Abstract Talk YoungInnovation

Tumor microenvironment (TME) is characterized by chemical properties well suitable to some pharmacological strategies, such as drug delivery systems. In this context, the tumor mass has received considerable attention for targeted therapy, because it presents several physiological features, such as acidic pH, high levels of reactive oxygen species, and enzymes overexpression, that are exploited as stimuli to induce specific changes in the nanocarrier structure, and thereby facilitate target-specific delivery of chemotherapeutic agents. Different nanotechnologies have been proposed for cancer management, due to their potential capacity to deliver the right drug concentration to the tumor site, avoiding its overall body distribution and thus reducing its toxicity. An approach to improve the efficacy of chemotherapeutics is the development of nanocarriers that can be triggered to release the drug in response to extracellular or intracellular chemical stimuli. The use of nanocarriers constituted of pH-sensitive substances could exploit the physiological differences between tumor microenvironment and physiological environment. Peptidyl-prolyl cis-trans isomerase NIMA-interacting 1 (Pin1) is a flexible, sequence-specific, and phosphorylation-dependent enzyme that recognizes, binds, and isomerizes phosphorylated serine/threonine-proline (pSer/Thr-Pro) motifs and that can impact the activity stability, and subcellular location of interacting proteins, playing important roles in cell cycle regulation, differentiation, immune regulation, stemness, and tumorigenesis. In particular, Pin1 is aberrantly overexpressed and/or overactivated in human cancers, exerting a critical influence on tumor initiation and progression via regulation of the biological activity, protein degradation, or nucleus-cytoplasmic distribution of its substrates. For all these reasons, Pin1 could be an attractive target for cancer therapy. Recently, it has been demonstrated that ATRA inhibited proliferation and invasion in ovarian cancer, liver cancer, and lung cancer. Moreover, it is noteworthy that ATRA induces the degradation of the peptidilprolylil-cis/trans isomerase Pin1 by binding its active site. Given that Pin1 is overexpressed in a wide range of tumors and it sustains several oncogenic pathways, these findings fostered the application of ATRA in the treatment of a great variety of solid tumors. Despite the promising results achieved in the pre-clinical phases for the treatment of solid tumors, it is well known that solid tumors could develop ATRA resistance during carcinogenesis (intrinsic resistance) or over the long-term treatment (acquired resistance). In addition to the above-mentioned drug resistance mechanisms, some ATRA properties may limit its clinical efficacy such as its hydrophobic nature, which does not allow parenteral administration, and the short biological half-life in humans caused by its metabolism regulation by CYP-450 in the liver. Additionally, since ATRA exhibits high photo-instability and chemical instability in the presence of heat and oxidants, its clinical use in cancer therapy is limited. To overcome the several limitations associated with current ATRA therapy, the use of drug-carriers could improve ATRA delivery while reducing side effects and to obtain higher concentration in the site of action with respect to free ATRA oral administration. The aim of this work is to design, prepare and characterize pH-sensitive niosomes to realize controllable release in the acidic TME. The pH-sensitive surfactant selected is Tween® 21 and Tween® 20 as control non-pH-sensitive surfactant. Niosomes were characterized in terms of hydrodynamic diameter, polydispersity index (PDI) and ζ-potential. The main objective of the present project is also to develop and compare nanocarriers prepared with two different techniques (Thin Layer Evaporation and Microfluidic Technique). In fact, while current manufacturing processes used to produce niosomes are generally complex multi-batch processes, leading to batch-to-batch variability and the production of particles with high dispersity, reproducible control of particle size and size distribution can be implemented thanks to microfluidics flow systems. Using hydrodynamic focusing in microfluidic channels, nano-sized niosomes with smaller size and narrower size distribution are easily formed by varying flow parameters. For example, niosome preparation processes adopted in the laboratory setting (e.g. TLE) do not offer easy translation to large scale production, which may delay the development and adoption of new niosomal systems. Preliminary evaluation of the physical-chemical properties of niosomes were carried out both at physiological pH and upon acidic pH by fluorescence analyses of niosomes loaded with specific fluorescent probes: Nile Red and Pyranine to assess pH-sensitivity. In addition, ATRA release rate and ATRA stability, free and loaded in pH-sensitive and non-pH-sensitive niosomes, over time were also evaluated. Moreover, ATRA-loaded niosomes in comparison to unloaded ATRA were studied *in vitro* on high-grade serous ovarian cancer (HGSOC) cell lines (i.e. Kuramochi, OVSAHO, OVCAR-3) to evaluate efficacy and determine whether the ATRA-loaded niosomes are more effective than free ATRA. The results of this project will help to design novel therapeutic strategies in ATRA ovarian cancer treatment.