Characterization of Trophoblast Derived Exosomes to Engineer an Immunomodulatory Macroencapsulation Device for Cell Therapies

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ABSTRACT

Trophoblasts are cells that populate the human placenta and release extracellular vesicles called exosomes, involved in cell-to-cell communication. Trophoblasts are believed to be involved in site specific immunomodulation and their exosomes may play a role in preventing immunological attack of the fetus despite presenting foreign antigens. Isolation of these exosomes and their encapsulation within hydrogel macroencapsulation matrices could allow for a sustained localized immunomodulation with potential applications in cell therapies such as islet transplantation to treat type 1 diabetes.

METHODOLOGY

The methodology followed in this project was mainly divided in two stages, exosome isolation and exosome characterization.
Our part of the project consisted in studying and identifying the key differences in the proposed exosome isolation methods which intended to isolate JAR and JEG-3 derived extracellular vesicles.
Total exosome isolation reagent kit; chemical isolation, and size exclusion chromatography; physical isolation, were the included and evaluated processes.
The second stage consisted in the characterization of the isolated exosomes with the objective of evaluating the performance and the obtained yield from the isolation processes. Dynamic Light Scattering (DLS) and MicroBCA Protein Assay were the characterization methods used for this purpose.

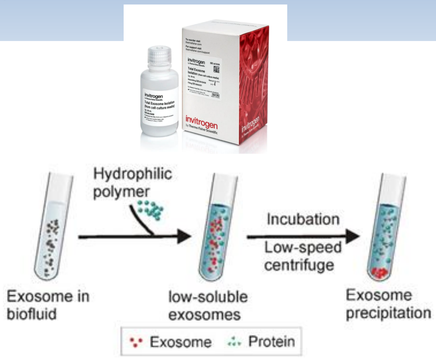

Figure 1:Total Exosome Isolation Reagent Kit and mechanism.
Source: ThermoFisher

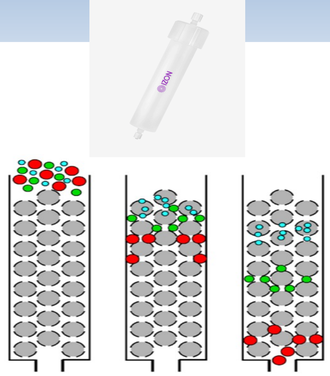

Figure 2:Size exclusion chromatography with qEV Isolation Column and mechanism.
Source: IZON

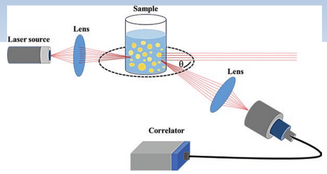

Figure 3: DLS mechanism
Source: Reference 5

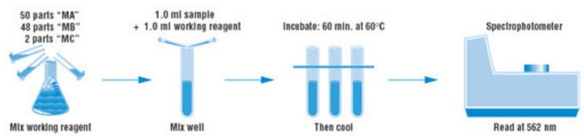

Figure 4: MicroBCA mechanism
Source: ThermoFisher

RESULTS

Following isolation methods proposed, evaluation of derived samples was challenging due to limitations in available working concentrations from the physical means of isolation and therefore a comparison of yield between the two methods did not reach a conclusive result.

CONCLUSION

Insufficient expression on exosomes which fall beneath the detection limits of the DLS.
Impossibility to conclude which elute fraction from the physically isolation sample contains the vesicle of interest.

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FUTURE LINE OF RESEARCH

Investigating about characterization processes which adjust to the exosome concentration of our sample.
NTA (Nanoparticle tracking analysis) is proposed as an adequate method for the characterization of the obtained samples.

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