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Abstract

Identification of arsenic chemical species at a sub-cellular level is a key to understand the mechanisms involved in arsenic toxicology and antitumor pharmacology. When performed with a microbeam, X-ray absorption near edge structure (µ-XANES) enables the direct speciation analysis of arsenic in sub-cellular compartments avoiding cell fractionation and other preparation steps that might modify the chemical species. This methodology couples tracking of cellular organelles in a single cell by confocal or epifluorescence microscopy with local analysis of chemical species by µ-XANES. Here we report the results obtained with a µ-XANES experimental setup based on Kirkpatrick-Baez X-ray focusing optics that maintains high flux of incoming radiation (> 10^{11} ph/s) at micrometric spatial resolution (1.5 x 4.0 µm²). This original experimental setup enabled the direct speciation analysis of arsenic in sub-cellular organelles with a 10^{-15}g detection limit. In a previous experiment (experiment report SC1773, and Baquart et al., 2007), using µ-XANES we evidenced that inorganic arsenite, $As(OH)_3$, is the main form of arsenic in the cytosol, nucleus, and mitochondrial network of cultured cancer cells exposed to As_2O_3. In this experiment, a predominance of As(III) species is observed in HepG2 cells exposed to $As(OH)_3$ with, in some cases, oxidation to a pentavalent form in nuclear structures of HepG2 cells (Bacquart et al., 2010). The observation of intra-nuclear mixed redox states suggests an inter-individual variability in a cell population that can only be evidenced with direct sub-cellular speciation analysis.

References

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