

## Cell death induced in tumoral epithelial cells: SR-IRMS analysis

Simona Sabbatini <sup>\*a</sup>, Elisabetta Giorgini<sup>b</sup>, Corrado Rubini<sup>c</sup>, Romina Rocchetti<sup>b,c</sup>, Carla Conti<sup>a</sup>, Elisa Mitri<sup>d</sup> and Lisa Vaccari<sup>d</sup>.

<sup>a</sup>Dipartimento di Scienze e Ingegneria della Materia, dell'Ambiente ed Urbanistica, Università Politecnica delle Marche, via Brecce Bianche, 60131 Ancona, Italy;

<sup>b</sup>Dipartimento di Scienze della Vita e dell'Ambiente, Università Politecnica delle Marche, via Brecce Bianche, 60131 Ancona, Italy;

<sup>c</sup>Dipartimento di Scienze Biomediche e Sanità Pubblica, Università Politecnica delle Marche, Via Conca, 71, 60126 Torrette (Ancona), Italy;

<sup>d</sup>Elettra – Sincrotrone Trieste, S.S. 14 Km 163.5, 34012 Basovizza (Trieste), Italy

Oxidative stress can cause cell death by nonphysiological (necrotic) or regulated (apoptotic) pathways.<sup>1</sup> FTIR microspectroscopy investigations carried out on biological samples, pointed out different spectral patterns, above all related to DNA damages, changes in plasma membrane permeability, protein content and folding, in healthy, apoptotic and necrotic cells.<sup>2</sup> Among all malignancies in human body, oral cavity cancer accounts for approximately 3%, with Oral Squamous Cell Carcinomas (OSCC) representing the main occurrence.

Oxidative Stress	OSCC	OSCC + H <sub>2</sub> O <sub>2</sub> (250 μM)
2 hs	OS_2CTRL	OS_2
4 hs	OS_4CTRL	OS_4
8 hs	OS_8CTRL	OS_8
12 hs	OS_12CTRL	OS_12

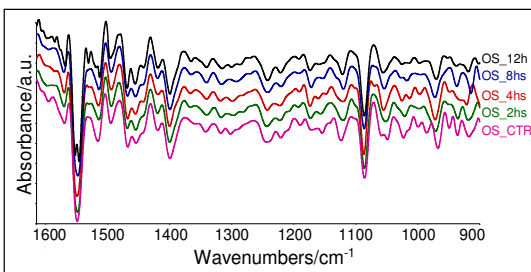


Figure 1: Average spectra of OSCC submitted to oxidative stress (OS\_2hs–OS\_12hs) and control sample (CTRL).

synchrotron light (SR-IRMS); repeated spectra were collected for 10 hours on a single cell, treated with H<sub>2</sub>O<sub>2</sub> (1000μM) (data not shown).

To evaluate the apoptotic and necrotic processes induced by oxidative-stress, to which cancer cells are highly susceptible, G3 OSCC living primary cell cultures were treated with different amounts of H<sub>2</sub>O<sub>2</sub> and analyzed in physiological solution by using a biocompatible IR-transparent microfluidic device (in-house build).<sup>3,4</sup>

In particular, height different aliquots of living OSCC were collected and divided as reported in Table 1. Four groups of OSCC were treated with H<sub>2</sub>O<sub>2</sub> (250 μM) for 2, 4, 8 and 12 hours, to induce the oxidative process; the remaining four aliquots were used as control (CTRL). By comparing meaningful band area ratios, spectral changes were detected in proteins, carbohydrates and nucleic acids (Fig.1).

A single cell analysis was also performed by using Infrared Microspectroscopy with

## References

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