

The presence of (NRPS) and (PKS) genes at the deep-sea hydrothermal field in the Aegean Sea



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INTRODUCTION

Deep-sea hydrothermal vents are characterized by extremely high concentrations of microorganisms in stark contrast to the surrounding sea bottom. Nevertheless, deep-sea consumers do not rapidly remove the high biomass of prey from these communities maybe due to vent microbes' chemical defenses which still remain largely unknown. Meanwhile, the detection of genes responsible for antimicrobial and cytotoxic activity such as non-ribosomal peptide synthases (NRPS) and polyketide (PKS) of deep-sea vent bacteria has not so far been attempted.

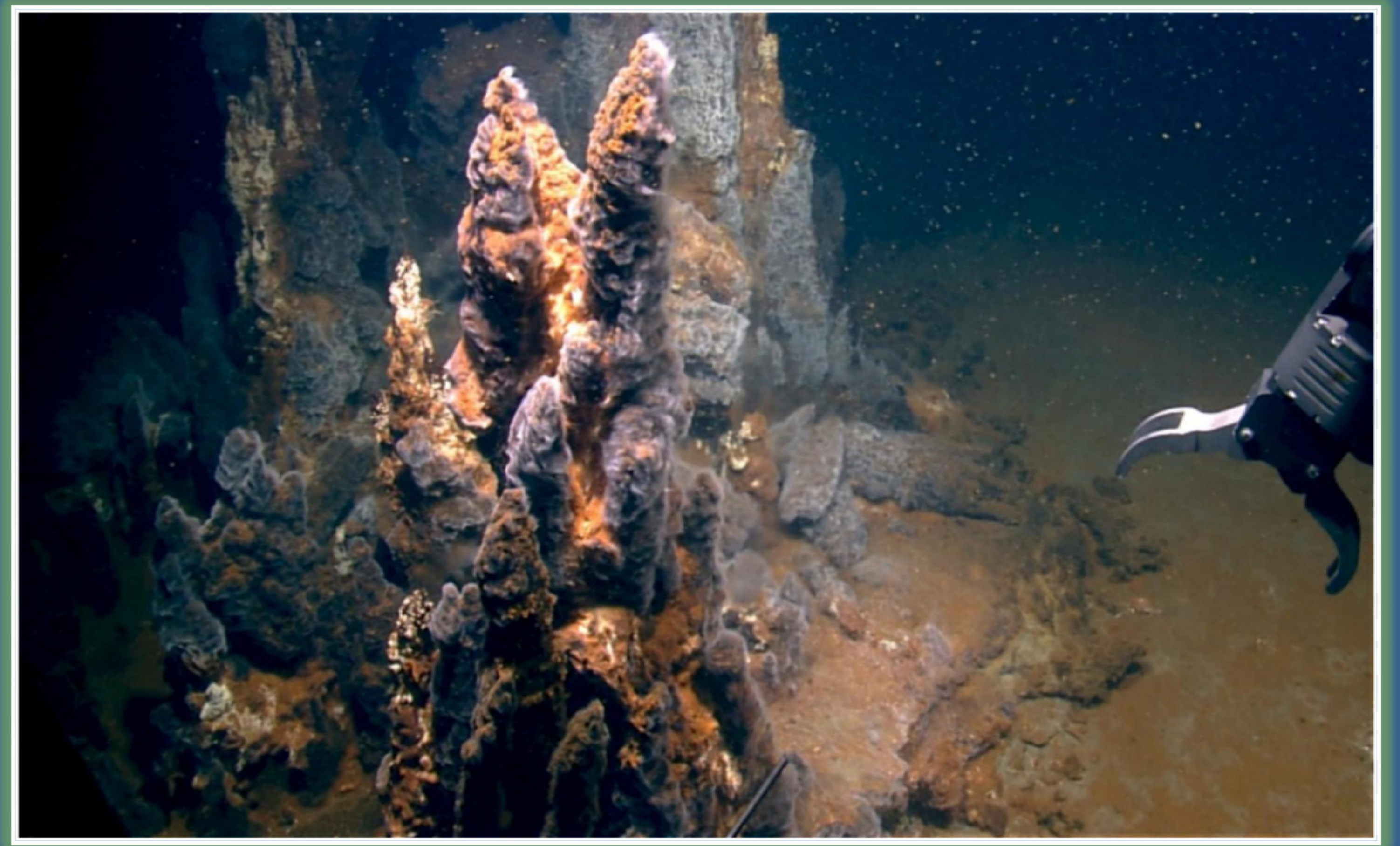


Fig. 1. Sampling of chimney with ROV manipulator arm

MATERIALS AND METHODS

In this study, sediment and chimney samples were collected from the hydrothermally active field of Kolumbo submarine volcano (500 m depth), in the Aegean Sea, during 2010 E/V *Nautilus* cruise (operated by the Ocean Exploration Trust) (Fig. 1 and 2). Samples were plated on selective media and incubated aerobically for 7 days at 25 °C. The isolated mesophylic bacteria were then tested for antimicrobial activity by diffusion method and strains which exhibited strong activity even after repeated transfer to fresh media, were selected for sequencing, phylogenetic analysis and screening for NRPS and PKS genes using degenerated primers.

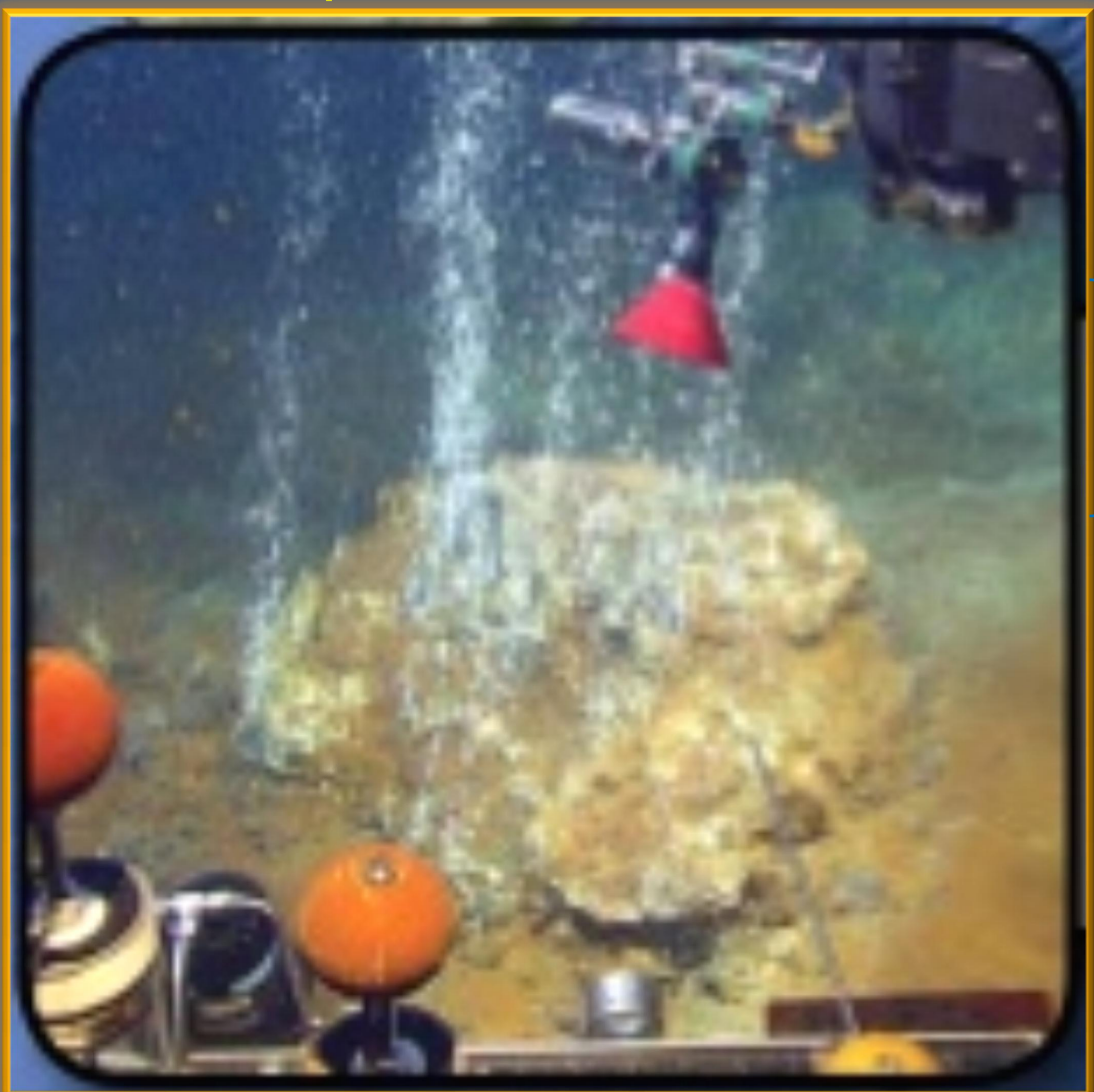


Fig. 2. Exploration of Hydrothermal Field on Kolumbo crater floor

RESULTS

230 mesophylic bacteria were isolated, 42 of which showed remarkably reproducible antimicrobial activity and were affiliated to *Bacillus* and *Proteobacteria*. Based on the conserved Adenylation domain of NRPS and Beta-keto-synthase of PKS, 19 non-ribosomal peptide synthases (Fig. 3a) and 6 polyketide synthesis genes, including *trans*-AT PKSs and hybrid PKS–NRPS, were detected in these bacterial isolates (Fig. 3b). The presence of surfactant and antibiotic biosynthesis-related to PKS and NRPS genes, suggested the potential ecological role of metabolites produced by the vent bacteria.

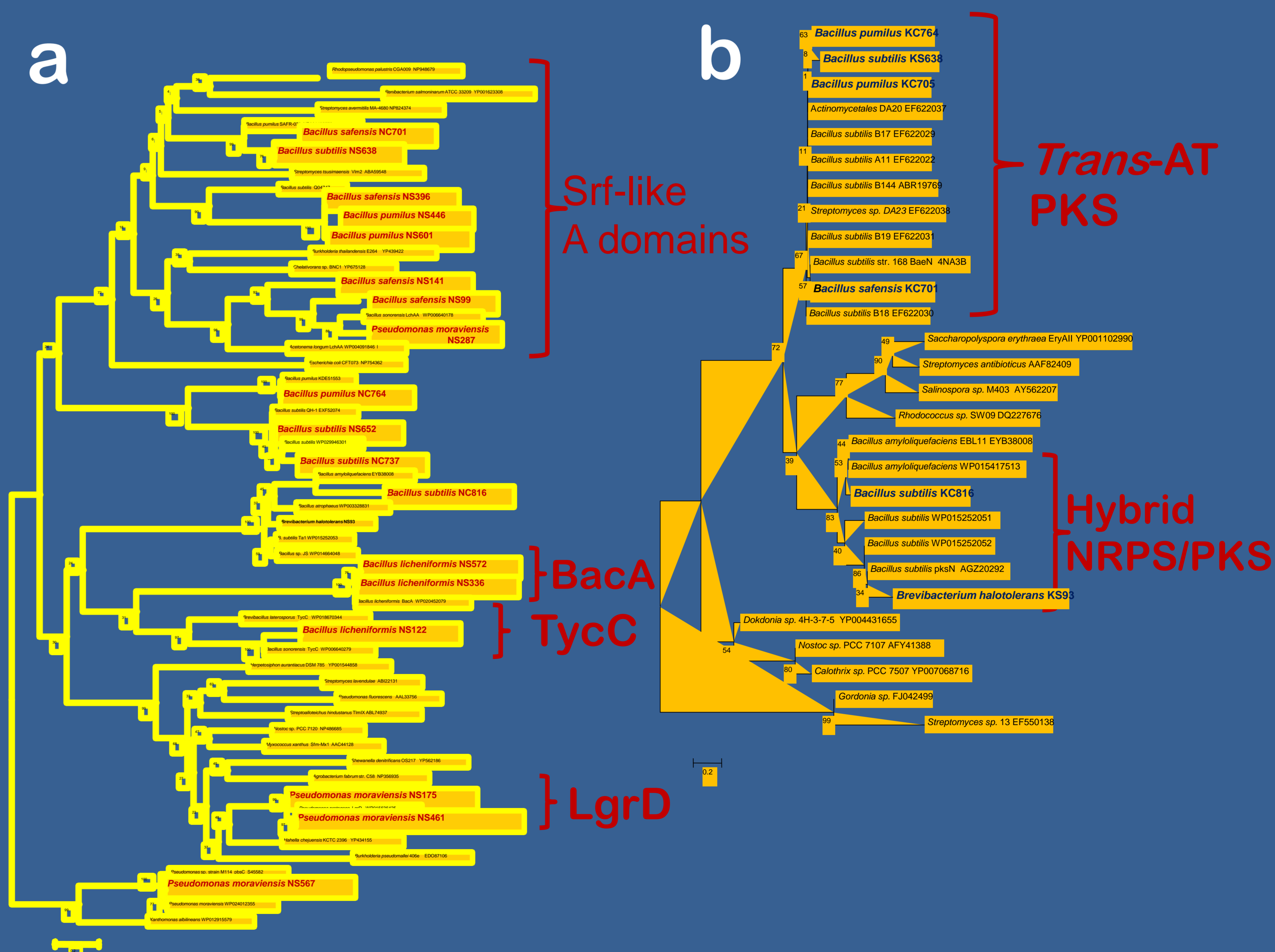


Fig. 3. Phylogenetic analysis of aa sequences of (a) (NRPS) and (b) (PKS) genes