

# Construction of a multifunctional DNA-based carrier system for Antisense Oligonucleotide delivery

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Single stranded DNA is suitable to construct three dimensional carriers by forming double strand DNA giving a predictable shape. This project deals with the design and assembly of DNA-based carriers for therapeutic applications. These carriers are going to be modified with diverse molecules for transfection e.g. of HeLa cells. Such molecules may simply be fluorescent dyes for localization and pathway studies inside the cell. On the other hand small cell penetrating peptides (CPPs) may be fused to the carrier to simplify its cellular uptake. Moreover these constructions are to be fused to short antisense oligonucleotides (ASO) that target distinct micro RNAs (miRNA) of living cells. With the help of these ASOs particular miRNAs, that are supposed to support cancer diseases somehow, might be knocked down by RNA interference. By confocal microscopy the success of transfection will be checked and in life activity assays the cell vitality before and after transfection can be evaluated - how far the knocked-down miRNA indeed plays a role in tumoral pathways. Until now the oligonucleotides were designed and successfully assembled to form the carrier system. Furthermore its free 5'-ends were functionalized using copper free click chemistry with CPPs and fluorescent dyes.

In near future we will try to use more nuclease stable nucleic acids e.g. phosphorothioate modified DNA (PTO). PTO is more exonuclease and endonuclease stable than DNA because of the introduction of sulfur into the phosphodiester backbone. In this way modified oligonucleotides can resist degradation in cells much longer after transfection. This issue could result in stronger effects by functionalized PTO-oligonucleotides (or functionalized PTO-carrier systems). This will be helpful if DNA-based carriers degrade too fast before the desired effect appears or the effect cannot be maintained for long.