SOFT X-RAY MICROSCOPY RADIATION DAMAGE ON FIXED CELLS INVESTIGATED WITH SYNCHROTRON RADIATION FTIR MICROSCOPY

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Radiation damage of biological samples remains a limiting factor in high resolution X-ray microscopy (XRM) [1]: the extent to which the lateral resolution can be pushed without unacceptable sample degradation is still an open question. The present work [2] reports on a novel study performed at Elettra - Sincrotrone Trieste where the impact of soft X-rays (1KeV) on formalin fixed Human Embryonic Kidney 293 cells exposed to different doses has been assessed not only with XRM, as often reported in relevant literature on the topic, but by coupling it with two additional independent non-destructive microscopy methods: Atomic Force Microscopy (AFM) and SR-FTIR Microscopy (FTIRM). The results reveal that cell morphology is not substantially affected even at higher exposure doses (~$6 \cdot 10^8$ Gy), while indeed nanometric pits and bulges increase in number and size when increasing the exposure time (See Fig.1, panels e-g). On the contrary, the biochemical response changes significantly also at the lower radiation dose (~$2 \cdot 10^6$ Gy), resulting in a progressive breakdown of the covalent bonding network (See IR chemical images in Fig. 1, panels b-d). Overall, FTIRM suggests that low-energy X-rays on formalin fixed cells primarily induce the oligomerization of bio-macromolecules and then affect their constitutive monomers down to the formation of small, and possibly volatile compounds. Consequently, unnatural ultrastructural details and modifications in elemental distribution could be induced by the X-ray probing source. Therefore special attention needs to be devoted to improve preparation techniques and acquisition strategies that minimize the risks of artifacts, especially when more radiation-sensitive hydrated species are imaged.

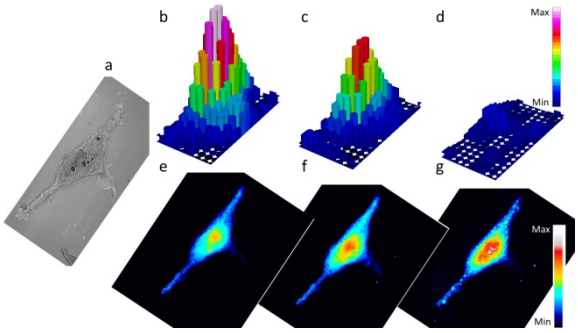

Fig 1: a) Optical image of a formalin-fixed HEK293T cell; b-d) Chemical images of the asymmetric stretching of phosphate moieties (area integral of the spectral band 1270-1190 cm^{-1}) for the same cell in a) upon air-drying and exposure to $2 \cdot 10^6$ Gy (compatible with low resolution STXM mapping) and $6 \cdot 10^8$ Gy (compatible with XRF mapping); e-g) AFM images of the same cell in a) following the same scheme of b-d.

REFERENCES

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