The successful application of new sample delivery methods for serial femtosecond crystallography (SFX) at XFEL facilities has warranted a fresh look at how experiments are conducted on storage-ring synchrotron sources. There are now publications from several storage-ring facilities describing serial millisecond crystallography (SMX) with monochromatic x-rays. Recently, we published the first serial, protein crystallography experiments with a polychromatic beam [1]. Monochromatic beam experiments were conducted at the GM/CA 23-ID-D beamline [2]. Micro-crystals of a wide variety of proteins including lysozyme, thaumatin, PSII, phycocyanin, human adenosine A2A receptor (A2A AR), beta-2 adrenergic receptor (β2 AR), KDO8PS, FLPP3, and proteinase K were screened. Crystals were delivered to the beam suspended in lipicd cubic phase, agarose or a high molecular weight PEO (MW=8,000,000), using a LCP injector. For each protein target, tens to hundreds of thousands of diffraction patterns were collected by a Pilatus 6M detector in shutterless mode at a repetition rate of 10 Hz, using a photon energy of 12 keV and 10 µm diameter beam size. From these experiments the structures of the A2A AR, phycocyanin, FLPP3, proteinase K and lysozyme were determined to 3.1 Å, 3.1 Å, 3.0 Å, 2.65 Å, and 2.05 Å resolution, respectively. Polychromatic beam experiments were conducted at the BioCARS beamline (14-ID-B) on micro-crystals in which each crystal was exposed to the X-rays from only a single ~100 picosecond bunch or 4 consecutive bunches separated by 153 ns for a total exposure time of 460 ns. The broad bandwidth (5%) yielded full reflections. Thus only a few thousand diffraction patterns were recorded to build a complete dataset of phycocyanin to 2.7 Å resolution, and over 60,000 images were collected from proteinases K with rates varying from 3.2% - 6.3%. The structure of proteinase K was determined to 1.8 Å resolution. These monochromatic and polychromatic beam experiments demonstrate the feasibility of serial data collection at the APS using micro-crystals. Upcoming developments in beamline optics will increase intensity by a factor of ten. In addition, the intensity will also be increased by another factor of ten to one-hundred from the planned APS-U upgrade enabled by the two orders of magnitude increase in Brightness. All these developments will enable the use of smaller micro-crystals as well as the SMX of larger macromolecules.

References
