 28 isolated from ripened soft cheese [11]; and *C. inhibens* subsp. *gilichinskiyi* isolated from Siberian permafrost [8, 12]. However, to date only one published report of a genome sequence from a type strain of *Carnobacterium* has appeared, from *C. jeotgali* strain MS3<sup>T</sup> isolated from salt-fermented shrimp [13]. As part of a larger project to determine the genome sequences of all type strains of the genus *Carnobacterium*, the present study determined the classification and features of *Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup> [8] as well as its genome sequence and gene annotations.

**Organism Information**

**Classification and features**

*Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup> (= DSM 13024<sup>T</sup> = JCM 16168<sup>T</sup>) is the type strain of...
the species *C. inhibens* [8, 14]. The strain was isolated from the intestine of an Atlantic salmon [14]. The species epithet was derived from the Latin verb *inhibeo*, meaning “to inhibit”, referring to the growth-inhibitory activity that the bacterium shows [14]. Recent discovery of *C. inhibens* strain WN1359 from Siberian permafrost [15] prompted a re-examination of strains K1<sup>T</sup> and WN1359, resulting in the proposal to rename the K1<sup>T</sup> type strain as *C. inhibens* subsp. *inhibens* and the permafrost isolate *C. inhibens* subsp. *gilichinskyi* [8].

*Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup> is a motile Gram-positive rod (Fig. 1). It is a psychrophile that lacks both catalase and oxidase, does not grow on acetate containing media, but grows at pH 9 and in Tryptic Soy Broth containing up to 6 % (w/v) sodium chloride. Strain K1<sup>T</sup> is facultatively anaerobic and tryptone as a sole source of nutrient promotes growth. The most abundant cellular fatty acid of strain K1<sup>T</sup> is oleic acid (18:1 cis9) [14]. Classification of strain K1<sup>T</sup> according to the MIGS recommendations published by the Genome Standards Consortium is presented in Table 1.

*C. inhibens* subsp. *inhibens* strain K1<sup>T</sup> [8] was obtained from the German Collection of Microorganisms and Cell Cultures as strain DSM 13024. The strain was sub-cultured once and was stored as a ~70 °C frozen glycerol stock in the corresponding author’s strain collection as strain WN1362. DNA isolated from strain WN1362 corresponding to 16S rRNA gene sequences was PCR amplified with universal bacterial primers B27F (5’-GAGTTTGA TCMTGGCTCAG-3’) and B1512R (5’-AAGGAGGTGA TCCANCCRC-3’) as described previously [16] and sequenced at the University of Florida Interdisciplinary Center for Biotechnology Research (UF-ICBR). The sequence was compared with those obtained using NCBI BLAST [17] with the default settings (only highly similar sequences). The most frequently occurring genera were *Carnobacterium* (17 %) and unidentified bacteria (83 %) (100 hits in total). The species with the Max score was *Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup> (NCBI Reference Sequence NR_036895) with a shared identity of 100.0 %, thus verifying the identity of strain WN1362 with the type strain. An updated 16S rRNA phylogenetic analysis of *Carnobacterium* spp. isolates including *C. inhibens* subsp. *inhibens* strain K1<sup>T</sup> is presented in Fig. 2 to supplement and expand upon those published previously [8, 14, 15].

### Table 1 Classification and general features of *Carnobacterium inhibens* strain K1<sup>T</sup> according to the MIGS recommendations published by the Genome Standards Consortium [20]

| MIGS ID | Property | Term | Evidence code<sup>a</sup> |
|---------|----------|------|---------------------------|
| Current classification | Domain: Bacteria | Phylum: Firmicutes | TAS [34] |
| | Class: Bacilli | TAS [35, 36] |
| | Order: Lactobacillales | TAS [35, 37] |
| | Family: Carnobacteriaceae | TAS [35, 38] |
| | Genus: Carnobacterium | TAS [14] |
| | Species: Carnobacterium inhibens | TAS [14] |
| | Subspecies: Carnobacterium inhibens subsp. inhibens | TAS [8] |
| | Type strain: K1<sup>T</sup> (DSM 13024) | TAS [8] |
| Gram stain | Positive | TAS [14] |
| Cell shape | Rod | TAS [8, 14] |
| Motility | Motile | TAS [14] |
| Sporulation | Non-spore forming | TAS [8, 14] |
| Temperature range | 0–37 °C | TAS [8] |
| Optimum temperature | 35 °C | TAS [8] |
| pH range; | 6–9; 8.2 | TAS [8] |
| Carbon source | Tryptone | TAS [14] |
| MIGS-6 | Habitat | Gastrointestinal tract of fish (Atlantic salmon) | TAS [14] |
| MIGS-6.3 | Salinity | Grows at 0–6 % NaCl (w/v) | TAS [8, 14] |
| MIGS-22 | Oxygen requirement | Facultative anaerobe; grows better in absence of O<sub>2</sub> | TAS [8, 14, 15] |
| MIGS-15 | Biotic relationship | Unknown | |
| MIGS-14 | Pathogenicity | Unknown | |
| MIGS-4 | Geographic location | Göteborg, Sweden | |
| MIGS-5 | Sample collection | Unknown | |
| MIGS-4.1 | Latitude | Unknown | |
| MIGS-4.2 | Longitude | Unknown | |
| MIGS-4.3 | Depth | Unknown | |
| MIGS-4.4 | Altitude | Below ocean surface | TAS [14] |

<sup>a</sup>Evidence codes - IDA Inferred from Direct Assay, TAS Traceable Author Statement (i.e., a direct report exists in the literature), NAS Non-traceable Author Statement (i.e., not directly observed for the living, isolated sample, but based on a generally accepted property for the species, or anecdotal evidence). These evidence codes are from the Gene Ontology project [39].

![Fig. 1 Scanning electron micrograph of *Carnobacterium inhibens* subsp. *inhibens* strain K1<sup>T</sup>. Size bar is 1 μm in length](image)
Genome sequencing information

Genome project history
This organism was selected for sequencing on the basis of its relevance to environmental issues in phylogenetic diversity, bioenergy, and bioremediation, and is part of the Community Sequencing Program at the U.S. Department of Energy, Joint Genome Institute for projects of relevance to agency missions (http://www.jgi.doe.gov). The project is registered in the Genomes OnLine Database [18] and the permanent draft genome sequence is deposited in GenBank. Draft sequencing and assembly were performed at JGI using state of the art sequencing technology [19]. A summary of the project information is shown in Table 2, which presents the project information and its association with MIGS version 2.0 compliance [20].

Table 2 Carnobacterium inhibens subsp. inhibens strain K1<sup>T</sup> genome sequencing project details

| MIGS ID | Property                      | Term                          |
|---------|-------------------------------|-------------------------------|
| MIGS-31 | Finishing quality            | Improved High-Quality Draft   |
| MIGS-28 | Libraries used               | PacBio                        |
| MIGS-29 | Sequencing platforms         | PacBio                        |
| MIGS-31.2| Fold coverage               | 273.1x                        |
| MIGS-30 | Assemblers                   | HGAP v.2.1.1                  |
| MIGS-32 | Gene calling method          | Prodigal 2.5                  |
|         | Locus Tag                    | BR65                          |
|         | Genbank ID                   | JQIV01000006.1                |
|         | Genbank Date of Release      | 16 August 2015                |
|         | GOLD ID                      | Gp0042S80                     |
|         | BIODIAGRAM PROJECT           | PRJNA234895                   |
| MIGS-13 | Source material identifier   | DSM 13024<sup>T</sup>         |
|         | Project relevance            | Environmental                  |
Growth conditions and genomic DNA preparation
Strain K1^T was grown to stationary phase by incubation for 36 h at 20 °C in TSY medium without shaking [8]. DNA was isolated from 100 mL of culture using a CTAB bacterial genomic DNA isolation method following the protocol recommended by JGI [21]. DNA fragment size and quality was confirmed by agarose gel electrophoresis and DNA was quantified by fluorometry (Qubit fluorometer, Invitrogen).

Genome sequencing and assembly
The draft genome of *Carnobacterium inhibens* K1 was generated at the DOE Joint genome Institute using the Pacific Biosciences sequencing technology [19]. A PacBio SMRTbell™ library was constructed and sequenced on the PacBio RS platform, which generated 252,358 filtered sub-reads totaling 752.5 Mbp. All general aspects of library construction and sequencing performed at the JGI can be found at (http://www.jgi.doe.gov). The raw reads were assembled using HGAP (version: 2.1.1) [22]. The final draft assembly contained six contigs in six scaffolds, totaling 2.7 Mbp in size. The input read coverage was 273.1 ×.

Genome annotation
The assembled sequence was annotated using the JGI prokaryotic annotation pipeline [23] and was further

![Fig. 3 Graphical map of the six scaffolds assembled for the genome of *Carnobacterium inhibens* K1^T, DSM 13024. From top to bottom, the scaffolds are: DSM 13024: DR65DRAFT_scf7180000000016_quiver.6, DSM 13024: DR65DRAFT_deg7180000000011.2, DSM 13024: DR65DRAFT_deg7180000000013.1, DSM 13024: DR65DRAFT_deg7180000000014.3, DSM 13024: DR65DRAFT_scf7180000000017, and DSM 13024: DR65DRAFT_scf7180000000019. From bottom to the top of each scaffold: Genes on forward strand (color by COG categories), Genes on reverse strand (color by COG categories), RNA genes (tRNAs green, rRNAs red, other RNAs black), GC content, GC skew.](image)
reviewed using the Integrated Microbial Genomes – Expert Review platform [24]. Genes were identified using Prodigal [25], followed by a round of manual curation using GenePRIMP [26] for finished genomes and Draft genomes in fewer than 10 scaffolds. The predicted CDSs were translated and used to search the National Center for Biotechnology Information nonredundant database, UniProt, TIGRFam, Pfam, KEGG, COG, and InterPro databases. The trnAScanSE tool [27] was used to find tRNA genes, whereas ribosomal RNA genes were found by searches against models of the ribosomal RNA genes built from SILVA [28]. Other non–coding RNAs such as the RNA components of the protein secretion complex and the RNase P were identified by searching the genome for the corresponding Rfam profiles using INFERNAL [29]. Additional gene prediction analysis and manual functional annotation was performed within the Integrated Microbial Genomes platform [23] developed by the Joint Genome Institute, Walnut Creek, CA, USA.

**Genome properties**

The genome includes five smaller contigs, for a total size of 201,043 bp, and one large contig of 2,547,565 bp (34.85 % GC content) (Fig. 3). For the genome, 2737 genes were predicted, 2621 of which are protein-coding genes. Of these, 2151 were assigned to a putative function with the remaining 470 genes annotated as hypothetical proteins. 1838 protein coding genes belong to paralogous families in this genome, corresponding to a gene content redundancy of 67.15 %. The statistics of the genome are summarized in Tables 3 and 4. Examination of the sequence data for the five small contigs revealed a variety of putative genes encoding plasmid functions such as: autonomous replication, mobilization, bacteriocin production and immunity, toxin-antitoxin systems, and Hg or Cd/Co resistance cassettes; therefore it is reasonable to assume that these five small contigs represent plasmids.

**Conclusion**

*Carnobacterium inhibens* is widely distributed in the environment, having been isolated from Atlantic salmon [14, 30], biogas slurry [31], a medicinal plant [32], and Siberian permafrost [8, 15]. In this communication we report an improved high-quality draft genome sequence of *Carnobacterium inhibens* subsp. *inhibens* strain K1T (= DSM 13024T = JCM 16168T). Genome analysis of this strain demonstrated a single presumed chromosome and at least five putative extrachromosomal elements.

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**Table 3** Genome statistics

| Attribute                  | Value     | % of Total |
|----------------------------|-----------|------------|
| Genome size (bp)           | 2,748,608 | 100.00     |
| DNA coding (bp)            | 2,356,497 | 85.73      |
| DNA G+C (bp)               | 957,950   | 34.85      |
| DNA scaffolds              | 6         | 100.00     |
| Total genes                | 2737      | 100.00     |
| Protein coding genes       | 2621      | 95.76      |
| RNA genes                  | 116       | 4.24       |
| Pseudo genes               | 66        | 2.41       |
| Genes in internal clusters | 515       | 18.82      |
| Genes with function prediction | 2151     | 78.59      |
| Genes assigned to COGs     | 1900      | 69.42      |
| Genes with Pfam domains    | 2196      | 80.23      |
| Genes with signal peptides | 113       | 4.13       |
| Genes with transmembrane helices | 691     | 25.25      |
| CRISPR repeats             | 0         | 0          |

**Table 4** Number of genes associated with general COG functional categories

| Code | Value | % age | Description                                           |
|------|-------|-------|------------------------------------------------------|
| A    | 25    | 1.20  | RNA processing and modification                      |
| B    | 19    | 0.91  | Chromatin structure and dynamics                     |
| C    | 71    | 3.39  | Defense mechanisms                                   |
| D    | 32    | 1.53  | Cell cycle control, Cell division, chromosome partitioning |
| E    | 163   | 7.79  | Carbohydrate transport and metabolism                |
| F    | 96    | 4.59  | Nucleotide transport and metabolism                  |
| G    | 76    | 3.63  | Coenzyme transport and metabolism                    |
| H    | 80    | 3.82  | Lipid transport and metabolism                       |
| I    | 102   | 4.87  | Inorganic ion transport and metabolism                |
| J    | 199   | 9.51  | General function prediction only                     |
| K    | 113   | 4.42  | Translation, ribosomal structure and biogenesis      |
| L    | 101   | 4.83  | Replication, recombination and repair                |
| M    | 113   | 4.42  | Cell wall/membrane biogenesis                        |
| N    | 51    | 2.44  | Cell motility                                        |
| O    | 61    | 2.91  | Posttranslational modification, protein turnover, chaperones |
| P    | 186   | 8.89  | Energy production and conversion                     |
| Q    | 156   | 7.45  | Function unknown                                     |
| R    | 837   | 30.58 | Not in COGs                                         |
| S    | 156   | 7.45  | Function unknown                                     |
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Authors’ contributions
WN supplied DNA and background information for this project and contributed to the assembly of the manuscript with CLD, AC, and NK. NS coordinated the project and all other authors were involved in either sequencing the genome and/or editing the paper. All authors read and approved the final manuscript.

Competing interests
The authors declare that they have no competing interests.

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References
1. Collins MD, Farrow JAE, Phillips BA, Fersus S, Jones D. Classification of Lactobacillus divergens, Lactobacillus piscicola, and some catalase-negative, anospore-forming, rod-shaped bacteria from poultry in a new genus, Carnobacterium. Int J Syst Bacteriol. 1987;37:310–6.
2. Schillinger U, Holzapfel WH. The genus Carnobacterium. In: Wood BJB, Holzapfel WH, editors. The Genera of Lactic Acid Bacteria. Volume 2. Springer Science+Business Dordrecht; 1995. p. 307–26.
3. Hammes WP, Hertel C. The genera Lactobacillus and Carnobacterium. In: Dworkin M, Falkow S, Rosenberg E, Schleifer KH, Staley JT, editors. The Prokaryotes. Volume 4. 3rd ed. Singapore: Springer; 2006. p. 320–403.
4. Leisner JJ, Laursen BG, Prevost H, Drider D, Dalgaard P. Carnobacterium: positive and negative effects in the environment and in foods. FEMS Microbiol Rev. 2007;31:592–613.
5. Franzmann PD, Höpfel P, Weiss N, Tindall BJ. Psychrotrophic, lactic acid-producing bacteria from anoxic waters in Ace Lake, Antarctica. Carnobacterium fundatum sp. nov. and Carnobacterium atterfundatum sp. nov. Arch Microbiol. 1991;156:255–62.
6. Snauwaert J, Hoste B, De Bruyne K, Peeters K, De Vuyst L, Willems P, Vandamme P. Carnobacterium iners sp. nov., a psychrophilic, lactic acid-producing bacterium from the littoral zone of an Antarctic pond. Int J Syst Evol Microbiol. 2013;63:1370–5.
7. Pilkut EV, Mans C, Bej A, Tang J, Kreader P, Hoover RB. Carnobacterium pleiicarbonate sp. nov., a novel psychrotolerant bacterium isolated from Siberian permafrost. Int J Syst Evol Microbiol. 2013;63:1370–5.
8. Nicholson WL, Zhalina K, Oliveira RR, Triplett EW. Proposal to rename Carnobacterium inhibens to Carnobacterium inhibens subsp. inhibens subsp. nov., and description of Carnobacterium inhibens subsp. gillinichkii subsp. nov., a novel psychrotolerant bacterium isolated from Siberian permafrost. Int J Syst Evol Microbiol. 2013;65:556–61.
9. Vogt S, Klippel B, Daniel R, Antranikian G. Complete genome sequence of Carnobacterium sp. 17–4. J Bacteriol. 2011;193:3404–3.
10. Leisner JJ, Hansen MA, Larsen MH, Hansen L, Ingmer H, Sorensen SJ. The genome sequence of the lactic acid bacterium, Carnobacterium maltaromaticum ATCC 35586 encodes potential virulence factors. Int J Food Microbiol. 2012;152:107–15.
11. Caillé-Gimel C, Chaillou S, Aiba-Mondoloni J, Loux V, Aïfaz M, Rahman A, Kergourly G, Champomer-Vergès MC, Zagorec M, Dalgaard P, et al. Complete chromosome sequence of Carnobacterium maltaromaticum LMA 28. Genome Announc. 2013;1. doi: 10.1128/genomeA.00115-12.
12. Leardini MT, Panayotova N, Faelen SG, Triplett EW, Neels A, Neelken WL. Complete genome sequence of Carnobacterium gillinichkii strain WN1359 (DSM 27470). Genome Announc. 2013;1. doi: 10.1128/genomeA.00985-13.
13. Whon TW, Hyun D-W, Nam Y-D, Kim MS, Song E-J, Jiang YK, Jung ES, Shin N-R, Oh JS, Kim PS, et al. Genomic and phenotypic analyses of Carnobacterium jeotgalii strain MS32, a lactate producing candidate biopreservative bacterium isolated from salt-fermented shrimp. FEMS Microbiol Lett. 2015;362(10). doi:10.1093/femsle/nvz088.
14. Joborn A, Dorsch M, Olsson JC, Westerdahl A, Kjelleberg S. Carnobacterium inhibens sp. nov., isolated from the intestine of Atlantic salmon (Salmo salar). Int J Syst Bacteriol. 1999;49:1891–8.
15. Nicholson WL, Krivushin K, Gilchinsky D, Schuerger AC. Growth of Carnobacterium spp. from permafrost under low temperature, pressure, and anoxic atmosphere has implications for Earth microbes on Mars. Proc Natl Acad Sci U S A. 2013; 110:6666–71.
16. Benardini JN, Sawyer J, Venkateswaran K, Nicholson WL. Spore UV and acceleration resistance of endolithic Bacillus pumilus and Bacillus subtilis isolates obtained from Sonoran desert basalt: implications for lithopsperma. Astrobiology. 2003;3:79–17.
17. Alsoush S, Madden T, Schaffer A, Zhang J, Zhang Z, Miller W, Lipman D. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Res. 1997;25:3389–402.
18. Reddy TB, Thomas AD, Stamatis D, Bertsch J, Ibsland M, Jansson J, Mallajosyula J, Pagan I, Lobos EA, Kyrpides NC. The Genomes Online Database (GOLD) v.5: a metadata management system based on a four level (meta) genome project classification. Nucleic Acids Res. 2015;43:D109–16.
19. Eil D, Fehr A, Gray J, Luong K, Lyle J, Otto G, Peluso P, Rank D, Baybayan P, Bettman B, et al. Real-time DNA sequencing from single polymerase molecules. Science. 2009;323:133–8.
20. Field D, Gamy B, Gray J, Morrison T, Selsengut J, Sterk P, Tatusova T, Thomson N, Allen MJ, Anguilloli SV, et al. The minimum information about a genome sequence (MIGS) specification. Nat Biotechnol. 2008;26:514–7.
21. DOE Joint Genome Institute user home. http://www.jgi.doe.gov.
22. Chin CS, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eckert EE, et al. Nontyphoid, finished microbial genome assemblies from king-rod swarming sequencing data. Nat Methods. 2013;10:563–9.
23. The Integrated Microbial Genomes (IMG) platform. http://img.jgi.doe.gov.
24. Markowitz VM, Mavromatis K, Ivanova NN, Chen IM, Chu K, Kyrpides NC. IMG ER: a system for microbial genome annotation expert review and curation. Bioinformatics. 2009;25:2271–8.
25. Hyatt D, Chen GL, Locht LF, Van D, Lamer WT, Hauershl J, Predigal: prokaryotic gene recognition and translation initiation site identification. BMC Bioinformatics. 2010;11:119.
26. Pati A, Ivanova NN, Mikhailov N, Oxchinnikova G, Hooper SD, Lykidis A, Kyrpides NC. GenePRIMP: a gene prediction improvement pipeline for prokaryotic genomes. Nat Methods. 2010;7:455–7.
27. Lowe TM, Eddy SR. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. Nucleic Acids Res. 1997;25:953–6.
28. Paez J, Quast C, Rittinger K, Fuchs BM, Ludwig W, Pedjes J, Glickner FO, Silva A, et al. Comprehensive online resource for quality checked and aligned ribosomal RNA sequence data compatible with ARB. Nucleic Acids Res. 2007;35:7188–96.
29. INFERNAL: Inference of RNA alignments. http://eddylab.org/infearnal/.
30. Accessed 6 Sept 2016.
31. Ringo E, Spersad S, Krugerof OF, KogdaI A. Use of 16S rRNA gene sequencing analysis to characterize cultivable intestinal bacteria in Atlantic salmon (Salmo salar) fed diets with cellulose or non-starch polysaccharides from soy. Aquac. Res. 2008;39:1087–100.
32. Feng N, Hao WH, Lei HX, Wei XH. Carnobacterium inhibens isolated from biogas slurry of Tianzhu county of Gansu province. In: National Center for Biotechnology Information (NCBI). 2015 edition. National Center for Biotechnology Information (NCBI), August 10, 2013 edition. 2013.
33. Gendrin P, Isailovic S, Auer M, et al. Real-time DNA sequencing from single polymerase molecules. Bioinformatics. 2009;25:2271–8.
34. Gottschall J, Meyer RM, Krugerof OF, KogdaI A. Use of 16S rRNA gene sequencing analysis to characterize cultivable intestinal bacteria in Atlantic salmon (Salmo salar) fed diets with cellulose or non-starch polysaccharides from soy. Aquac. Res. 2008;39:1087–100.