

Contribution ID: 7 Type: Invited

Rejuvenating online UV-Vis microspectrophotometry by monitoring dose- and time-resolved phenomena at both cryogenic and room temperature

Wednesday 4 June 2025 09:30 (30 minutes)

The French protein crystallography beamline BM07-FIP2 at the ESRF enables both cryogenic and room-temperature studies on single crystals, with precise control over the deposited X-ray dose thanks to a large, homogeneous top-hat beam. [1] In addition, its sample environment allows for easy integration of the EMBL/ESRF microspectrophotometer [2], enabling in crystallo UV-Visible absorption and fluorescence measurements in parallel with X-ray diffraction. This approach has allowed for the monitoring of the evolution of the absorbance of metal centres, cofactors, or chromophores as a function of X-ray dose, providing real-time insights into both the extent of radiation damage and the functional state of macromolecules. This presentation will focus on recent methodological developments that led studying the extent of radiation damage on various proteins, enabling the comparison between room and cryogenic temperature. Finally, the plans for the future sample environment of BM07-FIP2 fully integrating an improved microspectrophotometer will be described.

References

[1] McCarthy A. et al. (2025) J Synchrotron Radiat., 32, in press.

[2] McGeehan J, Ravelli RB, Murray JW, Owen RL, Cipriani F, McSweeney S, Weik M, Garman EF. (2009) J Synchrotron Radiat., 16, 163-172.

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Session Classification: Biological Studies Affected by Radiation Damage