## Crystal Structure of membrane-bound alcohol dehydrogenase from acetic acid bacteria 酢酸菌由来膜結合型アルコール脱水素酵素の結晶構造解析

## 後藤 勝<sup>1</sup>、広津 建<sup>2</sup>、外山博英<sup>3</sup>、足立収生<sup>4</sup>、松下一信<sup>4</sup>、〇宮原郁子<sup>2</sup> 東邦大・理<sup>1</sup>、大阪市大・理<sup>2</sup>、琉球大・農<sup>3</sup>、山口大・農<sup>4</sup>

Many Gram-negative aerobic bacteria can grow on alcohols and sugars as the sole carbon and energy sources. In the periplasm of acetic acid bacteria, quinoprotein alcohol dehydrogenases (ADH) containing pyrroloquinoline quinone (PQQ) instead of nicotinamide or flavin compounds as the prosthetic group catalyze the first step of acetic acid production, oxidation of ethanol to acetaldehyde. There are three types of quinoprotein ADHs. Type I ADH is a soluble, dimeric protein of identical subunits having a PQQ and a calcium ion in each active center, but no other redox cofactors. Type II ADH is a soluble, monomeric quinohemoprotein, having a PQQ-containing catalytic domain and an additional cytochrome *c* domain with a covalently bound heme *c*. Type III ADH is a quinohemoprotein–cytochrome complex with three nonidentical subunits that catalyzes the oxidation of ethanol and the subsequent reduction of ubiquinone, and attached on the cytoplasmic membrane of acetic acid bacteria.

We report here 3.0 Å crystal structure of the type III membrane-bound quinohemoprotein ADH from *Gluconobacter suboxydans* refined to *R*-factor 29 %. Our structure reveals that the enzyme contains a large subunit A similar to the type II quinoprotein ADHs which have a eight-stranded  $\beta$  propeller domain and a cytochrome *c* domain, a membrane-bound subunit B which has a novel three-heme cytochrome *c* structure, and a small subunit C which has unknown function. The PQQ is located near the axis of the propeller domain about 14 Å from the heme in subunit A. The shortest distances between four hemes are about 9 Å, 4 Å, and 8 Å, respectively.