Coflow SEC-SAXS at High Flux.

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Over the last three years we have been developing a sheath-flow method called coflow to minimise X-ray damage of sensitive solution samples such as proteins [1,2]. The method works by keeping the protein sample away from the region of slow flow velocities near sample cell walls inherent to laminar flow conditions, thereby narrowing the X-ray dose distribution received by the sample, whilst also better matching the sample geometry to the X-ray beam profile. Coflow is well suited to routine, high throughput solution SAXS and gives many advantages over the traditional flowing solution SAXS measurement for solution analysis of biological and other samples [1]. Efficient X-ray dosing maximises data quality per unit volume of sample and allows one to two orders of magnitude increase in X-ray flux at practical flow rates.

The limiting factor most commonly observed is the radiation resistance of buffers, which can be managed by buffer selection, radical scavenging additives such as glycerol and/or sodium azide, care with buffer composition (with respect to salts, reducing agents, de-gassing) and beam focal size. Coflow on protein solutions is routinely flux limited at the Australian Synchrotron SAXS/WAXS beamline. We have conducted an initial experiment on Petra III P12 beamline using a double multilayer monochromator to explore the full X-ray flux potential of coflow, for some model proteins with various buffers and additives. Results show that coflow allowed SEC measurements at 3.5 x 10^{14} ph/s at 10 keV with practical sample flow rates. The results support the design of a new high flux solution scattering beamline at the Australian Synchrotron.

The development of coflow offers a perhaps unprecedented ability to quantify and understand radiation damage on synchrotron sources, while improving the utility, quality and accessibility of SAXS measurements on routine and challenging samples alike.

References

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