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ESRF

ESRF	**Experiment title:** Combined microSAXS-Microscopy Studies on Colloidal Dispersions	**Experiment number**: **SC-2108**
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Report:

In this report we describe an improved setup of an optical tweezer (OT), which is able to fix a single sample entity in a liquid filled capillary and to perform scanning micro-diffraction experiments on it. The previous setup allowed only a 2D trapping due to the optical components used. This was overcome by applying new concepts in two beamtimes which allow fully 3D trapping and the full freedome of alignment in an 80 x 80 µm^2 rectangular capillary. The concept was tested on single starch granules and liposomes containing phospholipds.

The sketch of the new final setup is shown in Fig. 1. The rectangular, 80 x 80 µm large capillary connected to a remote controlled syringe pumpe and filled with the sample {0} is held in front of the microfocused beam. The single mode CW fiber laser (1064 nm) {1} is directed to spatial light modulator {2}, on which a computer generated diffrative optical element (DOE) is displayed. The DOE allows the control of the shape and the convergence of the laser beam. The dichroic mirror reflects the IR beam through the microscope, which focuses the beam in the capillary. Additional to the improved capillary configuration compared to the setup used in [1,2,3] the visible imaging in the new setup is performed through the same objective as the traping {3} and recorded on the CCD {5}. The drawbacks of the 1st beamtime was overcome in significant improvements for the 2nd beamtime and leaded to the results in this report.

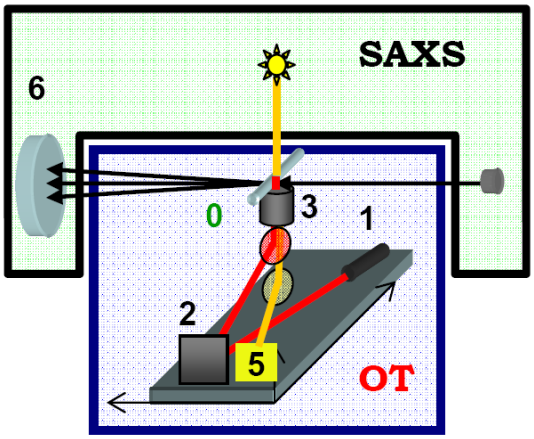
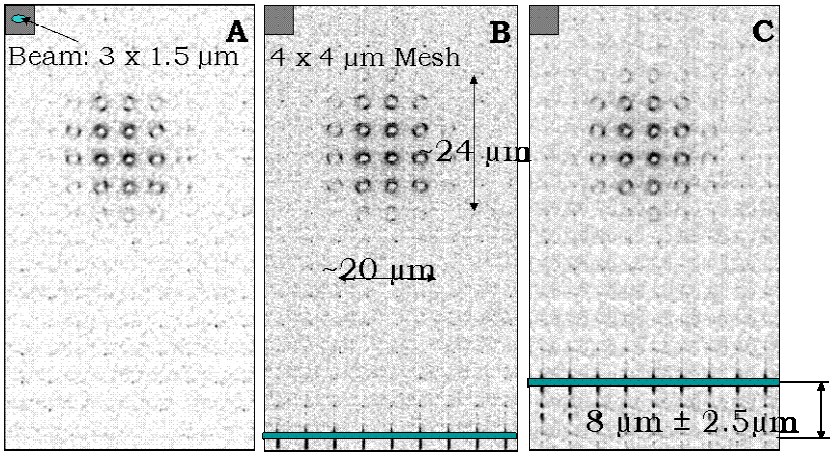


Fig. 1: OT setup 0–sample cell (capillary connected to fluidics),1- IR laser, 2-Phase Programmable Modulator (PPM), 3,4 - microscope objectives, 5,6-CCDs



Z_c=0 µm Z_c= +10 µm Z_c= +20 µm

Fig. 2: Scanning diffraction images of the starch ({001}-9 nm) after moving the capillary to position Zc. Line inidcates the increased scattering of the capillary wall

As mentioned before the new setup enables full 3D trapping. To demonstrate this feature the scanning diffraction images of the 9 nm reflection of the same 20 x 24 µm large starch granule have been taken at three different positions of the capillary (moved upwards 10 µm between each). In the diffraction images the movement of the capillary is clearly oberves from the appearance of the capillary boarder whereas the stability of the trap is recogonized from the same position of the starch granule. Moreover the positional stability is proofed that the images show the typical appearnce as expected from previous experiments on fixed samples [4]: outside - aligned fiber diffraction pattern in radial direction and in the center rotational average.

This method has been used to study the radiation damage of starch granules in water environment. A single strach granules has been trapped and succesively 50 ms exposures have been taken. After 200 ms the starch granules starts to loose long range order because of the radiation damage on one hand. On the other hand the SAXS scattering increases dramatically, which nature is to be revealed. The results obtained on starch granules and on liposomes will be published in [5].

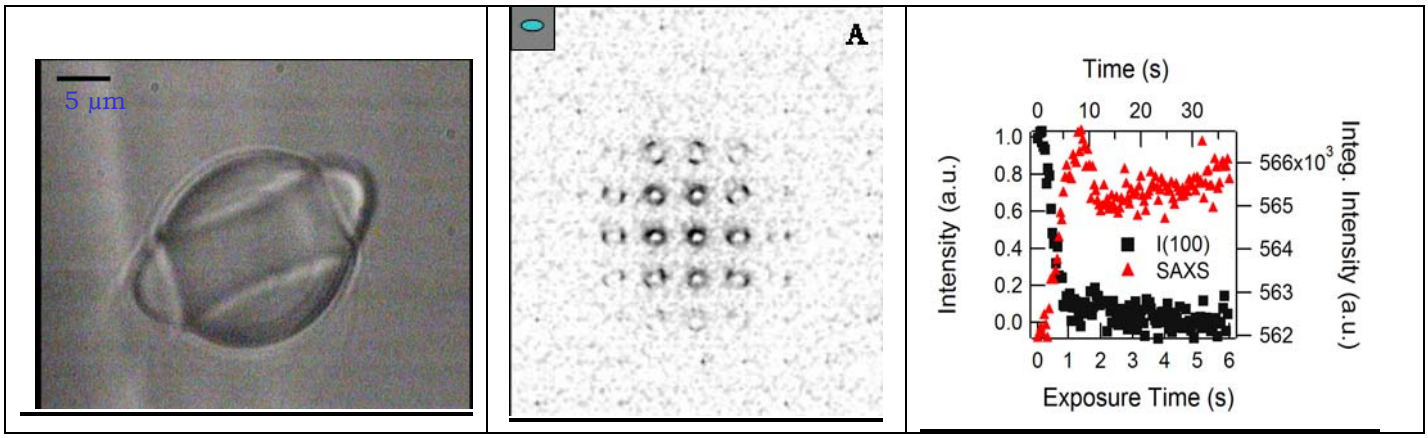


Fig. 2 (left) Microscope image of a single starch granuele with a hole formed by X-ray exposure. (middle) Zoomed scanning diffraction image taken from a 20 x 24 µm starch granules trapped in the LT (beamsize: 1.5 x 2 µm, step size (4 x 4 µm), visible the 9nm SAXS reflection, exposure time 50 ms. (right) Evolution of the integrated intensity of the (100) reflection and diffuse scattering during multiple 50 ms exposures.

[1] Amenitsch H., et al., 2007, AIP Conference Proceedings, 879, 1287-1290

[2] Cojoc D., et al., 2006,SPIE6326, 63261M

[3] Cojoc D., Amenitsch H., submitted to APL

[4] Buleon, A.; Pontoire et al., *Macromolecules* **1997**, *30*, 3952.Gebhardt, R.,et al., Biomacromolecules **2007,** 8, *2092.*

[5] Cojoc D., Amenitsch H., in preparation

Optical Tweezers for Sample Fixing in Micro-Diffraction Experiments

H. Amenitsch*,1, D. Cojoc**,1, et.al. **2007, AIP Conference Proceedings, 879, 1287-1290**

Abstract. In order to manipulate, characterize and measure the micro-diffraction of individual structural elements down to single phospholipid liposomes we have been using optical tweezers (OT) combined with an imaging microscope. We were able to install the OT system at the microfocus beamline ID13 at the ESRF and trap clusters of about 50 multilamellar liposomes (< 10 μm large cluster). Further we have performed a scanning diffraction experiment with a 1 micrometer beam to demonstrate the fixing capabilities and to confirm the size of the liposome cluster by X-ray diffraction.

Keywords: X-ray micro diffraction, Optical manipulation, Phospholipids, Single biological entity
PACS: 87.14.Cc, 87.64.Bx, 82.37.Rs

Combined laser trapping and small-angle X-ray scattering experiment for the study of liposome colloidal microparticles

Dan Cojoc, Heinz Amenitsch et.al. **, 2006,SPIE6326, 63261M**

ABSTRACT

We present and discuss a new experimental setup to perform small angle X-ray scattering and diffraction (SAXSD) of localized liposome colloidal microparticles. A home-built inverted infrared laser tweezers microscope is used to trap, manipulate and aggregate micron-scale liposome particles at single locations inside a 100 microns glass capillary. The micro-focused X-ray and the laser beams are aligned to intersect each other perpendiculary, allowing to associate the Xray diffraction signal to the micron-sized region of interest inside the capillary. Throughout the laser tweezer setup, using diffractive optical elements implemented on a spatial light modulator, we are able to manipulate small aggregates of colloidal particles (liposomes) and fix them in the optical path of the X-ray beam.
We present and discuss first scattering and diffraction experiments on phospholipid liposomes, at the ID13 microfocus beamline of the European Synchrotron Radiation Facility (ESRF). The results demonstrate that we can push the limit of measurable cluster size close to a single liposome.

Scanning X-ray micro-diffraction of optically manipulated liposomes

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ABSTRACT

We demonstrate optical tweezers manipulation of individual micron-sized samples in front of a synchrotron beam that enables nanostructure investigation by diffraction. The validity of this technique is demonstrated for clusters of multi-lamellar liposomes trapped in single and multiple positions in the optical path of a micro-focused X-ray beam (12.56 KeV) and analyzed in a micro-scanning mode. The signal to background ratio of the first order peak shows that single liposome measurements are feasible. Multiple trapping by means of diffractive optical elements is demonstrated as an effective manipulation tool for future X-ray diffraction studies of the interaction between different sample entities.

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