# Poly-extreme adaptation of early life in deep ocean: Minimum amino acid requirement for hyperthermophilic archaea, *Thermococcus eurythermalis, u*nder pH boundaries

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*Thermococcus eurythermalis*, a hyperthermophilic archaea isolated deep-sea hydrothermal vents at Guaymas basin, has exhibited tolerance to a broad range of different environmental stresses such as pH, temperature, salinity, and pressure. The poly-extreme adaptation of the species can potentially explain its dominance at its initial habitats, which is characterized by ephemeral and drastic gradient change of various chemo-physical factors. While the specific molecular mechanisms of the extreme adaptation remained unclear after intensive studies, physiological studies can provide crucial insight of the regulation of metabolic pathways under different extreme conditions. Here, we firstly examined the minimum amino acid requirement of the archaea under neutral and pH boundary conditions. For the experiment, cells were incubated in a defined medium, TRM basal plus each combination of 19 (out of 20) amino acids, with pH 4.5, pH 7.0, and pH 8.5, respectively. The amino acid was considered essential when the lacking of it could significantly affect the cell growth, and the essentiality was confirmed by incubation that only had selected amino acids added. Amino acids were used as the only carbon and nitrogen source in the medium. The result showed that there are 8 essential amino acids (trp, tyr, phe, his, thr, asp, arg, met) for pH 7, and the 12 essential amino acids (trp, tyr, phe, his, thr, asp, arg, lys, val, ile, leu) for both pH 4.5 and pH 8.5 include all the 8 amino acids for optimal condition plus four extra ones, three branch-chained (val, ile, leu) and one positively charged (lys). All the aromatic amino acids are essential in all the conditions, which might be explained by the easy availability of aromatic substances at the oil-immersed habitats. Comparison between these minimum amino acid requirements with Miller's amino acids provided important insight for the Heterotrophic Hypothesis regarding the origin of life.

Keywords: extremophiles; deep-ocean; hydrothermal vents; pH; adaptation