Is P-glycoprotein relevant for the release of microvesicles by tumor cells?

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Aim: In this study, we aimed to verify if MDR cells without expression of P-gp also produced more microvesicles and less exosomes than their DS counterpart cells.

Introduction: Cancer multidrug resistance (MDR) is a major cause of chemotherapy failure and is highly associated with overexpression of drug-efflux pumps such as P-glycoprotein (P-gp). The identification of mechanisms specific of P-gp overexpressing cells may contribute to the identification of biomarkers of MDR.

It was recently discovered that a drug-resistant phenotype may be horizontally transferred from drug-resistant (DR) to drug-sensitive (DS) cells, mediated by the cargo of extracellular vesicles (EVs) released by DR cells and captured by DS cells. These EVs include smaller exosomes and larger microvesicles. Our previous work showed that MDR cells with overexpression of P-gp released more microvesicles than exosomes, unlike their DS counterpart cells. However, it is not known if this phenomenon is restricted to MDR cells with overexpression of P-gp or if it is extensive to all DR cells (with other mechanism of drug resistance).

Methods: Drug-response curves of MDR and DS counterpart cells were obtained, using resazurin and trypan blue assays, to confirm the resistant or sensitive phenotype of the cell lines. Confirmation of their P-gp status was possible by Western-Blot. EVs released by both DS and MDR cells were isolated by ultracentrifugation and characterized by transmission electron microscopy, dynamic light scattering, nanoparticle tracking analysis and Western blot analysis.

Results: We confirmed that MDR cells without expression of P-gp release EVs with similar sizes to the ones released by their DS counterparts.

Conclusion: So, P-gp may be associated with the release of larger EVs by MDR cells. These results will be further confirmed by characterizing the EVs released by P-gp overexpressing MDR cell lines following downregulation of P-gp expression and the EVs released by DS cell lines following transfection of P-gp.