Introduction of newly developed cryo-FIB supporting CRYO ARM™ performance

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Cryo-EM, known as a tool for analyzing the three-dimensional structure of biological macromolecules, has achieved a great deal of success with single-particle analysis methods. Furthermore, with the advent and development of the cryo-FIB in recent years, it has become possible to observe thicker specimens such as cells and tissues that could not be directly observed by TEM in the past by thinning them. However, the preparation of samples for cryo-EM using cryo-FIB requires handling of a 3 mm diameter TEM grid in liquid nitrogen several times, which reduces the success rate.
To solve this problem, we developed a cryo-FIB system consisting of a cryo-FIB and a transport system.

The cartridge for the CRYO ARM™, a cryo-TEM equipped with an automatic transport system, can be mounted directly on the cryo-FIB. And the cartridge can be also mounted on the newly developed liqN2 cooling stage for fluorescence microscopy. Easy cryo-CLEM workflow without handling the TEM grid during transfer between devices can be achieved.
The cooling stage on this cryo-FIB is a thermal conduction system with liquid nitrogen, which provides extremely high stability of stage position and temperature, so it has the same level of stability as at room temperature, even when cooled. It's suitable not only for cryo-TEM lamella preparation but also FIB-tomography, which enables 3D analysis over a wider range than cryo-TEM tomography.

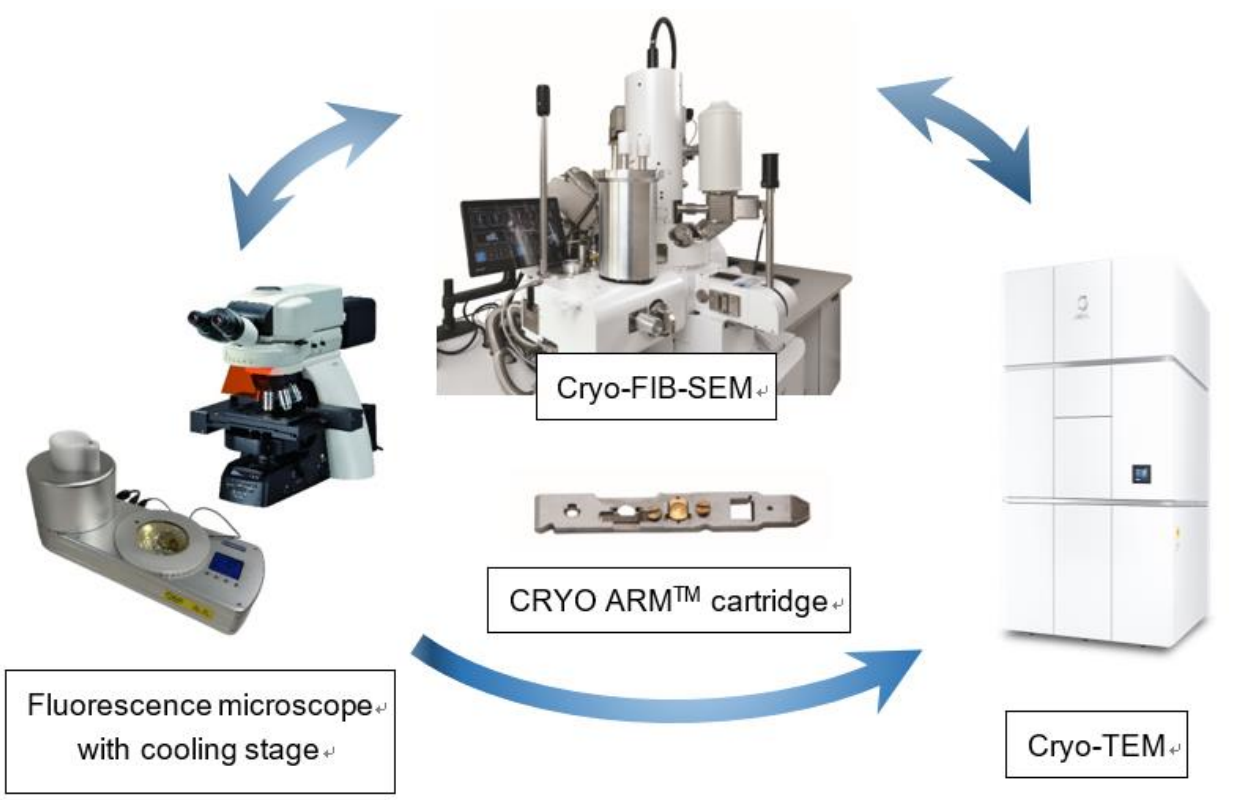

Fig. 1 Overview of cryo-CLEM system
Cryo-CLEM workflow with cryo-FIB consists of three microscopes; fluorescence microscope, cryo-FIB-SEM and cryo-TEM. Hydrated frozen specimen on the TEM grid is set on the CRYO ARM™ cartridge at the beginning of the workflow. The cartridge of can be mounted directly on these microscopes.

Keywords: cryo-FIB; cryo-TEM; analysis the three-dimenstional structure