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Review

Biological Functions and Applications of Exosomes in Drug Research

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Abstract: Exosomes have received increasing attention in recent years as an important substance for intercellular communication. Among the plethora of new research is their unique interaction with drugs, which is even more striking. Exosomes play essential roles in disease treatment either as extracellular vesicles to exert biological functions or as drug carriers to deliver therapeutic agents. We summarized the relationship between exosomes and drugs in the disease progression and treatment. Understanding how exosomes interact with drugs and exert their anti-inflammatory and pro-angiogenic effects, alongside a lipid peroxidation inhibitory result in different manners is essential for disease treatment.

Keywords: exosomes; drug-delivery; disease treatment

1. Introduction

During the past decades of cellular research, our understanding of cell communication remains at a stagnant molecular level of molecular communication, and within this time, exosomes have been found to play an important role in cell communication [1]. As substances that can communicate with cells, exosomes play an integral role in many aspects of human health and disease, including development, tissue homeostasis, immunity, neurodegenerative diseases, and cancer through this way [2–5]. In addition, the virus uses an exocrine biogenetic pathway to gather infectious particles and license hosts. The properties of exosomes generate a potential to evolve into therapeutic agents in a variety of disease models [6]. Compared to conventional nanocarriers, exosome-loaded drugs present more advantages. Drug interference through exosomes to achieve the desired therapeutic purpose presents an additional objective for progressive disease treatment in adjacent to drug development. This review highlights the role of exosomes in intercellular communication, targeting tumors and drug interactions, with further clinical significance as therapeutic targets.

2. Brief Introduction of Exosomes

Exosomes as the smallest extracellular vesicles, detected within a wide scope of cellular secretions, are widely distributed in body fluids with a diameter of 40–100 nm. Early secretory endosomes are formed when the plasma membrane of the cell is invaded by exosomes. A large paleosecular body (MVB) forms intracavitary vesicles (ILVs), and later acidification facilitates the maturation of endosomes. Lastly, release ILV as an exosome by fusing with the plasma membrane [7], which can transport rich proteins, lipids, DNA, RNA, and other substances. Now, numerous studies have demonstrated that exosomes are essential during immune responses and angiogenesis, in addition to osteogenesis and tumor-targeted therapy. Exosomes have shown their unique advantages not only as drugs but also as drug carriers and have progressed substantially

for targeted tumor therapy. Many studies have shown that a variety of drugs can modulate the release of exosomes and their contents, which can be modified to effectively treat disease more efficiently (Figure 1).

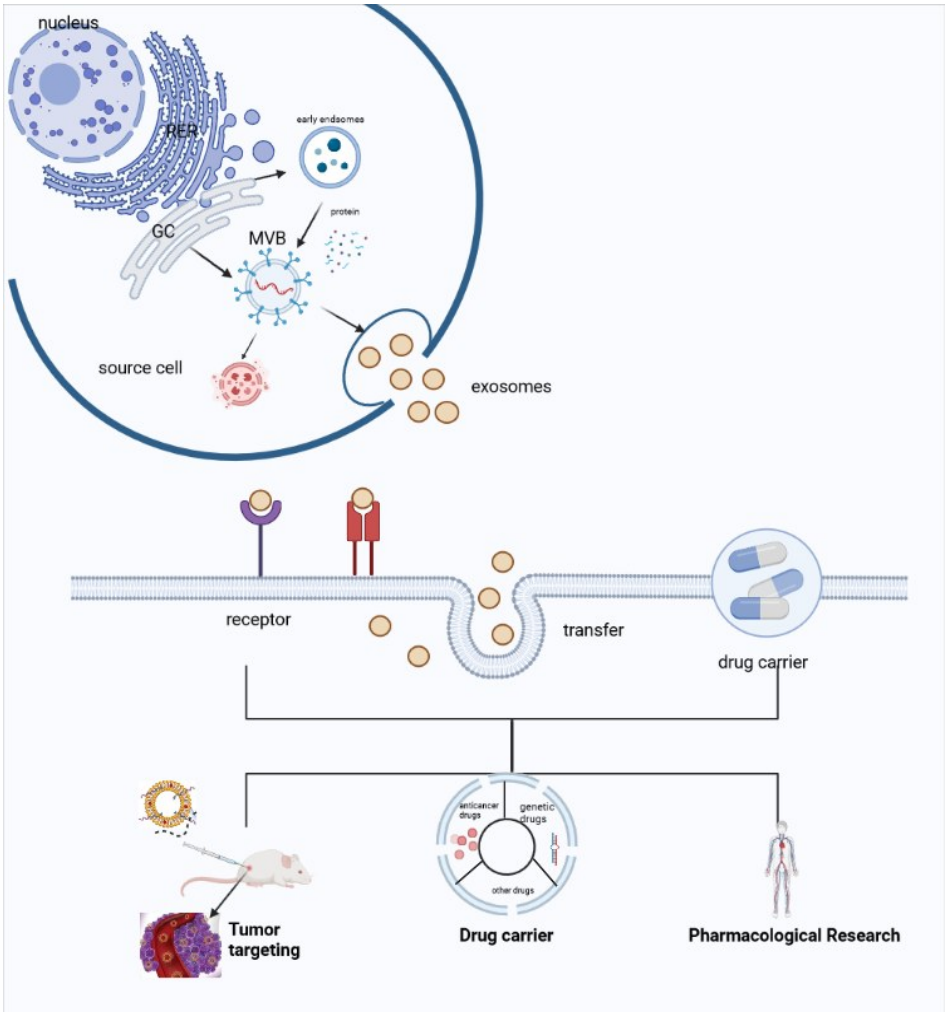

Figure 1. Exosomes are secreted to the outside of cells through endocytosis. They are absorbed by target cells once they have interacted with the extracellular receptors and play corresponding roles as materials for drug loading and physiological activities, as well as tumor management.

3. Therapeutic Potential of Exosomes

At the earliest time, exosomes were considered “garbage bags” of cells, enabling cells to get rid of some useless proteins. However, studies have recently found that the “goods” carried by exosomes have important biological significance. Compared with cell therapy, many diseases have been treated with exosomes since they are more convenient to store and can reduce safety risks.

In the cardiovascular system, microRNA or other contents carried by exosomes can promote angiogenesis and improve the repair of ischemic tissues. It is reported that exosomes derived from stem cells can act on the heart and blood vessels, and are considered to have protective effects against cardiovascular diseases using anti-inflammatory, antioxidant and apoptosis-resistant properties, alongside being able to prevent cardiovascular fibrosis, endothelial formation inhibition, whilst suppressing anti-vascular remodeling [8].

In inflammatory diseases, in terms of the progression of inflammatory bone disease, exosomes play a crucial role. Mechanistically, exosomes are involved in the onset and development of inflammatory bone disease and promote inflammatory osteolysis, in contrast, inhibition of this process is also possible when specific types of exosomes are utilized.

Emerging evidence has implicated that exosome play a therapeutic role in other systemic diseases. Exosomes derived from lung cancer cells can be harnessed in lung cancer treatment, as they can control metastasis, cell proliferation and can induce angiogenesis. Furthermore, they can regulate anti-tumor immunity and drug resistance during lung cancer treatment, which is currently considered to be an important part of liquid biopsy when detecting lung cancer [9]. In melanoma treatment, exosomes derived from natural killer (NK) cells have been reported to create melanoma xenografts from their interactions with melanoma cells, which may offer a potential approach to cancer immunotherapy [10]. In summary, exosomes are used in numerous diseases in multiple biological fields.

We have been studying exosomes secreted by endothelial progenitor cells (EPCs) to investigate the effect that they have in promoting post-infarct angiogenesis or vascular aging. Many studies have further demonstrated the contents of the exosome mediated mechanism, amongst which miRNA and IncRNA have been focused upon as the main research feature. Throughout the distinct contents, exosomes play a repairing role in acute lung injury, acute cardiomyopathy and neurological injury [11–15].

4. Drug Loading into Exosomes

The recent remarkable progress in the field of exocrine nanotechnology has provided an unprecedented opportunity for exocrine therapy in the field of diseases. The unique structure as a natural nanocarrier supports their special physiological and pathological effects.

Drugs such as antibiotics, herbal extracts and other specific targeted drugs can be loaded into the exosomes through incubation, ultrasound, electroporation, and other methods. For example, Sun et al. began to study the manufacturing process of engineered exosomes for the drug delivery system (DDS) in 2018. In 2010, curcumin was successfully encapsulated in exosomes for the first time. By packing curcumin inside exosomes, curcumin became more soluble and was able to circulate for longer, additionally being capable of maintaining drug therapeutic activity and delivering more medicine to the brain. They found that when curcumin is encapsulated in exosomes, its anti-inflammatory activity is enhanced [16–18]. Similarly, Tian et al loaded doxorubicin (Dox) into exosomes for cancer treatment, further confirming the role of exosomes as carriers for treating disease [19].

Compared with traditional drug carriers, exosomes exhibit their unique advantages, such as: (1) exosomes exhibit higher stability in blood, which enables them to carry out long-distance transmission in vivo under physiological and pathological conditions [20]. (2) Exosome carriers combine the advantages of cell-based drug delivery with nanotechnology to mutually induce potent drug efficacy. (3) Exosomes exhibit very low immunogenicity compared to systems where viruses are used to deliver drugs.

In view of this ability and characteristics, the application of exosome-delivered drugs has been operated to treat a wide range of illnesses, it has been shown that BCR-ABL siRNA loaded exosomes target chronic myeloid leukemia cells and inhibit their growth [21]. In addition, successfully coloaded imatinib and siRNA into the exosomes for cancer treatments, such as synergistic chemotherapy, proves the feasibility of this combined treatment method in regards to microstructure for use in heterogeneous therapy [22]. Researchers introduced two types of exogenous siRNA targeting RAD51 and RAD52 into exosomes derived from HeLa cells, which caused massive death of recipient tumor cells. The role of the exosome-based silencing gene in cancer therapy has also been verified [23]. Because of the promoting effect of miR-150 on VEGF expression, exosomes were used to transport siRNA targeting miR-150 in tumor cells. Studies have shown that this approach upregulates VEGF levels and attenuates angiogenesis in mice [24]. Exosome-mediated siRNA has also been applied to Alzheimer's treatment. Studies have shown that BACE1 expression at both mRNA and protein levels were strongly down-regulated in wild-type mice [25], proving the therapeutic potential of exosome-mediated delivery of siRNA.

In the process of building exosome-based nanoplatforms for therapeutics and diagnostics, a comprehensive explanation referring to the foundations of advanced nanotechnology, as well as the combination of the inherent advantages of exosomes as cell-derived vectors with advanced design methodologies in traditional medicine, will altogether unlock their innate power.

5. Drugs Interact with Exosomes

Exosomes may be exploited to treat diseases by interacting with other drugs in the human body. Drugs can affect the release of exosomes, or exosomes can mediate the interaction between autocrine cells and paracrine cells to change the distal and local microenvironment, thereby affecting the effect of drugs. There are few studies on the interaction between them, which may be a new direction for drug research.

Drugs can achieve a greater efficacy by acting on exosomes. Studies have pointed out that exosomes derived from drug-treated HepG2 cells exhibited potent anti-proliferative activity against induced immunogenic and HSP-specific NK cell responses in CFPAC-1 cells [26]. Some studies have shown that Manumycin A can inhibit the production and secretion of exosomes [27]. Additionally, a considerable proportion of traditional Chinese medicine (TCM) can affect the endocrine system of the body. Purple Perilla seed extract, for example, can inhibit endothelial cell secretion, thereby inhibiting its combined products of clothing factor II, VA and XA which can result in thrombosis. This extract can consequently protect the endothelial cells from vasculitis caused by infection and blood clots [27].

Exosomes are involved in the mechanism of drug action. Crenshaw et al. found that ethanol could affect the generation of microglia exosomes and changes their protein contents [28]. Recently, it has been found that ethanol increases the release of exosomes from primary cultured mouse astrocytes and the content of inflammation-related proteins (NF-κB-p65, NLRP3) and miRNA (such as miR-146a, miR-182) in a Toll-like receptor 4-dependent manner. Functional analysis of miRNA confirmed that the modified version was able to regulate the expression of genes that induced an inflammatory response [29].

Many drugs can promote each other when combined with exosomes. The synergized effect of melatonin and exosomes can together inhibit apoptosis and oxidative stress, which result in neuroprotective properties. It is possible to reduce vascular calcification and aging by using exosomes melatonin therapy, as well as gaining other beneficial effects. Datta et al found that in castrated resistant prostate cancer cells, Manumycin A can inhibit the production and secretion of exosomes by inhibiting the RAS/RASF/ERK1/2 signal pathway and hnRNP H1 protein channel [30]. A significant role of exosomes in the motility of cancer cells can be attributed to their ability to promote transient polarization states as well as adhesion and assembly of cancer cells. Recently, through screening the drug library approved by FDA, we identified the oral antimicrobial sulfa isoxazole (SFX) as a specific inhibitor of breast cancer cell biogenesis and extracellular vesicles (SEVs) secretion, which can effectively inhibit the growth and metastasis of breast cancer without obvious toxicity. Therefore, it can be inferred that inhibitors preventing tumor exosome secretion can inhibit both cancer progression and metastasis, as well as initiation, by eliminating exosomes secreted in situ from tumor areas, thus providing therapeutic benefits for cancer patients.

As mentioned above, studying the interaction between exosomes and drugs can maximize the potential of the new discovery of exosomes in order to promote the optimal effect of drugs, whilst minimizing their adverse reactions, in addition to seeking better therapeutic effects.

6. Conclusion

The rapid development of basic sciences related to multifunctional exosomes has furthered our understanding of the biology and pharmacology of exosomes alongside their interdependence with one another. The progress of exosome technology in various fields of biology, pharmacy, oncology and material science will promisingly benefit our health in the future by maximizing treatment effects while concurrently minimizing side effects. To sum up, we focus on the application of exosomes in disease treatment based on their consequential interactions as drug delivery vectors and as drugs. Although a great deal of research on exosomes has focused on the physiological and pathologically regulated biochemical properties of the original exosomes, the biology of exosomes has not yet been explored. In addition, our current understanding of how each functional unit on exosomes gives biological function is still limited. Since nanomedicines have developed in relation to exosomes over the last 20 years, a deeper exploration of biochemical principles is needed to uncover how exosomes are formed, with further clarification towards the physiological mechanisms that regulate the behavior of biological exosomes within their internal environment. In addition, a major barrier to further commercialization has been the time-consuming, inefficient, and expensive nature of currently available isolation methods and clinical transformation of exosomes. Understanding the optimal

method for developing exocrine therapy, when using the unique cell biology of exosomes, is a key problem to be solved.

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