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Genome sequence of *Enterobacter* sp. ST3, a quorum sensing bacterium associated with marine dinoflagellate

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ABSTRACT

Phycosphere environment is a typical marine niche, harbor diverse populations of microorganisms, which are thought to play a critical role in algae host and influence mutualistic and competitive interactions. Understanding quorum sensing-based acyl-homoserine lactone (AHL) language may shed light on the interaction between algal-associated microbial communities in the native environment. In this work, we isolated an epidermal bacterium (was tentatively named *Enterobacter* sp. ST3, and deposited in SOA China, the number is MCCC1K02277-ST3) from the marine dinoflagellate *Scrippsiella trochoidea*, and found it has the ability to produce short-chain AHL signal. In order to better understand its communication information at molecular level, the genomic map was investigated. The genome size was determined to be 4.81 Mb with a G + C content of 55.59%, comprising 6 scaffolds of 75 contigs containing 4647 protein-coding genes. The functional proteins were predicted, and 3534 proteins were assigned to COG functional categories. An AHL-relating gene, *LuxR*, was found in upstream position at contig 1. This genome data may provide clues to increase understanding of the chemical characterization and ecological behavior of strain ST3 in the phycosphere microenvironment.

Specification	
Organism	*Enterobacter* sp.
Strain	ST3
Sequencer	Illumina Hiseq 2000
Data format	Processed
Experimental factor	Microbial strain
Experimental features	Whole genome sequence of *Enterobacter* sp. ST3, assembly and annotation
Consent	N/A
Sample source location	Algae bloom, Yantian Port, Shenzhen, China.

1. Direct link to deposited data

http://www.ncbi.nlm.nih.gov/assembly/GCA_001469415.1/.

2. Introduction

As producers and decomposers, bacterioplankton are a key component of microbial food webs and play significant roles in biogeochemical cycles [1]. In "algae–bacteria" symbiosis, bacteria extensively participate in substance cycles, oxidation–reduction activities, and regulate the balance of phytoplankton ecosystems, which is the most basic and active link in the whole phycosphere habitat [2]. In recent years, chemical ecology has emerged as a new approach to evaluate the structural and functional diversity, as well as the dynamic equilibrium of the algae–bacteria symbiont [3]. Many key life processes, including food acquisition, movement behavior, defense, and signal communication are mediated by signal interactions at the individual, population, and community levels [4–5]. Understanding signal regulated ecological processes will help to define the crucial molecular interactions between algae and bacteria.

Infochemicals are frequent in the phycosphere, and various cross-talking behaviors of heterotrophic bacteria have been identified in algae–bacteria symbiosis relationships [6]. These processes are often controlled by cell density-dependent regulation of gene expression, which is mediated by diffusible signal molecules whose concentration correlates with the population density. This process is termed quorum sensing (QS), and one of the most well-known QS signals is N-acyl-homoserine lactone (AHL) [7]. AHLs can induce gene expression either directly by interacting with a transcriptional regulator, or indirectly by activating a signal cascade [8]. Bacteria use AHL to regulate a variety of phenotypes such as biofilm formation, exopolysaccharide production, virulence factor production, and motility, which are essential for the successful establishment of a symbiotic relationship with their eukaryotic hosts [9]. Previous studies have shown that alga-associated bacterial isolates (of epi- or endophytic origin) produce AHLs and exhibit various functions, including facilitating the settlement of zoospores

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Table 1
Genome and environmental features.

Item	Description
MIGS data	
Investigation type	Bacteria
Project name	NSFC (41476092)
Collected by	Jin Zhou
Collection date	05-23-2013
latitude/longitude	23°35′N, 114°17′E
Depth	0.5–1.0 m
Country	China
Environment	Surface sea water
Biotic relationship	Attached bacterium with algae
Trophic level	heterotrophic
Relate to oxygen	Aerobe
Isolation growth condition	LB medium
Sequencing method	Illumina MiSeq
Assembly	SPAdes v. 3.5.
Finishing strategy	Whole genome
Annot source	RAST/NCBI blastx
Estimated size	5–6 M bp
Geo_loc_name	Shenzhen, China
Sample material	Surface water from the Yantian Port
Temp	27 °C
Salinity	30.7 PSU
Motility	Yes
Genome assembly data	
Assembly name	ST3
Genome coverage	260×
Sequencing technology	Illumina HiSeq 2000

[10], regulating the morphogenesis pattern [11], affecting individual reproduction [12], and promoting the liberation of carpospores [13]. Recently, our group isolated an *Enterobacter* sp. ST3 strain from the dinoflagellate *Scrippsiella trochoidea* (the sample environmental features list in Table 1). The electron micrograph images showed that ST3 strain is a motile and long rod-shaped bacterium (Fig. 1). It has broad environmental suitability and the optimal growth temperature and pH values were 30 °C and 8.0, respectively (Fig. 2). As a member of γ-proteobacteria, *Enterobacter* sp. play multi-roles in symbiosis environment, such as exopolysaccharide produce, iron metabolism, heavy metals biosorption, and pollutant biodegradation [14–17]. In algae–bacteria symbionts, *Enterobacter* sp. is often found as a main species and participant in the matter cycles of the phycosphere [18]. Of particular note, this isolate ST3 possesses cross-talking language activities, and can secrete short-chain (C_6) AHL molecules (Fig. 3). Although the phenotype was previously observed, this type of functionality has not been elucidated at the gene level with genomic approaches. In addition, we speculated that its ecological function was regulated by quorum sensing, but there is a lack of direct evidence for confirmation on the genomic level. In order to better understand genetic underpinning of the bacterium roles, here, we performed whole-genome sequencing of this bacterium and searched for its AHL encoding gene(s).

3. Experimental design, materials and methods

The genomic DNA of *Enterobacter* sp. ST3 was extracted using the DNA extraction kit (Mo Bio, CA, USA) according to the manufacturer's instructions. The whole-genome shotgun project of *Enterobacter* sp. ST3 was performed using pair-end sequencing in an Illumina Miseq sequencing platform (Illumina, CA, USA), which was performed by Shenzhen Hengchuan Gene-Tech. Co., Ltd. The reads were assembled with SOAPdenovo (V.2.04) [19], and the sequence was annotated using the RAST annotation server [20]. tRNA and rRNA genes were predicted by tRNAscan-SE [21] and RNAmmer [22], respectively. Genes

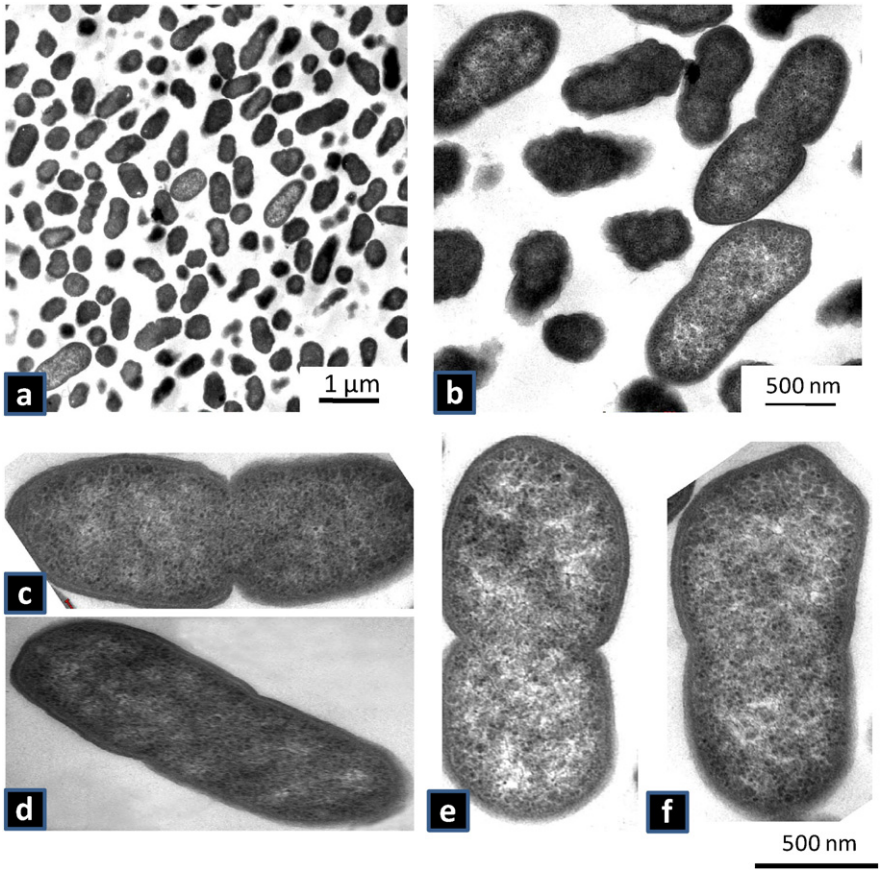

Fig. 1. Electron micrographs of cells of "*Enterobacter* sp. ST3". Preparation and EM conditions were as described by Hahnke et al. [25]. The magnification times: a (8000×), b (25,000×), c & d (40,000×), and e & f (60,000×).