Cardiac Targeting Peptide-Expressed Exosomes Recovered Ca2+ Release by miR-1 and 133 in Pacing Induced Tachycardia Model.

- Cardiac Specific Delivery of Exosome and Therapeutic effect of miRNA-loaded Exosome -

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Exosome: cell-to-cell communication

- Exosome is important for cell-to-cell communication.
- The effect of stem cell therapy is explained by mainly paracrine effect of extracellular vesicle including exosome.
- Since the immune response does not occur, it has **important advantages as a drug delivery system.**

Cardiac specific delivery of exosome

1. Exosome production

2. Exosome modification

3. Exosome delivery

- Recently, the idea of potentiating exosome is actively developed.
- However, to be used in cardiac disease, **cardiac specific delivery is needing to avoid off-target effect**
Exosome delivery of miRNA

- Functions of miRNA are RNA silencing and post transcriptional regulation of gene expression.
- It is known that these miRNAs are delivered by exosome and are involved in several heart diseases.

1) To develop **cardiac targeting exosome**

2) To evaluated the efficacy of cardiac specific delivery of **cardiac targeting exosome**

3) To develop specific **miRNA loaded exosome**

4) To evaluate whether the specific **miRNA loaded exosome** could effect Ca$^{2+}$ release in pacing induced tachycardia model.
Method
1. How to make cardiac target recombinant plasmid

- CTP-LAMP2B: Membrane Marker

2. Transfection

CTP-LAMP2B plasmid + CD81 labeled with mcherry plasmid

3. Producing CTP-Exo

- Lamp2b
- CTP
4. Make a genetically engineered exosome

- Multiple miRNAs, especially miR 1 and 133, related with calcium activity potentially contribute to electrical remodeling associated with AF.
4. Make a genetically engineered exosome

- For exosome modification, exosomes loaded with miRNAs by electroporation or transfection reagent.
5. Confirmation of delivery efficiency

- **in vitro**
  - HEK: Human kidney cells
  - H9C2: Rat cardiomyocytes

- **in vivo**
  - Mouse tail vein injection

6. Therapeutic effect

- Tachypacing
- HL-1
- Calcium confocal

**METHOD**
Cardiac Specific Delivery of Exosome: CTP - Exo
The successful development of cardiac targeting exosome was analyzed by Western blot and Nanoparticle Tracking Analysis.
Effectively delivery of CTP-Exo *in vitro*

- The delivery efficiency of CTP-Exo was 16% greater than that of Control (CTL)-Exo in cardiac cells.
- However, there was no difference in kidney cells.
Effectively delivery of CTP-Exo in vivo

- Compared with CTL-Exo, the in vivo delivery of exosomes to the heart, increased by 15% relative to that of CTP-Exo.
- However, the delivery of CTP-Exo was not different in other organs.
Therapeutic effect of miRNA-loaded Exosome
Development of miRNA loaded exosome

- miRNA loaded exosome was developed by electroporation and transfection reagent.
- Successfully delivered miRNA via exosomes was confirmed by Western blotting.
miR-1 or 133 loaded Exosome suppressed Ca\textsuperscript{2+} release

- In tachypacing model of HL-1 atrial cardiomyocytes, the exosome loaded with miR-1 or 133 reduced Ca\textsuperscript{2+} wave frequency and recovered Ca\textsuperscript{2+} transient amplitude.
1) Cardiac targeting exosome was successfully developed by genetic modification of CTP-Lamp2b.

2) Cardiac targeting exosome was delivered to cardiac cell and heart more efficiently than conventional exosome.

3) miRNA loaded exosome was successfully developed by electroporation and transfection reagent.

4) In tachypacing model, the exosome loaded with miR-1 or -133 reduced Ca\(^{2+}\) wave frequency and recovered Ca\(^{2+}\) transient amplitude.
Thank you for your attention