Development of a surface-switching theranostic lipid-PLGA hybrid nanoparticle platform

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Target audience: Researchers interested in novel, non-invasive imaging techniques to assess the efficacy of new drugs for cardiovascular disease smart MRI probes for targeted diagnosis and therapy.

Purpose: Surface functionalization of nanoparticles with targeting ligands such as antibodies, peptides or nucleic acids has shown significant advantages for target-specific imaging and therapy. However, in vivo, those moieties may result in an elevated recognition of the nanoparticles by the mononuclear phagocyte system or result in targeting of vascular markers, leading to their premature removal from the blood. To overcome the aforementioned limitations we have developed a new lipid-PLGA nanoparticle platform, which is decorated with targeting ligands that are shielded by a matrix metalloproteinase-2 (MMP-2) cleavable polyethylene glycol (PEG) coating.

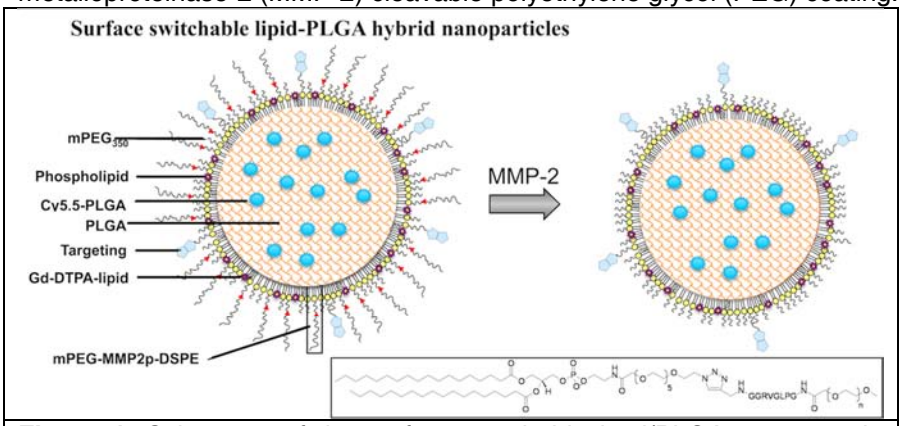

Figure 1. Schematic of the surface-switchable lipid/PLGA nanoparticle platform. Structure of the MMP2-cleavable lipid is depicted in the inset.

Methods and Results: The surface-switchable nanoparticle platform is composed of a near infrared (NIR) fluorescent PLGA core and a (paramagnetic) PEG-lipid coating (Figure 1). To allow surface-switching we synthesized a PEG350 phospholipid to which PEG2000 is conjugated via an MMP2 cleavable peptide unit (mPEG-MMP2p-DSPE using a state-of-the-art click chemistry (Figure 1, inset). The hybrid nanoparticles were synthesized by dripping a PLGA/acetonitrile mixture in hot water/ethanol containing PEG350 phospholipids (mPEG350-DSPE), PEG3000 phospholipid (mPEG3000-DSPE) to provide shielding and biotin or RGD functionalized PEG-phospholipids (biotin-PEG1000-DSPE or RGD-PEG750-DSPE) for targeting at a molar ratio of 0.85/0.10/0.05. To enable optical imaging 5% of the PLGA polymers were labeled with the NIR dye Cy5.5. The stability of the nanoparticles was improved by the inclusion of lecithin at 40 weight% of the total lipid content.

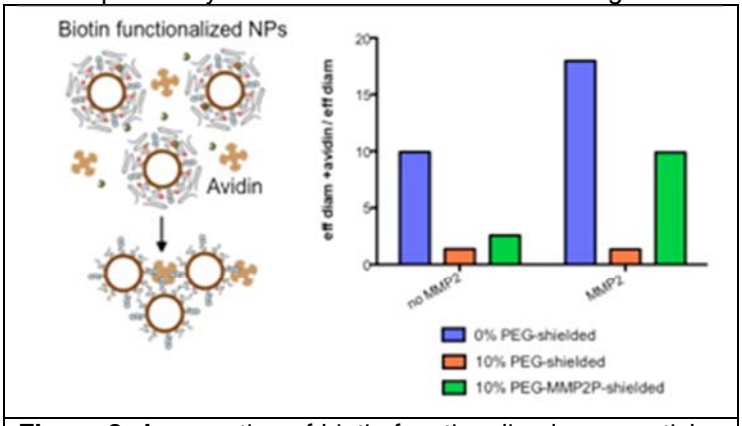

Figure 2. Aggregation of biotin functionalized nanoparticles through avidin-biotin interaction.

During MMP2 incubation long PEG chains are cleaved as conceptually depicted in Figure 1. We observed that mPEG-MMP2p-DSPE biotin-nanoparticles that were pre-incubated with MMP2 aggregated upon incubation with avidin, similar to the control nanoparticles with the freely exposed biotin-PEG1000-DSPE. Conversely, mPEG-MMP2p-DSPE nanoparticles not pre-incubated with MMP2 did not aggregate (Figure 2).

To demonstrate the versatility of mPEG-MMP2p-DSPE functionalized platform we included 25% paramagnetic Gd-DTPA-DSA lipids at the expense of mPEG350-DSPE. We observed the nanoparticles to have similar aggregation properties. The ionic relaxivies r1 and r2 were determined to be 18 $mM^{-1}s^{-1}$ and 32 $mM^{-1}s^{-1}$, respectively.

Discussion: In the current study we have demonstrated the synthesis of a lipid-PLGA hybrid nanoparticle with MMP2 cleavable PEG-lipid included it in the corona. It was observed that upon incubation with MMP2 the nanoparticle's targeting ligands become available for binding. The inclusion of paramagnetic lipids rendered this nanoparticle platform suitable for detection with MRI as well. Future studies will be aimed at evaluating this multifunctional nanoemulsion platform for specific targeting of extravascular cell types, without compromising its pharmacokinetics or inducing activation of the MPS.