New nanosized agents for imaging guided treatment of tumours

Maria Rosaria Ruggiero
University of Turin Saet S.p.A.
mariarosaria.ruggiero@unito.it
Tutors: Prof. Silvio Aime Ing. Ciscato Dario

Personalized medicine is defined, according to the U.S. National Institutes of Health (NIH), as 'a form of medicine that uses information about a person's genes, proteins, and environment to prevent, diagnose, and treat disease' [1]. The ultimate goal of this medicine is to furnish the proper treatment to the right person at right time, with potential benefits including increased response rates and survival, as well as reduced toxicity [1]. The impact of personalized medicine is contingent upon a systematic discovery of novel biomarkers that can be prognostic and predictive for patient outcome. It is well known that quantitative and selective biomarker detection and rapid profiling of diseased cells in unprocessed biological samples (biopsies, blood, needle and aspirate) is the key-point of this type of therapy. The urgent need is to take a step forward from the simple diagnosis of specific receptor positivity to a stratification of patients based on a quantitative estimation for seeking an improved matching to the responsiveness to one of the available targeted therapy. The available methods, mainly based on histological slide-tests, do not provide an adequate response to this medical need as they suffer for a poor reproducibility and a large heterogeneity in the regions of the tissue specimen [2]. In this context, a new diagnostic method, named R-ELISA (Relaxometric Enzyme Linked in Cells Suspension Assay) has been proposed in this PhD project, for the quantification of biomarkers of interest based on the combination of Nuclear Magnetic Resonance (NMR) relaxometric technology with an enzymatic amplification procedure, detecting the target epitopes in a way comparable to techniques like ELISA. This *in vitro* diagnostic method is based on the release of paramagnetic species from relaxometrically "silent" liposomes operated by the action of a phospholipase A_2 (PLA_2), previously targeted to the epitope of interest. The released paramagnetic species causes an increase of the longitudinal water proton relaxation rate proportional to the number of PLA_2 bound to the cell outer surface. The sensitivity of the herein proposed method, was attempted in the detection of folate receptor expression on human ovarian cancer cells by functionalizing PLA_2 with folic acid and validated by well-established spectrofluorimetric procedures [3]. In addition, for "in vivo" evaluation of biomarkers expression and "in real time" drug biodistribution, different nanoparticles (PLGA-NPs) have been proposed in this project, both based on use of PolyLactic and Glycolic Acid polymer and suitable for imaging guided drug delivery. Firstly, because the magnetic iron oxide nanoparticles (Fe-NPs) can be exploited in biomedicine as agents for magnetic fluid hyperthermia (MFH) treatments and as contrast enhancers in magnetic resonance imaging, new oleate-covered Fe-NPs have been prepared (either by co-precipitation or thermal decomposition methods) and incorporated into PLGA-NPs (PLGA-Fe-NPs) to improve their biocompatibility and in vivo stability. The NPs formulations are characterized by peculiar ^{1}H nuclear magnetic relaxation dispersion (NMRD) profiles that directly correlate with their heating potential when exposed to an alternating magnetic field. Furthermore, the PLGA-Fe-NPs have been loaded with paclitaxel to pursue an MFH-triggered drug release. By prolonging the magnetic field exposure to 30 min, a significant drug release was observed for PLGA-Fe-NPs in the case of the larger-sized magnetic NPs. [4]. Secondly, PLGA-NPs, coated with L-Ferritin, have been exploited for the simultaneous delivery of paclitaxel and an amphiphilic Gd based MRI contrast agent into breast cancer cells (MCF7). Ferritin moieties endow PLGA-NPs with targeting capability, exploiting SCARA5 receptors overexpressed by these tumour cells, that results in an increased paclitaxel cytotoxicity. Moreover, protein coating increased NPs stability thus reducing the fast and aspecific drug release before reaching the target. The theranostic potentiality of the NPs has been demonstrated by evaluating the signal enhancement on T1-weighted MRI images of labelled MCF7 cells, shown a good capability to target cancer cell line with higher level of SCARA5 receptors[5].

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