BMB
Reports

BMB Rep. 2023; 56(2): 71-77
www.bmbreports.org

Contributed Mini Review

Engineered adult stem cells: a promising tool for anti-cancer therapy

*Youngdong Choi, Hong Kyu Lee & Kyung-Chul Choi**
Laboratory of Biochemistry and Immunology, College of Veterinary Medicine, Chungbuk National University, Cheongju 28644, Korea

Cancers are one of the most dreaded diseases in human history and have been targeted by numerous trials including surgery, chemotherapy, radiation therapy, and anti-cancer drugs. Adult stem cells (ASCs), which can regenerate tissues and repair damage, have emerged as leading therapeutic candidates due to their homing ability toward tumor foci. Stem cells can precisely target malicious tumors, thereby minimizing the toxicity of normal cells and unfavorable side effects. ASCs, such as mesenchymal stem cells (MSCs), neural stem cells (NSCs), and hematopoietic stem cells (HSCs), are powerful tools for delivering therapeutic agents to various primary and metastatic cancers. Engineered ASCs act as a bridge between the tumor sites and tumoricidal reagents, producing therapeutic substances such as exosomes, viruses, and anti-cancer proteins encoded by several suicide genes. This review focuses on various anti-cancer therapies implemented via ASCs and summarizes the recent treatment progress and shortcomings. [BMB Reports 2023; 56(2): 71-77]

INTRODUCTION

Cancer is one of the most lethal diseases that has been treated since the existence of human beings. Approximately 19.3 million new patients and nearly 10.0 million cancer deaths were reported worldwide in 2020 (1). In the past few decades, before new therapies were discovered, a combination of conventional treatments such as surgical resection, chemo- and radiation therapies were first-line treatments for many types of primary cancers. Although conventional treatment methods are effective in the early stages of cancer, these treatments are non-selectivity and can cause adverse effects (2). For instance, lymphedema, which is the accumulation of lymph fluid in tissues, can be caused by damage to lymph nodes or blood vessels resulting from surgery or radiation therapy (3). Chemotherapy drugs can cause neutropenia by targeting fast-growing cells such as healthy white blood cells (4). Therefore, studies on a variety of therapeutic interventions for improving the selectivity of conventional cancer treatment are in progress. On that note, numerous anticancer therapies and reagents have emerged and stem cell-mediated therapies using adult stem cells (ASCs) are considered attractive vehicles for delivering anticancer agents (Fig. 1). ASCs, such as mesenchymal stem cells (MSCs), neural stem cells (NSCs), and hematopoietic stem cells (HSCs) are undifferentiated populations that exist in our bodies, and have tumor tropic effect and the potential to partially replace conventional treatments (5).

An important characteristic of adult stem cells is their outstanding migratory and homing ability (6). The migratory ability of ASCs was initially elucidated in a xenograft mouse model with brain tumors (7). The exact mechanism of ASCs tumor tropism is not fully elucidated, but several experimental evidence indicates that various chemokines such as monocyte chemoattractant protein-1, stromal cell-derived factor 1, and stem cell factor-1 are involved (8-11). The efficacy of NSC tropism has been reported in gliomas and other malignancies. It has been demonstrated that NSCs preferentially trace metastatic tumor foci in various mice organs. Intravenous injected NSCs or MSCs can migrate to metastatic nodes of breast cancer in the lungs (12-16). Because of these abilities, adult stem cells have been actively studied as intermediate carriers of anticancer substances that track and kill cancer from the past to the present.

STEM CELL-MEDIATED THERAPIES USING MSCs AND NSCs

Prodrug-enzyme therapy

Utilizing their migration ability to tumor sites, MSCs and NSCs are genetically engineered to express therapeutic genes. The suicide gene cytosine deaminase (CD), found in *Escherichia coli* or yeast, converts the prodrug 5-fluorocytosine (5-FC) to the widely used cytotoxic anticancer drug 5-fluorouracil (5-FU) (17-19), which subsequently converts to the activate metabolites, inhibiting RNA and DNA synthesis and functioning of thymidylate synthase in the tumor (18). CD expressing human MSCs, such as adipose tissue-derived MSCs and a clonal immortalized NSC HB1.F3.CD, are both genetically engineered to express

*Corresponding author. Tel: +82-43-261-3664; Fax: +82-43-267-3150; E-mail: kchoi@cbu.ac.kr

https://doi.org/10.5483/BMBRep.2022-0091

**Received 28 May 2022, Revised 28 July 2022,
Accepted 4 November 2022, Published online 31 January 2023**

Keywords: Anti-cancer effect, Engineered adult stem cells, Gene therapy

ISSN: 1976-670X (electronic edition)

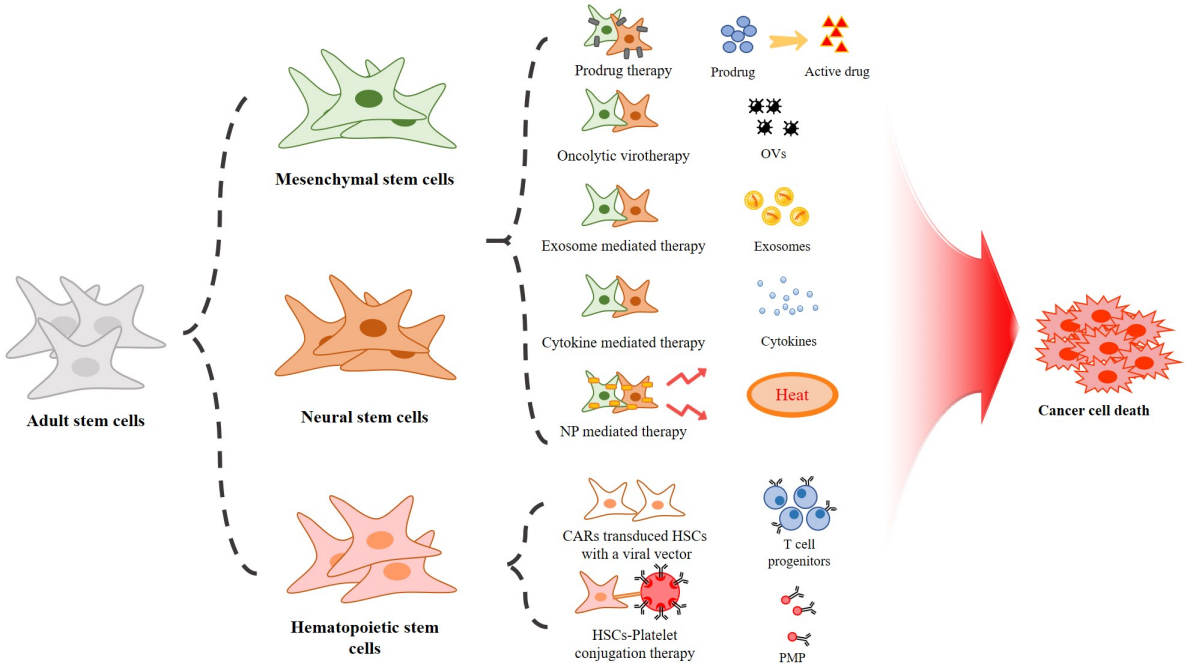

Fig. 1. Overview of the ASCs mediated anti-cancer treatments. ASCs mediated anti-cancer therapies are differentiated according to the use of MSCs, NSCs, and HSCs. Both MSCs and NSCs use the same carrier to deliver anti-cancer substances such as prodrug, oncolytic virus, exosomes, cytokines and photothermal heat. Viral transduced HSCs are localized in the bone marrow, and produce T cell progenitors that undergo normal immune cell maturation. Subsequently, immune cells expressing TCR that recognizes cancer-associated antigen, confer specific anti-cancer activity for a long time. Conjugation between HSCs and platelet adequately delivers immune checkpoint inhibitors to the bone marrow, and augments an anti-leukemia immune response.

the CD suicide gene (20, 21). CD-NSCs have been especially studied in malignant gliomas as a first-generation enzyme/prodrug system to overcome challenges associated with the delivery of anticancer compounds and the replacement of damaged tissues (22, 23). Research using yeast CD gene; yeast cytosine deaminase::uracil phosphoribosyltransferase (yCD::UPRT) has been studied and applied in MSCs (24). Numerous *in vitro* and *in vivo* studies have extensively demonstrated the safety and therapeutic efficacy of expressing yCD::UPRT and *E. coli* CD gene (25-27). Additionally, a clinical trial using CD and 5-FC to treat high-grade gliomas has also been applied and completed (NCT01172964 and NCT04657315) (28).

Likewise, carboxylesterases are enzymes that alter the prodrug CPT-11 (irinotecan) to its active form SN-38 (7-ethyl-10-hydroxycamptothecin), a robust inhibitor of topoisomerase I (29-31). The pro-drug CPT-11 is currently being used in combination with other anti-cancer drugs for neuroblastoma and has progressed well in phase I clinical trials (32). The adenovirus-transduced HB1.F3.CD (human NSCs) with human enzyme is around 70-fold more efficient at converting CPT-11 to SN-38, than the endogenous wild-type human isoform of CEs in the liver (33, 34). The herpes simplex virus thymidine kinase (HSV-TK) is an enzyme that is widely studied for another suicide gene strategy, using the pro-drug ganciclovir (GCV) (35, 36). Applying the same transduced HB1.F3.CD, HSV-TK phosphorylates GCV into a monophosphate active form, which has the potential to effectively eliminate cancer (37).

Oncolytic virotherapy

Oncolytic viruses (OVs) preferentially target, replicate in, and eliminate cancer cells, rather than other normal cells (38). OVs can induce both innate and adaptive immunity (39). These characteristics have given new directions for anti-cancer therapy. Several oncolytic viruses have been tested in tumor-bearing animal models and have garnered major scientific interest. Viral transduction of MSCs is frequently performed in adeno-, lenti-, and retro- viruses (40). The major obstacle of oncolytic virotherapy is viral destruction by host immunity. Several studies have shown that anti-viral destruction is mediated by endogenous interferon signaling (41). To overcome this limitation, MSC-mediated oncolytic virotherapy has been considered due to their tumor-tropic ability towards tumor foci and immunosuppressive functions (42, 43). MSCs produce several soluble factors and enzymes, such as cyclooxygenase pro-inflammatory cytokine interferon-γ (43). Previous studies have reported that MSCs specifically suppress immune cells, including the acti-

vity of T cells, B cells, and maturation of dendritic cells. These features provide robust shelters to safely deliver OVs to tumor sites (44). There has been a lot of research to improve the therapeutic efficacy of OVs. For example, Yoon AR *et al*. inserted Wnt-signaling inhibiting decoy acceptor sequences into an adenovirus, targeting hepatocellular carcinoma (HCC). This resulted in highly activating the Wnt signaling pathway and effectively destroying HCC. Additionally, reports indicate that MSC-mediated virotherapy prevents unpredictable side effects caused by MSCs due to the cells being lysed by adenovirus replication *in vivo* (45).

NSC-mediated virotherapy has been studied especially in gliomas and ovarian cancer. Aboody *et al*. reported that immortalized HB1.F3.CD21 (engineered neural stem cell line) in non-tumorigenic as well as enhancing tumor foci, inhibited the replication of adenovirus in glioma and ovarian cancer models. This oncolytic virus-loaded NSC model is currently in a clinical trial of glioma patients (46). In addition, injection of NSCs with oncolytic adenovirus (NSC-CRAd-S-pk7) successfully distributes the tumor area and allows the virus to deeper penetrate the tumor periphery (47).

Extracellular vesicle-mediated anti-cancer therapy

Extracellular vesicles (EV) are heterogeneous lipid bilayer particles that are produced by many types of cells including stem cells (48). Exosomes are extracellular membrane-bound vesicles ranging from 50-100 nm in diameter, and comprising protein, DNA, and RNA secreted by cells (49). As versatile therapeutic cargo, exosomes have several benefits. For instance, exosomes are involved in various biological processes such as antigen presentation, angiogenesis, and transferring horizontal miRNA (50). Alvarez-Erviti L *et al*. reported that in a mouse model, systemic injection of dendritic cell-derived exosome effectively delivered siRNA to the brain (51).

Unformulated antisense oligonucleotide (ASO) inhibitors have proven anticancer efficacy in multiple preclinical cancer models (52). ASO AZD9150 and CpG-STAT3ASO conjugates target the inhibition of oncogenic signal transducer and activator of transcription 3 (STAT3), which is an essential promoter of numerous human cancers (53). Adamus T *et al*. assessed the clinical efficacy of NSC-derived exosomes in the glioma as cargo for the delivery of oligonucleotides. Peritumorally administrated NSCs producing exosomes encapsulated CpG-STAT3ASO have demonstrated that NSC-mediated delivery significantly elevates the percentage of CpG-STAT3ASO and effectively reduces tumor growth, compared to the administration of oligonucleotides alone (54).

The previously mentioned CD/5-FC system has also been used as a therapeutic reagent of the exosome. It has been reported that MSCs engineered to express the CD gene released exosomes contained CD mRNA. The exosomes were internalized by cancer cells and triggers apoptosis due to the conversion of prodrug 5-FC to active drug 5-FU (55). MSC-EVs carrying therapeutic substances such as miRNA were also shown to have anti-cancer effects. In a previous study, it was shown that miRNA-148-3p transferred by human umbilical cord-derived MSC induce apoptosis by regulating tripartite motif 59 in MDA-MB-231 breast cancer xenografts (56).

Cytokine mediated therapy

Cytokines, small molecules normally produced by immune cells, are involved in the regulation of immune responses, and comprise interleukins, chemokines, tumor necrosis factors, and lymphokines (57). In the mid-1990s, a high dose of interleukin 2 was found to be an effective anti-cancer agent, leading to the commencement of cytokine-based therapy (58). Since then, a variety of cytokines have been evaluated for anti-cancer efficacy. However, systemic administration of immunomodulating cytokines demonstrated several shortcomings. Because of the short half-lives of cytokines, administration of high doses of cytokines can cause adverse side effects, including cardiac problems. To prevent unfavorable side effects, ASCs are an appropriate model for delivering cytokines precisely to the tumor environment. IFN-β-inserted MSCs were the first cytokine-engineered MSCs, which inhibited the growth of melanoma *in vivo*. Also, it was reported for the first time, that MSCs were positioned into the tumor and produced large quantities of IFN-β (59).

Subsequent studies confirmed that diverse cytokines and chemokines, such as the interleukin and interferon families, act as tumoricidal reagents when inserted in MSCs. Interleukin 7, interleukin 2, interleukin 12 (IL-12), interleukin 15, and interleukin 18 are reported as immunoregulatory cellular cytokines against cancer (60). Viral transduced MSCs of IL-12 could migrate to the tumor site, where the MSCs locally express the IL-12 gradually without disturbing other systems, thereby minimizing the inherent adverse side effects (61). MSCs overexpressing tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) showed a 6-fold increase in the ability to kill tumor cells, as compared to the non-modified MSCs (12). MSCs expressing the death receptors TRAIL-R1 and TRAIL-R2 resulted in TRAIL-induced apoptosis of tumor cells expressing the TRAIL death receptors (62). Additionally, a clinical study is being conducted on the therapeutic efficacy of TRAIL for patients (NCT03298763).

Nanoparticle-loaded ASCs anti-cancer therapy

The use of nanoparticle (NP) carriers as a delivery system for selective anti-cancer agents attenuates unfavorable side effects and improves chemotherapeutic pharmacologic properties by altering the biodistribution and pharmacokinetics (63). ASCs such as MSCs and NSCs are considered appropriate vehicles for delivering therapeutic NPs while overcoming drawbacks of traditional chemotherapy, such as suboptimal penetration and retention. Various cationic polymers that destruct tumors chemically or physically as ASC-NP conjugated or in an internalized form have been widely studied (64). For instance, gold nanorods (AuNRs) improve several drawbacks, including damage

to the surrounding healthy tissue due to the complex tumor geometries of photothermal therapy, *i.e.*, heat generation when a tumor is exposed to near-infrared radiation (65). Positioning the AuNRs within the tumor tissue converts the 810 nm non-harmful laser light into thermal energy, raising the local temperature to 28-60^{o}C and leading to tumor resorption (66). To overcome this positioning issue, NSCs were selected to deliver AuNRs to the tumor foci. NSC-AuNR conjugates resulted in enhanced tumor surrounding the ability of AuNRs, resulting in improved anti-cancer efficacy, compared to administration of AuNRs alone (67). Mesoporous silica nanoparticles can transport the drug to the tumor site effectively, because of their surface pores. The anti-cancer drug can be released by certain stimuli such as pH, temperature, magnetic fields, and ultrasound (68, 69). It has been reported that mesoporous silica-loaded MSCs release doxorubicin and induce apoptosis of cancer cells when the MSCs are stimulated by ultrasound (70).

STEM CELL-MEDIATED THERAPIES USING HSCs

HSCs engineered with chimeric antigen receptors for anticancer therapy

HSCs are promising candidates for anticancer therapy, especially leukemia, through the transduction of genes that encode chimeric antigen receptors (CARs) or T cell receptors (TCRs). CARs and TCRs recognize tumor-associated antigens directly, in a major histocompatibility complex (MHC)-independent manner (71). Although these methods have yielded unprecedented competency in B cell acute lymphoblastic leukemia, several challenges still need to be overcome, such as reduced T cell persistence and activity. Besides, the use of mature T cells has a possibility for mispairing, *i.e.*, a fusion of the introduced transgenic TCR chains with endogenous chains. Gene-editing technologies have enabled the elimination of the endogenous TCR and class-1 MHC-I to minimize graft-versus-host disease (GvHD) and allograft rejection (72). Another approach is the transplantation of HSCs. Using TCR- or CAR- transduced HSCs against cancers (especially leukemia) provides some advantages, which simultaneously resolves the challenges of both T cell persistence and TCR mismatching (73). First, the TCR- or CAR-transduced HSCs will constantly produce T-lymphocyte progenitors after transplantation, which thereafter undergo the natural T cell maturity process, developing potential immunity of the recipient. Next, unfavorable cytotoxicity of engineered TCR due to mispairing from transduced mature T cells is potently suppressed by HSCs that have not yet been rearranged. Giannoni F *et al*. reported that engraftment of transduced HSCs enhances the T cells expressing TCR recognizing tumor-associated antigen, which exerts specific anti-tumor activity (74).

HSC-platelet conjugation-mediated therapy

Hu *et al*. reported that monoclonal anti-programmed death-1 antibodies (aPD-1) are secreted from the HSC-platelet-aPD-1 (S-P-aPD-1) cellular complex due to platelet activation. The immune checkpoint inhibitor aPD-1 blocks programmed death-1 (PD-1) that are expressed in various immune cells and disturbs the immunosuppressing ability of cancer through PD-1 ligands (PD-L1) (75). S-P-aPD-1 effectively promotes the delivery of aPD-1, and leukemia-specific T cells induce tumor growth suppression in the bone marrow, which is the site of acute myeloid leukemia (76).

CONCLUSION

ASCs are attractive models and give new insight into anti-cancer therapy, but the application of ASCs will not completely replace previous traditional treatments such as surgery or radiation therapy. Stem cell therapy for cancer may be an attractive choice appropriate for patients diagnosed with metastasized malignancy, who have fewer treatment options than patients in the early stages. The Discovery of the tumor tropism of ASCs has led to the development of varied anti-cancer approaches. Of the various methods reported, engineered stem cell-mediated therapies have shown anti-cancer efficacy not only in various primary cancers, but also in metastatic malignancies (77). As a therapeutic agent, engineered ASC-secreted materials such as oncolytic viruses, cytokines, and proteins exert anti-cancer activity, and subsequently induce the shrinkage of tumor volume via cancer cell death. Although the outcome of ASC-mediated anti-cancer therapy is remarkable, several limitations and concerns about safety and shortcomings still remain. As with other anti-cancer therapies, a single treatment of ASC therapies does not guarantee complete eradication of the tumor and prevent tumor recurrence. Moreover, stem cells that migrate to the tumor site must remain only long enough to secrete the therapeutic agent, but the permanent alive immortalized ASCs should be avoided due to the possibility of them becoming tumorigenic. Portnow J *et al*. reported this based on first-in-human autopsy data, which stated that only infinitesimal NSCs remain longer than the required period of prodrug conversion. Additionally, it was reassuring to note that NSCs did not seem to undergo division (Proliferating cell nuclear antigen negative) and were positioned as single cells in the tumor (28). Another obstacle to cell-mediated therapy is unequal biodistribution in the tumor foci after stem cell administration. Stem cells trace tumor sites through leaky vessels of the tumor. Depending on the size and type of the tumors, varying degrees of blood vessel leakiness cause disruptions in the biodistribution and dose-response correlation. Moreover, engraftment of allogeneic immune cells and stem cells has the potential to possibly cause infections or GvHD (78, 79). To elucidate these concerns, the increasing number of preclinical and clinical researchers is aiding in developing and standardizing clinical trials. Advanced findings may guide investigators in the next phase of stem cell therapies through cutting-edge technologies, and a better understanding of cancer physiology.

ACKNOWLEDGEMENTS

This work was supported by the Basic Science Research Program (2020R1A2C2006060) and the Global Research and Development Center (GRDC) Program (2017K1A4A3014959) through the National Research Foundation (NRF) of Korea, funded by the Ministry of Science and ICT. In addition, this work was also supported by the Basic Research Lab Program (2022R1A4A1025557) through the National Research Foundation (NRF) of Korea, funded by the Ministry of Science and ICT.

CONFLICTS OF INTEREST

The authors have no conflicting interests.

REFERENCES

1. Sung H, Ferlay J, Siegel RL et al (2021) Global Cancer Statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 71, 209-249
2. Nurgali K, Jagoe RT and Abalo R (2018) Editorial: adverse effects of cancer chemotherapy: anything new to improve tolerance and reduce sequelae? Front Pharmacol 9, 245
3. Kayiran O, De La Cruz C, Tane K and Soran A (2017) Lymphedema: from diagnosis to treatment. Turk J Surg 33, 51-57
4. Harrold J, Gisleskog PO, Delor I et al (2020) Quantification of radiation injury on neutropenia and the link between absolute neutrophil count time course and overall survival in nonhuman primates treated with G-CSF. Pharm Res 37, 102
5. Gurusamy N, Alsayari A, Rajasingh S and Rajasingh J (2018) Adult stem cells for regenerative therapy. Prog Mol Biol Transl Sci 160, 1-22
6. Gage FH (2000) Mammalian neural stem cells. Science 287, 1433-1438
7. Aboody KS, Brown A, Rainov NG et al (2000) Neural stem cells display extensive tropism for pathology in adult brain: evidence from intracranial gliomas. Proc Natl Acad Sci U S A 97, 12846-12851
8. Erlandsson A, Larsson J and Forsberg-Nilsson K (2004) Stem cell factor is a chemoattractant and a survival factor for CNS stem cells. Exp Cell Res 301, 201-210
9. Widera D, Holtkamp W, Entschladen F et al (2004) MCP-1 induces migration of adult neural stem cells. Eur J Cell Biol 83, 381-387
10. Imitola J, Raddassi K, Park KI et al (2004) Directed migration of neural stem cells to sites of CNS injury by the stromal cell-derived factor 1alpha/CXC chemokine receptor 4 pathway. Proc Natl Acad Sci U S A 101, 18117-18122
11. Sun L, Lee J and Fine HA (2004) Neuronally expressed stem cell factor induces neural stem cell migration to areas of brain injury. J Clin Invest 113, 1364-1374
12. Loebinger MR, Eddaoudi A, Davies D and Janes SM (2009) Mesenchymal stem cell delivery of TRAIL can eliminate metastatic cancer. Cancer Res 69, 4134-4142
13. Kidd S, Spaeth E, Dembinski JL et al (2009) Direct evidence of mesenchymal stem cell tropism for tumor and wounding microenvironments using in vivo bioluminescent imaging. Stem Cells 27, 2614-2623
14. Frank RT, Edmiston M, Kendall SE et al (2009) Neural stem cells as a novel platform for tumor-specific delivery of therapeutic antibodies. PLoS One 4, e8314
15. Rachakatla RS, Pyle MM, Ayuzawa R et al (2008) Combination treatment of human umbilical cord matrix stem cell-based interferon-beta gene therapy and 5-fluorouracil significantly reduces growth of metastatic human breast cancer in SCID mouse lungs. Cancer Invest 26, 662-670
16. Joo KM, Park IH, Shin JY et al (2009) Human neural stem cells can target and deliver therapeutic genes to breast cancer brain metastases. Mol Ther 17, 570-575
17. Dachs GU, Tupper J and Tozer GM (2005) From bench to bedside for gene-directed enzyme prodrug therapy of cancer. Anticancer Drugs 16, 349-359
18. Longley DB, Harkin DP and Johnston PG (2003) 5-fluorouracil: mechanisms of action and clinical strategies. Nat Rev Cancer 3, 330-338
19. Kilstrup M, Meng LM, Neuhard J and Nygaard P (1989) Genetic evidence for a repressor of synthesis of cytosine deaminase and purine biosynthesis enzymes in Escherichia coli. J Bacteriol 171, 2124-2127
20. Kucerova L, Altanerova V, Matuskova M, Tyciakova S and Altaner C (2007) Adipose tissue-derived human mesenchymal stem cells mediated prodrug cancer gene therapy. Cancer Res 67, 6304-6313
21. Gutova M, Flores L, Adhikarla V et al (2019) Quantitative evaluation of intraventricular delivery of therapeutic neural stem cells to orthotopic glioma. Front Oncol 9, 68
22. Najbauer J, Huszthy PC, Barish ME et al (2012) Cellular host responses to gliomas. PLoS One 7, e35150
23. Ao Q, Wang AJ, Chen GQ, Wang SJ, Zuo HC and Zhang XF (2007) Combined transplantation of neural stem cells and olfactory ensheathing cells for the repair of spinal cord injuries. Med Hypotheses 69, 1234-1237
24. Ko TP, Lin JJ, Hu CY, Hsu YH, Wang AH and Liaw SH (2003) Crystal structure of yeast cytosine deaminase. Insights into enzyme mechanism and evolution. J Biol Chem 278, 19111-19117
25. Nouri FS, Wang X and Hatefi A (2015) Genetically engineered theranostic mesenchymal stem cells for the evaluation of the anticancer efficacy of enzyme/prodrug systems. J Control Release 200, 179-187
26. Ho YK, Woo JY, Tu GXE, Deng LW and Too HP (2020) A highly efficient non-viral process for programming mesenchymal stem cells for gene directed enzyme prodrug cancer therapy. Sci Rep 10, 14257
27. Amara I, Touati W, Beaune P and de Waziers I (2014) Mesenchymal stem cells as cellular vehicles for prodrug gene therapy against tumors. Biochimie 105, 4-11
28. Portnow J, Synold TW, Badie B et al (2017) Neural stem cell-based anticancer gene therapy: a first-in-human study in recurrent high-grade glioma patients. Clin Cancer Res 23, 2951-2960
29. Wierdl M, Morton CL, Weeks JK, Danks MK, Harris LC and Potter PM (2001) Sensitization of human tumor cells to CPT-11 via adenoviral-mediated delivery of a rabbit

liver carboxylesterase. Cancer Res 61, 5078-5082
30. Danks MK and Potter PM (2004) Enzyme-prodrug systems: carboxylesterase/CPT-11. Methods Mol Med 90, 247-262
31. Bachoud-Levi AC, Remy P, Nguyen JP et al (2000) Motor and cognitive improvements in patients with Huntington's disease after neural transplantation. Lancet 356, 1975-1979
32. DuBois SG, Marachelian A, Fox E et al (2016) Phase i study of the aurora a kinase inhibitor alisertib in combination with irinotecan and temozolomide for patients with relapsed or refractory neuroblastoma: a nant (new approaches to neuroblastoma therapy) trial. J Clin Oncol 34, 1368-1375
33. Wierdl M, Tsurkan L, Hyatt JL et al (2008) An improved human carboxylesterase for enzyme/prodrug therapy with CPT-11. Cancer Gene Ther 15, 183-192
34. Metz MZ, Gutova M, Lacey SF et al (2013) Neural stem cell-mediated delivery of irinotecan-activating carboxylesterases to glioma: implications for clinical use. Stem Cells Transl Med 2, 983-992
35. Miller WH and Miller RL (1980) Phosphorylation of acyclovir (acycloguanosine) monophosphate by GMP kinase. J Biol Chem 255, 7204-7207
36. Bhaumik S (2011) Advances in imaging gene-directed enzyme prodrug therapy. Curr Pharm Biotechnol 12, 497-507
37. Wang C, Natsume A, Lee HJ et al (2012) Neural stem cell-based dual suicide gene delivery for metastatic brain tumors. Cancer Gene Ther 19, 796-801
38. Davola ME and Mossman KL (2019) Oncolytic viruses: how "lytic" must they be for therapeutic efficacy? Oncoimmunology 8, e1581528
39. Lemos de Matos A, Franco LS and McFadden G (2020) Oncolytic viruses and the immune system: the dynamic duo. Mol Ther Methods Clin Dev 17, 349-358
40. Sage EK, Thakrar RM and Janes SM (2016) Genetically modified mesenchymal stromal cells in cancer therapy. Cytotherapy 18, 1435-1445
41. Ebrahimi S, Ghorbani E, Khazaei M et al (2017) Interferon-mediated tumor resistance to oncolytic virotherapy. J Cell Biochem 118, 1994-1999
42. Salmasi Z, Hashemi M, Mahdipour E, Nourani H, Abnous K and Ramezani M (2020) Mesenchymal stem cells engineered by modified polyethylenimine polymer for targeted cancer gene therapy, in vitro and in vivo. Biotechnol Prog 36, e3025
43. Gao F, Chiu SM, Motan DA et al (2016) Mesenchymal stem cells and immunomodulation: current status and future prospects. Cell Death Dis 7, e2062
44. Wang X, Zhao X and He Z (2021) Mesenchymal stem cell carriers enhance anti-tumor efficacy of oncolytic virotherapy. Oncol Lett 21, 238
45. Yoon AR, Hong J, Li Y et al (2019) Mesenchymal stem cell-mediated delivery of an oncolytic adenovirus enhances antitumor efficacy in hepatocellular carcinoma. Cancer Res 79, 4503-4514
46. Hammad M, Cornejo YR, Batalla-Covello J et al (2020) Neural stem cells improve the delivery of oncolytic chimeric orthopoxvirus in a metastatic ovarian cancer model. Mol Ther Oncolytics 18, 326-334
47. Morshed RA, Gutova M, Juliano J et al (2015) Analysis of glioblastoma tumor coverage by oncolytic virus-loaded neural stem cells using MRI-based tracking and histological reconstruction. Cancer Gene Ther 22, 55-61
48. Yanez-Mo M, Siljander PR, Andreu Z et al (2015) Biological properties of extracellular vesicles and their physiological functions. J Extracell Vesicles 4, 27066
49. Kalluri R and LeBleu VS (2020) The biology, function, and biomedical applications of exosomes. Science 367, eaau6977
50. Ha D, Yang N and Nadithe V (2016) Exosomes as therapeutic drug carriers and delivery vehicles across biological membranes: current perspectives and future challenges. Acta Pharm Sin B 6, 287-296
51. Alvarez-Erviti L, Seow Y, Yin H, Betts C, Lakhal S and Wood MJ (2011) Delivery of siRNA to the mouse brain by systemic injection of targeted exosomes. Nat Biotechnol 29, 341-345
52. Hong D, Kurzrock R, Kim Y et al (2015) AZD9150, a next-generation antisense oligonucleotide inhibitor of STAT3 with early evidence of clinical activity in lymphoma and lung cancer. Sci Transl Med 7, 314ra185
53. Huynh J, Chand A, Gough D and Ernst M (2019) Therapeutically exploiting STAT3 activity in cancer - using tissue repair as a road map. Nat Rev Cancer 19, 82-96
54. Moreira D, Adamus T, Zhao X et al (2018) STAT3 inhibition combined with CpG immunostimulation activates antitumor immunity to eradicate genetically distinct castration-resistant prostate cancers. Clin Cancer Res 24, 5948-5962
55. Altanerova U, Jakubechova J, Benejova K et al (2019) Prodrug suicide gene therapy for cancer targeted intracellular by mesenchymal stem cell exosomes. Int J Cancer 144, 897-908
56. Yuan L, Liu Y, Qu Y, Liu L and Li H (2019) Exosomes derived from MicroRNA-148b-3p-overexpressing human umbilical cord mesenchymal stem cells restrain breast cancer progression. Front Oncol 9, 1076
57. Berraondo P, Sanmamed MF, Ochoa MC et al (2019) Cytokines in clinical cancer immunotherapy. Br J Cancer 120, 6-15
58. Palmer PA, Vinke J, Evers P et al (1992) Continuous infusion of recombinant interleukin-2 with or without autologous lymphokine activated killer cells for the treatment of advanced renal cell carcinoma. Eur J Cancer 28A, 1038-1044
59. Studeny M, Marini FC, Champlin RE, Zompetta C, Fidler IJ and Andreeff M (2002) Bone marrow-derived mesenchymal stem cells as vehicles for interferon-beta delivery into tumors. Cancer Res 62, 3603-3608
60. Chulpanova DS, Kitaeva KV, Green AR, Rizvanov AA and Solovyeva VV (2020) Molecular aspects and future perspectives of cytokine-based anti-cancer immunotherapy. Front Cell Dev Biol 8, 402
61. Chen X, Lin X, Zhao J et al (2008) A tumor-selective biotherapy with prolonged impact on established metastases based on cytokine gene-engineered MSCs. Mol Ther 16, 749-756
62. Pan G, O'Rourke K, Chinnaiyan AM et al (1997) The receptor for the cytotoxic ligand TRAIL. Science 276, 111-113
63. Gao Z, Zhang L, Hu J and Sun Y (2013) Mesenchymal

stem cells: a potential targeted-delivery vehicle for anti-cancer drug, loaded nanoparticles. Nanomedicine 9, 174-184
64. Park JS, Na K, Woo DG et al (2010) Non-viral gene delivery of DNA polyplexed with nanoparticles transfected into human mesenchymal stem cells. Biomaterials 31, 124-132
65. von Maltzahn G, Park JH, Agrawal A et al (2009) Computationally guided photothermal tumor therapy using long-circulating gold nanorod antennas. Cancer Res 69, 3892-3900
66. Hirsch LR, Stafford RJ, Bankson JA et al (2003) Nanoshell-mediated near-infrared thermal therapy of tumors under magnetic resonance guidance. Proc Natl Acad Sci U S A 100, 13549-13554
67. Mooney R, Roma L, Zhao D et al (2014) Neural stem cell-mediated intratumoral delivery of gold nanorods improves photothermal therapy. ACS Nano 8, 12450-12460
68. Li Z, Clemens DL, Lee BY, Dillon BJ, Horwitz MA and Zink JI (2015) Mesoporous silica nanoparticles with pH-sensitive nanovalves for delivery of moxifloxacin provide improved treatment of lethal pneumonic tularemia. ACS Nano 9, 10778-10789
69. Guisasola E, Baeza A, Talelli M et al (2015) Magnetic-responsive release controlled by hot spot effect. Langmuir 31, 12777-12782
70. Paris JL, de la Torre P, Victoria Cabanas M et al (2017) Vectorization of ultrasound-responsive nanoparticles in placental mesenchymal stem cells for cancer therapy. Nanoscale 9, 5528-5537
71. Hong M, Clubb JD and Chen YY (2020) Engineering CAR-T cells for next-generation cancer therapy. Cancer Cell 38, 473-488
72. Liu X, Zhang Y, Cheng C et al (2017) CRISPR-Cas9-mediated multiplex gene editing in CAR-T cells. Cell Res 27, 154-157
73. Gschweng E, De Oliveira S and Kohn DB (2014) Hematopoietic stem cells for cancer immunotherapy. Immunol Rev 257, 237-249
74. Giannoni F, Hardee CL, Wherley J et al (2013) Allelic exclusion and peripheral reconstitution by TCR transgenic T cells arising from transduced human hematopoietic stem/progenitor cells. Mol Ther 21, 1044-1054
75. Tumeh PC, Harview CL, Yearley JH et al (2014) PD-1 blockade induces responses by inhibiting adaptive immune resistance. Nature 515, 568-571
76. Hu Q, Sun W, Wang J et al (2018) Conjugation of haematopoietic stem cells and platelets decorated with anti-PD-1 antibodies augments anti-leukaemia efficacy. Nat Biomed Eng 2, 831-840
77. Liu X, Hu J, Li Y et al (2018) Mesenchymal stem cells expressing interleukin-18 inhibit breast cancer in a mouse model. Oncol Lett 15, 6265-6274
78. Lagasse E, Connors H, Al-Dhalimy M et al (2000) Purified hematopoietic stem cells can differentiate into hepatocytes in vivo. Nat Med 6, 1229-1234
79. Dachs GU, Hunt MA, Syddall S, Singleton DC and Patterson AV (2009) Bystander or no bystander for gene directed enzyme prodrug therapy. Molecules 14, 4517-4545