In vivo optical tweezers-based X-ray elemental imaging of single cellular model organisms

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Introduction

Owing to its high sensitivity and non-destructive nature, synchrotron radiation (SR) based confocal X-ray fluorescence (XRF) imaging offers the unique potential of providing two- and three-dimensional information on the sample composition and elemental distributions with trace level detection limits [1]. With the increased availability of nanoscopic X-ray beams provided by 3rd generation SR sources, SR X-ray imaging methods pose important methodological challenges concerning sample preparation, non-contact sample manipulation and non-contact positioning.

Current XRF-related methodological challenges

- Preservation of the structure of biological organisms
- Special need for delicate mounting of microscopic samples onto a support that does not interfere with the X-ray measurement itself.
- Off-line and time-consuming sample preparation procedure prior to analysis at a synchrotron facility.
- Elemental analysis of biological samples that manifest/represent in a hydrated environment is ideally performed on fully hydrated samples.

Proposed methodology for solving challenges

- Optical tweezers (OT) for non-contact sample manipulation combined with
  - Highly sensitive, multi-elemental micro-XRF imaging
  - Organisms close to their natural, in vivo state
  - Free-standing samples in their natural, aqueous environment
  - Non-contact sample positioning and manipulation
  - Eliminate time-consuming and error prone sample preparation
  - Possibility of XRF tomography using multiple optical traps

Optical Tweezers Setup for X-ray Imaging at Synchrotron Facilities

Examine single cells

- Scripsiella trochoidea microalgae (0.30 µm)
- Contained within a quartz capillary filled with specimens & medium (0.100 µm, 10 µm wall).
- Optical manipulation using a 0.5 W laser power and translation to the upper capillary wall to prevent X-ray induced vertical sample movement during a progressing scan [3].

Experimental results

- Significant amounts of Mn, Fe and Zn are detected within the cells, reflecting their essential nature in photosynthesis processes [3]. Observation of an accumulation centre that most likely corresponds to an important organelle (e.g. nucleus, Golgi apparatus etc.).
- SAXS composite image shows a strongly ordered structure at the cell borders, possibly pointing towards the cellulose plates of the cell wall.
- NIST SRM 1577c (Bovine liver) is used for quantification purposes and for gaining insights into the elemental sensitivity. Corrections are applied for the water matrix and for the quartz capillary wall. LOD reaches a few ppm for the transition metals in a 0.5 s measurement.

Conclusions

The newly presented optical tweezers-based imaging approach allows for elemental analysis of biological model organisms and single cells in their natural, in vivo state. In particular, the methodology combines optical tweezers (OT) technology for non-contact, laser-based sample manipulation with synchrotron radiation confocal XRF micro-imaging. Several studies focusing on applications in environmental toxicology have been performed at ESRF-ID13, demonstrating the feasibility, repeatability and high throughput potential of the OT XRF methodology [3], while also providing insights into the methodological challenges [4].

References


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