A low background sample holder for fixed target serial crystallography experiments

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Serial crystallography has become a great success story and an increasing number of structures have been solved using this novel method [1]. In contrast to conventional crystallography, where a structure determination is ideally carried out by data collection from one large crystal in serial crystallography diffraction images from several thousands of small crystals are collected in a serial fashion. The individual datasets from all these crystals are then merged into one large and ideally complete data set from which the structure can be determined. Originally developed for X-ray Free Electron Lasers, serial crystallography has recently been applied at synchrotron sources.

Efficient sample delivery for serial crystallography remains challenging. Currently most of the experiments are conducted with liquid-jet systems with diameters down to 1 um. Drawbacks of all liquid jet delivery systems are high sample consumption and moderate hit rates of typically a few per cent. The use of high viscosity jets such as LCP or agarose tremendously reduces sample consumption but is paid with a large background scattering signal making this method impracticable for micron-sized crystals.

Another sample delivery technique for serial crystallography is the use fixed targets [2]. Here the crystals are loaded onto a solid support, which is then raster-scanned through the X-ray beam. We have developed a sample holder from single crystalline silicon and of about 2.5 x 4.5 mm in size, which is equipped with a periodic array of micro-pores with a diameter between 4 and 30 um [3]. For loading of the chips the micro-crystal-suspension is pipetted on the upper side of the chip. Mother-liquor is then soaked-off through the micro-pores side by using a piece of filter paper from the lower side of the chip. All micro-crystals larger then the pore diameter are retained by the chip and arrange themselves to periodic pore pattern. Drying out of the crystals is prevented by continuously exposing them to a stream of humidified air or helium during loading and data collection. For data collection at cryogenic temperatures the chips can be alternatively cryo-frozen by plunging them into liquid nitrogen or ethane after loading. Due to efficient removal of the mother liquor, the use of single crystalline silicon, and the absence of any cover foil, the chip allows for X-ray data collection with very low background. The chip has already been successfully used for several structure determinations at synchrotrons and also at the X-ray Free Electron Laser LCLS in Stanford.

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2. A. Zarrine-Afsar, A. *et al*. Crystallography on a chip. *Acta Cryst.* D**68**, (2012), 321-323.

3. P. Roedig et al., A micro-patterned silicon chip as sample holder for macromolecular crystallography experiments with minimal background scattering, *Sci. Rep.* 5, (2015), 10451.