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SPACE PROTEIN CRYSTALLIZATION: VAPOR DIFFUSION OR LIQUID/LIQUID DIFFUSION?

Abstract

Protein's function is closely related to its three dimensional structure. X-ray diffraction is one of the most efficient methods for the structure determination, which prerequisite is to obtain protein crystals of high quality. Protein crystallization becomes the limiting step of proteomics due to its complexity. The microgravity environment of space is favorable for growing protein crystals of high quality as it could eliminate the natural solutal convection and crystal sedimentation effectively. Both vapor diffusion (VD) and liquid/liquid diffusion (L/LD) methods are used to grow protein crystals. It is L/LD but not VD method which is considered to be suitable for space experiments although it is seldom used in ground laboratory. However, as the most frequently used method on ground, VD is less and less used in space experiments because of the existence of gas-liquid interface which would resulting in Marangoni convection. To make VD method applicable for space protein crystallization, the new growth chamber was developed which has the structure of immersed capillary tubes. After special treatment, the gas-liquid interface within the tube could keep flat, and the nearly one-dimensional vapor diffusion would not induce the non-uniform distribution of solute or temperature. In another words, this VD growth chamber could eliminate Marangoni convection. A fluorescent protein and a laser scanning co-focus microscope (LSCM) were adopted to study Marangoni convection during protein crystallization. The result showed that the Marangoni convection could be small enough to be neglected with fine controlling on the gas-liquid interface. Also the new VD L/LD growth chambers were successfully used for protein crystallization aboard Shenzhou 8 spaceship accompanying Simbox provided by Germany. Crystallization experiments of 14 proteins lasted 16.5 days. Their counterparts were executed in the ground laboratory with the same crystallization chambers. The results of space experiments showed that the proteins using VD chambers produced better results than those using L/LD chambers. The space-grown hen egg white lysozyme (HEWL) crystal with VD chamber has the diffracting resolution of 1.16Å; the Earth-grown one has 1.23Å only. Actually, the self-designed crystallization chamber could be used for VD or L/LD method with no structure change. In addition, the chamber group with crystallization solutions weighs about 110g, has the volume of 100ml, 120 wells, and needs no power-supply. Therefore, the new chamber is highly portable and of high-throughput.