

Biophysical Characterization of Hydrocarbon Transport Systems as a First Step in Marine Oil-Degradation by Hydrocarbonoclastic Bacteria

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Hydrocarbonoclastic (oil-eating) bacteria play an important role in the removal of petroleum hydrocarbons from polluted marine waters during oil spills. The bacterial biodegradation of hydrocarbons takes place in the cytoplasm and requires the passage of hydrophobic oil molecules across the hydrophilic periplasm. However, no hydrocarbon transporter was revealed from various genome sequences of hydrocarbonoclastic bacteria. Previous comparative genome analyses have identified Abo_0687 (T_Abo) and Abo_0688 (P_Abo) as proteins in the *Alcanivorax borkumensis* genome that comprise a putative periplasmic chain-like hydrocarbon transporter. The current study identified a homologous transport system in the arctic oil-degrading bacteria, *Oleispira antarctica* (T_Oan and P_Oan), but nothing was known about the nature of this hydrocarbon transport mechanism. Therefore, the purpose of the study was to employ recombinant proteins to understand the biochemical and biophysical bases of hydrocarbon transport. While recombinant T_Abo, T_Oan and P_Oan purified as soluble proteins, recombinant P_Abo was insoluble. Circular-dichroism spectroscopy revealed that both T (alpha-helical) and P (alpha-beta-mix) proteins were well folded, consistent with generated 3D models. Size-exclusion chromatography and analytical ultracentrifugation analyses demonstrated a strong interaction between the T and P proteins. The protein models and observed protein-protein interactions further supported the roles of T_Oan and P_Oan in periplasmic oil hydrocarbon transport. This new conceptual information gleaned from the bioinformatics and biophysical studies might be common to all hydrocarbonoclastic bacteria, thus having environmental and biotechnological implications in engineering microbes for bioremediation.

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