IVYSPRING INTERNATIONAL PUBLISHER

Journal of Genomics

2017; 5: 48-50. doi: 10.7150/jgen.16163

Short Research Communication

Whole-genome Sequencing of *Vibrio sinaloensis* T47, a Tropical Marine Isolate with Quorum Sensing Properties

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Published: 2017.03.09

Abstract

A large number of *Vibrio* sp. thrive in the marine environment and they are notable to cause food borne infection associated with undercooked seafood. In this study, we report the whole genome sequence of *Vibrio sinaloensis* T47 which was isolated from coastal marine water in Morib Beach, Hulu Selangor. The genome is made up of approximately 4.59 Mbp with 80 contigs and 46% G+C content. From the annotated genome, genes associated with quorum sensing (QS) were identified. This research provides a genetic basis for better understanding of QS pathway which contributes to the physiological traits of strain T47 to thrive in the marine environment.

Key words: *Vibrio sinaloensis*, whole genome sequencing, quorum sensing, autoinducer synthase, virulence factor

Introduction

Vibrio sp. is a very common bacterium which can be found in almost all water-borne environments including sea, estuary and fresh water. *Vibrio* sp. first made its debut into the scientific world through the discovery of bioluminescence properties. It was reported that *Vibrio fischeri* forms a symbiotic relationship with its host, the Hawaiian bobtail squid (*Euprymna scolopes*) and this underlying symbiosis is associated with quorum sensing (QS) [1]. QS is well known as a mechanism of virulence and colonization when the population in bacteria surpasses a threshold. This feature seems to be a common trait in the members of the genus *Vibrio*.

Since the development of advance taxonomical tools such as DNA-DNA hybridization, fluorescent amplified fragment length polymorphism and multilocus sequence analysis, the number of novel species from *Vibrio* family being discovered is constantly expanding [2, 3]. Among the vast members of *Vibrio* genus, a number of them were demonstrated to possess QS abilities such as *V. harveyi* [4], *V. cholera* [5], and *V. anguillarum* [6]. There is mounting data suggesting that QS is responsible for many unique traits such as pathogenicity, swarming abilities and biofilm production [7, 8, 9]. In this work, we study on *V. sinaloensis* strain T47 which was isolated from a tropical marine in Morib Beach, Selangor (2° 45' 2.7" N, 101° 26' 34.7" E). A water sample was collected approximately 15 cm from the water surface.

V. sinaloensis was first documented by Gomez-Gil and colleagues [10] from the spotted rose snapper (*Lutjanus guttatus*) which causes infection and vibriosis. In fact, this bacterium is a major threat to the aquaculture sector due to its pathogenicity properties.

The colonization of *V. sinaloensis* has been reported in crustaceans, for example, the white-leg shrimp, *Litopenaeus vannamei* [11]. Here, the sequencing strategy and data of the whole genome of strain T47 is presented to provide better understanding of the marine bacterium as well as insights to the physiological behaviors associated to QS activity.

V. sinaloensis strainT47 was cultured in aseptic condition on Luria Bertani Agar (LBA) with 3% NaCl concentration (w/v) and incubated at 28°C overnight. The genomic DNA of strain T47 was extracted using QIAamp DNA Minikit (Qiagen, Germany) according to the manufacturer's instructions. The quality of the extracted DNA was measured using NanoDrop Spectrophotometer (Thermo Scientific) and Qubit 2.0 fluorometer (Life Technologies). Next, Nextera DNA Prep Kit (Illumina Inc., CA) was used to prepare the sequencing library followed by whole genome sequencing using a personal sequencer, Illumina MiSeq (Illumina Inc., CA). The total reads were assembled into 80 contigs with 43.8 × coverage using CLC Genomic Workbench version 5.1 (CLC Bio, Denmark). The draft genome of strain T47 is made up of 4,599,504 bp with G+C content of 46.12%. The genome sequence has been deposited into GenBank under the accession number JXBJ00000000. The 16S rDNA sequence used in identification of strain T47 [12] was also deposited into NCBI under accession number KR058860.

Based on 16S rDNA sequence, strain T47 was found to have more than 99% similarity to several Vibrio sp. such as *V. variabilis*, *V. caribbeanicus*, and *V. sinaloensis*. On the other hand, annotations of both functional and predicted genes were performed using the Integrated Microbial Genomes (IMG-ER) platform and with GOLD-ID Ga0063884 [13]. As shown in Table 1, the genome was resolved into 4,105 protein coding genes (CDs) and a total of 127 RNA genes which consist of 8 genes responsible for 5S rRNA synthesis, 5 genes for 16S rRNA synthesis, 6 genes for 23S rRNA synthesis and 107 genes for tRNA. From the IMG-ER platform, cluster of orthologous groups (COG) categories showed that a large number of genes are responsible for basic life-sustaining needs of the bacterium. It was found that 310 genes were predicted to contribute to amino acid transport and metabolism, 245 genes are linked to carbohydrate transport and metabolism, 295 involves in signal transduction mechanisms, and 91 genes are related to the virulence and defense regulation (Table 2).

From the annotated genome sequences, a gene associated with QS was found in contig 14. The 1203 bp of *luxM* homologue is analogous to an *N*-acyl homoserine lactone (AHL) synthase, AinS, which can be also found in *V. fischeri* [14, 15]. Hence, it is highly postulated that the autoinducer synthase LuxM is responsible for the production of signaling molecules in strain T47. In this study, the availability of the sequence could contribute to a better understanding of QS system and its role in *V. sinaloensis*.

Table 1. Genome features of *V. sinaloensis* strain T47

Attributes	Number	% of Total
DNA, total number of bases	4, 599, 504	100.00
DNA coding number of bases	4, 053, 747	88.13
DNA G + C number of bases	2, 121, 494	46.12 [1]
DNA scaffolds	80	100.00
Genes total number	4232	100.00
Protein coding genes	4105	97.00
RNA genes	127	3.00
rRNA genes	19	0.45
5S rRNA	8	0.19
16S rRNA	5	0.12
23S rRNA	6	0.14
tRNA genes	106	2.50
Other RNA genes	2	0.05
Protein coding genes with function prediction	3428	81.00
Pseudo genes	65	1.54
Without function prediction	677	16.00
Protein coding genes with enzymes	1124	26.56
Without enzymes but with candidate KO based enzymes	5	0.12
Protein coding genes connected to Transporter Classification	614	14.51
Protein coding genes connected to KEGG pathways	1310	30.95
Not connected to KEGG pathways	2795	66.04
Protein coding genes connected to KEGG Orthology (KO)	2448	57.84
Not connected to KEGG Orthology (KO)	1657	39.15
Protein coding genes connected to MetaCyc pathways	954	22.54
Not connected to MetaCyc pathways	3151	74.46
Protein coding genes with COGs	3085	72.90
Chromosomal Cassettes	392	-
Biosynthetic Clusters	8	-
Genes in Biosynthetic Clusters	125	2.95
Fused Protein coding genes	136	3.21
Protein coding genes coding signal peptides	445	10.52
Protein coding genes coding transmembrane proteins	1034	24.43

Table 2. Cluster of orthologous groups for strain T47 IMG-ER platform

Name	Count	Percentage (%)
Amino acid transport and metabolism	310	8.8
Carbohydrate transport and metabolism	245	6.95
Cell cycle control, cell division, chromosome partitioning	39	1.11
Cell motility	135	3.83
Cell wall/ membrane/ envelope biogenesis	220	6.24
Chromatin structure and dynamics	1	0.03
Coenzyme transport and metabolism	173	4.91
Defense mechanisms	91	2.58
Energy production and conversion	198	5.58
Extracellular structures	49	1.39
Function unknown	207	5.87
General function prediction only	238	6.75
Inorganic ion transport and metabolism	177	5.02
Intracellular trafficking, secretion and vesicular transport	77	2.19
Lipid transport and metabolism	122	3.48
Mobilome: prophages and transposons	12	0.34
Nucleotide transport and metabolism	93	2.64
Posttranslational modification, protein turnover, chaperones	162	4.6
RNA processing and modification	1	0.03
Replication, recombination and repair	120	3.41
Secondary metabolites biosynthesis, transport and catabolism	64	1.82
Signal transduction mechanisms	295	8.37
Transcription	257	7.29
Translation, ribosomal structure and biogenesis	240	6.81
Not in COG	1147	27.1

Nucleotide sequence accession numbers

The draft genome sequence of *V. sinaloensis* strain T47 can be obtained from GenBank under the accession number JXBJ00000000. This version described in the paper is the first version, JXBJ00000000. The GenBank accession number for 16S rDNA nucleotide sequence for strain T47 is KR058860. This version described in the paper is the first version, KR058860.

Acknowledgement

This work was supported by the High Impact Research Grants, University of Malaya (UM-MOHE HIR Grant UM.C/625/1/HIR/MOHE/CHAN/14/1, No. H-50001-A000027; UM-MOHE HIR Grant UM.C/625/1/HIR/MOHE/CHAN/01, No. A000001-50001) awarded to Kok-Gan Chan which are gratefully acknowledged.

Competing Interests

The authors have declared that no competing interest exists.

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