

Thermococcus onnurineus NA1: a Lithoheterotroph as Revealed by Genome Analysis

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Deep-sea hydrothermal vents comprise a plethora of potential habitats, with its gradients of nutrient and extreme physicochemical conditions that include temperatures from 350 to 2°C, oxygen levels from highly reduced to oxygenated, and fluid velocities from high to low flow rates (Jannasch, 1995). Many multidisciplinary studies have been carried out to understand the complexities of hydrothermal vent systems. Biological studies have been also accomplished using samples collected from hydrothermal vent areas using culture-dependent and culture-independent techniques, revealing the presence of physiologically, metabologically, and phylogenetically diverse microorganisms (Karl, 1995). Among representative species of the Archaea, sulfur-reducing heterotrophs belonging to the order Thermococcales (encompassing the genera Thermococcus, Pyrococcus, and Palaeococcus) have been reported to be one of the predominant groups (Pledger and Baross, 1991; Marteinsson et al., 1995). Notably, members of the species of Thermococcus were found to be more abundant in the vent ecosystem, with such isolates more frequently reported than the Pyrococcus species (Orphan et al., 2000; Nercessian et al., 2003). Such large populations indicate some significance for *Thermococcus* to prevail in the microbial consortia that make up the microbial ecology of hydrothermal vent systems. In addition to ecological significance, the hyperthermophilic feature of Thermococcales has fascinated microbiologists interested in fundamental and/or application-based research. To date, the complete genome sequences of three Pyrococcus species, e.g., P. horikoshii (Kawarabayasi et al., 1998), P. furiosus (Robb et al., 2001) and P. abyssi (Cohen et al., 2003), and a Thermococcus strain, T. kodakaraensis KOD1 (Fukui et al., 2005), have been determined.

T. onnurineus NA1 was recently isolated from a deep-sea hydrothermal vent region in the PACMANUS

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field (3°14'S, 151°42'E) of the East Manus Basin (Bae *et al.*, 2006) that is a rapidly opening (~10 cm/yr) backarc basin set between the Manus and New Britain trenches. This isolate requires elemental sulfur as terminal electron acceptor for heterotrophic growth on peptides or amino acids mixture, and exhibits optimum growth at 80°C and pH 8.5 (Bae *et al.*, 2006).

To provide some insight into factors that contributed to the apparent successful competition of *Thermococcus* spp. in hydrothermal vent fields, the genome sequence of *T. onnurineus* NA1 was determined by combining random whole-genome shotgun sequencing with pyrosequencing. As a result, it was revealed that *T. onnurineus* NA1 has a single circular chromosome (1,847,607 bp) without any exochromosomal elements, and a total of 1,976 coding DNA sequences (CDSs) were identified. Synteny comparison by blastp-all-vs-all between *T. onnurineus* NA1 and other *Thermococcales* strains confirmed the high frequency of DNA rearrangement. The gene rearrangement between *T. onnurineus* NA1 and *T. kodakaraensis* KOD1 seemed to take place at the level of chromosomal segments, confirming more close phylogenetic relationship than to other *Pyrococcus* strains. Interestingly, we found that *T. onnurineus* NA1 retained the metabolic pathways not only for organotrophic growth but also for lithotrophic growth by analyzing the genome sequence.

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