

Research article

The indicative effect of structures of archaeal communities at deep-water sediment cores on natural gas hydrate: A case study from Station 973-4 in the Southwest Taiwan Basin, South China Sea

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Abstract

The formation and decomposition of submarine natural gas hydrate (hereinafter hydrate for short) are greatly influenced by methanogenesis and anaerobic methane oxidation of microorganisms, so it is necessary to conduct further analysis on the metabolic mechanism of archaeal communities and its indicative effect on hydrate reservoir formation. In this paper, the structures of archaeal communities at the cores of Station 973-4 in the Southwest Taiwan Basin of the South China Sea were studied by means of 16S rDNA molecular biological technologies, and then were compared with those at hydrate occurrence and non-occurrence zones in the Japan Sea and the Shenhu area of the South China Sea. It is shown that *Methanosarcina* is the predominant community at the cores from Station 973-4, with a percentage of 50% in the surface zone, 46.1% in the sulfate–methane transition zone (SMTZ) and 66.7% in the deep zone. *Methanomicrobiales* is the secondary predominant community, with a percentage of 28.3% in the surface zone, 30.7% in SMTZ and 11.1% in the deep zone. The percentage changing tendency of the above-mentioned communities are closely related with the variation of organic matter content and compositions during the early diagenetic process. It is concluded that the structures of archaeal communities are in accordance with the analysis results of bacteria, geochemistry and mineralogy, indicating the occurrence of hydrate at the bottom of the cores from Station 973-4. In addition, the structures of archaeal communities are characterized by diversity, so attention shall be paid to the synergy between geochemical parameters and geologic characteristics, so that the indicative effect on hydrate reservoir formation will be presented more accurately.

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Keywords: South China Sea; Southwest Taiwan Basin; Archaeal diversity; Japan Sea; Shenhu area; Natural gas hydrate; Methanogenesis; Anaerobic methane oxidation

The formation and decomposition of submarine natural gas hydrate (hereinafter hydrate for short) are greatly influenced by methanogenesis and Anaerobic Oxidation of Methane (AOM) of microorganisms [1,2]. The structures and indicative species of the microorganism communities can be used to study the hydrate reservoir formation, to which much attention has been paid by scientists in various countries. For instance,

Bidle et al. [3] found several types of sulfur metabolic bacteria, methanogenic bacteria (such as *Methanosarcinaceae*), methane-oxidizing bacteria (such as *Methylocaldum*) in Cascadia (Station ODP892B). Niemann et al. [4] found that there developed three major types of bacteria in the Haakon Mosby mud volcano: aerobic methane oxidizing bacteria *Methylococcales*, anaerobic methane oxidizing archaea ANME-2 and ANME-3; moreover, the diffusion of sulfate and free oxygen in porous fluid restrained the usability of electron acceptor of methane oxidizing bacteria, then limited its habitat scope. Based on the research results of Niemann et al. [4], Lösekann et al. [5], the 16S rRNA and fluorescence in situ hybridization

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technique were used to investigate the structures of the bacteria communities in the Haakon Mosby mud volcano. The search results of various scholars in various sea areas and habitats are useful for further understanding of the driving effect of the bacteria in the process of gas hydrate formation. However, it is necessary to conduct further studies on the metabolic mechanism of archaeal communities [6].

The South China Sea provides suitable conditions (e.g. source, temperature and pressure) for hydrate reservoir formation. Particularly, the passive continental margin in the northern South China Sea and the ocean basins and ocean trenches in the eastern South China Sea are the favorable areas for hydrate accumulation. After 2007, the Guangzhou Marine Geology Survey drilled and obtained hydrate samples again in the South China Sea in 2013, further promoting in-depth study on the mechanism of hydrate reservoir formation in the South China Sea by various scholars [7–12]. In the past few years, Zhang Yong et al. [13] have analyzed the archaeal diversity of surficial sediments in the Shenhu area of the northern South China Sea, and found that the major community of *Crenarchaeota* is C3, and the major *Euryarchaeota* is Marine Benthic Group (MBG)-D. Zhang Hao et al. [14] pointed out that *Crenarchaeota* is the major type in surficial sediments in Xisha Islands sea area, the main community is Marine Crenarchaeotic Group I (MGI), with apparently high diversity than that of the archaea. Shi Chunxiao et al. [15] found that *Proteobacteria* is the predominant community in the deposit cores near Jiulong methane reef in Dongsha Islands. Currently, though some progresses have been made in the surveys on the structures of archaeal communities in deposits in various sea areas of the South China Sea, the study on utilizing geomicrobiology to identify hydrate is still at its initial stage [16]. 16S rDNA molecular biological technologies were used to study the structures of archaeal communities at the cores of Station 973-4 (hereinafter 973-4 core for short) in the Southwest Taiwan Basin of the South China Sea. And those structures were compared with those in hydrate-occurrence and non-occurrence zones in the Japan Sea [17] and the Shenhu area [18] of the South China Sea so as to explore the indicative effect of archaeal communities on hydrate reservoir formation.

1. Materials and methods

1.1. The archaeal diversity of 973-4 core in the SW Taiwan Basin

The South China Sea is the biggest marginal sea in China. The Southwest Taiwan Basin is located on the passive continental margin in the northern South China Sea, adjacent to the Dongsha Uplift of the Pearl River Mouth Basin. Its north is the Penghu Beigang Uplift, and its east boundary is the Quchi–Laonong Fault on west fringe of the Taiwan Central Mountains, striking in NE direction. Its tectonic evolution underwent early extension and later compression. The sea area of the Southwest Taiwan Basin has plentiful terrigenous organic matters, with a high deposition rate, wide bottom-simulating reflector (BSR). The analysis results of geochemical features

of carbonate rocks, pyrite and pore water in the deposits indicate that this sea area is the optimum region of hydrate occurrence in the South China Sea [7,9]. The Station 973-4 is situated near the Jiulong methane reef in the Southwest Taiwan Basin, with water depth of 1666 m, from which 13.95 m cores were obtained (Fig. 1). This core was cut according to the max sampling density, then to measure the structures of archaeal communities in various depths. The concrete methods and steps are described below.

- 1) Take the 1 g deposit sample, and use the Power Soil DNA Isolation Kit reagent box to extract DNA.
- 2) Take the DNA sample as the template to do PCR amplification. The primer is 355F/1068R.
- 3) Connect the amplification product to 19T-Vector, and then transfer it into the DH5 α cell to select blue-white plaques; select positive clones by bacterial colonies PCR, and then send the positive clones to Sangon Biotech (Shanghai) Corporation to do sequencing.
- 4) Compare the measured archaeal sequences on NCBI, and search for bacterial types with higher homology.

Based on the measured Sulfate–Methane Transition Zone (SMTZ) data (<900 cm) [9] and the analysis result of bacterial diversity, the 973-4 core was divided into three zones within 12 various depths [15]: 20–382 cm in surface zones, 552–796 cm in SMTZ, and 862–1196 cm in deep zones, then the structures of the bacteria communities were explored (Fig. 2a).

1.2. Analysis of archaeal diversity in hydrate occurrence and non-occurrence zones

The hydrate occurrence zones in the Japan Sea are located in Umitaka Spur (the SW Joetsu Basin), including three

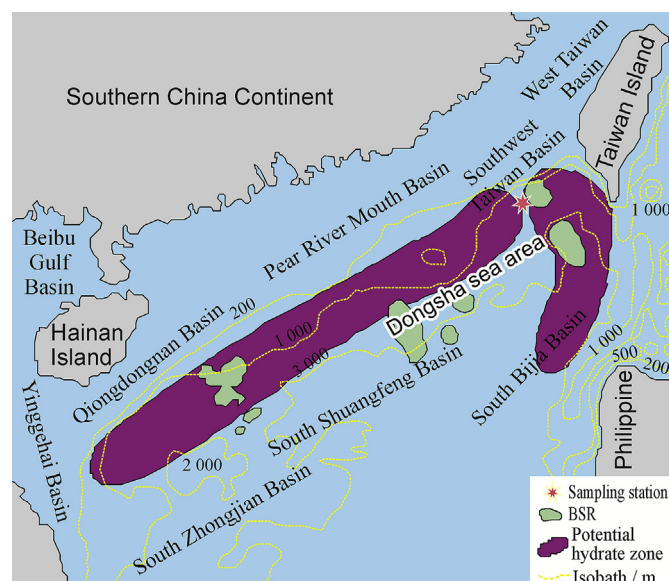


Fig. 1. Location of 973-4 core in the SW Taiwan Basin [15].

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