Regenerative Medicine

By Shyamasri Biswas, PhD

Executive Editor

The Emerging Potentials of Exosome-Based Drug Delivery

Intercellular Communication through Exosome Amplification: Reprogramming of Cells

Exosomes are ultra-small (50-150 nm) special membranous bio-vesicles. They are released from living cells into surrounding body fluids upon fusion of multi-vesicular bodies (MVB) and the plasma membrane [1]. These extracellular vesicles are known to be functional vehicles and carry cell-specific cargos of proteins, lipids, nucleic acids and genetic materials that can be selectively taken up by neighboring or distant cells that are located far from their release points. Subsequently, the recipient cells are reprogrammed through the triggered bioactivities of exosomes [2].

Studies have indicated that the bioactive molecules contained in exosomes could impact target cells by direct stimulation of target cells via surface-bound ligands and transferring of activated receptors to recipient cells. The subsequent epigenetic reprogramming of recipient cells can happen via the delivery of functional proteins, lipids, and RNAs (Figure 1) [2]. As Figure 1 schematically illustrates, the exosome amplification can potentially enable communication of parental cells with specific proximal or distant target cells [2]. In this way, exosomes represent an intercellular communication mode that can be implemented in many cellular processes including immune response, signal transduction and antigen presentation [2].

The regulated formation of exosomes, specific makeup of their cargo and cell-targeting specificity show high potential of exosomes for their use as next generation non-invasive diagnostic biomarkers, as well as therapeutic carriers [1, 2].

Therapeutic Ability of Exosomes

Exosomes' therapeutic and clinical application potentials have attracted a lot of attention, lately. One area of application that is being considered is cancer diagnosis and therapy. Studies have suggested that tumor-derived exosomes or TDEx regulate tumor microenvironment and assist in distant metastasis [1]. Researchers have shown the possibility of employing molecular markers on exosomes or TDEx that can be an

effective method as liquid biopsy indicator for cancer diagnosis and prognostic monitoring [1, 3].

Exosomes are also known for their ability to cross cytoplasmic membrane and the blood/brain barrier. This ability of exosomes can be leveraged for therapeutic delivery of drug molecules. Therefore, a great deal of current research has focused on using the potential of exosomes being a drug carrier due to exosomes' natural material transportation properties, intrinsic long-term circulatory capability, and excellent biocompatibility that are considered suitable to deliver a variety of chemicals, proteins, nucleic acids and gene therapeutic agents [1, 2]. To this end, the modified exosomes were shown to treat pancreatic cancer by delivering siRNA, and the CD47 protein on the exosomes prevented them from being phagocytosed by immune cells. This makes the exosomes more efficient than synthetic analogiliposomes. Researchers utilized the targeting effect of exosomes, and leveraged it to apply drugs such as adriamycin directly to tumor cells that limited the toxicity to normal cells [3].

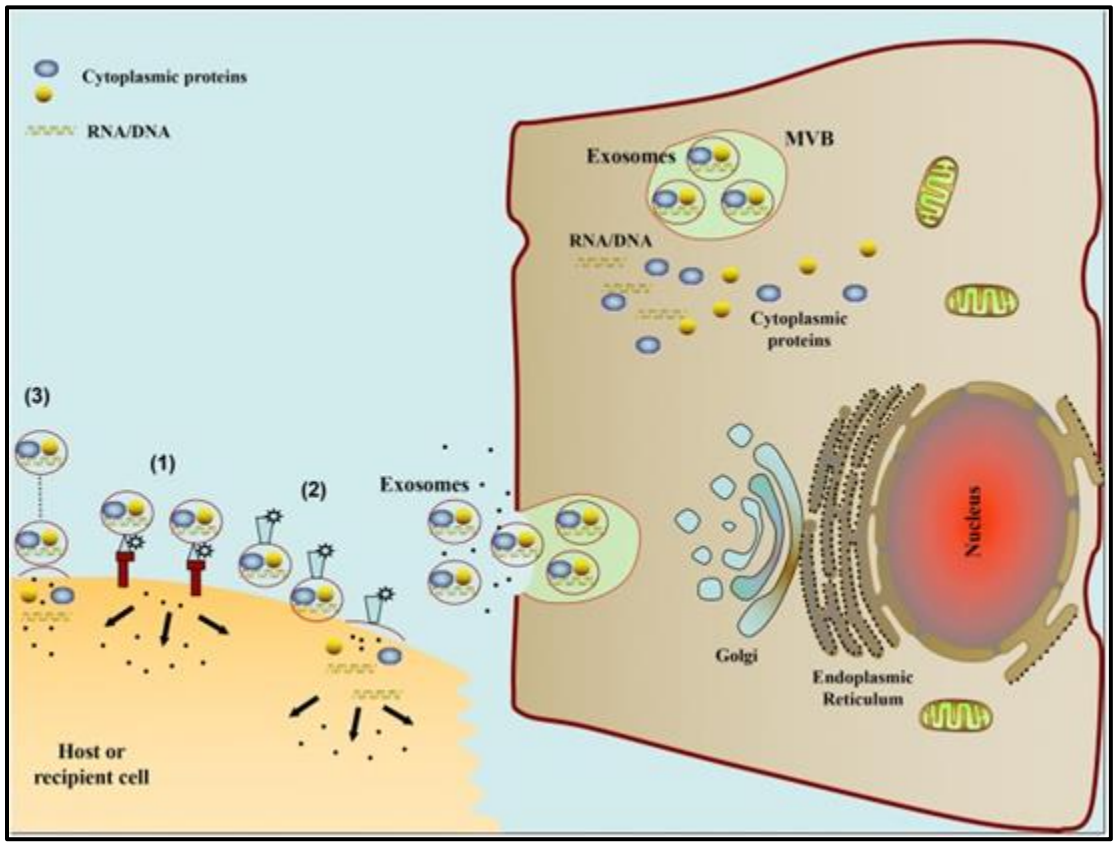

*Figure 1: The schematic illustration showing intercellular communication involved in exosome mediated process. (1) Direct surface-bound ligands by the recipient cells. (2) Transfer of activated receptors to recipient cells. (3) Epigenetic reprogramming of recipient cells [**Source:** Cell Biosci. 2019].*

Further, studies were conducted on the cellular immunity, angiogenesis, and regenerative effects in cell therapy mediated by mesenchymal stem cells (MSCs) and some other cells that were implemented by the exosomes released from these cells [4, 5]. The emerging field of exosome therapy has shown great application potential from oncology to regenerative medicine [1].

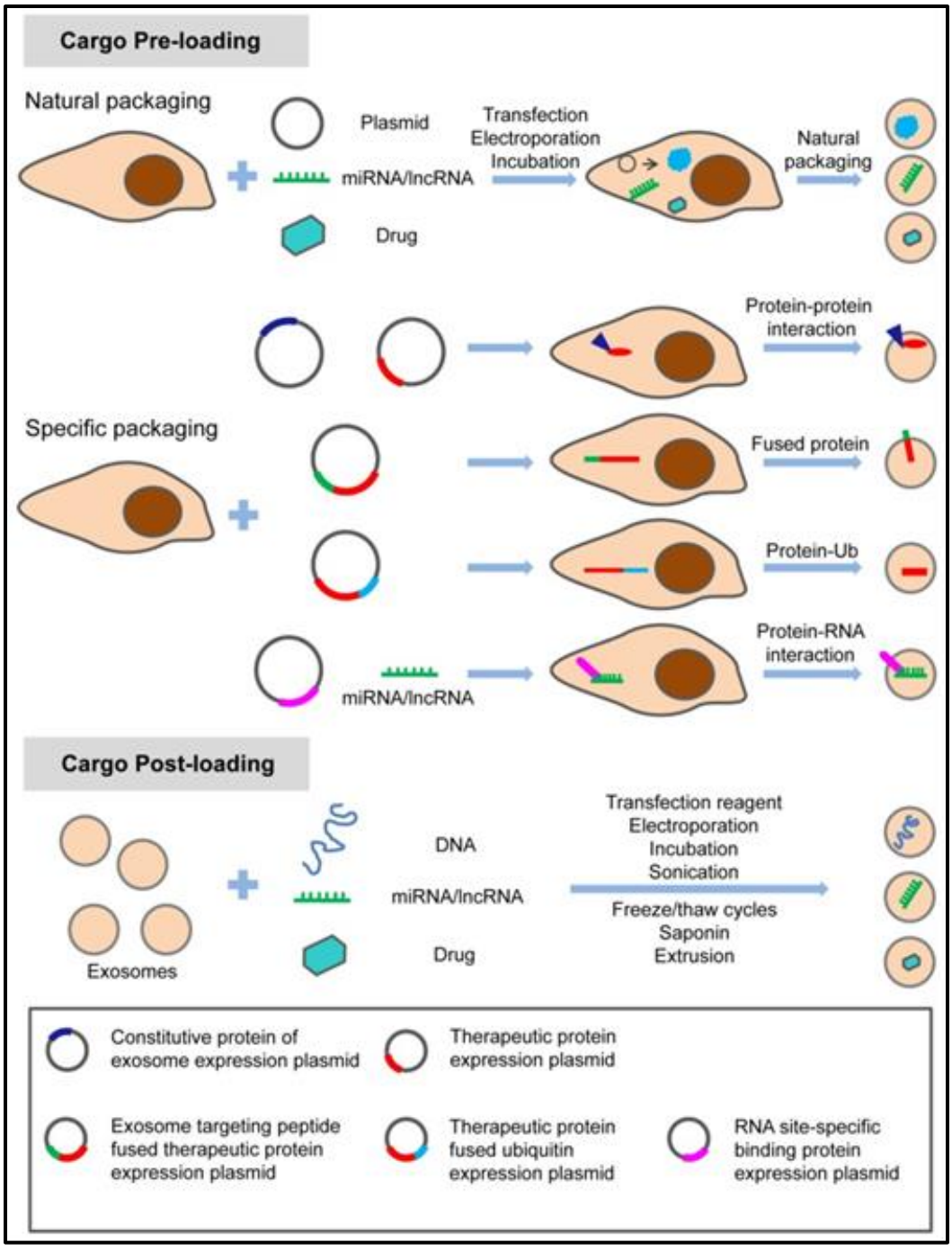

*Figure 2: Various methods that are employed for loading specific proteins, nucleic acids and small molecular drugs into exosomes [**Source:** Theranostics (2019)].*

Exosomes Loaded with Small Molecule Drugs

Exosomes can be employed as carriers of chemotherapy drugs. Researchers have developed methods of loading small molecule drugs into exosomes that include direct mixing, incubation, ultrasonic treatment, and eddy oscillation [1]. In addition to these methods, several other different ways of loading catalase into exosomes were demonstrated *ex-vivo* such as incubation, saponin permeabilization, freeze and thaw, sonication, and extrusion that were employed to treat Parkinson's disease. These methods have been demonstrated for their use to load exosomes with small molecule drugs as well.

The current state-of-the-art employs most of these passive loading methods for small molecule drugs that are loaded into exosomes. However, major drawbacks of these methods and approaches are the resulting loss and degradation of exosomes from multiple purification steps that are used in these methods. Also, a prolonged *in-vitro* treatment and the inherent physicochemical properties of drug molecules can affect the bioactivity and degrade the stability of exosomes. In view of these shortcomings, current research has focused on viable alternative methods that would enable the stability of exosomes.

Figure 1 summarizes the methods for loading the specific treating molecules (proteins, nucleic acids and small chemicals) into exosomes [1].

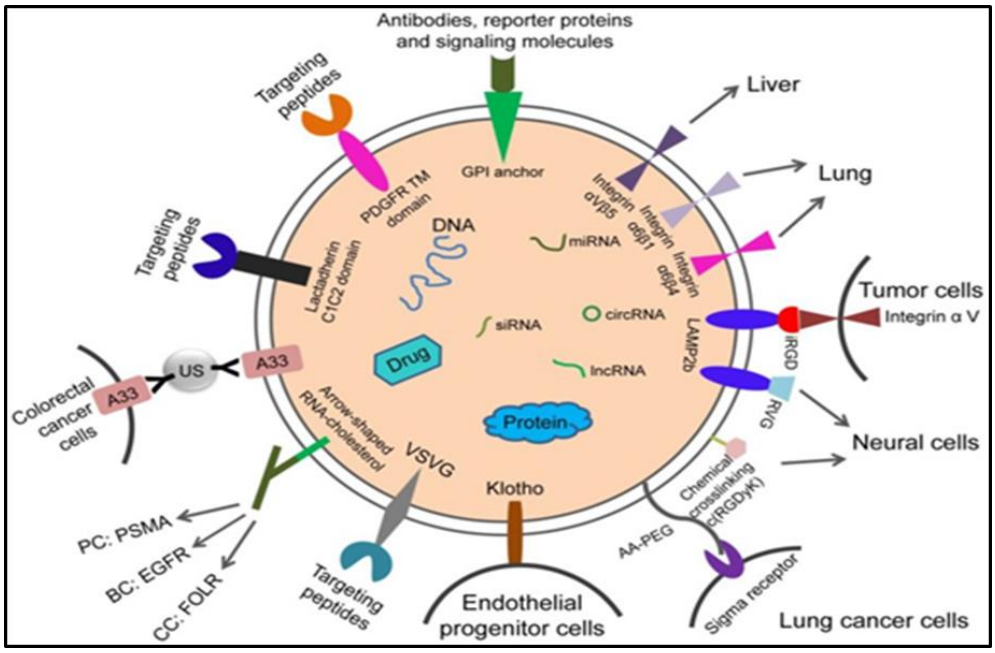

*Figure 3: A schematic presentation showing design strategies for therapeutic exosome targeting (AA-PEG: aminoethylanisamide-polyethylene glycol; GPI: glycosylphos-phatidylinositol; RVG: rabies virus glycoprotein; VSVG: vesicular stomatitis virus glycoprotein; A33Ab-US: the surface-carboxyl superparamagnetic iron oxide nanoparticles coated with A33 antibody, to bind to A33-positive exosomes; iRGD: an α V integrin-specific internalizing peptide; PDGFR TM domain: PDGFR transmembrane domain) [**Source:** Theranostics (2019)].*

Future Design Strategies for Therapeutic Exosome Targeting

Figure 3 shows a number of design strategies that are currently being developed for loading and targeting to realize the therapeutic potential of exosomes [1]. In the application of therapeutic exosomes, future designs will focus on accurately targeting a specific type of cells including tumor cells or a certain type of tissue (e.g., brain tissue) as opposed to broad distribution of exosomes to the liver, kidney and spleen. In view of this, researchers are focusing on to improving the targeting of exosomes through the study of exosome donor cells and the modification of surface molecules on exosomes [1].

Concluding Remarks

Studies have shown that exosomes can play a crucial role in both physiological and pathological processes including the capacity of exosomes to load a wide range of content including lipids, RNAs, and proteins to signal specific recipient cells or tissues. This makes them a promising diagnostic biomarker and therapeutic tool for treatment of cancers and other complex pathologies and offer exciting new possibilities in drug delivery. The early studies indicate the great clinical potential and value of exosomes due to their strong biocompatibility, offering a new frontier in drug delivery. We anticipate a rapid growth in research and developments in the future especially in the area of large scale production of exosomes for clinical use.

References

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