Rho0 cells and metabolomic approach to exploit mechanism of resistance to cisplatin in ovarian cancer cell lines

D. Catanzaro[1], M. Montopoli[1], M. Bellanda[2], C. Frezza[3], G. Froldi[1], E. Ragazzi[1], L. Caparrotta[1]

[1] Dept. of Pharmaceutical and Pharmacological Sciences, University of Padova, Largo E. Meneghetti 2, 35131 Padova, Italy
[2] Dept. of Chemical Sciences, University of Padova, via Marzolo 1, 35131 Padova, Italy
[3] Medical Research Council Cancer Cell Unit, Hutchison/MRC Research Centre, Hills Road, Cambridge CB2 0XZ, United Kingdom

The onset of resistance to cisplatin is one of the major limit in chemotherapy and, despite several mechanisms of resistance have been discovered, they are not still exhaustive[1]. Recently it has been shown that only approximately 1% of intracellular platinum is bound to nuclear DNA, while the great majority of the intracellular drug is available to interact with other nucleophilic sites including but not limited to phospholipids, cytosolic, cytoskeletal and membrane proteins, RNA and mitochondrial DNA[2]. mtDNA, unlike nDNA, does not possess efficient repair systems and is therefore more susceptible to the onset of mutations often associated to cancer development, loss of tumor suppressor, activation of oncogenes and mitochondrial dysfunctions often related with an increase of glycolytic activity. With the aim to identify alternative pathways exploited by ovarian cancer cells to escape cisplatin cytotoxicity in the present study the cytotoxicity of cisplatin was analysed in ovarian cancer cells sensitive and resistant to cisplatin (respectively 2008 and C13 cell lines) and in their derived rho0 cells depleted of mitochondrial DNA. Results indicated that the IC_{50} of cisplatin was significantly lower in 2008-rho0 than in 2008 cells (3.56 μM and 0.72 μM) but it was similar between C13-rho0 and C13 cells (IC_{50} 5.49 μM and 6.49 μM). Other experiments also revealed in C13 cells, as compared to 2008 cells, a significantly reduced respiratory chain activity which correlated to a lower mitochondrial mass and to a lower susceptibility to various metabolic stresses. [1] H-NMR spectroscopy evidenced higher basal content of intracellular GSH and mobile lipids (MLs) in C13 cells as confirmed by preliminary LC-MS experiments and Nile red staining that showed an higher lipid droplets content in C13 cells as compared to 2008.
These results support the hypothesis of a 'metabolic reprogramming' of cisplatin-resistant cells to a lipogenic phenotype with consistent changes in glycolytic rate and mitochondrial respiratory pattern[3]. Even if the complete metabolomic fingerprint of ovarian cancer cells remains to be further elucidated, data suggest that the C13 lipogenic phenotype might represent an innovative molecular rationale to design therapeutic agents useful to overcome cisplatin resistance.

1. Koberle B., Tomicic M.T., Usanova S., Kaina B. (2010). Cisplatin resistance: preclinical findings and clinical implications. *Biochim Biophys Acta.* 1806:172–82.
2. Fuertes M.A., Alonso C., Perez J.M. (2003). Biochemical modulation of Cisplatin mechanisms of action: enhancement of antitumor activity and circumvention of drug resistance. *Chem Rev.* 103(3):645–62.
3. Montopoli M., Bellanda M., F. Lonardoni, E. Ragazzi, P. Dorigo, G. Froldi, S. Mammi, L. Caparrotta (2011). 'Metabolic reprogramming' in ovarian cancer cells resistant to cisplatin. *Curr Cancer Drug Targets* 11:226-235.