International Journal of Nanomedicine

Dovepress
Taylor & Francis Group

Open Access Full Text Article

REVIEW

# Exosomes as Pivotal Mediators of Tumor-Immune Communication: Implications for Immunotherapy and Liquid Biopsy

Menglin Wei[1,2,*], Dongli Wang[1,2,*], Wenrong Xu[1,2], Xueyan Zang[1], Jiajia Jiang[1,2]

[1]Aoyang Institute of Cancer, Center of Clinical Laboratory, Affiliated Aoyang Hospital of Jiangsu University, Suzhou, Jiangsu, 215600, People's Republic of China; [2]Zhenjiang Key Laboratory of High Technology Research on Exosomes Foundation and Transformation Application, Jiangsu Key Laboratory of Medical Science and Laboratory Medicine, School of Medicine, Jiangsu University, Zhenjiang, Jiangsu, 212013, People's Republic of China

*These authors contributed equally to this work

Correspondence: Jiajia Jiang; Xueyan Zang, Email jiangjiajia_2001@163.com; Zangxy726@163.com



**Abstract:** Exosomes are membrane-bound vesicles secreted by almost all types of cells, including but not limited to immune cells, neurons, epithelial cells, and cancer cells. Exosomes carry DNA, RNA, lipids, metabolites, as well as cytoplasmic and cell surface proteins. Their role in cancer progression is dynamic and is related to the type of cancer, genetics, and stage. At the same time, exosomes have attracted widespread attention as key mediators of intercellular communication in the tumor immune microenvironment (TME). This comprehensive review delineates the pleiotropic roles of exosomes in tumor immunobiology, emphasizing their bimodal capacity to either foster immunosuppression or potentiate antitumor immunity. We systematically synthesize recent advancements in exosome-based immunotherapeutic regimens, with particular emphasis on their synergistic efficacy when integrated with established modalities, namely immune checkpoint blockade and adoptive cellular therapy. Furthermore, we critically appraise emergent technologies for exosome isolation and characterization, underscoring their transformative implications for liquid biopsy platforms in real-time immune surveillance and the development of predictive biomarkers. This review posits exosome-centric strategies as a paradigm-shifting frontier in precision immuno-oncology, furnishing innovative remedies for recalcitrant therapeutic hurdles and propelling the advancement of personalized oncology care.

**Keywords:** exosome, tumor immune microenvironment, cancer immunotherapy, biomarker, liquid biopsy

## Introduction

Exosomes are a type of membrane vesicle with a diameter of 40–100 nm, actively secreted by cells through the endocytosis-fusion-exocytosis process, and are widely found in bodily fluids such as blood, urine, and saliva.[1,2] Since exosomes were discovered in 1983, research has continued to deepen, and they are currently developing rapidly in clinical therapy. These nanosized carriers are abundant in biofluids, such as blood and saliva, and encapsulate diverse biomolecules, including proteins, nucleic acids, and lipids, reflecting their cellular origin and pathophysiological states. Growing evidence underscores their pleiotropic functions in intercellular communication through the horizontal transfer of bioactive molecules, playing critical roles in tumor progression by mediating immune evasion, metastatic niche formation, and therapeutic resistance.[3] The tumor microenvironment (TME) is a dynamically remodeled ecosystem composed of malignant cells, immunosuppressive components (eg, regulatory T cells, M2 macrophages), cancer-associated fibroblasts (CAFs), and a dysregulated extracellular matrix, collectively forming a self-reinforcing niche that facilitates tumor progression.[4] Accumulating studies have highlighted the powerful role of exosome within the TME, where they mediate information exchange between tumor cells and immune cells, which is a new direction for cancer immunotherapy in the future. However, current research on exosomes from various cell sources in the tumor immune microenvironment is relatively scattered and has not been systematically explained. In addition, the relationship between exosomes and immunotherapy, as an important direction for future immunological

https://doi.org/10.2147/IJN.S583756
Received: 23 November 2025
Accepted: 5 February 2026
Published: 24 February 2026

development, still has gaps that are worth exploring in depth. This article innovatively explores the mechanism of action of exosomes in the tumor immune microenvironment, as well as the latest technological advances in exosome isolation and identification and their prospects in clinical application. Finally, we will focus on the application value of exosomes in tumor immunotherapy. This will provide direction for future personalized cancer treatments.

## In TME, Tumor Cells and Immune Cells Exchange Information Through Exosomes

Exosomes contribute to angiogenesis, tumor growth, metastasis, and stromal cell activation.[5–7] Cancer cells not only release increased quantities of exosomes but also induce alterations in their local environment.[8] These contextual changes, in turn, influence the molecular composition of exosomal cargo. Therefore, a dynamic interplay exists between exosomes and the TME, collectively impacting cancer progression. Significant advances have recently been made in understanding how exosomes regulate tumor immunity. With the clinical approval of cellular immunotherapies and immune checkpoint inhibitors, immunotherapy has entered a phase of rapid development.[9] Tumor-derived exosomes modulate both innate and adaptive immune responses and can influence the efficacy of immunotherapies by regulating immune cell functions. Conversely, exosomes derived from immune cells also affect tumor behavior. Consequently, targeting exosomes represents a promising strategy for future therapies.[10,11] Based on the heterogeneity of exosome sources, we will explore how tumor-derived exosomes, immune cell-derived exosomes, and exosomes from other cell types shape the tumor microenvironment through signaling communication (Figures 1, 2 and Table 1).

### Tumor-Derived Exosomes Modulate Immune Cell Function and Shape the Immune Microenvironment

Tumor-derived exosomes carry diverse bioactive molecules and play a key role in regulating the tumor microenvironment, particularly through the modulation of immune cell activity (Figure 1A).

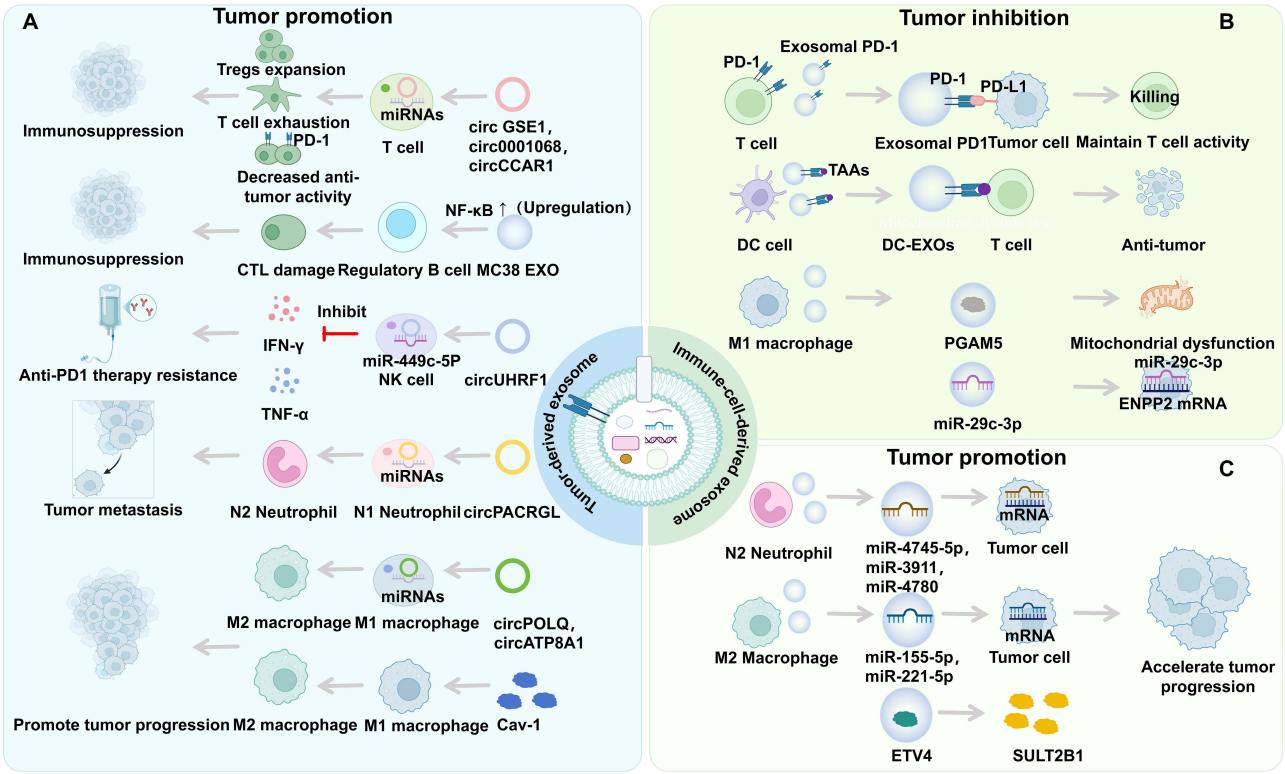


**Figure 1** Exosomes act as important messengers for communication between immune cells and tumors ((**A**) The role of tumor-derived exosomes in promoting tumor progression, (**B**) The role of immune cell-derived exosomes in promoting tumor inhibition, (**C**) The role of immune cell-derived exosomes in promoting tumor progression).

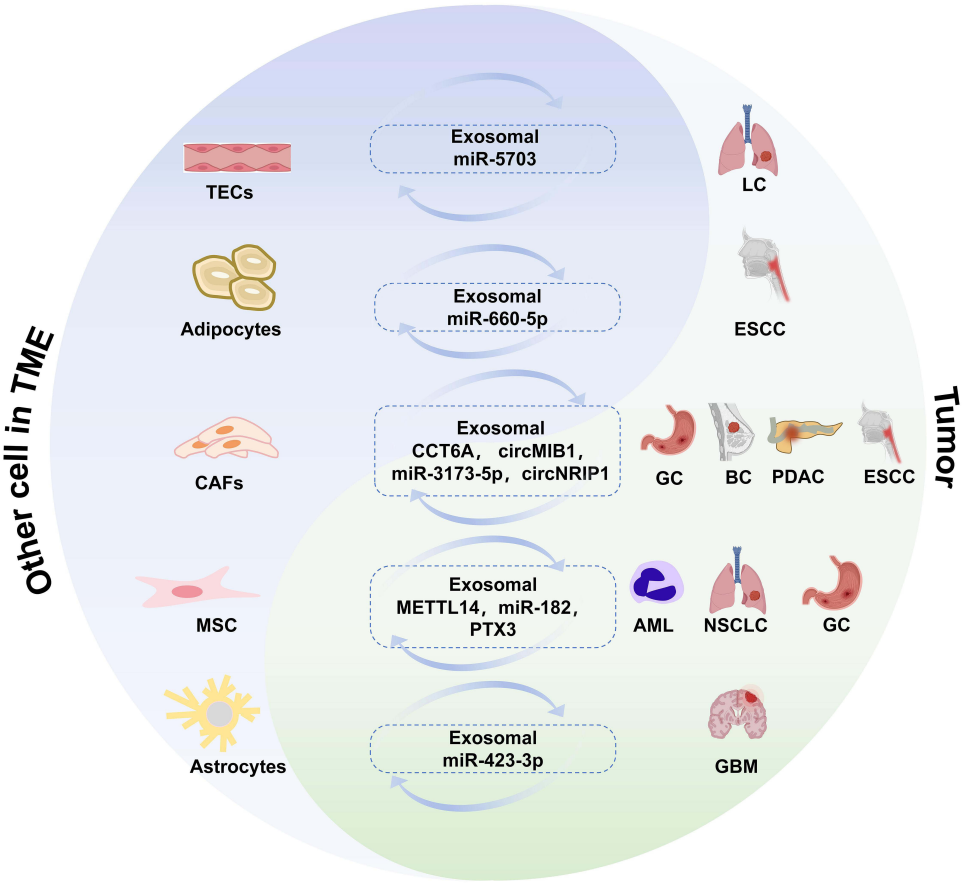


**Figure 2** Other cells (endothelial cells, stromal cells, etc.) communicate with tumor cells in the TME through exosomes (**A**) (TECs) Exosomes derived from TECs, (**B**) (Adipocytes) Exosomes derived from adipocytes, (**C**) (CAFs) Exosomes derived from CAFs, (**D**) (MSC) Exosomes derived from MSC, (**E**) (Astrocytes) Exosomes derived from astrocytes).

The heterogeneity of exosomal contents leads to different immune mechanisms coexisting for the same immune cells.

T lymphocytes include helper T cells (CD4+ T), regulatory T cells (Treg), cytotoxic T cells (CD8+ T), among others. For instance, exosomal circGSE1 from hepatocellular carcinoma (HCC) promotes immunosuppression and tumor progression by sponging miR-324-5p, activating the TGFBR1/SMAD3 pathway and upregulating FOXP3 to expand Tregs. This dual functionality identifies circGSE1 as a promising therapeutic target in HCC immunotherapy.[12] CircGSE1 is expected to serve as a tumor biomarker for multi-cancer screening. In ovarian cancer, exosomal circ-0001068 reshapes the T cell signaling network through the ceRNA mechanism by forming the "circ-0001068-miR-28-5p-PD1" axis, driving T cell exhaustion and immune evasion. This regulatory axis is a direction for immunotherapy, but the mechanism observed in mouse peritoneal models needs to be validated in human T cells to avoid clinical translation failures caused by species specificity.[14] Similarly, the exosomal circCCAR1/miR-127-5p/WTAP positive feedback loop and the circCCAR1-mediated dual-axis synergistic pathway that stabilizes PD-1 protein and enhances PD-L1 transcription collectively lead to CD8 T cell dysfunction and resistance to anti-PD1 therapy.[16]

The impact of exosomes on B cell-mediated anti-tumor immunity remains less understood. Zhang et al demonstrated that exosomes from murine colorectal cancer cell line MC38 (MC38-EXO) suppress B cell activity and promote their conversion into regulatory B cells (Bregs).[19] Then the Chronic lymphocytic leukemia (CLL)-derived exosomes inhibit hematopoietic progenitor proliferation and impair monocyte-derived fibroblast support. These exosomes contribute to B cell depletion and disease progression.[13]

Exosomal circUHRF1, mainly secreted by HCC cells, is associated with reduced NK cell proportion and impaired tumor infiltration.[21] In gastric cancer (GC), exosomal miR-552-5p suppresses NK cell activity via the PD-1/PD-L1 axis and promotes epithelial-mesenchymal transition (EMT). This underscores its role in GC progression and immune


https://doi.org/10.2147/IJN.S583756

**Table 1** The Role of Exosomes of Tumor Cells and Immune Cells in the Tumor Microenvironment

| Parent Cell | Target Cell | Content | Signaling Pathway | Functions | References |
|---|---|---|---|---|---|
| HCC cell | Treg cell | circ-GSE1 | circ-GSE1/miR-324-5p/TGFBR1/SMAD3 | Induce Treg cell expansion, promote immune evasion | [12] |
| CLL cell | T cell | | CD14/CD16/PD-1/CD160 | Reduce T-cell-mediated anti-CLL activity | [13] |
| OC cell | T cell | circ-0001068 | circ-0001068/miR-28-5p/PD1 | Increase the expression of PD1, deplete T cells | [14] |
| MDA-MB-231 cell | $CD4^+$T cell | | | Enhance $CD4^+$T cell activity | [15] |
| HCC cell | $CD8^+$T cell | circCCAR1 | circCCAR1/miR-127-5p/WTAP | Promote $CD8^+$T cell dysfunction | [16] |
| ccRCC cell | Treg cell, $CD8^+$T cell | PD-L2 | PD-1/TDE-PD-L2 | Increase the proportion of Treg cells, decrease the proportion of $CD8^+$T cells | [17] |
| HCC cell | $CD8^+$T cell, M1 macrophage | circARSB | TKI/AAV/circARSB /IFN-β | Promote $CD8^+$ T cell infiltration and M1 macrophage polarization | [18] |
| MC38 cell | B cell | | NF-κB/IL-10/TGFβ/IFNγ/TNFα/Granzyme B | Inhibit B cell activity, obtain Breg signature | [19] |
| CLL cell | CLL B cell | | | Eliminate B cells | [13] |
| CLL cell | B cell | S100A9 | S100A9/CNF-κB | Promote the progression of the disease | [20] |
| HCC cell | NK cell | circUHRF1 | circUHRF1/miR-449c-5p/TIM-3 | Reduce NK cell ratio and NK cell tumor infiltration | [21] |
| GC cell | NK cell | miR-552-5p | miR-552-5p/PD-1/PD-L1 | Inhibit NK cell activity | [22] |
| TSCC cell | NK cell | SNHG26 | HLA-DRA/STAT5 and TGFB1/Smad2 | Inhibit NK cell activity | [23] |
| CRC cell | Neutrophil | circPACRGL | circPACRGL/miR-142-3p/miR-506-3p-TGF-β1 | Promote neutrophil N2 polarization | [24] |
| GC cell | Neutrophil | HMGB1 | HMGB1/TLR4/NF-κB | Promote neutrophil N2 polarization | [25] |
| CRC cell | Macrophage | circPOLQ | circPOLQ/IL-10/STAT3 | Promote macrophage M2 polarization | [26] |
| GC cell | Macrophage | circATP8A1 | circATP8A1/miR-1-3p/STAT6 | Promote macrophage M2 polarization | [27] |
| BC cell | Macrophage | Cav-1 | Cav-1/PTEN/CCL2/VEGF-A | Promote macrophage M2 polarization | [28] |
| NSCLC cell | Macrophage | Hsa_circ_0003026 | Hsa_circ_0003026/Hsa-miR-1183/XRN2 | Promote macrophage M2 polarization | [29] |
| GC cell | Macrophage | BGN | BGN/CXCL10/JAK/STAT1 | Promote macrophage M2 polarization | [30] |
| PAC cell | Macrophage | miR-34a | MiR-34a/cytokine signaling 3 | Inhibit macrophage M2 polarization | [31] |
| CRC cell | Macrophage | HSP90B1 | HSP90B1/PMN | Promote macrophage M2 polarization | [32] |

https://doi.org/10.2147/IJN.S583756

| | | | | | |
|---|---|---|---|---|---|
| CC cell | Macrophage | circ_0020095 | circ_0020095/IGF2BP1/IRAK1 | Promote macrophage M2 polarization | [33] |
| RCC cell | Macrophage | miR-222-3p | miR-222-3p/Zeb1 | Promote macrophage M2 polarization | [34] |
| CRC cell | Macrophage | miR-92a-3p | miR-92a-3p/MAPK/ERK/EID2B | Promote macrophage M2 polarization | [35] |
| GBM cell | Macrophage | Rac1 | RAC1/AKT/NRF2 | Promote macrophage M2 polarization | [36] |
| OC cell | Macrophage | miR-205 | miR-205/PI3K/AKT/MTOR | Promote macrophage M2 polarization | [37] |
| DLBCL cell | Macrophage | NSUN2 | NSUN2/YBX1/PDL1 | Promote macrophage M2 polarization | [38] |
| HCC cell | Macrophage | SNORD5 | SNORD5/JAK2/STAT6 | Promote macrophage M2 polarization | [39] |
| T cell | Tumor cell | PD-1 | PD-1/PD-L1 | Enhance Treg cell activity | [40] |
| DC cell | T cell | TAAs, MHC-I/II, CD80, CD86 | | Present antigens to T cells, activate anti-tumor responses | [41] |
| NK cell | Tumor cell | Perforin, granzymes | | Inhibit tumor progression | [42] |
| N2 neutrophil | Tumor cell | miR-4745-5p/3911 | miR-4745-5p/3911/SLIT2 | Promote gastric cancer metastasis | [25] |
| M1 macrophage | Tumor cell | PGAM5 | | Induce mitochondrial dysfunction | [43] |
| M1 macrophage | Tumor cell | miR-29c-3p | miR-29c-3p /ENPP2 | Inhibit the invasion of cancer cells | [44] |
| M2 macrophage | Tumor cell | ETV4 | ETV4/SULT2B1 | Induce Growth, Glycolysis and Stemness in Hepatocellular Carcinoma | [45] |
| M2 macrophage | Tumor cell | mir-155-5p, mir-221-5p | miR-155-5p/miR-221-5p/E2F2 | Promote angiogenesis | [46] |
| M2 macrophage | Tumor cell | | | Promote the proliferation, migration and tubelization of HUVECs | [47] |
| M2 macrophage | Tumor cell | MDH1 | MDH1/Hippo/YAP | Promote cancer progression | [48] |
| M2 macrophage | Tumor cell | miR −3681-3p | miR −3681-3p/MLH1 | Increase cisplatin (DPP) resistance | [49] |
| M2 macrophage | Tumor cell | hsa-circ-0000326 | YY1/hsa-circ-0000326/miR-338-3p | Promote cancer progression | [50] |
| M2 macrophage | Tumor cell | miR-194 | miR-194/PTEN | Increase cisplatin (DPP) resistance | [51] |
| M2 macrophage | Tumor cell | NEAT1 | NEAT1/KLF5/Galectin-3 | Promotes immune evasion | [52] |
| M2 macrophage | Tumor cell | circ_0088494 | circ_0088494/KMT2D/STEAP3 | Inhibit ferroptosis | [53] |

https://doi.org/10.2147/IJN.S583756

evasion.[22] Exosomes derived from tongue squamous cell carcinoma containing SNHG26 mediate the immunosuppressive effects on NK cells through the HLA-DRA/STAT5 and TGFB1/Smad2 signaling pathways. It is worth mentioning that the clinical translation of SNHG26 still needs to be further validated in larger, independent patient cohorts. In addition, it is difficult for clinical tongue cancer patients to receive repeated intratumoral injections, so the development of systemic administration, such as lipid nanocarriers targeting exosomes, is needed.[23]

Tumor-derived exosomes can promote N2 neutrophil polarization. Exosomal circPACRGL derived from colorectal cancer (CRC) regulates neutrophil polarization via the miR-142-3p/miR-506-3p-TGF-β1 axis and facilitates CRC progression.[24] Additionally, exosomes from N2 polarized tumor-associated neutrophils (TANs) transfer miR-4745-5p/391 into gastric cancer cells, downregulating SLIT2 and promoting metastasis. In a feedback loop, gastric cancer cell-derived exosomal HMGB1 further reinforces N2 neutrophil polarization.[25] Targeting N2 polarization pathway, such as the NF-κB pathway, may therefore inhibit cancer progression.

Tumor cell-derived exosomes are also implicated in M2 macrophage polarization. In CRC, exosomal circPOLQ promotes metastatic nodule formation by enhancing M2 polarization.[26] Similarly, exosomal circATP8A1 from gastric cancer induces M2 macrophage polarization through the circATP8A1 / miR-1-3p/STAT6 axis. If circATP8A1 inhibitors are to be developed in the future, the issue of "targeted delivery" needs to be addressed — clinical gastric cancer patients cannot be treated via "intratumoral injection," so a systemic delivery-targeted vehicle needs to be developed.[27] In breast cancer (BC), exosomal Caveolin-1 (Cav-1) facilitates M2 polarization and contributes to tumor metastasis.[28]

Tumor-derived exosomes convey key information based on the heterogeneity of their contents, thereby playing various roles such as T cell exhaustion, tumor metastasis, and immune evasion.

Interestingly, tumor-derived exosomes are not only significant in tumor progression but can also have therapeutic effects in some immune-related diseases. Therefore, tumor-derived exosomes are not entirely harmful. For instance, exosomes derived from B16-F10 melanoma cells can regulate IL-17 signaling through miRNAs, thereby exerting therapeutic effects on psoriasis.[54]

Tumor-derived exosomes drive immune escape via immunosuppressive effects, offering therapeutic opportunities like blocking exosome activity or developing exosome-based vaccines. Yet their clinical application as biomarkers is hindered by limited validation data, inconsistent isolation methods, and lack of combined diagnostic approaches. Addressing this demands basic research breakthroughs and clinical translation systems, supported by interdisciplinary multi-center collaborations to advance standardization and low-cost technologies for precision medicine.

Based on the diverse roles of tumor-derived exosomes (TDEs), Li et al developed an integrated peptide-graphene sensing system for dual-mode detection of TDEs, which uses fluorescently labeled CD63-binding peptide CP05 and graphene oxide (GO) to achieve selective detection via fluorescence and resonance light scattering.[55] It provides a potential direction for the future personalized detection and screening of exosomes.

## The Dual Role of Immune Cell-Derived Exosome

Exosomes from immune cells play complex and context-dependent roles within the TME (Figure 1B and C). The dual role of exosome immune effects is affected by the heterogeneity of exosomes, including their composition and origin.

T cell-derived exosomes participate in intercellular signaling and anti-tumor immunity. For example, exosomal PD-1 from T cells can bind PD-L1 and enhance the activity of cytotoxic T cells.[40] Dendritic cell-derived exosomes enhance pMHC loading via the IFN-γ-JAK-STAT1 pathway, with cross-presentation by antigen-presenting cells as a key amplification step. Exosomes achieve effector functions through T cell activation and proliferation pathways and optimize therapeutic efficacy by regulating the immune microenvironment.[41,56,57]

NK cell-derived exosomes (NK-Exos) contain cytotoxic molecules such as perforin, granzymes, and microRNAs, which can be directly delivered to the cancer cells to induce apoptosis.[42]

Neutrophil-derived exosomes exhibit phenotype-specific functions. While the role of N1 neutrophil-derived exosomes remains underexplored, it is hypothesized that given the ability of N1 neutrophils to secrete antitumor cytokines such as CCL3, CXCL9 and CXCL10, their exosomes may similarly enhance anti-tumor immunity.[58] N2-TANs-derived exosomal miR-4745-5p/3911 inhibits the progression of gastric cancer in vivo (by subcutaneously injecting GC-803 cells into the backs of nude mice to establish a subcutaneous tumor model) and in vitro through the regulation of SLIT2.[25]

M1 macrophage-derived exosomes exhibit anti-tumor properties. In AML, M1-exo-derived PGAM5 is able to induce mitochondrial dysfunction and reduce inflammatory infiltration.[43] Similarly, M1-exos carrying miR-29c-3p inhibit melanoma invasion by targeting ENPP2.[44] Conversely, macrophage-derived exosomes (M2-exos) often promote cancer progression.[59–61] In HCC, M2-exo-transferred ETV4 promotes tumor proliferation and glycolysis via interaction with SULT2B1.[45] In pancreatic ductal adenocarcinoma (PDAC), M2-exo-derived miR-155-5p and miR-221-5p promote angiogenesis and growth by targeting E2F2.[46]

Thus, immune cell-derived exosomes play a dual role in TME. Anti-tumor exosomes (eg, antigen-loaded DC-Exos) may be harnessed as vaccines to stimulate immune response. Inhibition of protumor exosome secretion or function (eg, PD-L1-bearing TAM-Exos PD-L1-bearing TAM-EXOs) represents targets for intervention to reverse immunosuppression (Figure 1).

In summary, it can be seen that the communication of information between tumor-derived exosomes and immune cells, by carrying various contents, inhibits the ability of immune cells to perform anti-tumor functions (such as regulating the M2 polarization of macrophages and neutrophils), thereby promoting disease progression. Exosomes derived from immune cells play a dual role in both immune enhancement and immune suppression. T cells, DC cells, and M1-type macrophages play direct or indirect roles in the antitumor process. Exosomes derived from these immune cells can also inhibit cancer progression by regulating their contents. Tumor-associated immune cells, such as N2 neutrophils and M2 macrophages, carry RNAs, proteins, and other pro-cancer substances that accelerate tumor progression.

## In the TME, Other Cells Communicate with Tumor Cells Through Messenger Exosomes

The tumor microenvironment (TME) is composed of various cells and components, making the exchange of information between tumor cells and other cells an inevitable research direction (Figure 2).

Tumor-associated endothelial cells (TECs) are the core components oftumor neovasculature and proliferate abnormally in response to tumor-derived signals. Studies have found that miR-5703 is upregulated in lung cancer cells and patient-derived exosomes. It acts as an oncogene in lung cancer and can promote angiogenesis in TECs.[62]

Cancer-associated adipocytes (CAAs) have been found to exchange cytokines and lipids with tumor cells, leading to metabolic reprogramming and the acquisition of pro-inflammatory and invasive phenotypes.[63] For instance, plasma exosomes from obese patients with insulin resistance can exacerbate the progression of triple-negative breast cancer.[64] Exosomal miR-660-5p derived from adipocytes is highly expressed in the serum of ESCC patients, leading to poor prognosis in patients undergoing radiotherapy.[65]

Stromal cells are the main non-immune, non-tumor cell population in the TME, providing physical scaffolding and nutritional support for tumor growth, and the exosomes they secrete are the core carriers of signals in the microenvironment. Exosomes derived from cancer-associated fibroblasts (CAFs) can not only promote tumor progression but also reduce the effectiveness of tumor treatment.[66,67] CAF-derived exosomal CCT6A interacts with β-catenin to enhance chemoresistance and tumorigenesis in gastric cancer.[68] In triple-negative breast cancer (TNBC), CAF-derived exosomal circMIB1 promotes cancer metastasis by activating Notch signaling.[69] In PDAC, CAF-derived exosomal miR-3173-5p targets ACSL4, inhibiting ferroptosis while inducing resistance of pancreatic cancer cells to gemcitabine.[70] In esophageal squamous cell carcinoma, hypoxia-induced exosomal circNRIP1 activates CAFs to promote tumor migration and invasion.[71]

Mesenchymal stem cells derived from bone marrow, adipose tissue, umbilical cord, and other sources can migrate to the tumor microenvironment through the bloodstream and be "educated" into tumor-associated mesenchymal stem cells (TA-MSCs). Exosomes from AML-MSC co-delivering METTL14 stabilize ROCK1 expression via an m6A-IGF2BP3-dependent mechanism, thereby promoting AML cell proliferation and conferring radioresistance.[72] TA-MSCs promote the viability and invasiveness of NSCLC by delivering exosomal miR-182 through FBXW7-related AKT and ERK-dependent pathways.[73] It is worth mentioning that mesenchymal stem cells derived from human umbilical cord (hucMSCs) have broad prospects for cancer therapy.[74] Tang et al delivered PTX3 by fusing hucMSC-derived exosomes with neutrophil membrane vesicles, enhancing anti-tumor efficacy.[75]

Astrocytes are the most widely distributed type of cells in the mammalian brain and also the largest in volume among glial cells. In the TME, glioma cells can activate normal human astrocytes through the secretion of exosomal miR-423-3p, leading to poor prognosis in patients.[76]

As discussed, the TME is very complex and serves as a site for information exchange between various cells via exosomes. Therefore, targeting exosomes, the "couriers," is an important direction for future therapies.

## The Role of Exosomes in Liquid Biopsy

As previously discussed, exosomes transport diverse bioactive molecules (circRNAs, miRNAs, lncRNAs, proteins, lipids, DNA) that modulate immune responses upon entering immune cells. Specifically, circRNAs and lncRNAs often act via ceRNA or direct protein binding, miRNAs target mRNA 3'UTRs, proteins engage surface receptors, and lipids regulate metabolism, collectively influencing tumor immunity and inflammation. Importantly, they also serve as valuable biomarkers in liquid biopsy for cancer detection and monitoring. Exosomes dynamically reflect real-time disease status, offering broad application prospects in early screening, non-invasive diagnosis, treatment response monitoring, and guidance for adjuvant therapy.[3]

### Exosome Isolation and Detection Technologies

Exosome isolation technology is a core prerequisite for its clinical applications (such as liquid biopsy and drug delivery). The primary goal is to efficiently enrich exosomes (40–100 nm), remove impurities (cells, proteins, free nucleic acids, etc)., while preserving their structural integrity and biological activity. Currently, mainstream technologies can be divided into traditional methods and new integrated methods.[3] Traditional separation techniques are used for basic research and small-scale sample separation, including ultracentrifugation, density gradient centrifugation, immunoaffinity capture, polymer precipitation, size-exclusion chromatography, and ultrafiltration. Novel integrated separation technologies are commonly applied in clinical translation and high-throughput requirements, including microfluidic technology and tangential flow filtration (Table 2).

In the future, to improve the sensitivity, specificity, and clinical applicability of liquid biopsy, efforts can focus on multi-target synergistic capture, AI algorithms to exclude non-target vesicles, customized affinity ligands, and integrated automated devices.

### The Role of Exosomes as Biomarkers in Liquid Biopsy

Exosomes are intimately linked to gene mutations, serving as key mediators of oncogenic signaling. For instance, in colorectal cancer, PIK3CA-mutant tumor cells transmit oncogenic signals via exosome-derived arachidonic acid (AA), which induces H3K4 trimethylation and derives malignant transformation of intestinal epithelial cells (IEC).[85] As a tumor suppressor, p53 has received widespread attention from scientists. Xia et al used Escherichia coli Nissle 1917 (EcN) as a targeted delivery vehicle to deliver p53 and Tum-5 proteins to hypoxic tumor regions for cancer treatment.[86] Mutations in p53 are closely associated with the progression of cancer. In esophageal squamous cell carcinoma (ESCC), the p53-G245S mutation enhances exosome biogenesis, promoting cancer cell proliferation and metastasis.[87] These findings have spurred interest in targeting oncogenic mutations using exosome-based strategies. For example, mesenchymal stem cell-derived exosomes loaded with siRNA against KRASG12D (siKRASG12D-MSCExo) can specifically target KRASG12D-mutant cancer cells, inhibiting oncogene expression and suppressing tumor growth.[88] Similarly, engineered exosomes targeting the BRAFV600E mutation have been developed to carry specific siRNAs, blocking aberrant activation and tumor progression in colorectal cancer models, underscoring the role of exosomes in BRAF-driven tumorigenesis.[89]

Exosomes are present in various body fluids such as blood, urine, and saliva, enabling non-invasive or minimally invasive liquid biopsy approaches that improve patient compliance. Compared to traditional tissue biopsy, liquid biopsy using exosomes reduces procedural risks and is particularly valuable for patients with advanced disease or inaccessible tumors.[90] In hepatocellular carcinoma, plasma exosomal CDK1, FEN1 and PCNA have been used as diagnostic biomarkers, with machine learning models showing strong discriminatory power between HCC patients and controls.[91] Similarly, plasma-derived exosomal miR-15a-5p is upregulated in endometrial cancer and effectively distinguishes early-stage patients from healthy individuals (AUC =0.813).[92] Exosomal miR-92a-3p serves as a biomarker for

**Table 2** Advantages and Disadvantages of Traditional Exosome Separation Techniques

| Technology (Recycling Rate) | Method | Clinical Application | Advantages | Disadvantages | References |
|---|---|---|---|---|---|
| Ultracentrifugation (30–80%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Ultracentrifuge to enrich exosomes.<br>3. Centrifuge at 100,000×g for 70 minutes to remove residual free proteins and impurities, ultimately obtaining highly pure exosomes. | Preliminary screening and research of clinical samples<br>Testing in primary healthcare settings or resource-limited scenarios<br>Combining with other technologies to enhance clinical adaptability | Low cost, easy to operate, and widely available equipment | It takes a long time and requires professional ultracentrifugation equipment | [1] |
| Density Gradient Centrifugation (20–45%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Layer the supernatant on top of a density gradient medium and ultracentrifuge at 100,000–120,000×g for 16–20 hours (at 4°C); exosomes will migrate to the gradient layer matching their density.<br>3. Collect the exosome layer and centrifuge at 100,000×g for 70 minutes to pellet the exosomes. | Compatible with various clinical samples<br>Can handle plasma, serum, cerebrospinal fluid, urine, and other clinical samples, and is especially suitable for precious samples with complex impurities but limited volume, such as cerebrospinal fluid | High purity, high positive rate of exosome marker proteins<br>Isolation conditions are controllable, and result are highly reproducible | The operation is complex, requires special density gradient centrifuge tubes and centrifugation equipment, and the separation process takes a long time | [77] |
| Immunoaffinity Capture (40–70%) | 1. Immobilize exosome-specific antibodies.<br>2. Incubate the sample to form an "exosome-antibody-carrier" complex.<br>3. Wash away impurities and collect the exosomes. | Early tumor screening and early diagnosis (sensitivity over 80%, specificity 95%)<br>Assisting in the diagnosis of rare diseases and infectious diseases | High specificity and affinity, high purity of exosomes, isolation of specific types of exosomes | The cost of antibodies is high. The quality and specificity of different antibodies can vary, affecting the separation effect. | [78] |
| Size Exclusion Chromatography (50–80%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Add the supernatant to the chromatography column, elute, and collect the exosomes. | Rapid purification and detection of exosomes from clinical samples<br>Separation rate 40–60% | Gentle, high recovery and automatable | Purity is average, sample volume is limited | [79] |

(*Continued*)


https://doi.org/10.2147/IJN.S583756

**Table 2** (Continued).

| Technology (Recycling Rate) | Method | Clinical Application | Advantages | Disadvantages | References |
|---|---|---|---|---|---|
| Polymer Precipitation (60–90%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Use PEG 6000 or PEG 8000 to prepare the polymer solution.<br>3. Add exosomes to the PEG and incubate overnight at 4°C.<br>4. Centrifuge at 10,000–12,000×g for 30 minutes (4°C) to collect the exosomes.<br>5. Wash and purify. | Initial screening of clinical samples in primary medical institutions<br>Batch pre-processing of large-scale clinical samples<br>Emergency extraction of exosomes from trace samples | Easy to operate, fast and efficient, and low cost | Low purity and aggregation of exosomes | [80] |
| Microfluidics (50–85%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Use a microfluidic chip with surface-modified exosome-specific. Antibodies to sort exosomes in real time<br>3. Collect and elute. | Directly target and capture tumor-derived exosomes from minute clinical samples<br>Tumor treatment monitoring and relapse early warning | Low sample usage, fast and efficient, high degree of automation | High cost, low processing volume, high technical threshold. | [81] |
| Ultrafiltration (40–70%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Add the pre-processed supernatant to an ultrafiltration centrifuge tube and centrifuge at 3000–5000 × g for 10–20 minutes (4°C).<br>3. Add an appropriate amount of PBS to the ultrafiltration tube and centrifuge at 3000 × g for 10 minutes to wash and purify.<br>4. Collect the exosomes. | The recovery rate of clinical samples such as plasma and serum is approximately 40–60%<br>Rapid enrichment of large-scale clinical samples for tumor screening, epidemiological surveys, etc | Simple operation and low cost | The purity is average and the loss is high. | [82] |

(*Continued*)

**Table 2** (Continued).

| Technology (Recycling Rate) | Method | Clinical Application | Advantages | Disadvantages | References |
|---|---|---|---|---|---|
| Tangential Flow Filtration (70–80%) | 1. Centrifuge at 1300×g and 10,000×g to remove cell debris and large vesicles.<br>2. Load the TFF system and concentrate the exosomes.<br>3. Collect and concentrate the exosomes after washing and purification. | Separation rate 60–90%<br>Core Technologies for Preclinical Preparation of Stem Cell Exosomes | High productivity and recovery | High cost and complex operation | [83] |
| Fully integrated centrifugal microfluidic chip technology (70–85%) | 1. Select an integrated chip with matching functions, pre-load reagents, and set up the centrifuge platform.<br>2. Inject a small amount of body fluid into the chip's sample chamber.<br>3. Perform automated centrifugation separation.<br>4. The eluted exosomes flow into the chip's detection chamber, where markers (such as CD63) are detected using techniques like immunofluorescence, with real-time signal output.<br>5. Read the data. | Recovery rate of complex clinical samples such as plasma and serum is about 60–80%<br>Targeted detection of trace samples<br>Point-of-care testing (POCT) | High degree of automation and high purity | Small range of applications | [84] |

colorectal cancer. A ratiometric fluorescent biosensor developed by Sun et al accurately detected its levels in clinical samples, demonstrating significant diagnostic potential.[93] In gastric cancer, a combination of two lncRNAs (lncmstrg.2441832.8 and lncmstrg.2312697) in plasma exosomes achieved an AUC of 0.73 for diagnosis.[94]

Exosomal biomarkers also show great promise in prognostic prediction and recurrence monitoring. In GC, low blood levels of pre-miR-488 and mature miR-488-5p, detectable in plasma exosomes, correlate with poor prognosis and are independent predictors of overall survival. These miRNAs are inversely associated with genes involved in epithelial-mesenchymal transition and hypoxia.[95] Similarly, high expression of exosomal circATP8A1 in gastric cancer tissues and plasma-derived exosomes is associated with advanced TNM stage and worse outcomes, highlighting its utility as a prognostic marker.[27]

In recurrence monitoring, exosomes enable early detection of relapse across various cancer types. For instance, in cholangiocarcinoma, abnormal expression of hsa-circ-0000367, hsa-circ-0021647, and hsa-circ-0000288 in bile and serum exosomes can signal early recurrence.[96] In cervical cancer, elevated levels of exosomal DLEU1 in serum exosomes are an independent risk factor for postoperative recurrence and metastasis.[97]

With the advancement of immunotechnology, real-time immunomonitoring of extracellular vesicle biomarkers is crucial. Zhang et al developed an ultra-sensitive SERS detection method based on multivalent aptamer-linked tetrahedral DNA (MATD) assisted catalytic hairpin assembly (CHA). The multivalent aptamer specifically binds to the CD63 protein, allowing exosomes to be captured on the SERS sensing chip.[98] This method is a feasible strategy for the future monitoring of markers in auxiliary exosomes.

Notably, advances in artificial intelligence (AI) are enhancing exosome-based diagnostics. AI-driven microfluidic technology enables high-sensitivity detection of extracellular vesicles (EVs).[99] Furthermore, label-free analysis of plasma exosomes using AI and surface-enhanced Raman spectroscopy (SERS) permits simultaneous diagnosis of multiple cancer types.[100] The integration of exosome biomarkers with AI technologies holds great promise for future clinical applications in cancer diagnosis and prognosis (Figure 3 and Table 3).

However, exosomes face some challenges in terms of standardization in the field of liquid biopsy. The heterogeneity of exosomes is the primary obstacle to standardization, and there is still a lack of gold standard markers. There are difficulties in standardizing sample preprocessing and isolation techniques. The reproducibility of exosome liquid biopsies is affected by multiple factors, including technology, personnel, and samples. In terms of regulatory acceptance, there is a lack of validation data from large-scale, multicenter, prospective clinical trials, and the risk assessment and quality control systems for the technology are not well-developed, with unclear regulatory frameworks and classifications. Therefore, there is still considerable room for improvement in the standardization, reproducibility, and regulatory acceptance of exosomes in the future.

## The Role of Exosomes in Immunotherapy

Currently, clinically established tumor immunotherapies mainly include immune checkpoint inhibitors, adoptive cell therapy, cancer vaccines, and other emerging modalities. Exosomes, in turn, can be used in multiple ways to enhance or complement these immunotherapeutic strategies.

### Application of Exosomes in Tumor Vaccines

Tumor vaccines are characterized by their high specificity, favorable safety profile, and ability to induce long-lasting anti-tumor immune memory.

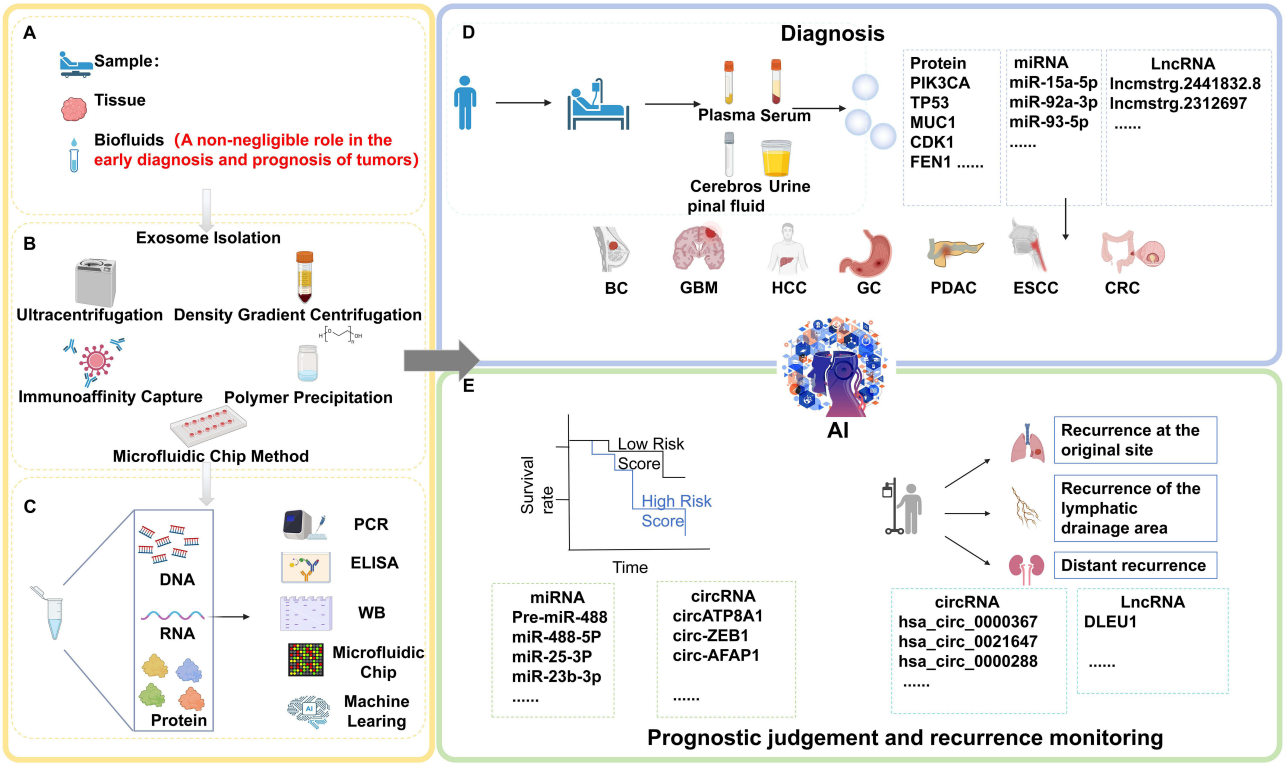


**Figure 3** The role of exosomes as biomarkers in liquid biopsy ((**A**) Source of exosomes, (**B**) Common methods for isolating exosomes, (**C**) Identification of exosome contents, (**D** and **E**) Applications of exosomes in clinical diagnosis and disease monitoring).

**Table 3** The Role of Exosomes as Biomarkers in Liquid Biopsy

| Functions | Molecular | Cancer | Source | Case Number | TNM | Reference |
|---|---|---|---|---|---|---|
| Genetic mutations | PIK3CA | CRC | Serum | 102 | Phase II | [85] |
| | P53 | ESCC | – | – | Phase I/II | [87] |
| | KRAS | PDAC | Blood | 173 | Phase I/II | [88] |
| | BRAF | CRC | Serum | 30 | Phase I/II | [89] |
| Diagnosis | MUC1 | GC | Serum | 20 | Phase I/II, Phase III | [98] |
| | CDK1 | HCC | Plasma | – | Phase I/II | [91] |
| | FEN1 | HCC | Plasma | – | Phase I/II | [91] |
| | PCNA | HCC | Plasma | – | Phase I/II | [91] |
| | miR-15a-5p | EC | Plasma | 258 | Phase I | [92] |
| | miR-92a-3p | CRC | Plasma | 115 | Phase I/II | [93] |
| | miR-93-5p | Glioma | Serum | 101 | Phase II | [101] |
| | lncmstrg.2441832.8 | GC | Plasma | 142 | Phase I | [94] |
| | lncmstrg.2312697 | GC | Plasma | 142 | Phase I | [94] |
| Prognostic judgement | Pre-miR-488 | GC | Plasma | 132 | Phase II/III | [95] |
| | miR-488-5P | GC | Plasma | 132 | Phase II/III | [95] |
| | miR-25-3P | ESCC | Plasma | 54 | Phase II/III | [102] |
| | miR-23b-3p | ESCC | Plasma | 54 | Phase II/III | [102] |
| | circATP8A1 | GC | Plasma | 30 | Phase II/III | [27] |
| | circ-ZEB1 | NSCLC | – | – | Phase II/III | [103] |
| | circ-AFAP1 | HCC | – | – | Phase II/III | [104] |
| Recurrence monitoring | Hsa_circ_0000367 | CCA | Serum Bile | 319 | Phase I/II | [96] |
| | Hsa_circ_0021647 | CCA | Serum Bile | 319 | Phase I/II | [96] |
| | Hsa_circ_0000288 | CCA | Serum Bile | 319 | Phase I/II | [96] |
| | DLEU1 | CC | Serum | 226 | Phase I/II | [97] |

Exosomes play multiple roles in tumor vaccine development, serving as antigen carriers, immune microenvironment modulators, and efficient delivery vehicles for vaccine components, thereby offering innovative strategies for cancer immunotherapy.[105,106]

Tumor-derived exosome vaccines represent a novel immunotherapeutic approach that leverages the natural ability of exosomes to carry tumor antigens and immunomodulatory molecules, thereby eliciting specific anti-tumor immune responses. Zou et al developed a homologous exosomal nanovaccine derived from GBM tumor cells, which dually targets lymph nodes and the brain. It elicited anti-tumor immunity in immunosuppressive CT2A-LUC GBM mice, extending survival; it also prevented brain metastasis and improved survival in B16F10-LUC melanoma mouse models. Although "induction of long-lasting protective immunity" is mentioned, the experimental observation period was short, and the vaccine's effect on long-term suppression of GBM recurrence was not evaluated (in clinical GBM patients, the

peak recurrence occurs 1–2 years post-surgery).[107] In another study, photosensitizer CyI and doxycycline (Doxy) were incorporated into heat-sensitive tumor-derived exosome-liposome hybrids (ECDL). In vitro and in vivo data show that ECDL homologously targets cancer cells, restricts mitochondrial respiration to alleviate tumor hypoxia, sustains oxygen supply to eliminate tumor cells and intracellular bacteria, and thus triggers in situ vaccine effects to suppress primary tumors, metastasis and recurrence.[108] A hybrid nanovaccine (Hy-M-Exo), fabricated by fusing tumor-derived exosomes (TEX) with dendritic cell membrane vesicles (DCMV), demonstrated significant therapeutic efficacy in a mouse model of head and neck squamous cell carcinoma (HNSCC).[109] Additionally, Ramazen et al loaded miR-124-3p mimics into exosomes isolated from CT-26 cells via a modified calcium chloride method, created a cell-free vaccine that promoted anti-tumor immunity in CT-26 tumor-bearing mice.[110]

DC-derived exosomes (DEX) modulate immune functions and promote immune cell-dependent tumor suppression. Clinical trials have confirmed the potential of DEX-based vaccines in advanced non-small cell lung cancer, melanoma, and colorectal cancer. Zhu et al constructed a MUC1-DEX conjugate vaccine via DBCO-NHS bioorthogonal ligation, which boosted DC activation and MUC1-specific immunity to inhibit tumor growth and extend survival.[111] Huang et al engineered breast cancer-derived exosomes to generate an orthotopic DC vaccine (HELA-Exos), which enhanced tumor responsiveness and elicited robust CD8 T cell responses in mouse models and human breast cancer organoids.[112] Yin et al developed a personalized DEX vaccine, named DEXP &A2&N. It induced significant tumor delay and tumor-specific immune responses in HCC mice with tumor burden.[113]

Immune cell-derived exosome vaccines are promising. Liu et al's γδ-T-EVs vaccine killed tumor cells, activated immunity, and achieved 78% complete remission in liver, lung and hematologic cancer trials, with high standardized production potential.[114]

In addition, in a triple-negative breast cancer model, extracellular vesicles released by M1 macrophages were combined with PLGA nanoparticles loaded with poly (I:C) to form a vaccine-like immunomodulatory system, which was used to study its antitumor activity by downregulating tumor immune evasion in the tumor microenvironment (TME) of an in situ tumor growth mouse model.[115] Cheng et al developed an inhalable exosome-based vaccine delivering IL-12 mRNA, which stimulated local interferon-γ (IFN-γ) production in the lung tumor microenvironment, activated systemic immunity, and established immune memory. This approach significantly inhibited lung cancer growth in mice with minimal systemic toxicity, providing a new strategy for treating metastatic lung cancer.[116]

In summary, exosomes hold great promise in tumor vaccine development due to their unique biological properties. However, challenges related to preparation standardization and safety must be addressed to facilitate the translation of these strategies from preclinical research to clinical application.

## Synergistic Effects of Exosomes with Immune Checkpoint Inhibitors

Immune checkpoints are a class of immunosuppressive molecules-small protein molecules produced by immune cells that regulate immune function. Immune checkpoint inhibitors (ICIs) are designed to specifically target these molecules and activate immune cells. Exosomes can influence the activity of ICIs in two contrasting ways: they may either interfer with therapeutic efficacy or enhance its potential.

Tumor-derived exosomes carry immune checkpoint molecules such as PD-L1 and CTLA-4.[117,118] Exosomal PD-L1 can bind to PD-1 on T cells and mimic the immunosuppressive signal of tumor cells. This interaction prevents ICIs from effectively blocking the communication between tumor cells and T cells, thereby reducing efficacy. Multiple research teams have found that PD-L1 is expressed on the surface of exosomes released by different cancer cells.[117,119,120] In addition, flow cytometry and immunofluorescence results show that PD-L1 is present not only on the surface of vesicles but also inside the vesicular structures.[120,121] In the tumor microenvironment, exosomal PD-L1 derived from hypoxic nasopharyngeal carcinoma cells can upregulate PD-L1 expression in macrophages, further enhancing $CD8^+$T cell suppression.[122] Compared with healthy individuals, NSCLC patients have significantly higher levels of Exo-PD-L1 than the healthy control group, with the increase being more pronounced in patients with stage III–IV. Exo-PD-L1 is closely associated with the disease progression of NSCLC and can reflect malignant features such as tumor size, lymph node metastasis, distant metastasis, and TNM staging.[123] Therefore, inhibiting exosomal PD-L1 (ExoPD-L1) secretion improves the clinical efficacy of PD-L1 antibodies. ApoA1-bExo/siRNA successfully inhibited the secretion of tumor-derived exosomal PD-L1 and

effectively enhanced the anti-tumor activity of T cells in vitro.[124] siRNA-loaded biomimetic exosome vesicles offer a new strategy for addressing PD-1/PD-L1 inhibitor resistance and enhancing immunotherapy efficacy by inhibiting tumor-derived Exo-PD-L1 secretion. Although currently at the preclinical stage, their targeting ability, safety, and synergistic effects have been validated. With future optimization of production processes and targeted clinical trials, they are expected to become the first combined immunotherapy approach for "Exo-PD-L1-targeted siRNA delivery".

### Exosomes Combined with Adoptive Cellular Immunotherapy

Exosomes derived from CAR-T cells have been shown to exhibit tumor-targeting properties and the ability to deliver cytotoxic cargo. Lip-CExo@ PTX is a hybrid nanoparticle constructed by fusing exosomes from dual-specific CAR-T cells targeting MSLN and PD-L1 with lung-targeted liposomes, which can prolong the survival of mice bearing CT-26 metastatic lung cancer (simulating the scenario of advanced human lung cancer with lung metastasis).[125] To address challenges such as the prolonged manufacturing time and difficulties in storage and transportation of CAR-T cells, Fan et al engineered tumor antigen-stimulated dendritic cell-derived exosomes (tDC-Exo) conjugated with anti-CD3 and anti-EGFR antibodies. This modification enhances T cell binding to tumor cells and improves the performance of CAR-T cell mimicking platforms.[126] Traditional antibody-drug conjugates (ADCs) primarily inhibit tumor growth through cytotoxic chemotherapy or immunomodulatory agents. CAR-M-derived exosome-drug conjugates enter Raji cells via CAR-mediated endocytosis, exerting immunotherapeutic effects through both SN38 chemotherapy and CXCL10-mediated antitumor immunity, with excellent in vivo antitumor activity.[127] The ExoCAR/T7@Micelle nanoplatform (comprising CAR-NK cell-derived exosomes and nanobomb micelles) provides a promising strategy for HER2-positive breast cancer brain metastases (HER2+ BCBM) via enhanced targeting and efficacy. ExoCAR/T7@Micelle demonstrates four major advantages in the $HER2^+$ BCBM model: "high blood-brain barrier penetration, precise targeting, low toxicity, and extended survival benefit," fully addressing the core challenges of clinical treatment. The next steps involve Phase I dose exploration and Phase II efficacy verification, and it is expected to become the first "HER2-targeted nanomedicine that can cross the blood-brain barrier," offering a new therapeutic option for patients with $HER2^+$ BCBM.[128] Additionally, loading antigen-presenting dendritic cell-derived exosomes with α-galactosylceramide (αGC) can activate invariant natural killer T (iNKT) cells, which subsequently stimulate NK cell activation and promote anti-tumor responses.[129]

ExoCAR-based platforms exhibit multiple anti-tumor mechanisms, including improved tumor targeting, release of cytotoxic components, and suppression of metastasis, suggesting potential to overcome current therapeutic limitations. Leveraging their natural biocompatibility and biofunctional properties, exosomes hold significant promise for advancing tumor immunotherapy. With continued progress in exosome isolation and targeted modification technologies, exosome-based therapies are expected to emerge as a powerful new modality in oncology. They may be applied in combination with immune checkpoint inhibitors and CAR-T cell therapies to further enhance the effectiveness of tumor treatment (Figure 4).

As natural carriers of intercellular information, exosomes show great potential in tumor immunotherapy, but there are still several key challenges in practical translation. Exosome yield is low, and exosomes derived from primary cells are difficult to produce on a clinical-scale. Exosomes have limited targeting capability to tumor tissues. They have a short circulatory half-life and are easily cleared. The long-term safety of exosomes is still unclear, including potential immunogenicity, accumulation toxicity, and off-target effects. Therefore, there is still a long way to go for their clinical translation in the future.

## Application of Exosome-Loaded Drugs

Enhancing immune responses through combination therapies and improving response rates to immunotherapy have become major focuses in cancer research. Chemotherapeutic agents are known to promote the release of tumor neoantigens and stimulate anti-tumor immunity, making them important partners for combination with immune checkpoint inhibitors.[130] Exosomes have emerged as a promising platform for modulating immune responses by delivering chemotherapy drugs in various formulations to regulate immune cell activity.

Yong et al developed a biocompatible biomimetic drug carrier based on porous silicon nanoparticles (PSiNPs) secreted by tumor cells. The DOX-loaded DOX@E-PSiNPs showed strong anticancer activity in subcutaneous, orthotopic, and metastatic tumor models, and also reduced cancer stem cells (CSC).[131] Aspirin has also been shown to possess

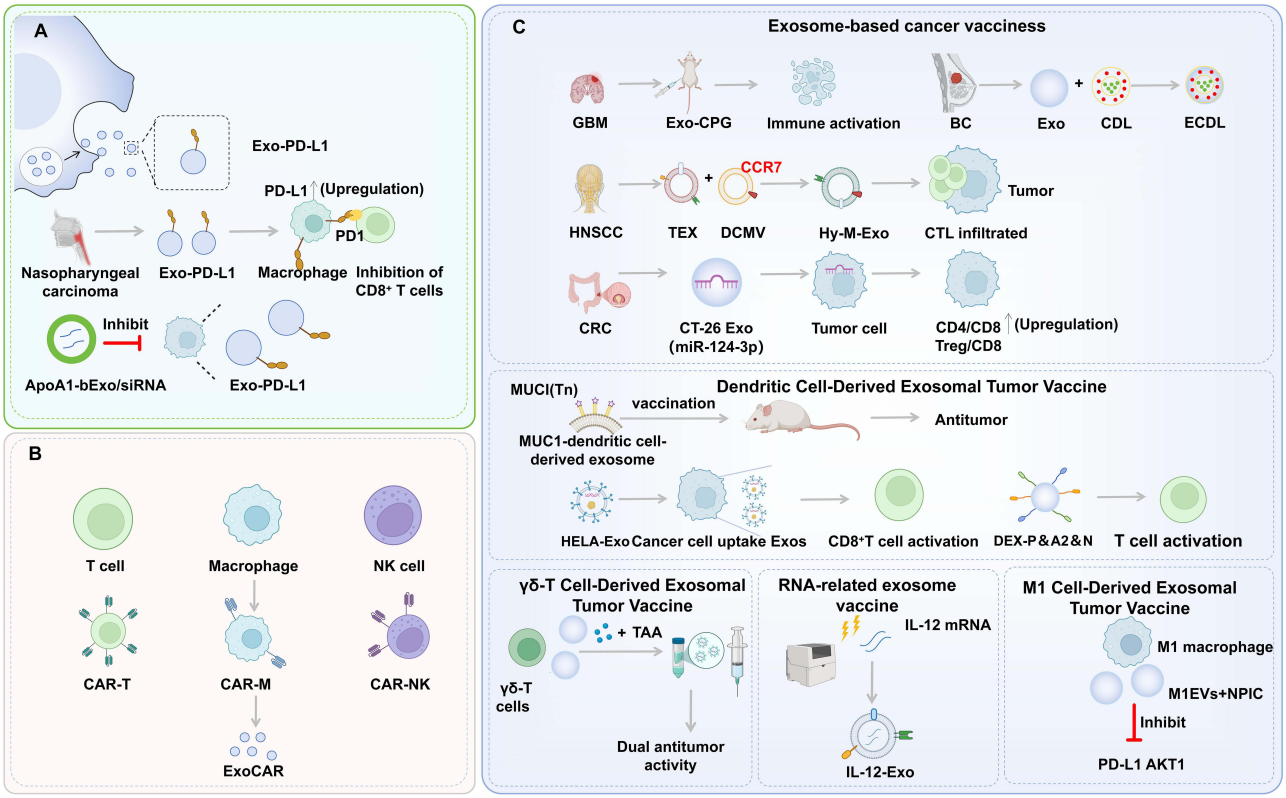


**Figure 4** The role of exosomes in tumor immunotherapy ((**A**) Synergistic effects of exosomes with immune checkpoint inhibitors, (**B**) Exosomes combined with adoptive cellular immunotherapy, (**C**) Exosomal vaccines).

anti-cancer properties.[132] Researchers engineered a biomimetic nanoplatform (TAFL) via fusion of tumor-derived exosomes and liposomes. TAFL enhances aspirin's efficacy against cancer stem cells (CSCs), lowers its effective dose to improve biosafety, and in vivo studies confirm that TAFL-mediated CSC depletion effectively suppresses tumor recurrence and metastasis post-FLASH-RT.[133]

Leveraging the inherent inflammatory chemotaxis and blood-brain barrier (BBB) penetrating ability of neutrophils, Wang and other researchers utilized the inherent inflammatory chemotaxis of neutrophils and their ability to penetrate the blood-brain barrier (BBB) to construct a neutrophil-derived exosome system loaded with doxorubicin (DOX). In a glioma mouse model, intravenous injection of NEs-Exos/DOX reduced tumor burden through a series of mechanisms, including crossing the blood-brain barrier, targeted accumulation in the inflammatory microenvironment, efficient drug release, inhibition of tumor proliferation, and improvement of the tumor microenvironment.[134] As previously discussed, exosomes derived from M1 macrophages exhibit potent anti-tumor effects. Gemcitabine (GEM), a first-line drug for bladder cancer, was loaded into M1 exosomes via ultrasound to form M1-Exo-GEM. The inflammatory factors carried by M1-Exo synergize with the chemotherapeutic effects of GEM, significantly enhancing the killing effect on bladder cancer cells by activating the endogenous apoptosis pathway, outperforming either drug alone or exosome treatment alone. In addition, M1-Exo-GEM can upregulate the levels of pro-inflammatory factors (such as TNF-α and IL-6) in tumor tissues, remodel the immunosuppressive microenvironment, activate the body's anti-tumor immunity, and synergistically inhibit tumor growth with chemotherapy.[135] Wang et al further enhanced the antitumor efficacy of chemotherapy in tumor-bearing mice by co-loaded paclitaxel (PTX) into exosomes via ultrasound.[136] In recent years, the clinical application of oxaliplatin (L-OHP) has been limited by its poor biocompatibility and severe side effects. To address this, a nanodrug delivery system was developed in which NK cell-derived exosomes were co-loaded with L-OHP using ultrasound (L-OHP-Exos). FasL carried by NK-Exos in L-OHP-Exos works synergistically with L-OHP to enhance the killing effect

by increasing ROS levels in tumor cells and activating the mitochondrial apoptosis pathway. This approach offers a promising strategy for the treatment of colorectal cancer with broad clinical application potential.[137]

Exosomes derived from human mesenchymal stem cells (MSCs) have demonstrated antitumor effects in various cancers.[138] Li et al isolated MSC-derived exosomes and loaded them with the first-line anti-cancer drug daunorubicin. This formulation effectively targets c-MPL AML cells while maintaining a favorable safety profile.[139] Ultrasound-mediated drug loading is a commonly used technique for encapsulating therapeutics into exosomes. For example, by using ultrasound to load doxorubicin (DOX) into exosomes derived from adipose-derived mesenchymal stem cells (ADMSCs), forming Exo-Dox, this complex inherits the tumor-homing ability of MSCs, efficiently targeting breast cancer cells (MDA-MB-231, MCF-7) and CAFs, reducing off-target effects and increasing local concentration, thereby lowering systemic toxicity.[140]

Beyond exosomes from mammalian cells, plant-derived extracellular vesicles have also gained interest as drug delivery vehicles. These natural nanoparticles offer advantages such as ease of mass production, low toxicity, and low immunogenicity.[141] For example, celery-derived exosome-like nanoparticles (CELNs) exhibit high cellular uptake efficiency, making them attractive drug carriers. Engineered CELNs loaded with DOX (CELNs-DOX) have been shown both in vitro and in vivo to outperform conventional synthetic vectors like liposomes in tumor treatment.[142] Similarly, Zhang et al developed ginger-derived nanovectors loaded with DOX (GDNVs). Dox-GDNVs increase intracellular Dox concentration, activate DNA damage response and mitochondrial apoptosis pathways, inhibit colon cancer cell proliferation, and induce apoptosis, showing better effects than free Dox.[143] Additionally, Lemon-derived extracellular vesicles delivering DOX can effectively overcome drug resistance in cancer cells via enhanced endocytosis.[144]

The heterogeneity of drug-loaded exosomes provides a reference for future engineered exosomes. Drug-loaded exosomes significantly reduce tumor burden in preclinical models and, with their advantages of biocompatibility and low immunogenicity, represent an important frontier in cancer therapy. Exosomes enable drug enrichment and precise release; protect drugs and reverse drug resistance; exert cytotoxic effects on tumor cells; and modulate the immune microenvironment. These functions are crucial for reducing tumor burden.

These findings highlight the broad prospects of exosome-based drug delivery systems. However, several challenges remain in their application for cancer therapy: isolation and purification technologies need improvement to obtain exosomes with high purity and bioactivity; drug loading efficiency is often low and release kinetics are difficult to control; distribution, metabolism, and potential side effects of exosomes require further investigation; and clear regulatory guidelines and standards are still lacking. These issues warrant further research (Figure 5).

## The Future Prospective

The dual role of exosomes means that on one hand, exosomes can activate cancer-promoting signaling pathways, leading to tumor proliferation, metastasis, drug resistance, and immune suppression (for example, in cancers such as gastric cancer, breast cancer, colon cancer, and liver cancer); on the other hand, they can play an immune-activating role, inhibiting cancer progression. Key functional molecules carried by exosomes include proteins, RNAs, and cytokines, which collectively influence immune cell states, immune checkpoint molecules, and metabolic pathways. These mechanisms open new avenues for dynamic monitoring and targeted therapeutic interventions. In combination with immune checkpoint inhibitors, adaptive immunotherapies, and cancer vaccines, exosomes are poised to become a critical breakthrough in cancer treatment.

Notably, exosomes are involved in multiple stages of the tumor immunity cycle, underscoring their potential as synergistic agents in immunotherapy. Promising applications include personalized exosome-based tumor vaccines, exosome-enhanced immune checkpoint inhibition and ExoCAR therapies. These approaches may define future directions in cancer immunotherapy. The combined therapeutic strategy of exosomes can achieve precise targeting, efficient drug delivery, and controlled release, holding promise to reshape the landscape of precision cancer treatment. Over the next 3–5 years, core breakthroughs are expected to focus on enhancing the in vivo efficacy of engineered exosomes, developing scalable production processes, and establishing standardized quality control systems, facilitating their rapid transition from the laboratory to clinical application and providing cancer patients with safer and more effective treatment options.

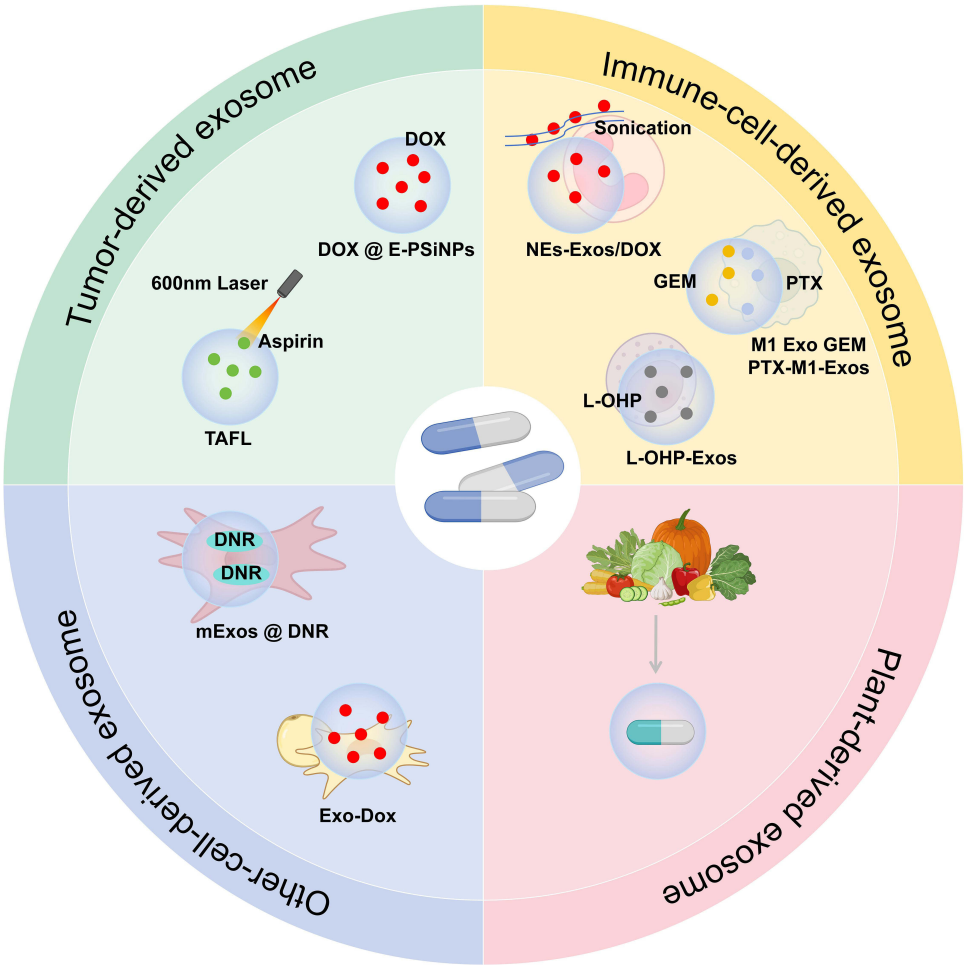


**Figure 5** Exosomes from different sources carry drugs.

However, due to the complex components and environment of the tumor immune microenvironment, the specific mechanisms by which exosomes affect itare still unclear and require further exploration. In addition, exosomes still have some limitations as biomarkers for cancer diagnosis and prognosis. Currently, research on exosomes as biomarkers still lacks a sufficient number of clinical samples. The lack of clinical trials remains an ongoing challenge in exosome research, and more multicenter, large-sample cohort studies are still needed. The large-scale production of exosomes is the primary bottleneck for their clinical application, mainly in terms of yield, purity, and cost. Exosomes have weak natural targeting ability, making it difficult to accurately deliver them to diseased tissues or cells, which is a key issue affecting their therapeutic efficacy. The biodistribution characteristics of exosomes after entering the body are complex and difficult to precisely control. The biosafety of exosomes is a core consideration for clinical translation, with potential risks mainly including immunogenicity, tumorigenicity, and cargo safety. The core of exosome pharmacokinetics is to balance "rapid clearance with targeted enrichment". In the future, it will be necessary to promote the transition from preclinical to clinical stages through engineered modifications and standardized evaluations. Engineering therapeutic strategies targeting exosomes must address issues of immunogenicity and standardization. Delivery via plant-derived exosomes is also an effective approach.

The stability and scalability of exosome preparations are key factors in determining whether they can transition from the laboratory to clinical application. To enhance the stability of exosome reagents, lyophilization can be used to improve storage stability, crosslinking agents can be employed to maintain the structural and compositional stability of exosomes, and targeted modifications can increase their in vivo stability. To achieve scalability of exosome reagents, large-scale production of exosomes is required, along with standardization and quality control, thereby increasing the likelihood of clinical translation.

## Conclusion

In summary, an increasing amount of evidence suggests that exosomes hold vast potential in the field of tumor therapy. The mechanisms of exosomes provide a new paradigm of "precise diagnosis and targeted therapy" for clinical practice. However, to achieve clinical application, three major core bottlenecks must first be overcome: clarifying the regulatory mechanisms of exosome functional heterogeneity, establishing standardized isolation and detection methods along with scalable production technologies, and improving clinical validation and regulatory systems. In the future, through the deep integration of basic research and clinical translation, these challenges need to be gradually addressed to promote the clinical application of exosomes in areas such as cancer and autoimmune diseases.

## Acknowledgments

Menglin Wei and Dongli Wang contributed equally as co-first authors for this study.

All images in this article are created by BioRender.

## Funding

This study is jointly supported by funding from the National Natural Science Foundation of China (Grant no. 82272179, 82302629); the Jiangsu Province's Major Project in Research and Development (BE2020680); the Technology Development Project of Suzhou (MSXM2024049, SYWD2025381).

## Disclosure

The authors report no conflicts of interest in this work.

## References

1. Thery C, Zitvogel L, Amigorena S. Exosomes: composition, biogenesis and function. *Nat Rev Immunol*. 2002;2(8):569–579. doi:10.1038/nri855
2. Trams EG, Lauter CJ, Salem N, Heine U. Exfoliation of membrane ecto-enzymes in the form of micro-vesicles. *Biochim Biophys Acta*. 1981;645(1):63–70. doi:10.1016/0005-2736(81)90512-5
3. Yu D, Li Y, Wang M, et al. Exosomes as a new frontier of cancer liquid biopsy. *Mol Cancer*. 2022;21(1):56. doi:10.1186/s12943-022-01509-9
4. Hanahan D, Coussens LM. Accessories to the crime: functions of cells recruited to the tumor microenvironment. *Cancer Cell*. 2012;21(3):309–322. doi:10.1016/j.ccr.2012.02.022
5. Yu X, Zhang Y, Luo F, Zhou Q, Zhu L. The role of microRNAs in the gastric cancer tumor microenvironment. *Mol Cancer*. 2024;23(1):170. doi:10.1186/s12943-024-02084-x
6. Gong H, Liu Z, Yuan C, et al. Identification of cuproptosis-related lncRNAs with the significance in prognosis and immunotherapy of oral squamous cell carcinoma. *Comput Biol Med*. 2024;171:108198. doi:10.1016/j.compbiomed.2024.108198
7. Wang D, Shen Y, Qian H, Jiang J, Xu W. Emerging advanced approaches for liquid biopsy: in situ nucleic acid assays of extracellular vesicles. *Theranostics*. 2024;14(19):7309–7332. doi:10.7150/thno.102437
8. Zhang F, Jiang J, Qian H, Yan Y, Xu W. Exosomal circRNA: emerging insights into cancer progression and clinical application potential. *J Hematol Oncol*. 2023;16(1):67. doi:10.1186/s13045-023-01452-2
9. Szeto GL, Finley SD. Integrative approaches to cancer immunotherapy. *Trends Cancer*. 2019;5(7):400–410. doi:10.1016/j.trecan.2019.05.010
10. Zhang Y, Zhang Z. The history and advances in cancer immunotherapy: understanding the characteristics of tumor-infiltrating immune cells and their therapeutic implications. *Cell Mol Immunol*. 2020;17(8):807–821. doi:10.1038/s41423-020-0488-6
11. Riley RS, June CH, Langer R, Mitchell MJ. Delivery technologies for cancer immunotherapy. *Nat Rev Drug Discov*. 2019;18(3):175–196. doi:10.1038/s41573-018-0006-z
12. Huang M, Huang X, Huang N. Exosomal circGSE1 promotes immune escape of hepatocellular carcinoma by inducing the expansion of regulatory T cells. *Cancer Sci*. 2022;113(6):1968–1983. doi:10.1111/cas.15365
13. Veletic I, Harris DM, Rozovski U, et al. CLL cell-derived exosomes alter the immune and hematopoietic systems. *Leukemia*. 2025;39(6):1380–1394. doi:10.1038/s41375-025-02590-x
14. Wang X, Yao Y, Jin M. Circ-0001068 is a novel biomarker for ovarian cancer and inducer of PD1 expression in T cells. *Aging*. 2020;12(19):19095–19106. doi:10.18632/aging.103706
15. Oztatlici M, Ozdemir AT, Oztatlici H, et al. Immunomodulatory effects of MDA-MB-231-derived exosome mimetic nanovesicles on CD4+ T cell line. *EJMO*. 2024;8(1). doi:10.14744/ejmo.2024.64067
16. Hu Z, Chen G, Zhao Y, et al. Exosome-derived circCCAR1 promotes CD8 + T-cell dysfunction and anti-PD1 resistance in hepatocellular carcinoma. *Mol Cancer*. 2023;22(1):55. doi:10.1186/s12943-023-01759-1
17. Liu T, Cheng S, Peng B, et al. PD-L2 of tumor-derived exosomes mediates the immune escape of cancer cells via the impaired T cell function. *Cell Death Dis*. 2024;15(11):800. doi:10.1038/s41419-024-07191-7
18. Song L, Cai J, Zhu G, et al. Hypoxia-related CircARSB modulates lipid metabolism and innate immune crosstalk to influence immune checkpoint inhibitor response in hepatocellular carcinoma. *Cancer Lett*. 2025;639:218188. doi:10.1016/j.canlet.2025.218188
19. Zhang Y, Yu Y, Gu X, Li Z, Zhou Y, Xiang J. Exosomes derived from colorectal cancer cells suppress B-cell mediated anti-tumor immunity. *Int Immunopharmacol*. 2025;148:114176. doi:10.1016/j.intimp.2025.114176

20. Uriepero-Palma A, Marquez ME, Payque E, et al. Targeting S100A9-mediated inflammation: a novel therapeutic approach for CLL. *Blood Adv*. 2025;9(20):5219–5233. doi:10.1182/bloodadvances.2025016061
21. Zhang PF, Gao C, Huang XY, et al. Cancer cell-derived exosomal circUHRF1 induces natural killer cell exhaustion and may cause resistance to anti-PD1 therapy in hepatocellular carcinoma. *Mol Cancer*. 2020;19(1):110. doi:10.1186/s12943-020-01222-5
22. Qin J, Yang J, Cui H, Feng C, Liu A. Exosomal miR-552-5p regulates the role of NK cells in EMT of gastric cancer via the PD-1/PD-L1 axis. *J Cancer*. 2025;16(2):406–416. doi:10.7150/jca.102360
23. Jiang Q, Xin Y, He F, Liu C, Qiu J. Exosomal SNHG26 mediates immunosuppression by impairing NK cells in tongue cancer. *NPJ Precis Oncol*. 2025. doi:10.1038/s41698-025-01185-0
24. Shang A, Gu C, Wang W, et al. Exosomal circPACRGL promotes progression of colorectal cancer via the miR-142-3p/miR-506-3p- TGF-β1 axis. *Mol Cancer*. 2020;19(1):117. doi:10.1186/s12943-020-01235-0
25. Zhang J, Yu D, Ji C, et al. Exosomal miR-4745-5p/3911 from N2-polarized tumor-associated neutrophils promotes gastric cancer metastasis by regulating SLIT2. *Mol Cancer*. 2024;23(1):198. doi:10.1186/s12943-024-02116-6
26. Sun Z, Xu Y, Shao B, et al. Exosomal circPOLQ promotes macrophage M2 polarization via activating IL-10/STAT3 axis in a colorectal cancer model. *J Immunother Cancer*. 2024;12(5):e008491. doi:10.1136/jitc-2023-008491
27. Deng C, Huo M, Chu H, et al. Exosome circATP8A1 induces macrophage M2 polarization by regulating the miR-1-3p/STAT6 axis to promote gastric cancer progression. *Mol Cancer*. 2024;23(1):49. doi:10.1186/s12943-024-01966-4
28. Wang Y, Li Y, Zhong J, et al. Tumor-derived Cav-1 promotes pre-metastatic niche formation and lung metastasis in breast cancer. *Theranostics*. 2023;13(5):1684–1697. doi:10.7150/thno.79250
29. Zhu X, Gu G, Shen Y, Abdurazik MH, Liu C, Sun G. Nsclc-derived exosomal hsa_circ_0003026 promotes tumor growth through macrophage M2 polarization via hsa-miR-1183/XRN2 axis. *Gene*. 2025;962:149557. doi:10.1016/j.gene.2025.149557
30. Li W, Wei H, Liu J, et al. Exosomal biglycan promotes gastric cancer progression via M2 polarization and CXCL10-mediated JAK/STAT1 activation. *Cancer Lett*. 2025;626:217758. doi:10.1016/j.canlet.2025.217758
31. Long K, Kui X, Zeng Q, Dong W. Cancer cell-derived exosomal miR-34a inhibits the malignant progression of pancreatic adenocarcinoma cells by restraining the M2 polarization of macrophages. *Eur J Histochem*. 2025;69(2). doi:10.4081/ejh.2025.4176
32. Li S, Fu X, Ning D, et al. Colon cancer exosome-associated HSP90B1 initiates pre-metastatic niche formation in the liver by polarizing M1 macrophage into M2 phenotype. *Biol Direct*. 2025;20(1):52. doi:10.1186/s13062-025-00623-0
33. Han Y, Zhou Z, Li R, Wang H. Tumor-derived exosomal circ_0020095 promotes colon cancer cell proliferation and metastasis by inhibiting M1 macrophage polarization. *J Biochem Mol Toxicol*. 2025;39(4):e70225. doi:10.1002/jbt.70225
34. Wang F, Li L, Sun X, et al. The feedback loop between miR-222-3p and ZEB1 harnesses metastasis in renal cell carcinoma. *Cell Death Discov*. 2025;11(1):97. doi:10.1038/s41420-025-02385-0
35. Zhao W, Wu Y, Wang Y, Li T, Liu Q, Hou Z. Exosomal miR-92a-3p modulates M2 macrophage polarization in colorectal cancer: implications for tumor migration and angiogenesis. *Med Oncol*. 2025;42(4):96. doi:10.1007/s12032-025-02635-2
36. Wu Q, Chen S, Xie X, et al. Glioblastoma- derived exosomes (GBM-Exo) regulate microglial M2 polarization via the RAC1/AKT/NRF2 pathway. *J Neurooncol*. 2025;172(2):447–460. doi:10.1007/s11060-024-04934-6
37. He L, Chen Q, Wu X. Tumour-derived exosomal miR-205 promotes ovarian cancer cell progression through M2 macrophage polarization via the PI3K/Akt/mTOR pathway. *J Ovarian Res*. 2025;18(1):28. doi:10.1186/s13048-025-01616-3
38. Ling H, Li Y, Wang P, Zhang Z, Yang Z. Diffuse large B-cell lymphoma cell-derived exosomal NSUN2 stabilizes PDL1 to promote tumor immune escape and M2 macrophage polarization in a YBX1-dependent manner. *Arch Biochem Biophys*. 2025;766:110322. doi:10.1016/j.abb.2025.110322
39. Zhang Y, Li B, Gu W, et al. Hepatoma cell-derived exosomal SNORD52 mediates M2 macrophage polarization by activating the JAK2/STAT6 pathway. *Discov Oncol*. 2025;16(1):36. doi:10.1007/s12672-024-01700-y
40. Qiu Y, Yang Y, Yang R, et al. Activated T cell-derived exosomal PD-1 attenuates PD-L1-induced immune dysfunction in triple-negative breast cancer. *Oncogene*. 2021;40(31):4992–5001. doi:10.1038/s41388-021-01896-1
41. Ghorbaninezhad F, Alemohammad H, Najafzadeh B, et al. Dendritic cell-derived exosomes: a new horizon in personalized cancer immunotherapy? *Cancer Lett*. 2023;562:216168. doi:10.1016/j.canlet.2023.216168
42. Alfawaz Altamimi AS, Arockia Babu M, Afzal M, et al. Exosomes derived from natural killer cells: transforming immunotherapy for aggressive breast cancer. *Med Oncol*. 2025;42(4):114. doi:10.1007/s12032-025-02647-y
43. Li W, Ma R, Fan X, Xiao Z. M1 macrophage-derived exosomes alleviate leukemia by causing mitochondrial dysfunction. *Ann Hematol*. 2024;103(12):5425–5438. doi:10.1007/s00277-024-06138-4
44. An B, Shin CH, Kwon JW, et al. M1 macrophage-derived exosomal microRNA-29c-3p suppresses aggressiveness of melanoma cells via ENPP2. *Cancer Cell Int*. 2024;24(1):325. doi:10.1186/s12935-024-03512-0
45. Xu Q, Chen X, Ma Z, Zhong H, Feng G, Gu S. Exosomal ETV4 derived from M2 macrophages induces growth, glycolysis and stemness in hepatocellular carcinoma by UpRegulating SULT2B1 expression. *Liver Int*. 2024;45. doi:10.1111/liv.16197
46. Yang Y, Guo Z, Chen W, et al. M2 macrophage-derived exosomes promote angiogenesis and growth of pancreatic ductal adenocarcinoma by targeting E2F2. *Mol Ther*. 2021;29(3):1226–1238. doi:10.1016/j.ymthe.2020.11.024
47. Xue Y, Chen J, Sun X, et al. Exosomes derived from M2 macrophage promote HUVECs proliferation, migration and tube formation in vitro. *Sci Rep*. 2025;15(1):17876. doi:10.1038/s41598-025-03113-5
48. Zhang J, Liu J, Liu Z, Guo L, Liu X. M2 macrophages-derived exosomal MDH1 drives lung adenocarcinoma progression via the Hippo/YAP signaling. *Pathol Res Pract*. 2025;269:155902. doi:10.1016/j.prp.2025.155902
49. Wei W, Li J, Huang J, et al. Exosomal miR-3681-3p from M2-polarized macrophages confers cisplatin resistance to gastric cancer cells by targeting MLH1. *Mol Med Rep*. 2025;31(4). doi:10.3892/mmr.2025.13459
50. Guan H, Tao H, Luo J, et al. Upregulation of YY1 in M2 macrophages promotes secretion of exosomes containing hsa-circ-0000326 via super-enhancers to facilitate prostate cancer progression. *Mol Cell Biochem*. 2025;480(6):3873–3888. doi:10.1007/s11010-025-05222-1
51. Zhou Y, Sun YC, Zhang QY, Wang J, Zhu XY, Su XY. Tumor-associated macrophage-derived exosome miR-194 confers cisplatin resistance in GC cells. *Eur J Med Res*. 2025;30(1):75. doi:10.1186/s40001-025-02329-5

52. Yuan W, Sun Q, Zhu X, Li B, Zou Y, Liu Z. M2-polarized tumor-associated macrophage-secreted exosomal lncRNA NEAT1 upregulates galectin-3 by recruiting KLF5 and promotes HCC immune escape. *J Cell Commun Signal*. 2025;19(1):e12060. doi:10.1002/ccs3.12060
53. Yin J, Pei Z, Wu C, et al. M2 macrophage-derived exosomal circ_0088494 inhibits ferroptosis via promoting H3K4me1 modification of STEAP3 in cutaneous squamous cell carcinoma. *Mol Carcinog*. 2024;64(3):513–525. doi:10.1002/mc.23862
54. Yang J, Zhu J, Lu S, Qin H, Zhou W. Transdermal psoriasis treatment inspired by tumor microenvironment-mediated immunomodulation and advanced by exosomal engineering. *J Control Release*. 2025;382:113664. doi:10.1016/j.jconrel.2025.113664
55. Lu JY, Guo Z, Huang WT, et al. Peptide-graphene logic sensing system for dual-mode detection of exosomes, molecular information processing and protection. *Talanta*. 2024;267:125261. doi:10.1016/j.talanta.2023.125261
56. Tang H, Wei Z, Zheng B, et al. Rescuing dendritic cell interstitial motility sustains antitumour immunity. *Nature*. 2025;645(8079):244–253. doi:10.1038/s41586-025-09202-9
57. Shpigelman J, Rao K. Dendritic cell-derived extracellular vesicles as therapeutic cancer vaccines: mechanisms and optimization strategies. *Immunology*. 2025. doi:10.1111/imm.70033
58. Fridlender ZG, Sun J, Kim S, et al. Polarization of tumor-associated neutrophil phenotype by TGF-beta: "N1" versus "N2" TAN. *Cancer Cell*. 2009;16(3):183–194. doi:10.1016/j.ccr.2009.06.017
59. Luo G, Zhou Z, Cao Z, et al. M2 macrophage-derived exosomes induce angiogenesis and increase skin flap survival through HIF1AN/HIF-1α/VEGFA control. *Arch Biochem Biophys*. 2023;751:109822. doi:10.1016/j.abb.2023.109822
60. Wang M, Zhao Y, Xu K, et al. Cancer-associated fibroblasts in clear cell renal cell carcinoma: functional heterogeneity, tumor microenvironment crosstalk, and therapeutic opportunities. *Front Immunol*. 2025;16:1617968. doi:10.3389/fimmu.2025.1617968
61. Liu L, Zhang S, Ren Y, et al. Macrophage-derived exosomes in cancer: a double-edged sword with therapeutic potential. *J Nanobiotechnology*. 2025;23(1):319. doi:10.1186/s12951-025-03321-1
62. Wen B, Tao R, Liu Y, Zhang Z. Investigating the role of exosomal microRNA-5703 in modulating tumor-associated endothelial cells in lung cancer. *Cytojournal*. 2024;21:77. doi:10.25259/Cytojournal_99_2024
63. Bouche C, Quail DF. Fueling the tumor microenvironment with cancer-associated adipocytes. *Cancer Res*. 2023;83(8):1170–1172. doi:10.1158/0008-5472.Can-23-0505
64. Llévenes P, Chen A, Lawton M, et al. Plasma exosomes in insulin resistant obesity exacerbate progression of triple negative breast cancer. *BMC Cancer*. 2025;25(1):1089. doi:10.1186/s12885-025-14447-8
65. Ge YY, Xia XC, Wu AQ, Ma CY, Yu LH, Zhou JY. Identifying adipocyte-derived exosomal miRNAs as potential novel prognostic markers for radiotherapy of esophageal squamous cell carcinoma. *World J Gastrointest Oncol*. 2025;17(2):98808. doi:10.4251/wjgo.v17.i2.98808
66. Mao X, Xu J, Wang W, et al. Crosstalk between cancer-associated fibroblasts and immune cells in the tumor microenvironment: new findings and future perspectives. *Mol Cancer*. 2021;20(1):131. doi:10.1186/s12943-021-01428-1
67. Xu W, Liu S, Ma L, et al. Identification of miRNA signature in cancer-associated fibroblast to predict recurrent prostate cancer. *Comput Biol Med*. 2024;180:108989. doi:10.1016/j.compbiomed.2024.108989
68. Sun H, Zhang T, Zhang X, et al. Exosomal CCT6A secreted by cancer-associated fibroblasts interacts with β-catenin to enhance chemoresistance and tumorigenesis in gastric cancer. *Adv Sci*. 2025;12:e06674. doi:10.1002/advs.202506674
69. Ye F, Liang Y, Wang J, et al. A novel peptide MIB1-223aa encoded by exosomal circMIB1 from cancer-associated fibroblasts drives triple-negative breast cancer metastasis and stemness via stabilizing MIB1 to activate Notch signaling. *J Adv Res*. 2025. doi:10.1016/j.jare.2025.06.023
70. Qi R, Bai Y, Li K, et al. Cancer-associated fibroblasts suppress ferroptosis and induce gemcitabine resistance in pancreatic cancer cells by secreting exosome-derived ACSL4-targeting miRNAs. *Drug Resist Updat*. 2023;68:100960. doi:10.1016/j.drup.2023.100960
71. Qiao G, Li C, Wang M, et al. Hypoxia-induced exosomal circNRIP1 activates cancer-associated fibroblasts to promote esophageal squamous cell carcinoma migration and invasion. *BMC Gastroenterol*. 2025;25(1):605. doi:10.1186/s12876-025-03978-w
72. Wang C, Song R, Yuan J, et al. Exosome-Shuttled METTL14 from AML-derived mesenchymal stem cells promotes the proliferation and radioresistance in AML cells by stabilizing ROCK1 expression via an m6A-IGF2BP3-dependent mechanism. *Drug Dev Res*. 2025;86(1):e70025. doi:10.1002/ddr.70025
73. Sun Y, Zhu X, Yu L, Dong H, Liu Z. Cancer-associated mesenchymal stem cell exosomes facilitate non-small cell lung cancer cell viability and invasiveness by delivering miR-182 in a FBXW7-related AKT and ERK-dependent pathway. *Oncol Lett*. 2025;30(4):487. doi:10.3892/ol.2025.15233
74. Li Y, Huang BM, Tao CC, Lan YY. Exosomes derived from human umbilical cord mesenchymal stem cells enhance cisplatin-induced apoptotic effects via the ROS-Fas pathway in human NPC-TW01 nasopharyngeal carcinoma cells. *Anticancer Res*. 2025;45(7):3117–3126. doi:10.21873/anticanres.17675
75. Tang Y, Shen Y, Zang X, et al. Neutrophil membrane engineered human umbilical cord MSC-derived sEVs enhance anti-tumor efficacy for gastric cancer via delivering pentraxin 3. *J Control Release*. 2025;383:113828. doi:10.1016/j.jconrel.2025.113828
76. Tang Z, Xue Z, Liu X, et al. Inhibition of hypoxic exosomal miR-423-3p decreases glioma progression by restricting autophagy in astrocytes. *Cell Death Dis*. 2025;16(1):265. doi:10.1038/s41419-025-07576-2
77. Temoche-Diaz MM, Shurtleff MJ, Schekman R. Buoyant density fractionation of small extracellular vesicle sub-populations derived from mammalian cells. *Biol Protoc*. 2020;10(15):e3706. doi:10.21769/BioProtoc.3706
78. Clayton A, Court J, Navabi H, et al. Analysis of antigen presenting cell derived exosomes, based on immuno-magnetic isolation and flow cytometry. *J Immunol Methods*. 2001;247(1–2):163–174. doi:10.1016/s0022-1759(00)00321-5
79. Vanderboom PM, Dasari S, Ruegsegger GN, et al. A size-exclusion-based approach for purifying extracellular vesicles from human plasma. *Cell Rep Methods*. 2021;1(3):136155. doi:10.1016/j.crmeth.2021.100055
80. Rider MA, Hurwitz SN, Meckes DG. ExtraPEG: a polyethylene glycol-based method for enrichment of extracellular vesicles. *Sci Rep*. 2016;6:23978. doi:10.1038/srep23978
81. Xu WM, Li A, Chen JJ, Sun EJ. Research development on exosome separation technology. *J Membr Biol*. 2023;256(1):25–34. doi:10.1007/s00232-022-00260-y
82. Lai RC, Arslan F, Lee MM, et al. Exosome secreted by MSC reduces myocardial ischemia/reperfusion injury. *Stem Cell Res*. 2010;4(3):214–222. doi:10.1016/j.scr.2009.12.003

83. Chen Y, Xu Y, Zheng Y, et al. Harnessing 3D cultured MSC exosomes through tangential flow filtration for enhanced diabetic wound healing. *Stem Cells Transl Med*. 2025;14(12). doi:10.1093/stcltm/szaf064
84. Zhao X, Liu X, Chen T, et al. Fully integrated centrifugal microfluidics for rapid exosome isolation, glycan analysis, and point-of-care diagnosis. *ACS Nano*. 2025. doi:10.1021/acsnano.4c16988
85. He B, Bie Q, Zhao R, et al. Arachidonic acid released by PIK3CA mutant tumor cells triggers malignant transformation of colonic epithelium by inducing chromatin remodeling. *Cell Rep Med*. 2024;5(5):101510. doi:10.1016/j.xcrm.2024.101510
86. He L, Yang H, Tang J, et al. Intestinal probiotics E. coli Nissle 1917 as a targeted vehicle for delivery of p53 and Tum-5 to solid tumors for cancer therapy. *J Biol Eng*. 2019;13:58. doi:10.1186/s13036-019-0189-9
87. Feng R, Yin Y, Wei Y, et al. Mutant p53 activates hnRNPA2B1-AGAP1-mediated exosome formation to promote esophageal squamous cell carcinoma progression. *Cancer Lett*. 2023;562:216154. doi:10.1016/j.canlet.2023.216154
88. Kamerkar S, LeBleu VS, Sugimoto H, et al. Exosomes facilitate therapeutic targeting of oncogenic KRAS in pancreatic cancer. *Nature*. 2017;546(7659):498–503. doi:10.1038/nature22341
89. Zhi J, Jia XJ, Yan J, et al. BRAFV600E mutant colorectal cancer cells mediate local immunosuppressive microenvironment through exosomal long noncoding RNAs. *World J Gastrointest Oncol*. 2021;13(12):2129–2148. doi:10.4251/wjgo.v13.i12.2129
90. Li J, Wang A, Guo H, et al. Exosomes: innovative biomarkers leading the charge in non-invasive cancer diagnostics. *Theranostics*. 2025;15(11):5277–5311. doi:10.7150/thno.113650
91. Huang H, Zhang M, Lu H, et al. Identification and evaluation of plasma exosome RNA biomarkers for non-invasive diagnosis of hepatocellular carcinoma using RNA-seq. *BMC Cancer*. 2024;24(1):1552. doi:10.1186/s12885-024-13332-0
92. Zhou L, Wang W, Wang F, et al. Plasma-derived exosomal miR-15a-5p as a promising diagnostic biomarker for early detection of endometrial carcinoma. *Mol Cancer*. 2021;20(1):57. doi:10.1186/s12943-021-01352-4
93. Sun Z, Li J, Yang Y, et al. Ratiometric fluorescent biosensor based on self-assembled fluorescent gold nanoparticles and duplex-specific nuclease-assisted signal amplification for sensitive detection of exosomal miRNA. *Bioconjug Chem*. 2022;33(9):1698–1706. doi:10.1021/acs.bioconjchem.2c00309
94. Wei Y, Hu X, Yuan S, et al. Identification of plasma exosomal lncRNA as a biomarker for early diagnosis of gastric cancer. *Front Genet*. 2024;15:1425591. doi:10.3389/fgene.2024.1425591
95. Tsuruda Y, Masuda T, Hiraki Y, et al. Circulating pre-microRNA-488 in blood is a potential prognostic biomarker in gastric cancer. *Anticancer Res*. 2025;45(1):123–133. doi:10.21873/anticanres.17399
96. Wen N, Peng D, Xiong X, et al. Cholangiocarcinoma combined with biliary obstruction: an exosomal circRNA signature for diagnosis and early recurrence monitoring. *Signal Transduct Target Ther*. 2024;9(1):107. doi:10.1038/s41392-024-01814-3
97. Chen Y, Cui F, Wu X, Zhao W, Xia Q. The expression and clinical significance of serum exosomal-long non-coding RNA DLEU1 in patients with cervical cancer. *Ann Med*. 2025;57(1):2442537. doi:10.1080/07853890.2024.2442537
98. Zhang J, Yan C, Xie L, et al. Multivalent aptamer-linked tetrahedron DNA assisted catalytic hairpin assembly for accurate SERS assay of cancer-derived exosomes in clinical blood. *Biosens Bioelectron*. 2025;282:117497. doi:10.1016/j.bios.2025.117497
99. Lin B, Lei Y, Wang J, et al. Microfluidic-based exosome analysis for liquid biopsy. *Small Methods*. 2021;5(3):e2001131. doi:10.1002/smtd.202001131
100. Shin H, Choi BH, Shim O, et al. Single test-based diagnosis of multiple cancer types using Exosome-SERS-AI for early stage cancers. *Nat Commun*. 2023;14(1):1644. doi:10.1038/s41467-023-37403-1
101. Zhang Y, Xie J, Zhang H, et al. Serum exosomal miRNA promote glioma progression by targeting SOS1 via abscopal effect of radiation. *Arch Biochem Biophys*. 2024;761:110138. doi:10.1016/j.abb.2024.110138
102. Jing Z, Guo Z, Zhang C. Plasma-derived exosomal miR-25-3p and miR-23b-3p as predictors of response to chemoradiotherapy in esophageal squamous cell carcinoma. *Technol Cancer Res Treat*. 2024;23:15330338241289520. doi:10.1177/15330338241289520
103. Wang Q, Ling S, Lv J, Wu L. circ-ZEB1 enhances NSCLC metastasis and proliferation by modulating the miR-491-5p/EIF5A axis. *Anal Cell Pathol*. 2025;2025:5595692. doi:10.1155/ancp/5595692
104. Han T, Chen L, Li K, et al. Significant CircRNAs in liver cancer stem cell exosomes: mediator of malignant propagation in liver cancer? *Mol Cancer*. 2023;22(1):197. doi:10.1186/s12943-023-01891-y
105. Lu Q, Kou D, Lou S, et al. Nanoparticles in tumor microenvironment remodeling and cancer immunotherapy. *J Hematol Oncol*. 2024;17(1):16. doi:10.1186/s13045-024-01535-8
106. Chen J, Verdiell A, Formoso C, Luciano M, Chen C, Thakur A. Tumor-derived extracellular vesicles: bridging communication and next-generation theranostics. *Biomed Pharmacother*. 2025;193:118815. doi:10.1016/j.biopha.2025.118815
107. Zou Y, Li S, Li Y, Zhang D, Zheng M, Shi B. Glioblastoma cell derived exosomes as a potent vaccine platform targeting primary brain cancers and brain metastases. *ACS Nano*. 2025. doi:10.1021/acsnano.4c14573
108. Lv B, Zhao Y, Li G, et al. Tumor-resident intracellular bacteria scavenger activated in situ vaccines for potent cancer photoimmunotherapy. *Adv Healthc Mater*. 2025;14:e2404271. doi:10.1002/adhm.202404271
109. Xu J, Liu H, Wang T, et al. CCR7 mediated mimetic dendritic cell vaccine homing in lymph node for head and neck squamous cell carcinoma therapy. *Adv Sci*. 2023;10(17):e2207017. doi:10.1002/advs.202207017
110. Rezaei R, Baghaei K, Hashemi SM, Zali MR, Ghanbarian H, Amani D. Tumor-derived exosomes enriched by miRNA-124 promote anti-tumor immune response in CT-26 tumor-bearing mice. *Front Med*. 2021;8:619939. doi:10.3389/fmed.2021.619939
111. Zhu H, Wang K, Wang Z, et al. An efficient and safe MUC1-dendritic cell-derived exosome conjugate vaccine elicits potent cellular and humoral immunity and tumor inhibition in vivo. *Acta Biomater*. 2022;138:491–504. doi:10.1016/j.actbio.2021.10.041
112. Huang L, Rong Y, Tang X, et al. Engineered exosomes as an in situ DC-primed vaccine to boost antitumor immunity in breast cancer. *Mol Cancer*. 2022;21(1):45. doi:10.1186/s12943-022-01515-x
113. Zuo B, Zhang Y, Zhao K, et al. Universal immunotherapeutic strategy for hepatocellular carcinoma with exosome vaccines that engage adaptive and innate immune responses. *J Hematol Oncol*. 2022;15(1):46. doi:10.1186/s13045-022-01266-8
114. Wang X, Zhang Y, Chung Y, et al. Tumor vaccine based on extracellular vesicles derived from γδ-T cells exerts dual antitumor activities. *J Extracell Vesicles*. 2023;12(9):e12360. doi:10.1002/jev2.12360

115. Souza SVP, Aguiar ACV, Albuquerque ECS, et al. M1 macrophage extracellular vesicles and TLR3 Agonist nanoparticles down-regulate immunosuppression and metastasis via AKT/TAM in triple-negative breast cancer. *Mol Carcinog*. 2025;64:1450–1461. doi:10.1002/mc.70003
116. Liu M, Hu S, Yan N, Popowski KD, Cheng K. Inhalable extracellular vesicle delivery of IL-12 mRNA to treat lung cancer and promote systemic immunity. *Nat Nanotechnol*. 2024;19:565–575. doi:10.1038/s41565-023-01580-3
117. Chen G, Huang AC, Zhang W, et al. Exosomal PD-L1 contributes to immunosuppression and is associated with anti-PD-1 response. *Nature*. 2018;560(7718):382–386. doi:10.1038/s41586-018-0392-8
118. Wang Y, Li P, Mao S, et al. Exosome CTLA-4 regulates PTEN/CD44 signal pathway in spleen deficiency internal environment to promote invasion and metastasis of hepatocellular carcinoma. *Front Pharmacol*. 2021;12:757194. doi:10.3389/fphar.2021.757194
119. Liu H, Zhao Z, Zhang L, et al. Discovery of low-molecular weight anti-PD-L1 peptides for cancer immunotherapy. *J Immunother Cancer*. 2019;7(1):270. doi:10.1186/s40425-019-0705-y
120. Xie F, Xu M, Lu J, Mao L, Wang S. The role of exosomal PD-L1 in tumor progression and immunotherapy. *Mol Cancer*. 2019;18(1):146. doi:10.1186/s12943-019-1074-3
121. Ratajczak K, Grel H, Olejnik P, Jakiela S, Stobiecka M. Current progress, strategy, and prospects of PD-1/PDL-1 immune checkpoint biosensing platforms for cancer diagnostics, therapy monitoring, and drug screening. *Biosens Bioelectron*. 2023;240:115644. doi:10.1016/j.bios.2023.115644
122. Yuan X, Liu X, Jiang D, et al. Exosomal PD-L1 derived from hypoxia nasopharyngeal carcinoma cell exacerbates CD8(+) T cell suppression by promoting PD-L1 upregulation in macrophages. *Cancer Immunol Immunother*. 2025;74(7):220. doi:10.1007/s00262-025-04047-7
123. Li C, Li C, Zhi C, et al. Clinical significance of PD-L1 expression in serum-derived exosomes in NSCLC patients. *J Transl Med*. 2019;17(1):355. doi:10.1186/s12967-019-2101-2
124. Zhang C, Wu Q, Gong Y, et al. Biomimetic exosomal vesicles loaded with siRNA improves antitumor immune responses by inhibiting the secretion of tumor-derived exosome PD-L1. *Int Immunopharmacol*. 2024;129:111659. doi:10.1016/j.intimp.2024.111659
125. Zhu T, Chen Z, Jiang G, Huang X. Sequential targeting hybrid nanovesicles composed of chimeric antigen receptor T-cell-derived exosomes and liposomes for enhanced cancer immunochemotherapy. *ACS Nano*. 2023;17(17):16770–16786. doi:10.1021/acsnano.3c03456
126. Fan M, Liu H, Yan H, et al. A CAR T-inspiring platform based on antibody-engineered exosomes from antigen-feeding dendritic cells for precise solid tumor therapy. *Biomaterials*. 2022;282:121424. doi:10.1016/j.biomaterials.2022.121424
127. Jiang Y, Xu X, Fan D, et al. Advancing tumor-targeted chemo-immunotherapy: development of the CAR-M-derived exosome-drug conjugate. *J Med Chem*. 2024;67(16):13959–13974. doi:10.1021/acs.jmedchem.4c00753
128. Tao B, Du R, Zhang X, et al. Engineering CAR-NK cell derived exosome disguised nano-bombs for enhanced HER2 positive breast cancer brain metastasis therapy. *J Control Release*. 2023;363:692–706. doi:10.1016/j.jconrel.2023.10.007
129. Wagner AK, Gehrmann U, Hiltbrunner S, et al. Soluble and exosome-bound α-galactosylceramide mediate preferential proliferation of educated NK cells with increased anti-tumor capacity. *Cancers*. 2021;13(2):298. doi:10.3390/cancers13020298
130. Wu Q, Qian W, Sun X, Jiang S. Small-molecule inhibitors, immune checkpoint inhibitors, and more: FDA-approved novel therapeutic drugs for solid tumors from 1991 to 2021. *J Hematol Oncol*. 2022;15(1):143. doi:10.1186/s13045-022-01362-9
131. Yong T, Zhang X, Bie N, et al. Tumor exosome-based nanoparticles are efficient drug carriers for chemotherapy. *Nat Commun*. 2019;10(1):3838. doi:10.1038/s41467-019-11718-4
132. Rothwell PM, Wilson M, Price JF, Belch JF, Meade TW, Mehta Z. Effect of daily aspirin on risk of cancer metastasis: a study of incident cancers during randomised controlled trials. *Lancet*. 2012;379(9826):1591–1601. doi:10.1016/s0140-6736(12)60209-8
133. Suo M, Shen H, Lyu M, et al. Biomimetic nano-cancer stem cell scavenger for inhibition of breast cancer recurrence and metastasis after FLASH-radiotherapy. *Small*. 2024;20(29):e2400666. doi:10.1002/smll.202400666
134. Wang J, Tang W, Yang M, et al. Inflammatory tumor microenvironment responsive neutrophil exosomes-based drug delivery system for targeted glioma therapy. *Biomaterials*. 2021;273:120784. doi:10.1016/j.biomaterials.2021.120784
135. Tang Z, Tang C, Sun C, Ying X, Shen R. M1 macrophage-derived exosomes synergistically enhance the anti- bladder cancer effect of gemcitabine. *Aging*. 2022;14(18):7364–7377. doi:10.18632/aging.204200
136. Wang P, Wang H, Huang Q, et al. Exosomes from M1-polarized macrophages enhance paclitaxel antitumor activity by activating macrophages-mediated inflammation. *Theranostics*. 2019;9(6):1714–1727. doi:10.7150/thno.30716
137. Han Y, Zheng W, Zhang Y, et al. Oxaliplatin-loaded natural killer cell-derived exosomes for a safe and efficient chemoimmunotherapy of colorectal cancer. *J Pharm Sci*. 2025;114(6):103783. doi:10.1016/j.xphs.2025.103783
138. Guo G, Tan Z, Liu Y, Shi F, She J. The therapeutic potential of stem cell-derived exosomes in the ulcerative colitis and colorectal cancer. *Stem Cell Res Ther*. 2022;13(1):138. doi:10.1186/s13287-022-02811-5
139. Li C, Wen Y, Wang J, et al. Human mesenchymal stem cell-derived exosomes as engineering vehicles of daunorubicin for targeted c-Mpl+ AML therapy. *Int J Nanomed*. 2025;20:5267–5289. doi:10.2147/ijn.S511713
140. Attar FA, Irani S, Oloomi M, Bolhassani A, Geranpayeh L, Atyabi F. Doxorubicin loaded exosomes inhibit cancer-associated fibroblasts growth: in vitro and in vivo study. *Cancer Cell Int*. 2025;25(1):72. doi:10.1186/s12935-025-03689-y
141. Zheng Y, Wang T, Zhang J, et al. Plant-derived nanovesicles: a promising frontier in tissue repair and antiaging. *J Agric Food Chem*. 2025. doi:10.1021/acs.jafc.5c01547
142. Lu X, Han Q, Chen J, et al. Celery (Apium graveolens L.) exosome-like nanovesicles as a new-generation chemotherapy drug delivery platform against tumor proliferation. *J Agric Food Chem*. 2023. doi:10.1021/acs.jafc.2c07760
143. Zhang M, Xiao B, Wang H, et al. Edible ginger-derived nano-lipids loaded with doxorubicin as a novel drug-delivery approach for colon cancer therapy. *Mol Ther*. 2016;24(10):1783–1796. doi:10.1038/mt.2016.159
144. Xiao Q, Zhao W, Wu C, et al. Lemon-derived extracellular vesicles nanodrugs enable to efficiently overcome cancer multidrug resistance by endocytosis-triggered energy dissipation and energy production reduction. *Adv Sci*. 2022;9(20):e2105274. doi:10.1002/advs.202105274