ESRF	**Experiment title:** On-line measurement of cellular activity under hard X-rays irradation	**Experiment number**: LS-2712
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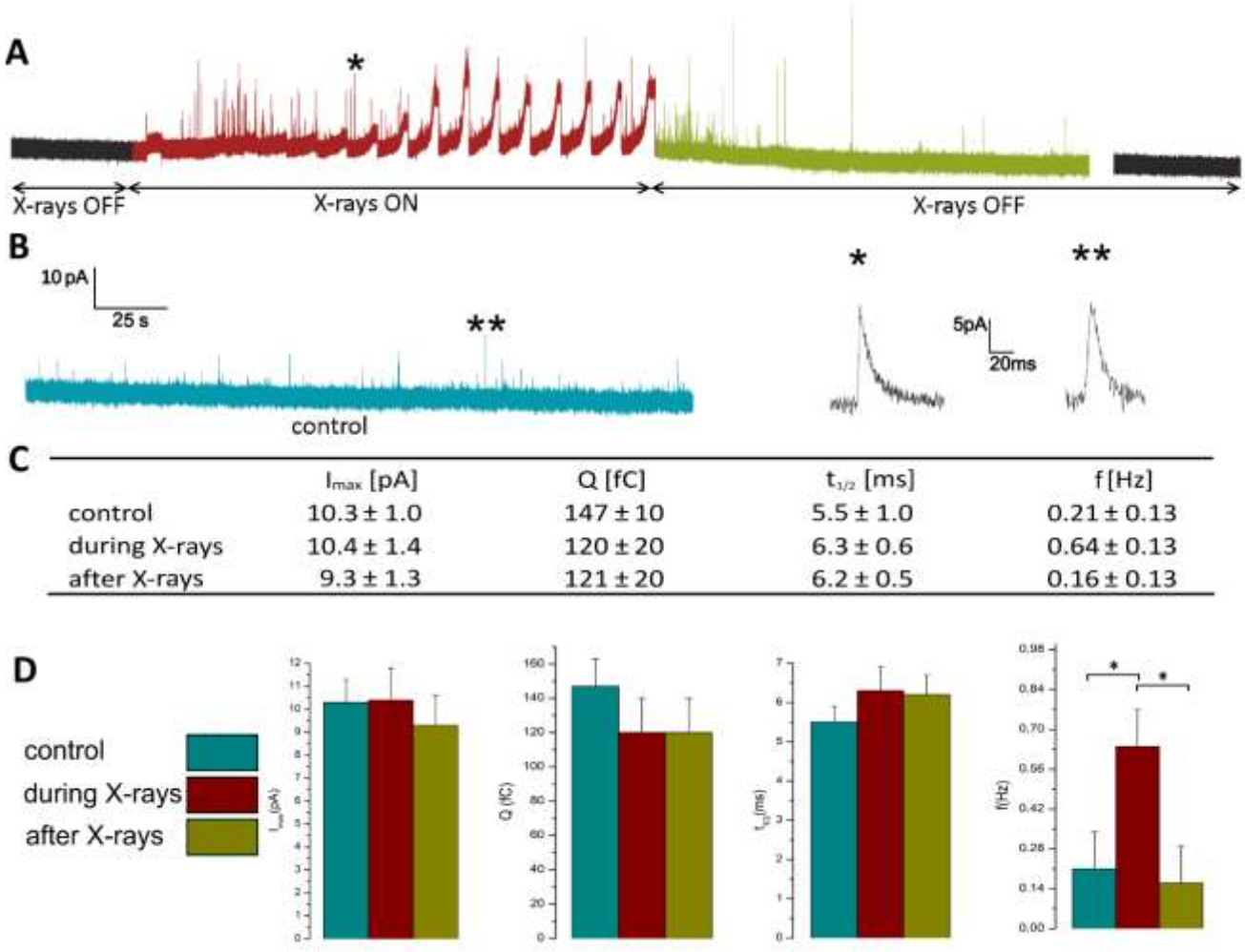
Names and affiliations of applicants (* indicates experimentalists):

M. Truccato[1*], L. Mino[1*], V. Bonino[1*], F. Picollo[1*], C. Lamberti[2], A. Agostino[2], V. Carabelli[3], G. Tomagra[3*]

[1] Department of Physics and NIS Centre, University of Turin, Turin, Italy
[2] Department of Chemistry and NIS Centre, University of Turin, Turin, Italy
[3] Department of Drug Science and Technology, University of Turin, Turin, Italy

Report:

The employment of ionizing radiation is a powerful tool in cancer therapy but beyond targeted effects, many studies have highlighted the relevance of its off-target consequences. An exhaustive understanding of the mechanisms underlying these effects is still missing and no real-time data about signals released by cells during irradiation are presently available. We have employed the X ray nano-beam at ID16B-NA to perform the first real-time simultaneous measurement of both X-ray irradiation and in vitro neurotransmitter release from individual adrenal phaeochromocytoma (PC12) cells plated over a diamond based multi-electrode array. The experiment was performed at 17.4 keV with 55 nm × 60 nm spot size and high flux (up to $\approx 7 \times 10^{10}$ photons s^{-1}) in air. The sample was positioned in the focal plane of a conventional optical microscope, which was aligned to almost coincide with the focal plane of the X-ray beam. By irradiating individual cells at a flux of 7×10^{8} photons s^{-1}, we ensured to keep the cells alive for at least few tens of seconds. The chronoamperogram reported in Fig. 1 shows the activation of an intense exocytotic pattern after starting the raster scanning of the X-ray beam across a single cell that was initially inactive. In this case, many amperometric spikes overlap with the broad and intense peaks associated with the detection of the photocurrent. On the other hand, the parameters associated with the kinetics (i.e. full width half maximum $t_{1/2}$) and intensity (i.e. maximum peak current I_{max} and total peak charge Q) of the exocytotic spikes detected from irradiated cells are statistically consistent with the control dataset ($p > 0.05$, ANOVA followed by Bonferroni post hoc comparison, Fig. 1b - c), while exocytosis frequency presents a significant increment only during the irradiation [1].



	I_{max} [pA]	Q [fC]	$t_{1/2}$ [ms]	f [Hz]
control	10.3 ± 1.0	147 ± 10	5.5 ± 1.0	0.21 ± 0.13
during X-rays	10.4 ± 1.4	120 ± 20	6.3 ± 0.6	0.64 ± 0.13
after X-rays	9.3 ± 1.3	121 ± 20	6.2 ± 0.5	0.16 ± 0.13

Figure 1. X-ray stimulated neurotransmitters release. **A**: amperometric recording of X-ray-induced exocytosis with zoom-in of a spike (labelled as *): the switching-on and off of the beam is highlighted; **B**: Typical chronoamperogram of spontaneous exocytotic events from PC12 cells with zoom-in of a spike (labelled as **); **C**: Table reporting the characteristic parameters of the exocytotic events for both X-ray stimulated and spontaneous events. **D**: Histograms illustrating the data reported in Table C.

In this way we have demonstrated that, in specific conditions, X-rays can alter cell activity by promoting dopamine exocytosis: such effect is potentially very attractive for a more effective treatment of tumours.

Reference

[1] F. Picollo, G. Tomagra, V. Bonino, V. Carabelli, L. Mino, P. Olivero, A. Pasquarelli, M. Truccato, "Triggering neurotransmitters secretion from single cells by x-ray nanobeam irradiation", submitted