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Quantifying nuclear import of lipoplex-deliveredpDNA in A10 and MDCK cells

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Cationic lipids spontaneously bind, condense and coat DNA resulting in the formation of lipid/DNA complexes, so-called lipoplexes [1]. These complexes transduce plasmids into cells causing expression of the genes (transfection).

Low levels of transfection circumvent therapeutical efficacy of non-viral strategies. Nuclear accumulation of plasmid DNA is among the major obstacles of non-viral gene delivery [2].

This project aims to analyse nuclear transport of lipoplex-released pDNA as a potential transfection barrier in two cellular models, A10 and MDCK, which have been characterized regarding their endocytic profile and transfectability in previous work [3]. To study nuclear import of pDNA, a novel strategy called "quantitative imaging" is applied, a combination of confocal laser scanning microscopy and image-based analysis using the open source software CellProfiler. This technique enables to track cy3-labeled complex DNA inside the cell and to quantify the amount of nuclear-accumulated complex released pDNA.

These studies reveal nuclear entry of the pDNA to represent a significant transfection barrier in MDCK cells. The investigated cellular models differ significantly in the amount of nuclear-accumulated complex DNA: nuclear transport is by far more efficient in A10 compared to MDCK cells.

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