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Response of microbial communities to bioturbation by artificially introducing macrobenthos to mudflat sediments for in situ bioremediation in a typical semi-enclosed bay, southeast China

Ying Ma^a, Anyi Hu^b, Chang-Ping Yu^b, Qingpi Yan^a, Xizhu Yan^a, Yongzhong Wang^a, Fei Deng^a, Hejian Xiong^{c,*}

^a Key Laboratory of Healthy Mariculture for the East China Sea, Ministry of Agriculture, Fisheries College of Jimei University, Xiamen 361021, China

^b Key Laboratory of Urban Pollutant Conversion, Institute of Urban Environment, Chinese Academy of Sciences, Xiamen 361021, China

^c Bioengineering College of Jimei University, Xiamen 361021, China

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ABSTRACT

Although microbes play important roles during the bioremediation process using macrobenthos in degraded environments, their response to macrobenthos bioturbation remains poorly understood. This study used 16S rRNA gene-Illumina Miseq sequencing to investigate the microbial communities and their response to bioturbation by artificially introducing macrobenthos to the mudflat of Sansha Bay, southeast China. A total of 56 phyla were identified, dominated by δ - and γ -Proteobacteria, with a total percentage of over 50%. Others, such as Acidobacteria, Chloroflexi, Bacteroidetes, Planctomycetes and Alphaproteobacteria occupied 4–7% respectively. Eighteen genera indicating the microbial communities response to bioturbation and seasonal change were identified. Bioturbated samples contained more ecologically important genera, and untreated samples contained more genera ubiquitous in marine environments. The physicochemical characteristics did not change significantly probably due to the short time of bioremediation and low survival rate of macrobenthos, confirming that microbial communities are more sensitive and can serve as sentinels for environmental changes.

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1. Introduction

Semi-enclosed bays are among the most important marine ecosystems in the world (Peng et al., 2013). Because of its good capability of shelter condition and wave attenuation, semi-enclosed bay is often the first selection for port construction and marine aquaculture. It is also the natural habitats, breeding and nursery areas for many marine organisms. However, semi-enclosed bay features narrow mouth and wide abdomen, in poor conditions of water exchange. It is prone to environmental issues such as organic materials sedimentation, metal contamination (Dell'Anno et al., 2002; Lim et al., 2012) and long-term pollutant accumulation (Hong et al., 2009; Hu et al., 2011). Besides, high levels of nutrients in sediments will induce eutrophication and algal blooms when released to the water. Therefore, the environmental protection and remediation in semi-enclosed bays are essential for maintaining ecological balance, biological diversity, and sustainable economic development in the region.

Compared with physico-chemical methods, bioremediation is a more effective, versatile, economical and environment friendly alternative to slow down the eutrophication and eliminate the pollutants by living organisms. The use of macrobenthos, such as shellfish and polychaetes, as nutrient extractors and transformers has proven to be valid way and was mostly used in organic rich fish-farm sediments (Tsutsumi et al., 2005; Niu et al., 2014) and aquaculture wastewater (Jones et al., 2001; Marinho-Soriano et al., 2011). Microorganisms have been widely used in organics decomposition and pollutants metabolism (Fabiano et al., 2003; Bao et al., 2012; Kang, 2014), and they also play important roles during the bioremediation process using macrobenthos. Macrofaunal bioturbation, such as feeding, excretion, and burrowing, reshapes the physical, chemical and biological properties of aquatic sediments (Reise, 2002; Volkenborn et al., 2007), stimulate the process of nitrification and denitrification (Gilbert et al., 2003; Banks et al., 2013) and influence degradation rates of organic detritus (Gribsholt and Kristensen, 2002). Bioturbation has been shown to increase total microbial abundance and viable microbial biomass (Steward et al., 1996), and lead to establishment of specific microbial communities in burrows of

* Corresponding author. Tel./fax: +86 592 6180470.

E-mail address: hjxiong@jmu.edu.cn (H. Xiong).

macroorganisms (Marinelli et al., 2002; Matsui et al., 2004; Papaspyrou et al., 2005; Pischedda et al., 2011). Bioturbating activities of sediment-dwelling polychaetes can also affect the release and distribution of PAHs (Grossi et al., 2002) and favor the development of bacteria which may help the bioremediation processes in oil contaminated sediments (Cuny et al., 2007). However, most of the works only investigated particular microbial groups such as sulfate-reducing bacteria and ammonia- and nitrite-oxidizing bacteria (Matsui et al., 2004; Satoh et al., 2007), and the studies were mainly restricted to the burrow associated environment. The bioturbation responses on overall bacterial communities in a larger scale environment have not been thus far extensively characterized. Furthermore, previous studies were mainly based on traditional molecular techniques, such as terminal restriction fragment length polymorphism (T-RFLP) (Laverock et al., 2010), rRNA gene fingerprinting (Lucas et al., 2003) and cloning (Matsui et al., 2004; Papaspyrou et al., 2005, 2006; Laverock et al., 2010; Pischedda et al., 2011), or phospholipid fatty acid analysis (PLFA) (Steward et al., 1996; Marinelli et al., 2002). Because of the inherent limitations of traditional techniques, those studies provide limited information about phylogenetic structure of microbial communities and how they respond to the bioturbation. The emergence of high-throughput sequencing technology has provided us important opportunity for fully and deeply understanding environmental microbes because of its high data throughput (10^3 – 10^6 sequences per sample), high accuracy and low cost. This technique enables us to perform in-depth analyses for studies of comparative microbial ecology (Binladen et al., 2007; Caporaso et al., 2011).

Sansha Bay is located on the northeast coast of Fujian Province, southeast China. It is a typical semi-enclosed tidal inlet bay, with a wet area of 262 km² (Wang et al., 2011). Sansha Bay is studded with numbers of islands and surrounded by hills. It has only one narrow mouth, named Dongchong Mouth, connecting the East

China Sea, the mouth is the only path for waters inside to exchange with that of the open sea (Fig. 1). Since its open degree (the ratio of entrance width to shoreline length of a bay) is less than 0.01, some documents (Yu et al., 2014) called it enclosed bay. For its deep water, low wind and small waves, it is a naturally excellent site for harbor construction, migratory fishes spawning and nursing and marine aquaculture. Sansha Bay is the only inner bay for large yellow croaker (a famous local fish in China) spawning, and the largest marine cage breeding base is located inside the bay. The Bulletin of China's Marine Environmental Status in 2007 reported that the total aquaculture product is 3.22×10^5 tons in Sansha Bay. Because of its location at the northeast wing of the West Coast Economic Zone, Sansha Bay has undergone severe habitat degradation in recent years mainly due to substantial influences by aquaculture, coastal industries, urbanization, and land-sourced pollution.

One ongoing Ocean Public Welfare Research Project in our group is to restore the polluted mudflat sediments in Sansha Bay by introducing polychaetes clamworm and bivalves cardiidae to the mudflat, and the traditional physical and chemical indexes were monitored to assess the bioremediation effects. No information are currently available on the response of bacterial assemblages to the in situ bioremediation by introducing macrobenthos to the mudflat sediments. Nevertheless, effects on microbial processes are key to many aspects of the functioning of the ecosystem. The present study here aims to investigate the response of microbial communities to in situ bioremediation through the high-throughput sequencing technique. The primary objectives of this study were (1) to illuminate the microbial community composition in mudflat sediments in Sansha Bay; (2) to investigate the response of microbial communities to bioturbation of macrobenthos introduced to the mudflat sediment (3) to establish the relationships between the microbial community structure and the traditional physiochemical indexes.

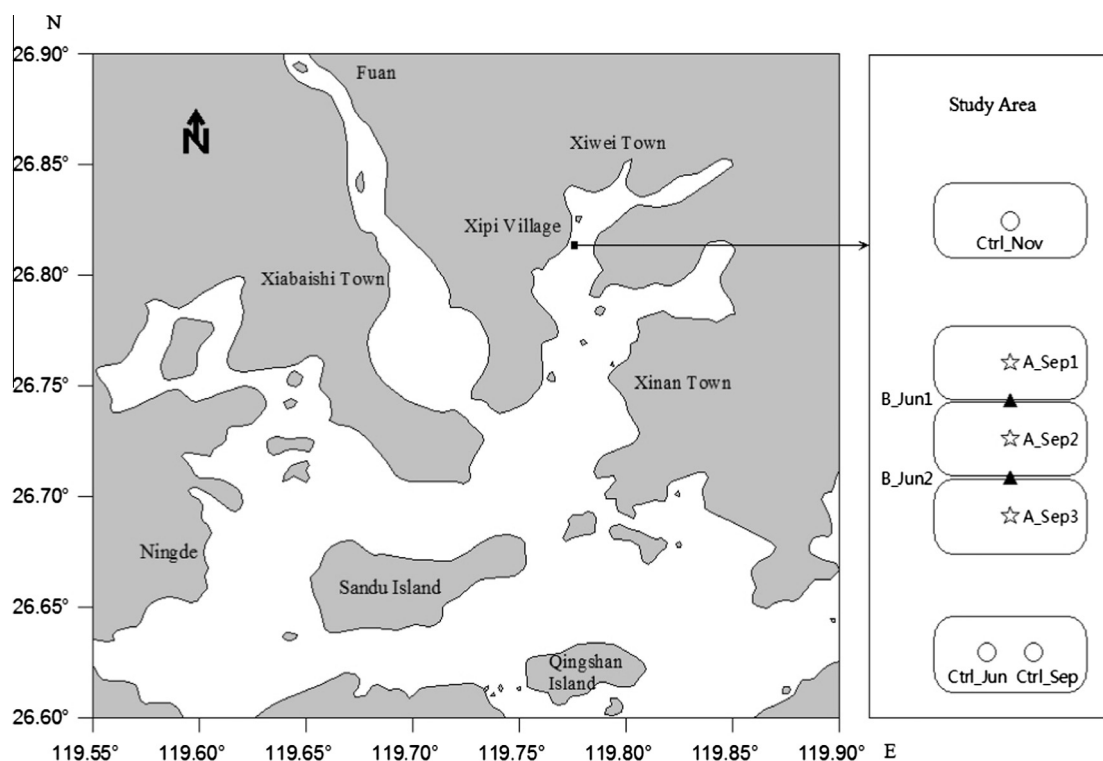


Fig. 1. Study area and experimental sketch map.

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