Moelcular Phylogenetic Analyses of G1P Dehydrogenase and G3P Dehydrogenase Suggest the Late Origin of Archaea-Type Membrane

Yokobori, S., Y. Nakajima, S. Akanuma, & A. Yamagishi. Laboratory of Extremophiles, Department of Applied Life Sciences, School of Life Sciences, Tokyo University of Pharmacy and Life Sciences, Horinouch, Hachioji, Tokyo 192-0392, Japan. Email address: yokobori@ls.toyaku.ac.jp (to S. Y.)

Introduction: All extant terrestrial life is classified into three domains, Bacteria, Archaea, and Eukarya, although the relationship among three domains is still under debate [1-3]. The membrane component before the appearance of Bacteria and Archaea is one of the foci of the argument, because membrane lipids that divide inside and outside of the cell are essential for life [4-5].

Various lipid structures are found in the three domains. However, all cellular organisims have a glycerol backbone as the common structure, with the exception of the stereostructure. The stereostructure of the glycerol backbone in polar lipids of Bacteria and Eukarya is *sn*-glycerol-3-phosphate (G3P), while in polar lipids of Archaea, it is *sn*-glycerol-1-phosphate (G1P) [5-6]. G3P and G1P are generated from dihydroxyacetone phosphate (DHAP) by different enzymes: G3P dehydrogenase (G3PDH) and G1P dehydrogenase (G1PDH), respectively. There is no sequence similarity between G3PDH and G1PDH at gene and protein levels. It was proposed that division of Bacteria and Archaea have been caused by the cellularization with different type of membrance lipids synthesized by G3PDH and G1PDH with different origins [6].

Wächtershäuser proposed a model [7] that incorporated the Koga model [6] and the precell theory [8]. In his hypothesis, in the earliest stage, precell had heterochiral membrane lipids. Under the assumption that the homochiral membrane is more stable than heterochiral membrane, the heterochiral membrane is thought to have evolved toward homochiral membrane. After invention of G3PDH in the cells with G3P-rich membrane, Bacteria was thought to have appeared. After invention of G1PDH in the cells with G1P-rich membrane, Archaea was thought to have appeared. However, Shimada and Yamagishi suggested that the membrane with heterochiral lipids is stable as that with homochiral lipids [9]. This suggests that the cells with the membrance with heterochiral lipids could have existed not only in the earliest cellular stage but also in later stages. For example, the replacement of the membrane with G1P by that with G3P, or the replacement of the membrane with G3P by that with G1P, could occurred during the evolution of cellular cells, and could directly been related to the origin of either of Bacteria and Archaea.

Proteins with G1PDH activity have been reported from certain bacterial lineage [10-11]. If they were not originated by the horizontal gene transfer from arracheal species after the separation of Bacteria and Archaea, the common ancestor of Bacteria and Archaea (or LUCA/Commonote) could have had G1P in its membrane. Proteins with G3PDH activity have also been reported from certain archaeal lineage [12]. If they were not originated by the horizontal gene transfer from bacterial species after the separation of Bacteria and Archaea, the common ancestor of Bacteria and Archaea (or LUCA/Commonote) could have had G3P in its membrane.

To understand the early evolution of cellular membrane, we reconstructed the molecular phylogenetic trees of G1PDH and G3PDH separately. First, we collected G1PDH and G3PDH sequences from Bacteria and Archaea together with the sequences of their homolog proteins. G1PDH and its homolog protein sequences were aligned carefully, and then used for the maximum likelihood (ML) analysis and Bayesian (BI) analysis. G3PDH and its homolog protein sequences were also used for similar phylogenetic analyses.

Results and Discussion: The ML and BI analyses suggested that the bacterial G1PDHs were originated from crenarchaeal G1PDHs by the horizontal gene transfer. However, archaeal G3PDHs form the separate group from bacterial G3PDHs.

Our phylogenetic analyses of G1PDH and G3PDH suggested that the common ancestor of Bacteria and Archaea (or LUCA/Commonote) had cellular membrane with G3P that were formed by G3PDH. During the appearance of Archaea, G1PDH was acquired by the archaeal ancestry, and then the membrane with G3P was replaced with that with G1P. Since the heterochiral membrane may not be unstable [9], the period with heterochiral membrance might have existed during the establishment of Arcahea, even though Arcaeal common ancestor is thought to have been thermophile/hyperthermophile [13].

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