Nanocarrier-mediated co-delivery of chemotherapeutic drugs and gene agents for cancer treatment

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Abstract The efficacy of chemotherapeutic drug in cancer treatment is often hampered by drug resistance of tumor cells, which is usually caused by abnormal gene expression. RNA interference mediated by siRNA and miRNA can selectively knock down the carcinogenic genes by targeting specific miRNAs. Therefore, combining chemotherapeutic drugs with gene agents could be a promising strategy for cancer therapy. Due to poor stability and solubility associated with gene agents and drugs, suitable protective carriers are needed and have been widely researched for the co-delivery. In this review, we summarize the most commonly used nanocarriers for co-delivery of chemotherapeutic drugs and gene agents, as well as the advances in co-delivery systems.

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Abbreviations: γ-CD, γ-cyclodextrin; ANG-CLP, angiopep-2 modified cationic liposome; CMC, critical micelle concentration; CPLA, cationic polylactide; DOTAP, 1,2-dioleoyl-3-trimethylammonium-propane; FA, folic acid; FCAP, ferrocenium capped amphiphilic pillar[5]arene; GSH, glutathione; miRNA, micro-RNA; OEI, oligoethylenimine; PAMAM, poly(amido amine); PAsp(AED), poly(N-(2,2'-dithiobis(ethylamine))aspartamide); PCL, poly(ε-caprolactone); PDMAEMA, poly(dimethylaminoethyl methacrylate); PDPA, poly(2-(diisopropyl amino)ethyl methacrylate); PEG, polyethylene glycol; PEI, poly(ethyleneimine); PEI-Pc, ferrocene modified poly(ethyleneimine); PEI-PChLg, poly(ethylene imine)-poly(γ-cholesterol-ε-glutamate); PEI-PCL, poly(ethyleneimine) and poly(ε-caprolactone); PLA, polylactic acid (or polylactide); PLGA, poly(lactic-co-glycolic acid); PnBA, poly(n-butyl acrylate); PPEEA, poly(2-aminoethyl ethylene phosphate); RNAi, RNA interference; siRNA, small interfering RNA; siVEGF, VEGF-targeted siRNA; SNPs, supramolecular nanoparticles; SSTRs, somatostatin receptors poly(N-(2,2'-dithiobis(ethylamine))aspartamide)

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1. Introduction

Cancer is one of the most devastating diseases and a leading cause of death in the world. According to the mortality data from the National Center for Health Statistics in 2013, one in four deaths in the United States is due to cancer. Chemotherapy is a treatment choice for many types of cancers, but its success is often hampered by development of drug resistance after repeated administration. Drug resistance has a genetic basis and it is caused by abnormal gene expression. There are several types of drug resistance, including efflux pumps which reduce the cellular concentration of the drug, alterations in membrane lipids that reduce cellular uptake, increased or altered drug targets, metabolic alteration of the drug, inhibition of apoptosis, repair of the damaged DNA, and the alteration of cell cycle checkpoints.

RNA interference (RNAi) is a special mechanism which occurs normally in most eukaryotic cells. RNAi mediated by small interfering RNA (siRNA) and microRNA (miRNA) have emerged as the most promising strategies for anti-cancer therapy, since siRNA and miRNA can induce gene-specific cleavage through their complementary pairing with mRNA, resulting in degradation of mRNA. For example, siRNA targeting the MDR1 gene can reduce the formation of efflux transporters in cell membrane, resulting in an increase in cellular drug concentration. Survivin is a protein that plays a crucial role in the survival pathway. Therefore, the silencing of the gene will open a window of time in which the resistant cells transiently become sensitized to the anti-cancer drug, thereby overcoming multi-drug resistance. On the other hand, since tumor suppressor gene, such as p53, can induce cell growth arrest or apoptosis, plasmid DNA encoding p53 can also be delivered for cancer therapy. All the RNA interference agents and plasmid DNA are known as gene agents.

Combination therapy is emerging as a promising approach for the treatment of cancer. Rational drug combinations aim to exploit either additive or synergistic effects arising from the action of several species with the final goal to maximize therapeutic efficacy. It has been shown that an appropriate combination of chemotherapeutic drugs and gene agents can improve the therapeutic outcome and patient compliance due to reduced dose and decreased development of drug resistance.

However, the biggest challenge in co-delivery drugs and gene agents is to find applicable carriers, since gene agents have higher molecular weight and negatively charged surface, while most frequently used anti-cancer drugs are hydrophobic small molecules. During the recent years, there has been a remarkable progress in a co-delivery system. The objective of this article is to review various nanocarriers that have been researched for the co-delivery of chemotherapeutic drugs and gene agents for tumor therapy, and to make suggestions for further design the in co-delivery system.

2. Co-delivery nanocarriers for drugs and gene agents

Since the physicochemical properties of drugs and gene agents are drastically different from those of small molecular weight drugs, separate mechanisms are usually required to encapsulate these two distinct payloads. The small-molecule drugs can be enclosed within the nanocarriers via hydrophobic force, electrostatic interaction or chemical conjugation, whereas gene agents are usually compressed by the carriers through electrostatic force.

To meet the above requirements, traditional nanocarriers, such as liposome and micelle, and novel nanocarriers, including dendrimer and a supramolecular system, have been used to delivery chemotherapeutic drugs and gene agents, as demonstrated in Table 1 and 2. The following section will systematically review the commonly and recently employed organic nanocarriers for the co-delivery of gene agents and drugs.

2.1. Traditional nanocarrier

2.1.1. Liposome-based nanocarrier

Liposomes represent one of the most successful drug vehicles, as well as in the co-delivery of chemotherapeutic drugs and gene agents. Recently, many researchers reported their achievements in co-delivery systems using modified cationic liposomes, as shown in Table 1. 1,2-Dioleoyl-3-trimethylammonium-propane (DOTAP) is the most commonly utilized cationic lipid. Cationic liposome-based nanosystems are usually prepared through simple electrostatic interaction between the positively charged cationic lipids and the negatively charged phosphate backbones of oligonucleotides. The drugs can be loaded via hydrophobic force, as exhibited in Fig. 1a. However, the liposome usually has poor physiological stability compared to other polymeric vectors.

Saad and colleagues demonstrated a cationic liposome-based co-delivery system, which consisted of cationic lipids, doxorubicin, and siRNA targeted to MRP1 and BCL2 mRNA (suppressors of pump and nonpump cellular resistance, respectively). The drug vehicle provided an effective co-delivery approach to induce cell death and to suppress cellular resistance in MDR lung cancer cells. Sun and colleagues reported an angiopep-2 modified cationic liposome (ANG-CLP) for the efficient co-delivery of a therapeutic gene encoding the human tumor necrosis factor-related apoptosis-inducing ligand and paclitaxel for glioma. The dual targeting co-delivery system improved uptake and gene expression not only in U87 MG cells and BCECs, but also in the glioma bed and infiltrating margin of intracranial U87 MG glioma-bearing models.

To improve tumor therapy efficacy, Feng and colleagues built a vaperotide-modified core-shell type nanoparticle co-encapsulating VEGF-targeted siRNA (siVEGF) and paclitaxel. Vaperotide is a somatostatin analog possessing high affinity to somatostatin receptors, which are overexpressed in many tumor cells. The nanoparticle core was a negatively charged ternary complex composed of siRNA, chondroitin sulfate and protamine, and could be coated with cationic lipid shell. As a result, the mixed liposome had significantly stronger drug distribution in tumor tissues via receptor-mediated targeting delivery, accompanied by substantial inhibition of neovascularization induced by siVEGF silencing.

2.1.2. Micelle based nanocarrier

To achieve simultaneous delivery of chemotherapeutic drugs and gene agents, the carriers must be able to protect the contents from degradation and prevent premature release. Micelleplexes consisting of amphiphilic block copolymers are the most commonly reported examples of co-delivery carriers, as shown in Table 1. Usually, the micelle self-assemblies with the hydrophobic blocks to form the interior of the micelle and with hydrophilic blocks to form the micelle shell. The hydrophobic interior acts as a reservoir for the poorly soluble hydrophobic drugs. Hydrophilic blocks on the shell mask the payloads. The most frequently used mask is polyethyleneglycol (PEG). Other commonly-used hydrophilic blocks are cationic polymers that can condense complex
Copolymers consisted of linear poly(ethyleneimine) and poly(ε-caprolactone) and paclitaxel. Cao and colleagues synthesized a diblock copolymer that could be applied for the delivery of VEGF siRNA and pDNA. The copolymer formed nano-sized micelles in water with positively charged surface that could be applied for the delivery of VEGF siRNA and paclitaxel. Micelle-based nanoparticles are frequently used cationic blocks. The most popular hydrophobic polymers are poly(ε-caprolactone) (PCL), poly(γ-butyl acrylate) (PNBA), polylactide (PLA) and poly(lactic-co-glycolic acid) (PLGA). The outer shell may be further decorated with targeting ligands, such as folate, to enhance the active targeting-ability of the carrier in most cases, as exhibited in Fig. 1b. Micelle-based nanocarriers are tunable, biocompatible, and physiologically stable owing to their low critical micelle concentration (CMC); the preparation process of functional block copolymers is always complicated. Zhu and co-workers prepared a biodegradable cationic micelle for tumor-targeted siRNA and drug co-delivery. The unique delivery system exhibited excellent stability and tumor-targeting triggered by the up-regulated tumoral MMP2. This system achieves enhanced cell internalization after MMP2-activated exposure of the previously hidden PEI. Chen et al. developed a reduction and pH dual-sensitive nanocarrier for synergistic cancer therapy. A ternary block copolymer PEG-poly(2-aminoethyl ethylene phosphate) (PPEEA) was synthesized by Zhang et al. for the co-delivery. PCHLG played an analogous role to lipoproteins in terms of drug delivery, and had high drug loading. PEI-PCHLG was able to assemble into micelles with high drug and gene loading efficiency. Amphilic chimeric peptide, for example, (Fmoc)2KH7-TAT, and Ac-(AF)6-H5-K15-NH2(FA32), can also be used for drug and gene co-delivery.

Stimuli-responsive micelle systems have also been developed for co-delivery. Zhu et al. presented a simple but multifunctional micellar platform constructed by a matrix metallproteinase 2 (MMP2)-sensitive copolymer (PEG-poly(earihtinicotinate) (PPEG)-PEI) via self-assembly for tumor-targeted siRNA and drug co-delivery. The unique delivery system exhibited excellent stability and tumor-targeting triggered by the up-regulated tumoral MMP2. This system achieves enhanced cell internalization after MMP2-activated exposure of the previously hidden PEI. Chen et al. developed a reduction and pH dual-sensitive nanocarrier for synergistic cancer therapy. A ternary block copolymer PEG-PAsp(AED)-PDPA contained pH-sensitive poly(2-(diisopropyl amino)ethyl methacrylate) (PDA), reduction-sensitive poly(N-(2,2-dithiobis(ethylamine)) aspartamide) (PAsp(AED)) and PEI. The copolymer assembled into a core–shell structural micelle, which encapsulated doxorubicin in its pH-sensitive core and BCL2 siRNA in a reduction sensitive interlayer. The dual stimuli-responsive design of micellar carrier allowed microenviroment-specific rapid release of both doxorubicin and BCL2 siRNA inside acidic compartments, resulting in improved drug efficacy.

### Table 1: Traditional Cationic micelles of chemotherapy drugs and gene agents in recent researches.

| Carrier type                   | Composition of carrier | Drug              | Gene agent          | Cell line    | Ref. |
|-------------------------------|------------------------|-------------------|---------------------|--------------|-----|
| Cationic solid lipid nanoparticles (cSLN) | Paclitaxel             | MCL1 siRNA        | KB                  | 16           |     |
| PLGA/FPL                     | Paclitaxel             | pEGFP-biTRAIL     | U87                 | 17           |     |
| Cationic liposome             | Doxorubicin            | pEGFP             | MDA-MB-231          | 18           |     |
| Vapreotide-modified core–shell liposome | Paclitaxel             | VEGF siRNA        | MCF-7, HCT15        | 19           |     |
| Lipid nanocapsules functionalized with PEI | Paclitaxel             | pDNA              | MCF-7               | 20           |     |
| Thermosensitive magnetic cationic liposomes | Doxorubicin            | SATB1 shRNA       | MKN-28              | 21           |     |
| Nanostructured lipid carrier  | Doxorubicin or paclitaxel | MRP1 and BCL2 siRNA | A549              | 22           |     |
| Micelle                       | PEGylated liposome     | Docetaxel         | BCL2 siRNA          | A549         | 23           |     |
| Amphiphilic chimeric peptide  | Docetaxel              | p53 plasmid       | A549, 293T, Hela    | 24           |     |
| (Fmoc)2KH7-TAT                | Doxorubicin            | BCL2 siRNA        | SKOV-3              | 25           |     |
| PEG-PCHLG                    | Doxorubicin            | pDNA              | HEK293              | 26           |     |
| PDMAEMA-PCL-PDMAEMA           | Paclitaxel             | GFP siRNA         | MDA-MB-435          | 27           |     |
| PS5-PEI/TPGS                 | Paclitaxel             | Survivin siRNA    | A549                 | 28           |     |
| ABP-PEG1,53-paclitaxel        | Paclitaxel             | gWiz-Luci         | MCF-7, A549         | 29           |     |
| FA-PEG-PGA and PEI-PCL        | Doxorubicin            | BCL2 siRNA        | C6                   | 30           |     |
| PEO-b-PCL                    | Doxorubicin            | MDRI siRNA        | MDA-MB-435          | 31           |     |
| Oligopeptide amphiphile       | Doxorubicin            | Luc siRNA         | HepG2               | 32           |     |
| PDP-PAHA                     | Doxorubicin            | Surviving siRNA   | MCF-7               | 33           |     |
| PEG-p-polycation              | Paclitaxel             | Surviving siRNA   | A549                 | 34           |     |
| MPEG-PCL-g-PEI               | Doxorubicin            | Msurvivin T34A gene | B16F10, MCF-7, CT26 | 35           |     |
| PEOz-PLA-g-PEI               | Doxorubicin            | mcDNA             | MCF-7               | 36           |     |
| PEG-PLL-PLL-Leu               | Docetaxel             | BCL2 siRNA        | MCF-7               | 37           |     |
| Cationic core–shell nanoparticles | Paclitaxel             | IL-2 plasmid BCL2 siRNA | MDA-MB-231, 4T1 | 38           |     |
| mPEG45-b-PCL80-b-PPEEA10      | Paclitaxel             | polo-like kinase 1 (Plk1) specific siRNA | MDA-MB-435 | 39           |     |

with negatively charged DNA or RNA. Poly(ethyleneimine) (PEI) and poly(2-aminoethyl ethylene phosphate) (PPEEA) are the most frequently used cationic blocks. The most popular hydrophobic polymers are poly(ε-caprolactone) (PCL), poly(n-butyl acrylate) (PNBA), polylactide (PLA) and poly(lactic-co-glycolic acid) (PLGA). The outer shell may be further decorated with targeting ligands, such as folate, to enhance the active targeting-ability of the carrier in most cases, as exhibited in Fig. 1b. Micelle-based nanocarriers are tunable, biocompatible, and physiologically stable owing to their low critical micelle concentration (CMC); the preparation process of functional block copolymers is always complicated.

Zhu and co-workers prepared a biodegradable cationic micelle with PDMAEMA-PCL-PDMAEMA triblock copolymer, which formed nano-sized micelles in water with positively charged surface that could be applied for the delivery of VEGF siRNA and paclitaxel. Cao and colleagues synthesized a diblock copolymer consisted of linear poly(ethyleneimine) and poly(ε-caprolactone) (PEI-PCL), and the amphiphilic polymer assembled into micelles for co-delivery of BCL2 siRNA and doxorubicin. Folic acid was conjugated to the polyanion and further coated onto the surface of the cationic PEI-PCL nanoparticle pre-loaded with siRNA and doxorubicin, potentiating a ligand-directed delivery to human hepatic cancer cells. This hierarchical assembly strategy was beneficial for active targeting. Another dual-functional poly(ethyleneimine)-poly(γ-cholesterol-l-glutamate)(PEI-PCHLG) copolymer was synthesized by Zhang et al. for the co-delivery. PCHLG played an analogous role to lipoproteins in terms of drug delivery, and had high drug loading. PEI-PCHLG was able to assemble into micelles with high drug and gene loading efficiency. Amphilic chimeric peptide, for example, (Fmoc)2KH7-TAT, and Ac-(AF)6-H5-K15-NH2(FA32), can also be used for drug and gene co-delivery.

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lyso-some with enriched reducing agent. This resulted in synergis-
tically-enhanced apoptosis of human ovarian cancer SKOV-3 cells,
thereby dramatically inhibiting tumor growth.

2.2. Non-traditional nanocarrier

2.2.1. Dendrimer based nanocarrier

Dendrimers are hyperbranched and monodispersed macromole-
cules which have defined molecular weights and host–guest
entrapment properties. More importantly, dendrimers can interact
with drug and gene molecules by simple encapsulations, electro-
static interactions and covalent conjugations since they possess
empty internal cavities and a much higher density of surface
functional group as shown in Fig. 1c. Therefore, monodispersal
and high drug-loading capacity are prominent advantages of
dendrimers. However, dendrimers still have some safety–toxicity
issues according to comprehensive statistics.

Several polyamine polymers have been explored as carriers for
drug delivery. For example, poly(amido amine) (PAMAM), a
cationic dendrimer which introduces ammonia as the core, has
been investigated as non-viral delivery vector for efficient siRNA
delivery. Han and co-workers employed peptide HAIYPRH
(T7)-conjugated PEG-modified PAMAM dendrimer (PAMAM-
PEG-T7) for the co-delivery of pDNA and doxorubicin. In
comparison with single doxorubicin or pDNA delivery system,
this co-delivery system induced apoptosis of tumor cells in vitro
and inhibited tumor growth in vivo more efficiently. Combining
PAMAM with other amphiphilic block copolymers was also an
approach for co-delivery. Biswas et al. modified PAMAM with
poly(ethylene glycol)-1,2-dioleoyl-sn-glycero-3-phospho-ethanol-
amine to form a new construct G(4)-D-PEG-2K-DOPE. This G(4)-
PAMAM dendrimer was utilized as a cationic source for efficient
siRNA condensation; DOPE provided optimum hydrophobicity
core and poly(L-lysine) dendron arms for doxorubicin and MMP-9
drug/gene co-delivery. Qian et al. constructed dendrimer
analog with three amphiphilic star-branched copolymers comprising
polylactic acid (PLA) and polydimethylaminoethyl methacrylate
(PDMAEMA) for microRNA and doxorubicin transport. By testing
architectures with different repeat degrees, they found that (AB3)1
architecture exhibited the highest transfection efficiency. Ma and
colleagues designed a star-shaped porphyrin-arginine-functionalized
poly(L-lysine) copolymer (PP-PLLD-Arg) for photo-enhanced drug
and gene co-delivery. Results with this copolymer demonstrated that
PP-PLLD-Arg with suited irradiation was a promising non-toxic and
photo-inducible effective drug and gene delivery strategy.

2.2.2. Supramolecular nanocarrier

The development of self-assembly techniques has permitted the
introduction of supramolecular nanoparticles (SNPs), such as
host–guest architectures, as drug and non-viral gene carriers. The
host–guest system is a complex in which one chemical compound
(the “host”) forms a cavity in which molecules of a second “guest”
compound are located. In drug delivery system, the most
frequently used host is γ-cyclodextrin (γ-CD), which contains a
torus-like structure with a hydrophobic cavity, and can form
inclusion complexes with chemotherapeutic drug, as demonstrated
in Fig. 1d. Rational inclusion complexes exhibit excellent serum
stability and promising application. However, perfectly matched
host/guest materials are not easy to find.

Recently, Yang and colleagues designed a pH-responsive
drug/gene co-delivery nanoplatform by means of host–guest
chemistry. γ-CD/doxorubicin complexes were attached onto
phenylboronic-acid-modified oligoethylenimine (PEI1.8K-PB2.9) at
neutral conditions. The drug is detached from PEI1.8K-PB2.9
under acidic conditions owing to the acidity-labile feature of

Figure 1 Schematic illustration of four major types of nanocarriers to co-delivery gene and chemotherapeutic drug. (a) Cationic liposome, the most frequently used cationic lipid and general lipid are DOTAP and DOPE, respectively, and PEG modified with PE can prolong the cycle
time in the circulation system. (b) Micelle system, hydrophilic block is usually positively charged, such as PEI, polyamino acid and so on, PCL, PLA and PE are employed as hydrophobic core. (c) Dendrimer system, PAMAM is the most commonly used dendrimer for co-delivery. (d) A supramolecular system, γ-CD can form inclusion complexes with chemotherapeutic drugs.
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boronate linkage, thereby facilitating drug release. Moreover, \( \text{PEI}_{1.8k} \cdot \text{PB}_{2.9} \cdot \gamma - \text{CD} \) conjugates demonstrated significantly improved cell-biocompatibility and DNA transfection activity by overcoming serum-susceptible drawbacks frequently associated with synthetic gene carriers. Zhao and co-workers\(^5\) also employed \( \gamma - \text{CD} \) and multiple oligoethylenimine (OEI) arms with folic acid (FA) as co-delivery materials for paclitaxel and pDNA.

In another study, Fan and colleagues\(^6\) designed a SNP consisted of host PEI-CD (as gene vector) and guest adamantane conjugated groups (as chemotherapeutic agent carriers) for co-delivery of drug and gene. The adamantane-conjugated doxorubicin as the guest Ad-Dox component assembled with the host PEI-CD into supramolecular PEI-CD/Ad-Dox, which could further interact with plasmid DNA to form drug- and gene-loaded PEI-CD/Ad-Dox/pDNA SNP. The in vitro data in different cell lines indicated that such SNP could ensure that both drug and gene can interact with plasmid DNA to form drug- and gene-loaded PEI-CD/Ad-Dox/pDNA SNP. The in vitro data in different cell lines indicated that such SNP could ensure that both drug and gene can be delivered to the same cancer cell, providing the feasibility of combinational tumor treatment. Hu et al.\(^7\) conducted synergistic treatment of ovarian cancer by co-delivery of survivin shRNA and paclitaxel via a similar supramolecular micellar assembly.

2.2.3. Novel nanoformulation

Chang et al.\(^8\) constructed a redox-responsive system for drug/siRNA co-delivery based on ferrocenium capped amphiphilic pillar[5]arene (FCAP). Pillar[5]arenes are a new class of macrocyclic compounds which possess a hydrophobic core sandwiched between two functional rims and can self-assemble to cationic vesicles in aqueous solution. The ferrocenium cation, which is sensitive to glutathione (GSH), is a redox-responsive bond, and the positive charge of ferrocenium makes possible for the loading of negatively charged siRNA onto nanocarriers. Therefore, FCAP allowed building an ideal GSH-responsive drug/siRNA co-delivery system for rapid drug release and gene transfection in cancer cells in which higher GSH concentration existed.

Chen et al.\(^9\) reported a unique architecture, cationic polymeric nanocapsule, which had well-defined covalently stabilized biodegradeable structures and can function as a potentially universal and safe therapeutic nanocarrier for co-delivery of doxorubicin and siRNA targeting interleukin-8. This nanocapsule was synthesized from allyl-functionalized cationic poly(ethylene) (CPLA) by a highly efficient UV-induced thiol-ene interfacial cross-linking in transparent miniemulsions. Liu and co-workers\(^10\) adopted a double-emulsion solvent evaporation technique to prepare intelligent gelatinases-stimuli nanoparticles for the co-delivery of miR-200c and doxetaxel. This miniemulsion was able to inhibit cancer stem cells and non-cancer stem cells and showed promise for cancer therapy.

Dr. Hammond's group\(^11\) developed a layer by layer nanoplatform for systemic co-delivery of doxorubicin and siRNA for potential triple-negative breast cancer treatment. The layer by layer nanoparticle could be divided into three parts in structure: drug-loaded core, siRNA/polycation–loaded middle film and tumor targeting outer shell. The advantage of this unique architecture was that it provided a modular platform for a broad range of controlled multidrug therapies customizable to the cancer type in a singular nanoparticle delivery system. Meanwhile, Sun et al.\(^12\) presented a system with multilayers for co-delivery of doxorubicin and DNA. Ferrocene modified poly(ethyleneimine) (PEI-Fc) formed micelles in solution and trapped DNA and drug to form PEI-Fc–DOX-DNA nanocomplexes, and such cationic nanocomplexes were further used to construct multilayers through layer by layer assembly with

| Carrier type | Composition of carrier | Drug | Gene agent | Cell line | Ref. |
|--------------|-----------------------|------|------------|-----------|------|
| Dendrimer    | T7-modified dendrigraft/poly-t-lysine | Doxorubicin | pTRAIL | U87 | 40 |
|              | PAMAM-PEG-T7          | Doxorubicin | pORF-hTRAIL | Bel-7402 | 41 |
|              | b-cyclodextrin core and poly(t-lysine) dendron arms | Doxetaxel | pMR3 | HNE-1 | 42 |
|              | PLA-\(\beta\)-PDMAEMA | Doxorubicin | miR-21 | LN229 | 43 |
|              | Porphyrin-arginine Functionalized poly(t-lysine) copolymer | Doxetaxel | MMP-9 shRNA | HNE-1 | 44 |
|              | Poly(t-lysine) dendrimers with a silsesquioxane cubic core | Doxorubicin | luciferase siRNA | U87 | 45 |
| Supramolecular system | Host PEI-CyD (PC) guest adamantine conjugated PTX | Doxorubicin | siGFP | A549 | 46 |
|              | Host PEI-CyD (PC) guest adamantine conjugated DOX | Doxorubicin | pTRAIL | SKOV3 | 46 |
|              | \(\beta\)-CD and OEI-FA | Paclitaxel | p53 | KB, A549 | 48 |
|              | \(\text{PEI}_{1.8k} \cdot \text{PB}_{2.9} \cdot \gamma - \text{CD} \) | Doxorubicin | pDNA | 293T, HeLa | 49 |
| Novel nanoformulation | Amphiphilic[5]arene capped with ferrocenium | Doxorubicin | MDR1 siRNA | 293T, HeLa | 50 |
|              | Aptamerconjugated PEI-PEG | Doxorubicin | Bcl-\(\beta\)-shRNA | PC3, LNCaP | 51 |
|              | Chitosan-graft-PEI | Candesartan | p53 | PANC-1 | 52 |
|              | Hyaluronic acid and chitosan | Doxorubicin | miR-34a | MDA-MB-231 | 53 |
|              | Layered double hydroxide | S-fluorouracil | Allstars Cell Death siRNA | MCF-7, U2OS and HCT-116 | 54 |
|              | CholsiRNA/LDL-coupled N-succinyl chitosan | Doxorubicin | MDR1 siRNA | HepG2 | 55 |
|              | PEG-Pep-PCL copolymer | Doxetaxel | miR-200c | BGC-823 | 56 |
|              | PLGA nanoformulation | Doxorubicin | MDR1 siRNA | MCF-7 | 57 |
|              | PEI-Fc | Doxorubicin | DNA | HepG2 | 58 |
|              | Cationic polymeric nanocapsules | Doxorubicin | IL-8 siRNA | MCF-7 | 59 |
|              | PEI-PEG based nanoparticles | Doxorubicin | DNA | HUVE, HepG2, MCF-7 | 60 |
negatively charged dextran sulfate. The multilayers could be potentially applied to the biomedical devices for cancer treatment, regenerative medicine, etc. Some other novel co-delivery nanoformulations are displayed in Table 2.

3. Conclusions and future perspectives

The co-delivery of chemotherapeutic drugs and gene agents provides a promising strategy to overcome drug resistance in cancer therapy. According to recent research, it is clear that combination delivery of gene and drug using nanocarriers is indeed helpful in inhibiting tumor growth compared to gene or drug alone. Although various nanocarriers have been developed for co-delivery, most carriers just focus on successful co-delivery of gene and drug. This approach has often resulted in functional materials, such as PEG, PEI and PLGA et al., being used repeatedly in different permutation and combination, without paying attention to the rational ratio of gene and drug or the interaction between them in the vehicle. Development of new materials and technologies affords the opportunity to discover and produce novel drug delivery systems. Presently, an ideal co-delivery carrier should be biocompatible and biodegradable, and demonstrate circulatory stability, thereby facilitating transport of the cargos to the targeting sites. The ideal carrier will also be multifunctional, with the ability to transport simultaneously both chemotherapeutic drugs and gene agents to cancer cells, releasing the payloads in a controlled manner and accurate dose, thereby achieving a maximum effect of the combination therapy for treating drug resistant tumors. Further studies should focus on the interaction between drugs and gene agents, as well as the interaction between therapeutic agents and carriers. Continuous development of such combination delivery systems will ultimately lead toward availability of effective therapies for cancer.

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