Stable aqueous nanocolloids of Platicur: photocytotoxicity in visible light and metabolomics studies in cancer cells

Federica De Castro,[1] Viviana Vergaro,[1,2] Michele Benedetti,[1] Francesca Baldassarre,[1,2] Laura Del Coco,[1] Maria Michela Dell'Anna,[3] Piero Mastrorilli,[3] Giuseppe Ciccarella, [1,2] Francesco Paolo Fanizzi.[1]

[1] Dipartimento di Scienze e Tecnologie Biologiche ed Ambientali, Università del Salento & UdR INSTM di Lecce, Via Monteroni, I-73100 Lecce, Italy

[2] CNR NANOTEC - Istituto di Nanotecnologia c/o Campus Ecotekne, Università del Salento, Via Monteroni, 73100 Lecce, Italy

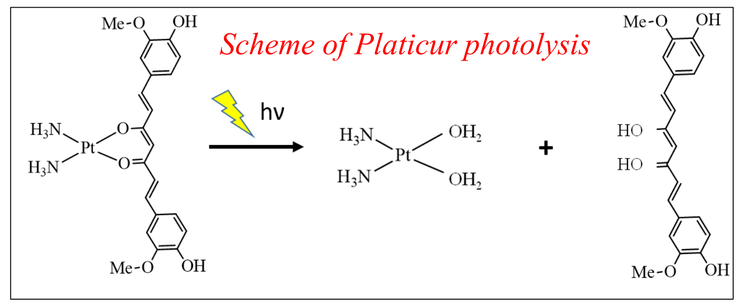
[3]DICATECh, Politecnico di Bari, via Orabona 4, 70125, Bari, Italy

Introduction

Platicur is a *cis*-diamine-platinum(II) complex linked to curcumin, low soluble in water solution. Previous studies have shown that the photoactivation of Platicur, with visible light, is responsible for the production of active species of platinum(II) and a photoactive curcumin. The active species formed act synergistically causing an interesting anticancer activity. [1]

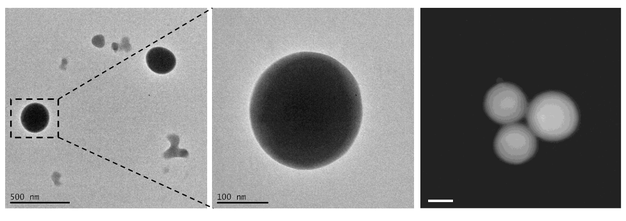
Nanotechnology-based drug delivery to the cancer tissue offer better therapeutic properties as they specifically target cancer cells, also increasing the transport of promising drugs characterized by low water solubility in aqueous solutions.

The use of a metabolomic approach based on NMR spectroscopy in the research of the mechanism of action or for the evaluation of the tumor response to the anticancer metal drugs is a new tool. The side effects induced by metal based drugs, the prediction of the response to treatment and the key information on the mechanism of action (for known and new compounds) could be easily obtained using NMR-based metabolomics. [2, 3]



Experimental

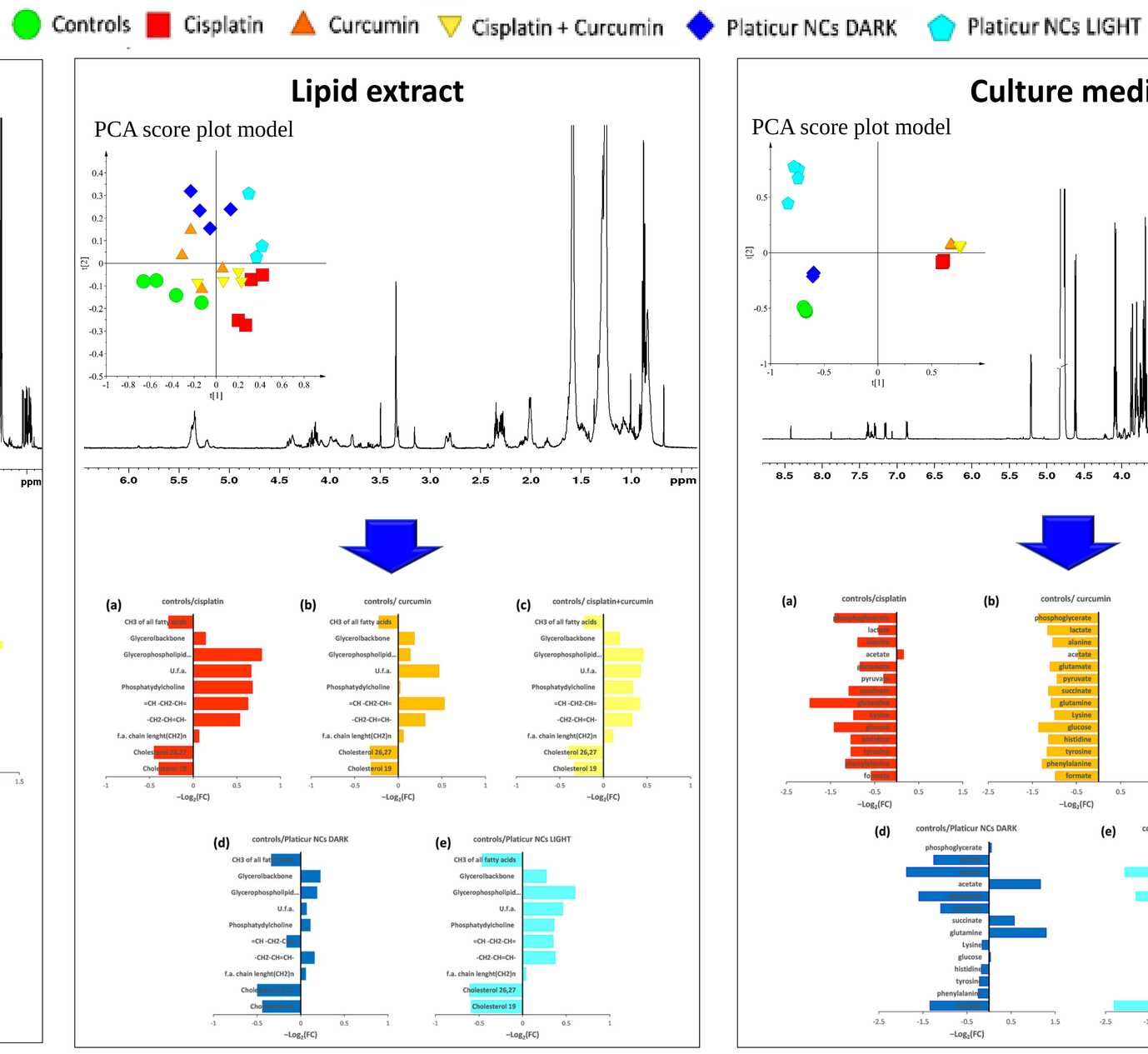
This study reports the synthesis and characterization of new chitosan based nanocolloids of Platicur with spherical shape and size of 120nm. Cytotoxicity assays on HeLa cells exhibited an enhanced efficacy of Platicur when delivered by nanocolloids in DARK (non photoactivated) and LIGHT (photoactivated) form. Photoactivation occur through a Xenon visible light source. Interestingly photoactivated nanocolloids of Platicur reach an IC_{50} dose in the nanomolar order (IC_{50}= 50nM) with respect the micromolar of the dark form (IC_{50}=75µM)



TEM and STEM characterization of Platicur NCs

Multivariate Data Analysis (MVA) of 600 MHz ^{1}H NMR spectra

In order to probe the responses of Hela cells to photoactivated Platicur nanocolloids exposure, a multivariate spectroscopic data analysis was performed. The both intracellular extracts and extracellular medium of Hela cells at the 24 h of treatment, at IC_{50} dose of treatment, were analyzed using chemo-metric and pattern recognition techniques, PCA (Principal Component Analysis) and OPLS-DA (Orthogonal Partial Least Squares Discriminant Analysis) models of analysis. All treatments (cisplatin, curcumin, cisplatin-curcumin, Platicur nanocolloids DARK and Platicur nanocolloids LIGHT) are performed at the IC_{50} dose.



Conclusion

In this work, chitosan-pectin polysaccharides were chosen as Platicur delivery systems obtaining Platicur nanocolloids of about 100 nm, by ultrasonication assisted Layer by Layer Technology (LbL). Studies of different formulations of Platicur nanocolloids have been performed and the (CHI/PE)2,5 formulation appeared the best in term of controlled release and enhanced bioavailability. The light activated Platicur NCs showed a very low IC_{50} value (IC_{50} 50nM) with respect to the Platicur NCs in the darkness (IC_{50} 75µM), as demonstrated on HeLa cancer cell line (cervix cancer cell line).A ^{1}H-NMR metabolomics approach was used to study the metabolic alterations induced by Platicur NCs LIGHT and DARK with those of Cisplatin, curcumin and Cisplatin+curcumin (as co-administered drugs) on HeLa cells.

Very similar metabolic alteration were induced by Platicur NCs LIGHT treatment and the combined treatment Cisplatin+curcumin (characterized by an higher IC_{50} and low solubility due to the presence of curcumin). The metabolomics investigation strongly support the hypothesis that photoactivation of Platicur NCs induces curcumin and Cisplatin aqua species release. Moreover, the observed metabolic alterations due to Platicur NCs LIGHT treatment are typically attributed to apoptotic phenomena. The use of Platicur NCs and their stability under physiological conditions could allow a controlled drug release. Moreover, the non-toxicity in the darkness and the very low IC50 are very interesting properties for possible side effects reductions, therefore promoting Platicur NCs as very attractive photo-chemotherapeutic drug.

References

[1]Mitra K., Gautam S., Kondaiah P., Chakravarty A. R. Angew Chem Int Ed Engl. 2015; 54 (47): 13989-13893.

[2] De Castro F., Benedetti M., Antonaci G., Del Coco L., De Pascali A. S., Muscella A., Marsigliante S., Fanizzi P. F. Molecules. 2018, 23 (9): 2301/1-2301/21.

[3]De Castro F., Benedetti M., Del Coco L., Fanizzi P. F. Molecules. 2019, 24(12):2240/1-2240/14.