

Methods of growing high-quality crystals as seen from crystallogensis studies

Mitsuo Ataka

National Institute of Advanced Industrial Science and Technology (AIST), Kansai Center

The accuracy and reliability of the three-dimensional structure of protein molecules determined by X-ray crystallography depend on the quality of the single protein crystal used to collect the X-ray diffraction data. The growth of high-quality single crystal and the methods of controlling the crystal quality are thus critically important for structure determination.

We wanted to contribute to this problem from the studies on crystallogensis. Physical, chemical, and biological approaches have been considered.

As a physical means to control the crystalline quality, we considered the use of a superconducting magnet. Adenosine deaminase crystals improved the quality when grown in a magnetic field. Magneto-Archimedes levitation of lysozyme crystals was realized in a magnetic field as low as 3.8 T by using a paramagnetic gadolinium salt as a crystallizing agent. The crystals so obtained in a floating state were superior in quality to the controls as judged by optical microscopy and white X-ray topography.

As a chemical means, crystallization in the presence of two kinds of salt was tried for cytochrome *c* purified from four different organisms. Horse, bovine and tuna cytochrome *c* crystals diffracted X-ray sufficiently well, and the reason behind was speculated to be a stabilizing role of a nitrate ion bound to the flexible terminal region. Protease A from *Aspergillus niger* could form a single crystal when an amphiphilic agent was added in which the proportion of hydrophobicity/hydrophilicity was optimized.

As a biological means, we are considering the comparison of the genomic information and deletion of a part of the molecule.

We expect that a rational way of improving the quality of protein crystals is to some extent to be found by combining the considerations as described above.