DNA Nanoparticles for Cardiac Regeneration with miR-199a-3p

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Cardiovascular diseases are the leading cause of death globally. Myocardial infarction (MI) accounts for almost 50% of these deaths. If it is not lethal, the infarcted tissue remodels and may progress to heart failure. To date, there is no cure for heart failure nor treatments capable of inducing post-MI cardiac regeneration [1].

miRs therapy (with mimics or anti-miRs) has shown curative potential in heart disease, including post-MI treatment in large animals [2]–[4]. In particular, miR-199a-3p induced cardiac regeneration in mouse, rat, and pig MI models by promoting cardiomyocyte (CM) proliferation [4]–[6]. However, therapy with miRs has a series of associated obstacles such as the lack of stability in circulation, the targeting of the therapy to the target organ or the difficulty cross the vasculature. In this regard, the use of synthetic nucleic acids for the preparation of biocompatible nanostructures is an emerging and promising alternative since it can house a large amount of miRs in its structure, and be functionalized to target a specific organ by promoting their parenchymal accumulation across vessels.

Here we present a nanohydrogel-type nucleic-acid based nanostructure (NANs) harboring miR-199a-3p. NANs are formed by self-assembly of a DNA three-way junction construct with complementary RNA-based linkers [7]. NANs have been characterized by gel electrophoresis, dynamic light scattering and transmission electron microscopy. NANs effectively internalize HEK293 with almost 100% of cells loaded with nanostructures. The miR is actively released through the action of intracellular RNase H. Furthermore, since one of the most common techniques to increase the stability of the RNA against serum nucleases is the use of chemical modifications, we have studied how these modifications and the combinations of them affect the functionality of the miR. From all the combinations, 6 different functional linkers were selected to study their activity in the NANs obtaining 3 potential NANs that demonstrated to be able to deliver a functional miR in a HEK293 model (labelled as NAN1, 2 and 3 in figure 1). Finally, we have established a myocardial endothelial barrier model, enabling future studies on the transfer of ligand-decorated NANs through vascular layers to boost cardiac accumulation, a key limitation in their potential *in vivo* efficacy.

References

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Figures

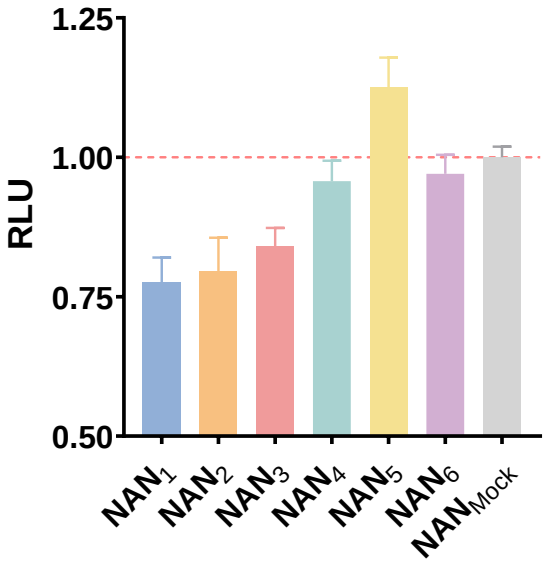

Figure 1. *Activity of 6 different NANs bearing different miR-199a-3p sequences and/or RNA modifications by luciferase reporter assay in HEK293 cells and with a miR-199a-3p sponge. Relative Luciferase Units (RLU) to the non-functional NAN (NAN_{Mock}).*