41.3.10 - Drugs, Drug Targets

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# Modified mRNA delivery to the heart using Lipid Nanoparticles

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Myocardial infarction is a global health burden for which there is no treatment available that aims to recover the damaged tissue after the ischemic event. Lipid nanoparticles (LNPs) represent a well characterized class of mRNA delivery systems, which were recently approved for clinical usage in their application for mRNA-based covid-19 vaccines.

After myocardial infarction, endogenous mechanisms that enable repair of the functional damaged tissue can be triggered by modified mRNA (modRNA) delivery, locally in the infarcted area. As a first step, in order to optimize the LNP formulation for effective myocardial delivery and study cellular tropism of the LNPs in the heart, different LNPs formulations will be evaluated as delivery systems in a murine healthy heart model.

Different LNP formulations varying in type and amount of helper lipid were used as delivery systems for modRNA encoding the reporter genes luciferase or eGFP. In vitro, LNPs were evaluated for modRNA delivery in a human endothelial cell line (HMEC-1), induced pluripotent stem cell-derived cardiomyocytes (iPS-CMs) and induced pluripotent stem cell -derived fibroblasts (iPS-FBs). In vivo, modRNA delivery was evaluated in C57BL-6 mice, undergoing open chest heart surgery under general anaesthesia in order to infuse LNPs into the left ventricular wall. For determination of luciferase expression levels, animals were infused with luciferin substrate intraperitoneally 24 hrs after injection. Heart, liver, lungs, spleen and kidneys were extracted for imaging in a bioluminescence imaging system. The organs were then stored in liquid nitrogen for further ex-vivo modRNA delivery analysis. For determining cellular tropism, histology was performed on mice treated with eGFP modRNA.

Both bioluminescence imaging and luminescence analysis in tissue lysates showed that mRNA transfection is achieved in the myocardium 24 hours after LNP intramyocardial administration. However, all LNP formulations also resulted in high expression levels in other organs, including liver and spleen. Changes in type or amount of helper lipid in LNPs strongly affected transfection levels. Histology of the treated hearts revealed a distinct transfection pattern. The targeted, interstitial cells were negative for CD31 (marker for endothelial cells and monocytes) and Troponin I3 (marker for cardiomyocytes) (Figure 1).

We show that, using an optimized LNP formulation, a significant degree of modRNA local transfection of the heart can be achieved. However, despite the local route of administration (into the left ventricular wall), the highest LNP transfection is shown in remote organs such as liver and spleen. More improvements of the LNP formulations must be done to increase their tropism towards the heart tissue for their optimization as cardiac delivery systems. Determining which cell types are being targeted is also important in order to establish a therapeutic target when applying the LNPs for cardiac therapy.

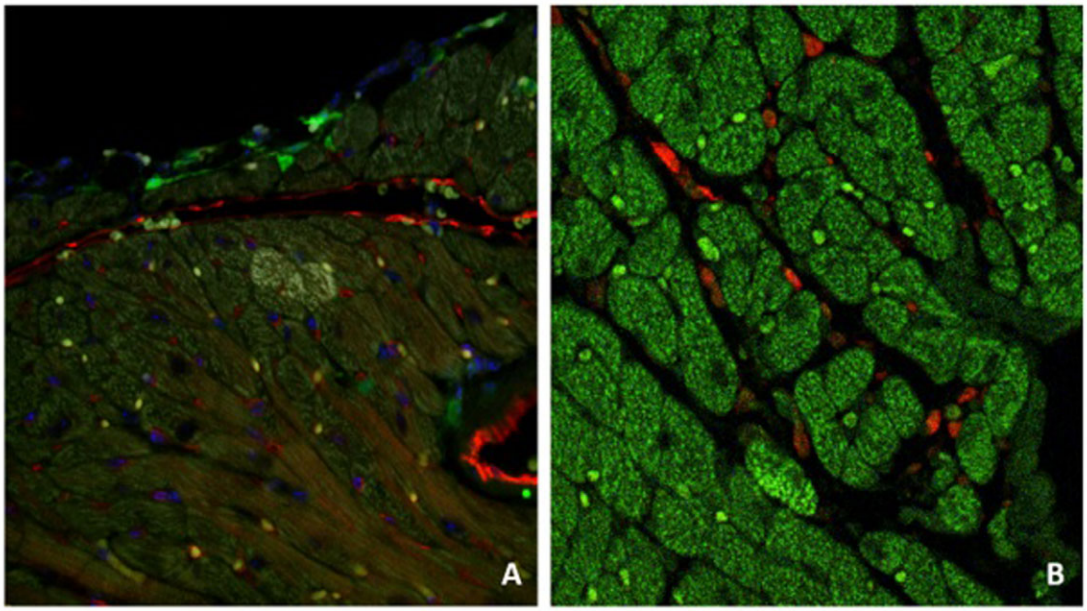


Figure 1: Immunofluorescence images of heart tissue sections of C57BL-6 mice 24hs after intramyocardial injection of eGFP encoding mRNA-LNPs. A: CD31 is shown in red and eGFP in green. B: Troponin I3 in shown in green and eGFP in red.

Immunofluorescence of heart sections